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**A STUDY OF THE INTERACTIONS BETWEEN HOLSTEIN-  
FRIESIAN GENOTYPES AND FEEDING SYSTEMS, WITH  
EMPHASIS ON SYSTEM PERFORMANCE AND COW GRAZING  
ABILITY**

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of the requirements for the degree of:

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**Dedicated to Sonia, Martín and Dominique**

## ABSTRACT

**Rossi, J.L. 2006.** *A study of the interactions between Holstein – Friesian genotypes and feeding systems, with emphasis on system performance and cow grazing ability.* PhD Thesis, Massey University, Palmerston North, New Zealand.

Imported genetic material of the Holstein-Friesian breed from overseas (OS), mainly from North America, has been used in New Zealand (NZ) since the late 1960's. This has diluted the genetic base of the former NZ Friesian genotype selected under intensive seasonal pasture-based systems. As a result, increased concerns have been raised about the negative influence of these overseas genes on the modern NZ Holstein-Friesian, as it is apparent that the OS Holstein-Friesian has a lower capacity to perform on grazed pasture. The objective of the present thesis was to investigate differences in production performance between three Holstein-Friesian genotypes farmed at different feed allowances (FA) on pasture-based systems; in addition, to investigate differences in the grazing process between strains under contrasting managements and sward conditions, and so to identify animal and pasture factors that affect the herbage intake ( $DMI_H$ ) and performance of the grazing cow. An accurate procedure was also established to estimate  $DMI_H$  for cows fed forage and maize supplements, grazing in groups. Two modern high breeding worth (BW) Holstein-Friesian strains from NZ (NZ90) or overseas (OS90) origin and a low BW 1970's NZ Friesian genotype (NZ70) were farmed in two field experiments: (1) a long-term 'system' study that compared the yield performance of these genotypes in a range of systems with different feed allowance (FA) per cow, and a (2) a short-term 'component' study that compared the grazing capacity of the strains under contrasting sward conditions but at a common daily herbage allowance. The differences in productive performance between genotypes increased as the study progressed in the system study, with the largest observed in the last season. The mean milk solids (MS) yield per cow and per hectare were higher in NZ90 (395 kg cow<sup>-1</sup> and 1,236 kg ha<sup>-1</sup>) than in the NZ70 (336 kg cow<sup>-1</sup> and 1,093 kg ha<sup>-1</sup>) and the OS90 (377 kg cow<sup>-1</sup> and 1,154 kg ha<sup>-1</sup>). The higher production of NZ90 cows was supported by their higher mean daily MS yield than the NZ70 (1.45 vs. 1.21 kg MS cow<sup>-1</sup>day<sup>-1</sup>) and more days in milk than OS90 cows (271 vs. 257 DIM). The lower lactation length of the OS90 strain occurred due to its lower body condition score (BCS) in late lactation, which determined an early dry-off for these cows. The lowest BCS of OS90 at the nadir (irrespective of FA), during lactation and at dry-off indicate these cows mobilised greater amount of body reserves and partitioned most of the energy ingested to yield. Genotype by FA interactions for milk and lactose yields, protein content in the milk and BCS were observed in the second and third seasons of the 'system' study. Milk yield increased as FA increased to a greater extent in OS90 than in the two NZ strains, whereas the content of solids in milk, particularly protein, increased to a greater extent for NZ90 than in both OS90 and NZ70. During lactation  $DMI_H$  was higher for NZ90, intermediate in OS90 and lower in NZ70 (14.5, 13.9 and 12.6 kg DM cow<sup>-1</sup>day<sup>-1</sup> respectively for NZ90, OS90 and NZ70, as measured with n-alkanes), and declined as lactation progressed, with a smaller difference for the total intake achieved (15.5, 15.2 and 13.1 kg DM cow<sup>-1</sup>day<sup>-1</sup> respectively) due to the increased supplement consumption. These results indicate that the OS90 needs more feed with a higher proportion of supplement in the diet to improve productive

performance on pasture-based systems; the NZ90 would perform better when cow nutrition is mainly supported by grazing pasture, although further increments in performance could be expected from strategic supplementation, but requiring more feed than NZ70. The  $DMI_H$  per unit of live weight ( $DMI_H/LW$ ) was highest in NZ90 strain in both the 'system' and in the short sward of the 'component' study (31.5 and 31.1 g DM  $kg^{-1}$ DM in NZ90 vs. 28.9 and 28.6 g DM  $kg^{-1}$ DM for OS90 in 'system' and 'component' studies respectively). The higher intake of NZ90 on pasture was sustained by a higher capacity to graze short swards than NZ70 and OS90, and to deal with the herbage of higher bulk density and lower quality present at the base of taller swards. The NZ90 can maintain  $DMI_H$  in swards with different structures, indicating higher flexibility to perform under different managements and sward conditions. The size of the jaw is smaller in NZ90 than OS90 (88.4 vs. 92.4 mm) with effects on bite area and bite size, and this flexibility to adapt the size of the bite to swards of different structure may improve bite penetration under constraining sward conditions. The reduced ability of the OS90 to adjust ingestive behaviour to different swards would limit the capacity of this strain to perform on pasture. The fact that OS90 cows increased  $DMI_H$  and  $DMI_H/LW$  substantially in a leafy and taller sward (up to 21.6 kg  $cow^{-1}$  and 40.8 g DM  $kg^{-1}$ LW vs. 19.2 kg  $cow^{-1}$  and 41.0 g DM  $kg^{-1}$ LW in NZ90 during early lactation) suggests that yield performance can be improved in these cows even on pasture, by fine-tuning pasture management.

**Keywords:** Pasture-based dairy systems, Holstein-Friesian, genotype environment interactions, grazing ability, grazing behaviour, pasture management.

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By the time I arrived at Hamilton in late March 2002 the 'Strain Trial' had already started at the old Dexcel No 2 dairy. Although I was involved in the trial since my arrival, it took me time to settle. Soon after arrival John Milburn was appointed farm manager and this made my time at the 'No 2' a lot easier. John is not only an excellent manager, he became a good friend and I really enjoyed sharing time with him and the rest of the staff at the farm. Hopefully they also found enjoyable having me around, requiring their frequent help.

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# LIST OF TERMINOLOGY AND ABBREVIATIONS

## Abbreviations

ADF	acid detergent fibre	H <sub>R</sub>	height reduction
AFRC	Agricultural and Food Research Council	<i>i...ii</i>	content of C <sub>31</sub> & C <sub>33</sub> odd natural <i>alkanes</i>
am	ante meridian	IA	artificial insemination
B	bite	IR	intake rate
BCS	body condition score	<i>j</i>	content of C <sub>32</sub> dosed <i>n-alkane</i>
B <sub>D</sub>	bulk density	J <sub>Z</sub>	jaw size
BR	bite rate	k <sub>1</sub>	efficiency of energy utilisation for milk
BV	breeding value	k <sub>m</sub>	efficiency of energy utilisation for maintenance
BW	breeding worth	LC	milk lactose concentration
BZ	bite size	LIC	Livestock Improvement Corporation
C <sub>27...C<sub>35</sub></sub>	odd natural <i>alkanes</i> , different carbon chain	L <sub>0</sub>	lowest feed allowance (equal to FA1)
C <sup>3</sup>	winter grasses, C <sup>3</sup> photosynthetic pathway	LW	liveweight
C <sub>32</sub>	dosed of pellets with C <sub>32</sub> - <i>alkanes</i>	LW <sup>0.75</sup>	metabolic LW
C <sup>4</sup>	summer grasses, C <sup>4</sup> photosynthetic pathway	LW <sub>E</sub>	empty LW
CIDR <sup>®</sup>	controlled internal drug-releasing device	M	measurement period
C <sub>0</sub>	concentrate feed	M <sub>E</sub>	dietary metabolisable energy
CP	crude protein	ME	metabolisable energy
Cr <sub>2</sub> O <sub>3</sub>	chromic oxide	MJ ME	megajoule of ME
CRC	controlled release capsule	MJ	megajoule
CSR	comparative stocking rate	MS	milk solids
DH	herbage in-vitro digestibility	NDF	neutral detergent fibre
DHA	daily herbage allowance	NE <sub>m</sub>	net energy maintenance
DHA/LW	daily herbage allowance per LW unit	NIRS	Near Infrared Reflectance Spectroscopy
DIM	days in milk	NPO	nominal pasture offered
DM	dry matter	NSO	nominal supplement offered
DMD	dry matter digestibility	NTF <sub>0</sub>	nominal total feed offered
DMI	daily dry matter intake	NZ	New Zealand
DMI <sub>H</sub>	daily herbage DMI	NZ70	NZ Holstein-Friesian dairy cow of the 1970's
DMI <sub>LW</sub>	daily herbage DMI per LW unit	NZ90	NZ Holstein-Friesian dairy cow of the 1990's
DMI <sub>MZ</sub>	daily maize silage or maize grain DMI	OF	oesophageal fistulated animals
DMI <sub>PS</sub>	daily pasture silage DMI	OM	organic matter
DMI <sub>S</sub>	daily supplement DMI	OMD	organic matter digestibility
DMI <sub>T</sub>	daily total DMI	OS	overseas
DMI <sub>T</sub> /LW	daily total dry matter intake per LW unit	OS90	OS Holstein-Friesian dairy cow of the 1990's
DMI <sub>V</sub>	daily dry matter intake visual	P	parity
D <sub>0j</sub>	dosed n-alkane <i>j</i>	PADI	<i>Paspalum dilatatum</i> poir.
DS	supplement in-vitro digestibility	PC	milk protein concentration
DTA	daily total allowance	P <sub>E</sub>	experimental period
E	eating supplement	pm	post meridian
e	error term	P <sub>0</sub>	pasture offered
E <sub>NBAL</sub>	energy balance	PR <sub>E,XP</sub>	pre-experimental period
E <sub>NM</sub>	energy required for maintenance	PU	pasture utilisation
E <sub>NO</sub>	energy in milk	q <sub>m</sub>	dietary ME/G <sub>E</sub>
E <sub>T</sub>	eating time	R	ruminating
F	faeces	ratio	Relation C <sub>31</sub> /C <sub>33</sub> in H or F
FA	feed allowance	Rel H/F	ratio C <sub>31</sub> /C <sub>33</sub> in H to the same ratio in F
FA1...4	feed allowance level from 1 (lowest) to 4 (highest)	R <sub>i...ii...j</sub>	Recovery of <i>i...ii...j-alkanes</i> in faeces
FC	milk fat concentration	RPM	Rising Plate Meter
Fi...ii...j	<i>i...ii...j-alkanes</i> concentration in faeces	RT	ruminating time
FO	faecal output	SAS	Statistical Analysis System <sup>®</sup>
FO <sub>H</sub>	herbage faecal output	SC	sward condition
FO <sub>S</sub>	supplement faecal output	SC	sward condition
FO <sub>T</sub>	total faecal output	S <sub>F</sub>	proportion of supplement in faeces
G	grazing	SH	sward surface height
GE	genotype of Holstein-Friesian	SHORT	short sward
G <sub>E</sub>	gross energy	SH <sub>POST</sub>	post-grazing sward surface height
GLU	concentration of glucose in blood samples	SH <sub>PRE</sub>	pre-grazing sward surface height
G <sub>T</sub>	grazing time	Si...j	content of n-alkane in the supplement
G <sub>T</sub> /R <sub>T</sub>	grazing to ruminating ratio	SR	stocking rate
H	herbage	TALL	tall sward
HAR	herbage accumulation rate	TMR	total mixed ration
HERD	cows allocated to experimental mobs	TS <sub>0</sub>	total supplement offered
H <sub>F</sub>	proportion of herbage in faeces	v.s.	versus
H <sub>I</sub>	highest feed allowance (equal to FA4)	Y	age of the cow (in years)
Hi...ii...j	<i>i...ii...j-alkanes</i> concentration in herbage	δ <sup>13</sup> C	ratio of the natural <sup>12</sup> C and <sup>13</sup> C isotopes in the herbage or feed
HM	herbage mass	δ <sup>13</sup> C <sub>F</sub>	ratio <sup>12</sup> C and <sup>13</sup> C in faeces
HM <sub>POST</sub>	post-grazing herbage mass	δ <sup>13</sup> C <sub>S</sub>	ratio <sup>12</sup> C and <sup>13</sup> C in supplement
HM <sub>PRE</sub>	pre-grazing herbage mass		

## LIST OF SYMBOLS

### Weights, volumes, measures and statistical terms

P	probability
*	significant at $P < 0.05$
**	significant at $P < 0.01$
***	significant at $P < 0.001$
°C	degree centigrade
CORR	correlation
g	gram
kg	kilogram
km	kilometre
L	litre
m	meter
m <sup>2</sup>	square meter
mg	milligram
n	number of observations
ha	hectare
MJ	megajoule
mm	millimetre
t	tonne
h	hour
min	minute
MIXED	mixed
PROC	procedure
$r$	coefficient of correlation
$r^2$	coefficient of determination
REG	regression
RMSE	root mean square error
SD	standard deviation
SE	standard error of the mean
SED indicated)	standard error of the difference (maximum indicated)
CV	coefficient of variation

# SECTION I

## CHAPTER 1

### GENERAL INTRODUCTION

#### 1.1. INTRODUCTION

The pastoral dairy industry in New Zealand (NZ) is based on efficient grazing and conversion of the growing herbage into milksolids (MS). A concentrated spring calving pattern that allows farmers to match pasture growth rate with herd requirements and the use of relatively high stocking rate (SR) are the keys to the high utilization of pasture in these systems. According to the World Animal Review (Steinfeld & Maki-Hokkonen, 1995), grazing systems produce only about 10% of the world's milk. Consequently most dairy cattle have not been selected in grazing systems (Buckley *et al.*, 2005) but on diets that are very different from grazed pasture, for example a total mixed ration (TMR). Hence, it is possible that modern Holstein-Friesian dairy cows have lower capacity to graze effectively despite their higher yield potential and intake capacity. The ability to harvest pasture by grazing was considered the main limiting factor in NZ dairy systems (McMeekan, 1961).

The cow's potential for milk production has increased during the last fifty years due to genetic improvement. In NZ, this was achieved in part by the high use of overseas Holstein-Friesian genetics since the late 1960s, to take advantage of the high protein yield of this genotype (Harris *et al.*, 1999). However, differences in breeding policies meant that overseas dairy cows were selected for increased milk yield in predominantly feedlot feeding systems, whereas NZ dairy cows are sustained on grazed pastures with little or no concentrate supplementation (Harris & Kolver, 2001). The introduction of overseas Holstein-Friesian genetics diluted the genetic base of the former NZ Friesian strain (Harris & Kolver, 2001), in which selection had been focused on high capacity to produce in seasonal intensive pasture-based systems.

Against this background, concerns have been expressed about the ability of the high-potential genotypes selected under conventional dairy systems in Europe, and particularly in the USA, to demonstrate their potential under the nutritional and environmental constraints of intensive pasture-based systems (Veerkamp *et al.*, 1994; Holmes, 1995; Veerkamp & Emmans, 1995; Oldham *et al.*, 1996; Buckley *et al.*, 2005). These concerns led to the establishment of a series of trials to investigate the productive performance of the Holstein-Friesian genotype of high genetic yield potential in different parts of the world (e.g. by Teagasc, Moorepark, in Ireland and by Dexcel, Hamilton, New Zealand). In general these trials demonstrated a significant genotype by environment interaction effect on productive and reproductive performance, where the environmental contrasts were extreme (Kolver *et al.*, 2002), but interactions have been less evident where the nutritional contrast are less extreme (Freeman, 1975; Holmes, 1995; Buckley *et al.*, 2000).

This experiment, that provides the basis for the present thesis, was set up as one part of an international trial to investigate the effects of different feeding systems on production and reproduction efficiency of different Holstein-Friesian genotypes. The genotypes utilised in the present thesis represent the ‘traditional’ NZ dairy cow of 30 years ago (NZ70), a modern ‘pasture based’ dairy cow, which reflects the progress made within the NZ dairy herd (NZ90), and a modern ‘TMR’ based dairy cow with a high proportion of North American genetics in its ancestry (OS90)(see Appendix IV for details about genotypes characteristics).

The trial was set up as a long-term systems study, carried out at Dexcel N°2 dairy farm, Hamilton, New Zealand, and involved 11 systems (herds) representing a range of levels of nutrition and grass/supplement balance in self-contained mini farms, over a three year period (between 2001 and 2004). The other part of the international trial was carried out at Moorepark Research Centre, County Cork, Ireland.

Within the context of the NZ trial, the primary objective was to focus attention on the productivity and efficiencies in systems managed with different genotypes at different levels of feed allowance, with particular emphasis on genotype by feeding level interactions and the relationships between grazing behaviour, herbage and supplementary forage intakes, milk production and estimated energy balance of the three genotypes when managed on intensive pasture-based systems.

It is hypothesized that in systems managed on relatively low feed allowance per cow on pasture, genotypes with different potentials for yield would produce similarly, as cows with higher yield potential would be more constrained than cows of low genetic potential for yield and lower energy requirements, when low amount of feed is available. However,

production would increase substantially in cows with high genetic potential when the level of feed is increased.

The use of whole systems as treatments provided a very effective basis for evaluating practical aspects of the management required by the genotypes used in this trial. However, it presented difficulties in dealing with the confounding variations in pasture condition, supplementary feed and animal status, so limiting the validity of assessment of the direct relationships between sward conditions and herbage intake by the individual genotypes. Accordingly, the project was completed with a ‘component’ grazing experiment in which all three genotypes were managed under closely controlled and contrasting sward conditions after preliminary ‘standardization’ periods.

The central importance of accurate estimation of herbage intake to the understanding of the performance of grazing systems has been repeatedly emphasized (Dove & Mayes, 1991; Dove & Mayes, 1996; Penning, 2004), but this depends upon the availability of reliable and flexible techniques for measuring aspects of intake and grazing behaviour. Thus, the first part of this thesis is concerned with the establishment and evaluation of procedures for measuring intake in the particular conditions of a system study where cows were fed forage and maize supplements as well as grazing pasture during the whole season, and where pasture and supplement conditions changed regularly.

## **1.2. THESIS OUTLINE**

The structure of this thesis takes the aspects described above into account. It is divided in five sections. The first section is a general introduction (Chapter 1), which is followed by three sections related to the main aspects of this thesis, then the last section for general conclusions. In Section II (Chapters 2 to 4), the methodologies to estimate the individual intake of grazing pasture, forages and supplements for grazing animals supplemented in a group context were reviewed and the more accurate combination of methods identified. The two following sections (III and IV) report results from field studies. Section III deals with the results of a long-term ‘system’ trial where three different genotypes of Holstein-Friesian dairy cows were compared in a range of systems farmed at different feed allowance per cow. This section is divided in three chapters, the first one (Chapter 5) reports the productivity of these systems and cow genotype – nutrition interactions on pasture-based systems, the second (Chapter 6) deals with differences in  $DMI_H$  and cow nutrition between genotypes and feeding levels, and the last one (Chapter 7) compares the  $DMI_H$  and grazing behaviour of the genotypes under contrasting feeding management in early and late lactation. Section IV (Chapter 8) reports the  $DMI_H$  and productive performance achieved during early lactation when the same three genotypes grazed

swards differing in structural characteristics, but allocated to a similar daily herbage allowance (DHA). In Section V (Chapter 9) the main factors affecting the differences in performance and grazing ability between genotypes are discussed. The design of more efficient systems by combining the differences in the grazing ability of the genotypes and feeding conditions is also discussed.

**Section I: General introduction.**

Chapter 1. Introduction

**Section II: Intake studies. The use of *n*-alkanes in estimating herbage intake, digestibility and diet composition.**

Chapter 2. A review of the *n*-alkane method and its accuracy.

Chapter 3. Effect of grazing preference on the estimation of herbage intake with *n*-alkanes.

Chapter 4. Estimation of herbage and supplement intakes of individual cows grazing in a group context: testing of field procedures.

**Section III: System studies. Productive performance of contrasting Holstein-Friesian genotypes in pasture-based systems.**

Chapter 5. Interactions between cow genotype and management.

Chapter 6. Forage and supplement consumption, and cow nutrition.

Chapter 7. Herbage intake and grazing behaviour of three Holstein-Friesian genotypes managed at low or high feed allowance in early and late lactation

**Section IV: Component study. Herbage intake and grazing ability of contrasting Holstein-Friesian genotypes.**

Chapter 8. Herbage intake and productive performance of three Holstein-Friesian genotypes grazing under controlled sward conditions.

**Section V: Final discussion.**

Chapter 9. Final integrated discussion and conclusions.

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## SECTION II

### THE USE OF N-ALKANES IN ESTIMATING HERBAGE INTAKE, DIGESTIBILITY AND DIET COMPOSITION IN GRAZING ANIMALS

Daily herbage dry matter intake ( $DMI_H$ ) determines the nutritional status and milk production of a dairy cow fed on pasture and is a factor contributing to most of the productive and behavioural responses and efficiencies to be estimated. It is also necessary to understand the factors that determine cow productivity and differences between individual animals. In grazing situations the  $DMI_H$  of individual cows is generally difficult to measure, and when obtained, a large error is often associated with the estimate (Mayes & Dove, 2000). Direct measurement techniques usually disturb the cow and changes in her grazing behaviour potentially affect  $DMI_H$ . Another approach is to measure faecal output (FO), which can be indirectly estimated by using Chromic Oxide ( $Cr_2O_3$ ) as a marker, but the calculation of  $DMI_H$  is then dependent on the accurate measurement of the digestibility of the herbage ingested ( $D_H$ ), obtained from the analysis of one *in-vitro* sample that is assumed would give a measure of the digestibility of the diet of the cow. This requires that the sample of herbage must closely represent what the cows have eaten. Additionally, the estimation of  $DMI_H$  when grazing animals are fed supplement is also based on the *in-vitro* of both feeds, even though  $D_H$  values could differ when they are fed separately or in combination (Dillon, 1993).

The wax present in the plant cuticle is a complex chemical containing n-alkanes, which are suitable for use as markers in nutritional studies. The first results from  $DMI_H$  estimated using the n-alkane method were presented by Mayes et al. (1986b), who suggested that these markers could also be used to estimate the '*in-vivo*' digestibility of the diet (DMD). A pasture sample that represents accurately what the cows have eaten is also crucial with this method despite the fact that DMD can be also estimated by using these natural markers, which would increase the precision of the estimate obtained.

However, data from Mayes et al. (1996b) indicate incomplete recovery of the natural markers in animal's faeces, nevertheless, as recognised by Dove and Mayes (1991), the main contribution of Mayes and co-workers was to point out that the incomplete recovery would not affect the estimate of  $DMI_H$ , if both of the odd and even alkanes used have similar faecal recovery rates. Since then, others (Dillon & Stakelum, 1989; Dillon, 1993) have applied the same principle to estimate  $DMI_H$  and DMD in dairy cows, moreover other uses for the n-alkanes were also proposed (Dove & Mayes, 1991).

To estimate total nutrient intakes, understand differences in the animal's response to extra feed and estimate efficiencies, other information in addition to  $DMI_H$  is needed. This includes diet composition and DMD as the most important parameters to be determined. The main interest in using n-alkanes is that this provides an independent estimate of  $DMI_H$  and DMD for each individual cow in a herd, so differences between individuals can be identified.

The three chapters in this section deal with aspects of the evaluation and utilisation of methods for estimating the  $DMI_H$  and diet composition of grazing animals, based on modern techniques associated to natural markers.

Chapter 2 deals principally with a review of literature on the use of the n-alkanes methodology (Mayes *et al.*, 1986b; Dove & Mayes, 1991; Dove, 1996; Mayes & Dove, 2000), but also introduces information on the use of the stable isotope discrimination method in the specific case of diets containing a combination of  $C^3$  and  $C^4$  plant components (Jones *et al.*, 1979).

Chapter 3 examines in more detail the specific problem of cow selectivity and its effects on the accuracy of the  $DMI_H$  estimated from n-alkanes. The possibility of reducing this problem by utilising the two main natural markers in the herbage consumed is analysed in detail to improve the estimation of both the individual  $DMI_H$  of grazing dairy cows and the subsequent estimate of diet composition.

Chapter 4 concentrates more on the combination of methods utilised in the field experiments developed for the present thesis, focusing on estimating the individual  $DMI_H$  and diet composition of grazing dairy cows also fed forage and supplement in the paddock, managed in a group feeding context or 'system'.

## CHAPTER 2

# A REVIEW OF THE N-ALKANE METHOD AND ITS ACCURACY

### 2.1. INTRODUCTION

The main interest in using *n-alkanes* is that it provides an independent estimate of herbage dry matter intake ( $DMI_H$ ) and diet digestibility (DMD) for each individual animal in a grazing herd, so differences between individuals can be identified. The objective of this chapter is to review the advantages and accuracy of the *n-alkanes* methodology to estimate  $DMI_H$  in a strip-grazing situation with dairy cows in relation to other methods that use the same principle. In addition, to analyse those factors that should be considered to improve the accuracy of the  $DMI_H$  estimate, furthermore, to explore how to use a combination of methodologies to estimate forage and supplement consumption in grazing dairy systems.

### 2.2. DESCRIPTION OF METHODS

#### 2.2.1. The faecal output-diet digestibility method

One of the most successful animal based methods to estimate individual intake in grazing animals is the faecal output ~ diet digestibility [or Chromic Oxide ( $Cr_2O_3$ ) ~ *in-vitro*] method where  $DMI_H$  is calculated from the measurement of total faecal output (FO) and the *in-vitro* digestibility of the herbage consumed ( $D_H$ )(Dillon, 1993; Dove & Mayes, 1996).

**Equation 2.1:**  $DMI_H = FO / (1 - D_H)$

where  $DMI_H$  is the herbage dry matter intake, FO is the total faecal output and  $D_H$  is herbage digestibility.

The accuracy of the method relies on both the estimation of the FO for each individual animal and the measurement of  $D_H$ . Faecal output can be measured directly by collecting the total faeces produced daily and over a number of days, using bags attached to a

harness. On FO is estimated using the harness, the *in-vitro* digestibility of a sample representing what the animal consumed can be used to calculate  $DMI_H$ . This procedure is usually used indoors where complete collection is more practicable, in contrast, the use of the harness on a grazing cow could modify her natural behaviour and negatively affect both  $DMI_H$  and daily production. Diet digestibility is defined as the portion of the diet absorbed by the animal and not excreted in faeces; therefore, the direct estimation of  $D_H$  for each animal requires the measurement of intake and FO, which is not possible for a grazing cow. To solve this problem indirect methods have been developed.

A widely used indirect method is to determine FO from the dilution in faeces of a known amount of an indigestible marker, dosed daily.

**Equation 2.2:**  $FO = (D_{Oj} * R_j) / F_j$

where  $D_{Oj}$  is the amount of the daily dose of the marker ( $g \text{ day}^{-1}$ ),  $R_j$  is the recovery rate of the marker and  $F_j$  is the mean concentration of the marker in faeces ( $g \text{ kg}^{-1} \text{ DM}$ ).

The marker must be administered over a period of 10-14 days with faeces sampled over the last 5-7 days, once the concentration of the marker in faeces reaches equilibrium (steady state). Different markers have been used (Mayes & Dove, 2000),  $Cr_2O_3$  being one of the first widely mentioned in the literature - probably because of its high recovery rate in faeces. Recovery is defined as the weight of the marker excreted (measured with total faeces collection) expressed as percentage of the weight of the marker given (Curran *et al.*, 1967). Mean recovery values of 96.5% for cattle and sheep (Le Du & Penning, 1982) and 97.2% for dairy cows (Corbett *et al.*, 1958) have been reported, with a coefficient of variation (CV%) between animals of 7.2% for  $Cr_2O_3$  impregnated paper and 9.4% when  $Cr_2O_3$  is given in capsules (Langlands *et al.*, 1963).

Although the use of a marker like  $Cr_2O_3$  allows prediction of the individual faecal output, expressed in dry matter basis (DM), of each animal in a group, the  $DMI_H$  calculated still relies on the estimate of the  $D_H$  for the diet. Digestibility can be measured using *in-vitro* methods but this value does not always represent the actual value of the diet, which is difficult to obtain for each animal in a group grazing together. Thus, estimation of the  $D_H$  value is considered to be the main cause of concern when using  $Cr_2O_3$ , either due to accuracy of the estimation of  $D_H$ , especially with diets of higher digestibility where the error in  $DMI_H$  caused by errors in  $D_H$  increases (Dove & Mayes, 1996; Mayes & Dove, 2000), or because it is unlikely that the mean pasture sample collected from the pasture actually represents the diet of each cow in the group.

In grazing experiments usually one hand clipped pasture sample represents what the group of cows are consuming, and is the origin of  $D_H$  (*in-vitro*). It is assumed that a common digestibility value applies to all the cows in the group and no allowance is made for differences in grazing activity, capacity to exert selection and differences in digestion between cows. Factors like animal species, age or feeding level are not considered in the estimate of  $D_H$  when the *in-vitro* technique is used (Dillon, 1993). The digestibility value obtained *in-vitro* is calibrated against *in-vivo* measurements, usually obtained with sheep fed at maintenance level of feed. This calibration is then applied to other types of animals fed different levels of feed, which can cause also errors. An additional problem is the prediction of the whole-diet digestibility when animals consume a combination of feeds with different digestibility, such as pasture and concentrates (Dixon & Stockdale, 1999; Mayes & Dove, 2000), making this procedure invalid for use with supplemented animals (Dove & Mayes, 1996).

### **2.2.2. Obtaining a representative sample of the herbage consumed**

As for other methods requiring analysis of the diet, the sample of feed (herbage or a combination of herbage plus supplement) must represent what the animal actually consumed (Mayes & Dove, 2000). Nevertheless, and despite the effort involved, this sample is usually different from what each animal ingested. A principal reason for this is that grazing animals are highly selective individuals, while opportunities for selectivity are increased in heterogeneous swards. Because of this problem, the digestibility obtained *in-vitro* is more affected by errors associated with the sampling procedure of the sward than by errors in the analytical methods used.

In order to obtain one herbage sample that is similar to what the animals consume two main procedures are used: a) the hand clipping technique and b) the use of oesophageal fistulated animals (OF). The first is based on a close observation of what species and plant-parts the animals are mainly eating and then the equivalent parts of similar plants are plucked or cut from the pre-grazing sward. This procedure is easy to implement in a rotational grazing system by observing the paddocks recently grazed to determine grazing height and then sampling the herbage from the same grazing height in the next paddock to be grazed. If residual mass after grazing is low and evenly distributed, the sample taken pre-grazing will closely represent the mean characteristics of the herbage ingested by the group of cows. In contrast, when an uneven post-grazing residual mass combining long and short patches is left after grazing due to lax pasture management, the probability of obtaining a pre-grazing sample that accurately represents what the cows will consume is reduced. The identification of those areas, species, or plant parts that the animal will avoid when grazing will result in improving the sample taken. Thus, the sampling

procedure is simple but subjective, and the species or plant parts sampled could be different from those actually consumed; in addition, the operator could bias the sample obtained in heterogeneous situations. In this case the difference between 'diet' and 'sample'  $D_H$  is likely to increase.

In the OF method, a number of surgically prepared animals (with a fistula in the oesophagus), similar in breed, age and productivity to those used in the grazing study, are required. In these fistulated animals the herbage consumed passes directly through the fistula into a sample collection bag. The use OF animals was claimed to overcome the problems mentioned with the hand clipped method, however, these animals can select a diet that is not similar to what the group is eating (Dove & Mayes, 1991). For example, a short period of grazing (minutes) is assumed to represent what the animal consumes during the whole day, even though it has been recognised that animal preference changes during the day. Additionally, contamination of the sample with saliva and/or digestive enzymes can increase the digestibility of the sample by 1.5-3% (Le Du & Penning, 1982). Coates et al. (1987) suggested that OF animals should not be used because the botanical composition of the extrusa collected from them did not provide a good estimate of the diet consumed by resident animals.

It is evident that sampling problems are present in both sampling procedures, but are reduced in uniformly sown pastures (Dove & Mayes, 1996) where a representative sample can be achieved more readily from either hand gathered or extrusa samples (Vulich *et al.*, 1991). A representative sample of the herbage consumed is required to provide an *in-vitro*  $D_H$  value that accurately represents the animal's diet. It seems that, despite every effort in the sampling procedure, the actual  $D_H$  values of individual animals are likely to differ from the *in-vitro*  $D_H$  from the average sample.

### **2.2.3. The *n-alkanes* method to estimate dry matter intake and diet digestibility in grazing animals**

Herbivore nutritionists have long sought that one indigestible marker substance in the diet (internal marker) could be used to estimate the *in-vivo* digestibility of the diet (DMD) from its concentrations in diet and faeces. This method is based on the knowledge of the concentration of a substance in the feed and in the faeces of the animals, so that digestibility can be estimated according to the following equation:

**Equation 2.3:**  $DMD = 1 - (H_i / F_i)$

where  $H_i$  is the concentration of the marker (i) in the herbage and  $F_i$  is the concentration of the marker i in the faeces.

The substance to be used as a marker must be indigestible and should not be altered during passage through the digestive tract. A number of substances in the herbage have been suggested as potential markers: lignin, chromogen, silica, potentially indigestible cellulose and indigestible acid detergent fibre have all been mentioned. These substances, however, have been shown to be inaccurate for different reasons (Dillon, 1993). On the other hand, wax in the plant cuticle is a complex chemical mixture of saturated hydrocarbons (mainly *n*-alkanes) which have the necessary properties for use as natural markers (Dove & Mayes, 1991).

**Table 2.1: Mean *n*-alkane concentrations of some temperate and tropical species, forage and concentrates.**

Species	<i>n</i> -alkanes (mg 100 g <sup>-1</sup> DM)								Reference
	C <sub>27</sub>	C <sub>28</sub>	C <sub>29</sub>	C <sub>30</sub>	C <sub>31</sub>	C <sub>32</sub>	C <sub>33</sub>	C <sub>35</sub>	
<i>Lolium perenne</i> L.	1.9	0.50	7.3	0.90	13.70	0.9	11.6	1.8	Dove and Mayes, 1991, 1996
	2.9		9.3		11.90		7.9	1.4	Dove and Mayes, 1991, 1996
	3.6	0.60	14.2	1.20	22.00	0.7	9.9	0.9	Dove and Mayes, 1991, 1996
	2.6	0.70	16.3	1.40	26.10	0.8	11.0	0.7	Dove and Mayes, 1991, 1996
	2.0		10.9		21.50		14.1	1.2	Dove and Mayes, 1991, 1996
	2.9	1.10	15.9	1.80	31.70	1.0	14.9	1.1	Dove and Mayes, 1991, 1996
	2.58	0.56	10.4	0.85	15.32	0.86	9.62	1.08	Dillon, 1993
	2.06	0.50	9.07	0.92	14.39	0.73	10.52	1.09	Hameleers and Mayes, 1998
	2.6	0.70	19.0	1.5	27.50	0.7	8.9	0.3	Laredo <i>et al.</i> , 1991
	4.02	1.12	23.1	1.17	24.23	0.59	5.70	0.65	Dillon, 1993
<i>Dactylis glomerata</i> L.	2.0	0.20	3.8	0.20	5.80	0.2	2.1	0.0	Dove and Mayes, 1991, 1996
	0.99	0.09	1.46	0.09	5.15	0.32	5.57	0.57	Dillon, 1993
<i>Festuca rubra</i> L.	2.16	0.33	21.6	0.65	27.30	0.47	4.98	0.75	Dillon, 1993
<i>Trifolium repens</i> L.	3.8	0.7	10.9	0.50	6.70	0.1	0.7	0.0	Dove and Mayes, 1991, 1996
	1.9		7.5		6.60		0.5	0.0	Dove and Mayes, 1991, 1996
	1.2	0.60	8.8	0.40	5.50	0.3	2.6	0.5	Dove and Mayes, 1991
	2.84	0.40	5.12	0.32	4.23	0.38	3.42	0.76	Dillon, 1993
	2.99	0.58	7.22	0.42	5.04	0.28	0.85	0.05	Hameleers <i>et al.</i> , 1998
	3.0	1.10	40.8	0.50	5.70	0.10	1.10	0.0	Dove and Mayes, 1991, 1996
<i>Trifolium pratense</i> L.	3.4		37.6	0.30	4.20		0.80	2.0	Dove and Mayes, 1991, 1996
	3.6	0.90	20.2	1.20	32.4	0.70	2.10	0.0	Dove and Mayes, 1991, 1996
<i>Medicago sativa</i>	5.5	0.60	20.7	1.30	10.4	0.30	0.80	0.0	Dove and Mayes, 1991, 1996
<i>Paspalum dilatatum</i> poir	0.4	0.20	1.4	0.60	10.4	0.60	3.9	0.2	Dove and Mayes, 1991
<i>Pennisetum</i> <sup>(1)</sup>	1.64		2.62	0.83	20.47	1.04	24.07	10.59	Reeves <i>et al.</i> , 1996
<i>Sorghum</i> spp.	4.8	0.60	12.0	0.40	6.3	0.30	2.8	1.7	Laredo <i>et al.</i> , 1991
<i>Hordeum vulgare</i>	1.25	0.20	1.37	0.18	0.83	0.09	0.14	0.04	Hameleers <i>et al.</i> , 1998
Grass pasture	2.48	0.56	7.39	0.81	11.7	0.663	8.83	1.25	Hameleers <i>et al.</i> , 1998
Grass silage (*)	3.12	0.54	1.41	1.03	20.2	0.72	12.89	1.45	Hameleers <i>et al.</i> , 1998
Concentrate (**)	0.37	0.06	0.73	0.06	0.80	0.05	0.15	0.0	Hameleers <i>et al.</i> , 1998

<sup>(1)</sup>*Pennisetum clandestinum*; (\*) Ensiled from the grass based pasture which *n*-alkanes profile is previously indicated; (\*\*) Oven-dry matter: 870 g kg<sup>-1</sup>; ME: 12 MJ kg<sup>-1</sup> DM; CP: 200 g kg<sup>-1</sup>.

The *n*-alkanes have mostly odd numbers of carbon atoms in the range C<sub>25</sub>–C<sub>35</sub>-alkanes. The length of the carbon-chain identifies the type of *n*-alkanes present; shorter carbon-chain (under C<sub>25</sub>) are present in small quantities and are difficult to detect, whereas those in the range of C<sub>25</sub> (pentacosane) to C<sub>35</sub> (pentatriacontane) are usually present in most of the temperate pastures species and easily detectable. Within the range of C<sub>25</sub>–C<sub>35</sub>-alkanes, odd-numbered carbon chain *n*-alkanes are present in much greater amounts than even-numbered alkanes, with C<sub>29</sub>-alkane (nonacosane), C<sub>31</sub>-alkane (hentriacontane) and

C<sub>33</sub>-alkane (tritriacontane) being predominant and showing different absolute and relative content between species (Tables 2.1 and 2.2).

Grace and Body (1981) were the first to mention the potential use of these cuticular plant wax components of long-chain (C<sub>19</sub>–C<sub>32</sub>) carbons as markers in nutritional studies. They also demonstrated *n*-alkanes are highly indigestible (Grace & Body, 1981). Subsequently it was suggested these markers could be used to estimate DMD, however it was observed that faecal recovery was incomplete and declined as carbon-chain length decreased (Mayes et al., 1986b). Incomplete recovery occurs due to absorption from the small intestine (Dove & Mayes, 1996); consequently the estimation of DMD obtained must include a correction for the incomplete recovery of the *n*-alkanes used. Faecal recovery in cattle (see Table 2.3) appears to be lower and more erratic than in sheep (Dove & Mayes, 1991) so errors in the estimate of DMD may be greater in cattle.

**Table 2.2: Mean *n*-alkane concentration of leaves and stems of three temperate pasture species.**

Species	<i>n</i> -alkane (mg 100 g <sup>-1</sup> DM)							
	C <sub>27</sub>	C <sub>28</sub>	C <sub>29</sub>	C <sub>30</sub>	C <sub>31</sub>	C <sub>32</sub>	C <sub>33</sub>	C <sub>35</sub>
<i>Lolium perenne</i> L. (leaves)	2.49	0.61	10.39	0.95	15.94	0.85	10.99	1.44
<i>Lolium perenne</i> L. (stems)	2.41	0.37	8.9	0.52	11.54	0.58	5.33	0.83
<i>Lolium multiflorum</i> L. (leaves)	4.35	1.21	32.37	1.44	32.63	0.78	7.69	0.73
<i>Lolium multiflorum</i> L. (stems)	2.79	0.79	19.33	0.94	22.36	0.44	3.94	0.28
<i>Trifolium repens</i> L. (leaves)	2.89	0.47	4.75	0.16	0.86	0.23	0.45	0.43
<i>Trifolium repens</i> L. (stems)	2.21	0.12	0.75	0.0	0.46	0.20	0.52	0.49

All data from Dillon, 1993.

To estimate DMD using the marker – ratio technique, Equation 2.3 should be used. The only requirement is to know the concentration of the odd-chain alkane “i” in the herbage and the concentration in faeces of the same *n*-alkane (mg kg<sup>-1</sup> DM). As the recovery rate of the *n*-alkanes in faeces is not complete, a correction for incomplete recovery has to be applied (R<sub>i</sub>). This is shown in Equation 2.4:

$$\text{Equation 2.4: } \text{DMD} = 1 - (\text{H}_i \cdot \text{R}_i) / \text{F}_i$$

This is similar to Equation 2.3 with H<sub>i</sub> being the concentration of the *n*-alkane (i) in the herbage, R<sub>i</sub> is the recovery of the same *n*-alkane (i) in the faeces and F<sub>i</sub> is the concentration of the *n*-alkane (i) in the faeces.

In contrast to the Cr<sub>2</sub>O<sub>3</sub> – *in-vitro* method, Equation 2.4 provides an estimate of the DMD of the diet for each animal within a group grazing the same pasture while differences in the accuracy of the digestibility value between *in-vitro* and *n*-alkanes will depend on the relative variability of the *n*-alkanes profile of the ingested herbage in comparison to the *in-vitro* estimate from the average pasture sample. Comparisons between the Cr<sub>2</sub>O<sub>3</sub> – *in-*

*vitro* and the *n*-alkanes methods show the superiority of the *n*-alkanes methodology for cattle and sheep (Malossini *et al.*, 1996; Mayes & Dove, 2000) and indicate that the discrepancy between methodologies depends on the *in-vitro* being representative of the *in-vivo* digestibility.

Herbage dry matter intake per cow can be estimated in addition to DMD, by dosing a daily known amount of an even-chain *n*-alkane as an indigestible marker (usually C<sub>32</sub>-alkane, which is normally present in a very low concentration or not present in the pasture). This double capacity to estimate DMI<sub>H</sub> and DMD simultaneously is a great advantage compared with the Cr<sub>2</sub>O<sub>3</sub> – *in-vitro* method from the expansion of the Equations 2.2 and 2.4 herbage dry matter intake (DMI<sub>H</sub>) can be estimated:

**Equation 2.5:**  $FO = (D_{Oj} * R_j + DMI_H * H_j * R_j) / F_j$

Equation 2.5 is derived from Equations 2.2 and 2.4, with D<sub>Oj</sub> being the total daily amount of the dosed alkane 'j' (g day<sup>-1</sup>), R<sub>j</sub> the recovery of the dosed alkane 'j', H<sub>j</sub> and F<sub>j</sub> the concentration of the alkane 'j' in the herbage and in faeces respectively (mg kg<sup>-1</sup>DM). It has to be noted that although the higher proportion of the 'j' alkane should come from the daily dose, a small proportion could be present in the pasture and considered in the calculation (H<sub>j</sub>).

**Equation 2.6:**  $DMI_H = [(D_{Oj} * R_j + DMI_H * H_j * R_j) / F_j] / [H_i * R_i / F_i]$

Which considers the dosed odd chain *j*-alkane plus the content of the same *j*-alkane in the pasture, both affected by its recovery in faeces.

**Equation 2.7:**  $DMI_H * F_j * H_i * R_i = R_j (D_{Oj} + DMI_H * H_j) * F_i$

if the recovery of alkanes 'j' and 'i' in faeces is similar, they cancel out in Equation 2.8.

**Equation 2.8:**  $DMI_H * F_j * H_i = F_i * D_{Oj} + F_i * DMI_H * H_j$

**Equation 2.9:**  $DMI_H * (F_j * H_i - F_i * H_j) = F_i * D_{Oj}$

**Equation 2.10:**  $DMI_H = F_i * D_{Oj} / (F_j * H_i - F_i * H_j)$

**Equation 2.11:**  $DMI_H = F_i * D_{Oj} / [F_j * (H_i - (F_i / F_j) * H_j)]$

**Equation 2.12:**  $DMI_H = (F_i / F_j) * D_{Oj} / [H_i - (F_i / F_j) * H_j]$

Equation 2.12 is the equation presented by Dove and Mayes (1996), where  $DMI_H$  is the calculated herbage dry matter intake expressed in  $kg\ cow^{-1}day^{-1}$ . To estimate  $DMI_H$  for each cow using Equation 2.12, the concentrations of the natural alkanes 'i' and 'j' in H and F respectively are required, plus the total amount of the dosed alkane 'j'.

As indicated by Mayes et al. (1986b) the use of a pair of *n*-alkanes with similar recovery rates will give unbiased intake estimations. This method solves most of the problems associated with the use of other markers. However, it still relies on the accurate measurement of the *n*-alkanes profile of the pasture sample eaten, therefore the sampling problems mentioned still apply to this methodology. Nevertheless, individual values for  $DMI_H$  and DMD are estimated for each animal in the herd, which is an advantage compared with the use of the  $Cr_2O_3$  – *in-vitro* method.

When  $DMI_H$  has to be estimated for cows eating a combination of different feeds with very different *n*-alkanes content for instance pasture and maize silage, the last one with very low content of *n*-alkanes (see Table 4.1), the faecal concentrations of the natural and dosed *n*-alkanes are reduced. This effect of dilution does not affect their faecal recovery rate (Mayes et al., 1986b) and the estimate of  $DMI_H$  is still accurate.

The best estimation of  $DMI_H$  is usually obtained with the pair  $C_{33}$ – $C_{32}$  (Mayes *et al.*, 1986b; Dove & Mayes, 1991; Vulich *et al.*, 1991; Dillon, 1993; Reeves *et al.*, 1996; Unal & Garnsworthy, 1999; Berry *et al.*, 2000) while other possible pairs (e.g. either  $C_{29}$ – $C_{32}$  or  $C_{31}$ – $C_{32}$ ) underestimated the actual intake due to a reduction in the recovery rate of  $C_{29}$  and  $C_{31}$ -alkanes relative to  $C_{33}$ . This discrepancy was not affected by diet type, feeding level, or method of administering the dosed *n*-alkanes (Mayes et al., 1986b), although errors may be expected to be larger in grazing animals and more related to the difficulty of collecting a representative sample of the ingested herbage (Mayes et al., 1986b). However, if accurate estimates are to be obtained, it has been recommended that the concentration of the natural *n*-alkane in the forage exceed  $5\ mg\ 100\ g^{-1}\ DM$  (Laredo *et al.*, 1991). It should be noted that  $C_{33}$  is not present in sufficient quantity in some tropical species (Laredo et al., 1991) like *Zea mays* L. (maize) or *Sorghum saccharatum* L. (sorghum), frequently used as supplements in pasture based systems. Laredo et al. (1991) suggested that a shorter chain-length *n*-alkane should be used in tropical species, but recognised a higher bias due to the lower recovery rate between dosed and natural *n*-alkane. On the other hand, Hamelers and Mayes (1998a) suggested the use of the  $C_{31}$  instead of  $C_{33}$ -alkane for animals grazing *Trifolium repens* L. (white clover). As the content of  $C_{31}$  in this species is higher, a better estimate of  $DMI_H$  could be expected using the pair  $C_{31}/C_{32}$  instead of the pair  $C_{33}/C_{32}$ .

As mentioned, the ratio of the faecal concentrations of natural and dosed *n*-alkanes is needed to estimate  $DMI_H$  (Mayes et al., 1986b). Thus, in addition to the pasture sample, a representative sample of faeces should be collected. Within-day variations in such ratios are relevant when faecal grab samples are taken at regular intervals. In addition, day-to-day variation in the ratio could also affect the estimate of dry matter intake. A significant diurnal variation in faecal *n*-alkanes concentration in dairy cows has been reported for once or twice daily dosing (Dove & Mayes, 1991). In contrast, Mayes et al. (1986b) did not find any evidence of systematic variation throughout the day and between days. They suggested one daily dose and a grab faecal sample will be sufficient for accurate estimation of  $DMI_H$  and DMD; similar values for  $DMI_H$  have been estimated from am or pm faecal samples (Reeves et al., 1996). Malossini et al. (1996) found a smaller variability of the *n*-alkanes concentration in faeces than for  $Cr_2O_3$ , which allowed the number of samples to be reduced with the *n*-alkanes method. However, errors could arise since it is assumed that grab samples are representative of the total daily FO of the sampled animal. Dove and Mayes (1991) indicated that the diurnal variation in the ratio of the faecal concentration of herbage and dosed *n*-alkanes is more important than the variation of the absolute marker concentration which is the case when  $Cr_2O_3$  is used; thus it is possible to have temporal variability in the absolute faecal concentration of each of the *n*-alkanes selected, but a constant ratio in their faecal concentrations (Dove & Mayes, 1991). Dillon and Stakelum (1989) found that diurnal variation in the faecal ratios was greater when dosing once rather than twice daily, with the dosed *n*-alkanes being the main contributor to the diurnal variation of the ratio.

In order to reduce the variability in the faecal ratio caused by dosing once or twice daily and the additional work involved in the faecal sampling procedure, a controlled release capsule (CRC) containing synthetic *n*-alkanes has been designed (Mayes & Dove, 2000), which is commercially available (Captec Alkane<sup>TM</sup>, Captec Ltd, Auckland, New Zealand). This device is administered once orally at the start of the experiment and releases intra-ruminally  $C_{32}$  and  $C_{36}$ -alkanes at a steady rate of 388 – 386 mg day<sup>-1</sup> respectively, with an expected time-span at a constant release rate of 20±3 days (Berry et al., 2000). Using the CRC the optimal time for a once a day grab sample was in the early morning using the ratio of  $C_{33}/C_{32}$  to estimate  $DMI_H$  (Berry et al., 2000).

Weekly pooled samples from faeces were highly correlated to the linear regression of the daily intakes estimates and the recovery estimated from the weekly bulk of daily faecal samples was similar to the estimated mean of the same samples, which indicates that the method produce an accurate estimate when a weekly bulk sample is used (Berry et al., 2000). Vulich and Hanrahan (1995) estimated a bias of around 3 and 9% in  $DMI_H$  based on the analysis of an individual grab sample, if it was a pooled sample or a number of

individual daily samples respectively, using a daily dose (am) of the C<sub>32</sub>-alkane. Taking grab samples twice daily would reduce the magnitude of the bias in DMI<sub>H</sub> and it is reasonable to expect that simultaneously sampling and dosing would reduce the bias (Vulich & Hanrahan, 1995), particularly when realised at the same time every day (Unal & Garnsworthy, 1999).

#### 2.2.4. Recovery of the marker in faeces

Published recovery values in faeces for the main *n*-alkanes present in the pasture consumed from studies with dairy cows are presented in Table 2.3. As mentioned, faecal recovery of alkanes is not complete (Mayes et al., 1986). When both of the *n*-alkanes in the pair used to estimate DMI<sub>H</sub> had similar recovery rates in faeces, the incomplete recovery errors R<sub>j</sub> and R<sub>i</sub> cancel out in the numerator and denominator of Equation 2.7 because they are affected by similar biases (Mayes *et al.*, 1986b; Dove & Mayes, 1991; Hameleers & Mayes, 1998a). Additionally, it was mentioned that the estimated error arising from a different recovery rate between two *n*-alkanes is lower than the discrepancy that arises from a similar error in digestibility when the Cr<sub>2</sub>O<sub>3</sub> is used as the marker. This error increases if the digestibility of the feed consumed is high (Dove & Mayes, 1991; Dove & Mayes, 1996).

**Table 2.3: Mean faecal recovery rates (proportion of ingested recovered in faeces) of odd and even chain *n*-alkanes present in the herbage from experiments with dairy cattle.**

Reference	<i>n</i> -alkane								
	C <sub>27</sub>	C <sub>28</sub>	C <sub>29</sub>	C <sub>30</sub>	C <sub>31</sub>	C <sub>32</sub>	C <sub>33</sub>	C <sub>35</sub>	C <sub>36</sub>
Dillon, 1993	0.62±0.34	0.84±0.05	0.81±0.03	0.80±0.03	0.83±0.03	0.88±0.03	0.87±0.04	0.91±0.03	0.91±0.02
Dillon, 1993	0.76±0.05	0.81±0.04	0.79±0.03	0.80±0.04	0.83±0.02	0.87±0.04	0.85±0.02	0.94±0.04	0.86±0.03
Dillon, 1993	0.55±0.07	0.57±0.07	0.69±0.10	0.65±0.09	0.72±0.11	0.74±0.09	0.73±0.09	0.78±0.11	0.76±0.07
Dillon, 1993	0.63±0.03	0.69±0.04	0.73±0.02	0.77±0.02	0.78±0.02	0.86±0.03	0.84±0.02	0.88±0.02	0.87±0.04
Dillon, 1993	0.62±0.03	0.65±0.03	0.76±0.04	0.73±0.04	0.80±0.05	0.83±0.05	0.82±0.05	0.86±0.05	0.84±0.05
Unal and Garnsworthy, 1999						0.95±0.04	0.94±0.03		0.95±0.04
Berry <i>et al.</i> , 2000					0.76±0.02	0.87±0.02	0.85±0.02		0.81±0.03

Data from Dillon (1993), estimated from different experiments with dairy cows fed pasture alone or combined with supplements. C<sub>32</sub> and C<sub>36</sub> were dosed *n*-alkanes.

On the other hand, for the *n*-alkanes to provide an accurate DMD, a correction for incomplete recovery is required (Dove & Mayes, 1991). Thus, the prediction of the recovery rate of the *n*-alkanes used will define the accuracy of the DMD predicted. As indicated by Mayes and Dove (2000), the C<sub>35</sub>-alkane is a satisfactory marker for DMD, with a recovery of at least 95%. However, herbage content of C<sub>35</sub> is not high enough to allow its use in both DMI<sub>H</sub> and DMD estimates, and mainly due to this the faecal C<sub>33</sub>/C<sub>32</sub> ratio is usually recommended (Mayes *et al.*, 1986a; Dove & Mayes, 1991; Vulich & Hanrahan, 1995; Reeves *et al.*, 1996; Unal & Garnsworthy, 1999; Berry *et al.*, 2000).

Different recovery rates have been recorded for sheep and cattle, with greater differences for short than long chain *n*-alkanes (Dove & Mayes, 1991). Differences in recovery between adjacent pairs decrease as chain length increases (Dove & Mayes, 1991) so the use of short chain *n*-alkanes will increase the difference between the value estimated and the actual intake. Errors in the estimated DMD may be greater in cattle as faecal recovery is usually lower and more erratic than in sheep (Dove & Mayes, 1991). However, it has been demonstrated that faecal recovery of *n*-alkanes increases with increasing carbon-chain length in both animal species (Mayes *et al.*, 1986b; Dove & Mayes, 1991; Dove, 1992; Dove & Mayes, 1996).

The relative proportions of the individual *n*-alkanes in the diet and faeces of ruminants suggest that the microbial population in the rumen has little effect on these natural markers (Mayes *et al.*, 1986b). Hence, although some degree of absorption in the digestive tract was suggested, the profile of the long chain *n*-alkanes in faeces can be used as a good indication of the diet of the animal.

### **2.2.5. Differences in *n*-alkanes content between species and plant components**

The concentrations and patterns of *n*-alkanes in the pasture are known to change with pasture composition and state of growth of each component (Laredo *et al.*, 1991; Dillon, 1993; Hameleers & Mayes, 1998b; Genro *et al.*, 2001), the total content of *n*-alkanes being lower in some species (e.g. particularly white clover but also in *Paspalum dilatatum* poir.) than in others (Tables 2.1 and 2.2). These differences between species and plant parts can be used to estimate the composition of the available and consumed herbage. Hameleers and Mayes (1998b) used this principle to estimate the proportion of herbage and grass silage consumed by individual cows (Hameleers & Mayes, 1998a). It was also interesting that despite the fact that grass silage was originated from the same pasture that cows grazed during the experimental period its *n*-alkanes profile was different because the different stage of maturity between feeds, which allowed a successful discrimination between them. As this profile is also different between plant parts (Dillon, 1993), the same principle could be used to know which species or parts of plants are present in the diet of different animals.

It was mentioned before that for the accurate estimates of  $DMI_H$  and DMD the pasture sample should represent exactly what the animals consumed. Pasture composition can be estimated using simultaneous equations (Dove & Mayes, 1991; Dove, 1992; Newman *et al.*, 1995) or the least square procedure (Dove & Moore, 1995). The accuracy of the estimate is related to the concentrations of *n*-alkanes in the species to be identified in the diet of the cow, as it is more difficult to estimate the proportion of those species with lower contents of *n*-alkanes. Moreover, the accuracy of the estimate is likely to decline

with increases in the number of components to be identified (Dove & Mayes, 1996). In addition, the faecal recovery of each *n*-alkanes used in the procedure must be known.

Other approaches are also valid, for instance the ratios of individual *n*-alkanes could provide an alternative means of determining the legume content in the diet of a cow (Dove, 1992). However, Mayes et al. (2000) indicated that use of the *n*-alkanes with the highest absolute concentration should be the most suitable procedure for the estimation of diet composition.

Dove and Moore (1995) utilised the least squares procedure to develop a programme called 'Eatwhat' to estimate the best solution, normalised into proportions of each species in the pasture. This solution provided the concentration of each *n*-alkane in the diet of the animal. Theoretically, the fact that diet composition can be estimated from faecal *n*-alkanes removes the need to obtain the ideal pasture sample, or to use OF animals. Mayes and Dove (2000) suggested this could be the major advantage of the method as diet composition is estimated from the *n*-alkanes profile of the main species in the pasture and faeces of each animal, therefore once the *n*-alkanes pattern in the diet is known, the individual  $DMI_H$  can be estimated. However, this procedure relies on the accuracy of the pasture sample obtained, which should represent the actual diet of the animal during the faecal collection period.

Although theoretically valid, it is difficult to find examples in the literature where this approach was utilised in grazing experiments, probably because some bias could be introduced when choosing the reference species or plant parts to which the individual faeces should be compared. As individual cows could differ greatly in their capacity to select a diet different from the mean composition of the pasture grazed, using the pasture sample *n*-alkanes profile as a baseline for the diet of all the cows in the group means that the  $DMI_H$  obtained for each cow is not independent from others in the same group (Mayes & Dove, 2000). In addition, errors in  $DMI_H$  estimate are almost directly proportional to any errors in the estimate of the key *n*-alkanes consumed by the animal (Friend *et al.*, 1995).

#### **2.2.6. Stable carbon isotope discrimination**

The ratio of stable isotopes  $^{13}C$  and  $^{12}C$  in plant tissues differs between plants with different photosynthetic pathways ( $C^3$  or  $C^4$  species). The relation between the ratio of these isotopes in the feed and in the resultant faeces was studied and a technique developed to estimate the proportion of  $C^3$  or  $C^4$  species selected by grazing animals (Jones et al., 1979). The ratio of stable isotopes  $^{13}C$  and  $^{12}C$  in a sample of herbage is expressed as the difference between the ratios of the number of atoms of  $^{13}C$  to the

number of atoms of  $^{12}\text{C}$  in a standard sample ( $\delta^{13}\text{C}$ ). Thus by knowing the  $\delta^{13}\text{C}$  in the feed or pasture, the proportion of each  $\text{C}^3$  and  $\text{C}^4$  species ingested by the animal can be calculated from the  $\delta^{13}\text{C}$  of the faeces. The accuracy of this method is high, with a few exceptions mainly attributed to contamination of material from the previous feeding regime of the animals. This contamination can be avoided as a new equilibrium was reached for cattle after a 6-day period, which was considered necessary for the complete passage of the previous feed through the rumen.

The technique depends upon the fact the  $\delta^{13}\text{C}$  is about  $-28$  and  $-11$  for  $\text{C}^3$  and  $\text{C}^4$  species respectively, so if the  $\delta^{13}\text{C}$  of both components of the pasture are known, the proportion of the  $\text{C}^3$  and  $\text{C}^4$  species in the pasture or faeces will be in a direct proportion to the amount of these components in the pasture or diet (Jones et al., 1979). The discriminated components are first expressed on organic matter (OM) basis and then a correction by ash content must be used to express them on dry matter basis (DM).

Values for  $\delta^{13}\text{C}$  in faeces were  $0.4 - 2.0\%$   $\delta^{13}\text{C}$  units lower (more negative) than the feed eaten (Jones et al., 1979). Coates et al. (1991) showed that large within-day variations in dietary  $\text{C}^3 - \text{C}^4$  proportions had little effect on faecal  $\text{C}^3$  or  $\text{C}^4$  proportions provided there is no change in diet composition. In addition, sufficiently prolonged changes in the  $\text{C}^3$  or  $\text{C}^4$  proportions in the herbage will result in changes to the  $\delta^{13}\text{C}$  of the faeces, but abrupt changes in the diet are transformed into progressive and gradual changes in the faeces (Coates *et al.*, 1991). They also concluded that changes of a regular nature, for example those associated with the diurnal grazing behaviour, are not likely to cause significant changes in the  $\delta^{13}\text{C}$  of the faeces. These effects should not affect FO or the composition of the faeces if the relative composition, quality and other characteristics of the herbage offered to the herd are maintained when  $\text{DMI}_H$  and the *in-vitro* digestibility are measured, despite changes in the daily pattern of activity due to management (for example milking or supplementation time). This is in agreement with the lack of systematic variation within and between days for the dosed *n*-alkane in faeces (Mayes et al., 1986b).

Another interesting use of the stable carbon isotope discrimination is the use of animal tissues such as hair (Schwertl *et al.*, 2003) to provide an indication of the dietary ratios of the  $\text{C}^3$  or  $\text{C}^4$  species or pasture – supplement proportions in the diet over a long period (Mayes & Dove, 2000). This information in combination with the rate of intake ( $\text{kg DM day}^{-1}$ ) on specific dates would be useful to compare the feeding pattern of cows farmed in different systems.

### 2.2.7. Estimate of dry matter intake and diet digestibility of pasture and supplement consumed together

If supplements are individually offered to the animals and the quantity consumed is exactly known, the concentration of the marker in the supplement must be determined to calculate  $DMI_H$  (Mayes *et al.*, 1986c; Dove & Mayes, 1991), hence this information must be included in Equation 2.12 to generate Equation 2.13.

**Equation 2.13:**  $DMI_H = [(Fi / Fj) * (D_{Oj} + DMI_S * S_j) - DMI_S * S_i] / [Hi - (Fi / Fj) * H_j]$

where  $DMI_H$  is the herbage intake and  $DMI_S$  is the supplement intake (both in kg DM day<sup>-1</sup>); and  $S_j$  and  $S_i$  are the concentration of the natural alkane 'j' and 'i' in the supplement.

This equation can only be used when the quantity of supplement consumed by each cow is accurately measured. However, in most of the situations involving farm or systems studies based on pasture, the supplement is usually offered in the paddock or in troughs in a group context, then consumption must be estimated indirectly. One way to solve this problem is to consider the *n*-alkanes profile of the supplement as if it was another pasture species. The proportion of each feed in the diet can be estimated using the pattern of *n*-alkanes in the faeces of each animal and the pattern shown by each of the dietary components, as was described previously for the identification of different species in the pasture. Consequently, the differentiation between two feeds is possible, dependent on the *n*-alkanes patterns in each feed being different. The supplement could be treated as another forage or species of a mixture and the proportion of each component estimated (Dove, 1992; Hameleers & Mayes, 1998a).

The low *n*-alkanes contents of some supplements, for example tropical species such as sorghum and maize (Bianchi & Avato, 1984; Laredo *et al.*, 1991), could limit the accuracy of estimates by this procedure. One solution is to label the supplement with a marker and monitor its concentration in the faeces; for instance Hameleers and Mayes (1998a) used the C<sub>36</sub>-alkane (hexatriacontane) diluted in heptane, which was mixed with oven-dried soyabean meal and this was then mixed with the supplement (grass silage). The procedure was successful but as the supplement was offered in two meals during the day, there was potential diurnal variation in the excretion pattern of the *n*-alkanes in the faeces. This could explain the higher standard error obtained in the estimate when compared with using the *n*-alkanes profile discrimination (Hameleers & Mayes, 1998a). This indicates that for a better estimation of pasture and forage (e.g. pasture silage) or supplement intakes using *n*-alkanes, the forage should contain naturally occurring *n*-alkanes at measurable concentrations with a pattern different from that of the pasture

(Hameleers & Mayes, 1998a). In contrast, Unal and Garnsworthy (1999) mentioned the benefit of mixing the dosed *n*-alkanes with the supplement, suggesting that it should be fed in the morning to ensure it is eaten. Another marker successfully added to the supplement was Ytterbium acetate (Curtis *et al.*, 1994), which was offered to sheep successfully; however, total faecal collection during a period of 7 days was needed, making this technique difficult to use for grazing dairy cows.

A second option, appropriate in the case of supplementing a temperate pasture with sorghum or maize (tropical grasses with a C<sup>4</sup> photosynthetic pathway), is to use the ratio of natural <sup>12</sup>C and <sup>13</sup>C isotopes ( $\delta^{13}\text{C}$ ) in faeces to estimate the proportion in the diet between the C<sup>3</sup> and C<sup>4</sup> species (Jones *et al.*, 1979; Coates *et al.*, 1991), then use a combination of the *n*-alkane -  $\delta^{13}\text{C}$  methods to estimate intake of each component separately (Garcia *et al.*, 2000) as follows:

$$\text{Equation 2.14: } (H_F * \delta^{13}\text{C}_H) + (S_F * \delta^{13}\text{C}_S) = \delta^{13}\text{C}_F$$

where  $H_F$  and  $S_F$  are the unknown proportions of herbage and supplement in the faeces (constrained to vary between 0 and 1). The  $\delta^{13}\text{C}_H$  and  $\delta^{13}\text{C}_F$  are the values in the herbage and faeces as explained previously. Results must be adjusted for the ash content of the herbage and supplement, expressed as proportions of faecal dry matter (Jones *et al.*, 1979).

$$\text{Equation 2.15: } \text{FO}_H = \text{DMI}_H * (1 - D_H)$$

where  $\text{FO}_H$  is the faecal output estimated from the herbage consumed,  $\text{DMI}_H$  is the herbage intake estimated from Equation 2.8 and  $D_H$  is the *in-vitro* digestibility of the herbage sample.

The  $\text{DMI}_H$  is first estimated using the Equation 2.12, and then  $\text{FO}_H$  is estimated from  $D_H$  using Equation 2.15; this is probably the principal disadvantage of this method as the  $\text{FO}_H$  value relies on the *in-vitro* analysis. Finally, the values of the faecal organic matter ratio (ratio herbage – supplement in faeces) from Equation 2.14, adjusted for ash content of the herbage and the supplement, should be combined with the  $\text{FO}_H$  value allowing the total FO ( $\text{FO}_T$ ) (from herbage plus supplement) to be estimated using Equation 2.16. Then the supplement FO ( $\text{FO}_S$ ) is calculated as the difference between  $\text{FO}_T$  and  $\text{FO}_H$ .

$$\text{Equation 2.16: } \text{FO}_T = \text{FO}_H / (\% H_F)$$

where  $\text{FO}_T$  is the total faecal output (of the herbage plus the supplement) and  $H_F$  is the proportion of the herbage in the faeces estimated as in Equation 2.14.

**Equation 2.17:**  $FO_S = FO_T - FO_H$

where  $FO_S$  is the faecal output of the supplement.

Finally, the supplement dry matter intake ( $DMI_S$ ) is derived from the  $FO_S$  and the supplement *in-vitro*, expressed in  $kg\ DM\ day^{-1}$ .

**Equation 2.18:**  $DMI_S = FO_S / (1 - D_S)$

where  $DMI_S$  is the estimated supplement intake and  $D_S$  is the *in-vitro* digestibility value from a representative supplement sample, which is usually more reliable than the value of the pasture.

The supplement  $DMI_S$  depends on the previously estimated  $DMI_H$  and the  $FO_T$  calculated, so estimates of  $DMI_H$  using the *n*-alkanes method will affect the estimated intake of the supplement. Once the intake of supplement is known, the Equation 2.13 proposed by Mayes and Dove (2000) can be used to improve both  $DMI_H$  and  $DMI_S$  (Garcia et al., 2000), however accuracy is mostly determined by those previously mentioned factors affecting each of the methods combined in the estimate.

### 2.3. CONCLUSIONS

The *n*-alkanes method has been demonstrated to be a useful tool to estimate  $DMI_H$  and DMD of individual grazing cows. Intakes of pasture alone or in combination with supplements (e.g. grass silage, maize grain, or maize silage) have been determined accurately when compared to the actual intake measured directly. When the supplement is derived from a  $C^4$  species, the *n*-alkanes –  $\delta^{13}C$  methodology has been successfully used. This procedure is based on the *in-vitro* of the herbage sample to estimate  $FO$ , which was demonstrated to be less accurate than using *n*-alkanes, and also on the *in-vitro* of the supplement sample. However, it is the only procedure shown experimentally to give accurate estimates for intakes of both the pasture and supplement, when the supplement was maize silage. The same pasture and faecal sample can be used for the analysis of *n*-alkanes or  $\delta^{13}C$ .

The collection of a representative pasture sample has a great influence on the accuracy of the estimate of  $DMI_H$ , and DMD must represent what the animals are consuming. A close observation of what plant species and plant-parts cows are consuming is needed and this will help to improve the sampling procedure when hand plucking is used. In a strip grazing condition with dairy cows grazing a temperate sown pasture, a close observation of post-grazing height, if uniform, will help to improve the pre-grazing sampling. In a

more heterogeneous situation the use of cages could help, however, this will involve additional work and can still be influenced by operator bias. The fact that faecal *n*-alkanes can be used to determine diet composition may remove the use of the pasture sample as being representative of the diet for all the individual cows in the experiment. This possibility should be further tested, as it is also dependant on obtaining a sample of those pasture components and plant parts actually being consumed.

The best pair of *n*-alkanes to estimate  $DMI_H$  has been recognised to be  $C_{33}$ – $C_{32}$ , however, the  $C_{31}$ -alkane content in most of the temperate pasture species is higher than the content of the  $C_{33}$ -alkane. When species with low  $C_{33}$  but high  $C_{31}$  are present and selected by the cows (e.g. white clover or *Paspalum* spp.), the use of the pair  $C_{31}$ – $C_{32}$  could be a better estimator of the intake of these species. Discrepancies between actual and estimated intakes could be partly explained by the consumption of those pasture components with low  $C_{33}$  being underestimated when the pair  $C_{33}$ – $C_{32}$  is used. Similar recovery rates have been described for both  $C_{33}$  and  $C_{32}$ -alkanes, however there is a difference in the recovery rate between the  $C_{31}$  and  $C_{32}$ -alkanes that must be corrected.

Sampling faeces twice daily by taking a grab sample from each animal, always at the same times each day and coincident with the dose of even *n*-alkanes, have been suggested to reduce variation within and between days. A CRC eliminates the error in the daily dose and reduces the work involved in the procedure, while it produces a steady release of the dosed *n*-alkanes during the experimental period. Additionally, because both the  $C_{32}$  and  $C_{36}$ -alkanes are dosed simultaneously by the CRC,  $DMI_H$  and DMD are estimated with higher accuracy; however, the constant release rate of the even *n*-alkanes in the capsule dosed should be confirmed in practice for an accurate  $DMI_H$  to be estimated from the procedure.

Laboratory errors have been reported to be less important than sampling errors, hence, great advantages have been mentioned from improving pasture sampling and dosing procedures. The accuracy of the average values of  $DMI_H$  and DMD obtained depend upon the number of sampling units used.

## CHAPTER 3

# EFFECT OF GRAZING PREFERENCE ON THE ESTIMATION OF HERBAGE INTAKE WITH N-ALKANES

### 3.1. INTRODUCTION

The *n*-alkane methodology proposed by Mayes et al. (1986) has been used successfully to estimate the individual daily herbage dry matter intake ( $DMI_H$ ) of ruminants (Dove & Mayes, 1991; Dove, 1992; Dillon, 1993; Dove & Mayes, 1996; Hameleers & Mayes, 1998b; Hameleers & Mayes, 1998a) by applying the Equation 3.1

**Equation 3.1:**  $DMI_H = [F_i * D_o] / [(F_j * H_i) - (F_i * H_j)]$

This equation is similar to Equation 2.10, where  $DMI_H$  is the dry matter intake of herbage; H and F are the concentration in the herbage and faeces of the natural alkane 'i' and the dosed alkane 'j' respectively; and  $D_o$  is the daily dose of 'j'.

Many authors mentioned the differences in the concentration of the natural *n*-alkanes between pasture components, parts of the same component, or with plant maturity (Dove & Mayes, 1991; Laredo *et al.*, 1991; Dillon, 1993; Dove & Mayes, 1996; Hameleers & Mayes, 1998b; Genro *et al.*, 2001). A number of studies have indicated that accuracy of the individual estimate depends on the pasture sample being representative of the diet of each animal (Mayes *et al.*, 1986b; Dove & Mayes, 1991; Dove & Mayes, 1996). In fact, *n*-alkanes composition of the diet was proposed as a requirement to estimate intake (Duncan *et al.*, 1999) and it is unlikely that a hand-plucked sample would reflect the herbage consumed by each animal grazing a complex pasture (Dove & Mayes, 1991).

In addition, to accurately estimate  $DMI_H$  the natural *n*-alkane must be present in high concentration in the feed and be recovered in faeces at a similar rate to the dosed *n*-alkane. The lowest discrepancy was obtained using the ratio of the natural  $C_{33}$  and the dosed  $C_{32}$ -alkanes in Equation 3.1 (Mayes et al., 1986b; Vulich et al., 1991; Dillon, 1993; Berry et al., 2000). However, the  $C_{31}$ -alkane has been proposed as a better alternative than the  $C_{33}$ -alkane under particular conditions where pasture components had

low concentration of the C<sub>33</sub>-alkane (Malossini *et al.*, 1996; Hameleers & Mayes, 1998a; Berry *et al.*, 2000), and despite the fact that there is a difference in the faecal recovery of the natural C<sub>31</sub>-alkane and the dosed C<sub>32</sub> alkane (see Chapter 2).

If the concentration of the *n*-alkanes in the herbage can be accurately measured and differences in faecal recovery corrected, the calculated intake should be similar by using the C<sub>33</sub> or C<sub>31</sub> alkanes. This would be true if the ratio C<sub>31</sub>/C<sub>33</sub> in faeces is similar to the ratio in the herbage offered however, if the animal has a preference for one component of the pasture this would be reflected in the faecal ratio C<sub>31</sub>/C<sub>33</sub>, and neither C<sub>31</sub> nor C<sub>33</sub> considered alone paired with the C<sub>32</sub>-alkane would give an accurate estimate. To solve this problem it was suggested to use first the *n*-alkanes method to determine the composition of the diet and its *n*-alkanes concentration (Dove & Moore, 1995), and then to estimate DMI<sub>H</sub> (Mayes & Dove, 2000). This improvement has increased the opportunity for the use of *n*-alkanes in nutritional studies (Mayes & Dove, 2000). However, the difficulties to obtain the reference herbage pools needed would still affect the accuracy of dietary estimated proportions.

A possible alternative would be to consider the total concentration of natural *n*-alkanes in faeces and herbage instead of only one of them. In theory, the total *n*-alkanes recovered in faeces are equal to the total *n*-alkanes ingested with the herbage adjusted by the proportion of them recovered in faeces.

**Equation 3.2:**  $DMI_H * [Hi+ii]_R = FO_H * [Fi+ii]$

**Equation 3.3:**  $[DMI_H / FO_H] = [Fi+ii] / [Hi+ii]_R$

where DMI<sub>H</sub> and FO<sub>H</sub> are the herbage intake and faecal output from the herbage consumed; H and F are the concentration in the herbage and faeces respectively of the natural alkanes 'i' and 'ii'; and R is the faecal recovery of these *n*-alkanes.

As already stated, C<sub>33</sub> and C<sub>31</sub> have been used to estimate intake because they are present in most of the pasture components. Additionally, their faecal recovery has been measured and is known to be close to the recovery of C<sub>32</sub>, although not similar for C<sub>31</sub> (see Table 2.3 in Chapter 2). Considering Equation 3.3, the increment in the concentration of these two natural *n*-alkanes in the faeces relative to the herbage sample would be determined by the digestibility of the diet ingested, as it affects faecal output. Thus, if a grazing animal selects a diet with a higher digestibility than the average of the pasture (for instance, by consuming more leaves), the concentration of both odd and even alkanes in faeces would change. However, a change in the digestibility of the diet caused by the

selection of different plant components would generate a non proportional change in faeces of the main two *n*-alkanes of the herbage sample (e.g. C<sub>31</sub> and C<sub>33</sub>).

It is known that the *n*-alkanes compositions of different plant components are different (see Chapter 2). Then a change in the diet of the animal, and consequently in the *n*-alkanes composition, would be reflected in the *n*-alkanes composition of the faeces, principally affecting the concentration of those *n*-alkanes that are present in higher concentration (e.g. C<sub>31</sub> and C<sub>33</sub>). Thus, a change in the composition of the diet would be indicated by a change in the ratio C<sub>31</sub>/C<sub>33</sub> in faeces, that would be different to the mean ratio of the pasture grazed. The different increase in the faeces of one *n*-alkane relative to the other would indicate the preference of the animal for one particular component or plant-part within the components available in the pasture or a limitation to increase the ingestion of the more desirable components of the pasture. As a result, if in Equation 3.1 only one of the main *n*-alkanes is considered (e.g. C<sub>33</sub>) when there was a change in the diet that was reflected mainly in the other (e.g. C<sub>31</sub>), the DMI<sub>H</sub> estimated would not be accurate as the change in the diet of the animal is not completely reflected by the *n*-alkane being considered in the calculation.

When the sum of these two *n*-alkanes is used, changes in the faecal ratio C<sub>31</sub>/C<sub>33</sub> relative to the pasture ratio are considered as in Equation 3.4 and 3.5 and the accuracy of the estimate of DMI<sub>H</sub> is then improved.

**Equation 3.4:**  $[F_{i+ii}] = [Ratio_F * F_{ii}] + [F_i / Ratio_F]$

**Equation 3.5:**  $[H_{i+ii}] = [Ratio_H * H_{ii}] + [H_i / Ratio_H]$

where F and H are faecal and herbage concentrations of the natural *n*-alkanes; 'i' is the C<sub>31</sub>-alkane; 'ii' is the C<sub>33</sub>-alkane; 'i+ii' is the sum C<sub>31</sub>+C<sub>33</sub>-alkanes; and the Ratio is C<sub>31</sub>/C<sub>33</sub> in H and F.

The objective of this study is to compare the DMI<sub>H</sub> estimated by using the pair [C<sub>31</sub>+C<sub>33</sub>]-C<sub>32</sub> and the usual pairs C<sub>31</sub>-C<sub>32</sub> or C<sub>33</sub>-C<sub>32</sub>, all adjusted by differences in recovery rate, with data from the literature. In addition, to analyse the concentration of *n*-alkanes in faeces of grazing dairy cows in relation to the *n*-alkanes concentration of the pasture being grazed, and the difference between the DMI<sub>H</sub> estimated by using the different pairs previously mentioned.

### 3.2. MATERIALS AND METHODS

The validity of the use of the pair  $[C_{31}+C_{33}]$ – $C_{32}$  instead of the usual pairs  $C_{31}$ – $C_{32}$  or  $C_{33}$ – $C_{32}$  was tested using data of Mayes et al. (1986), Dillon (1993) and Berry et al. (2000). The  $DMI_H$  was estimated using Equation 3.1. To consider the two natural *n*-alkanes simultaneously the concentration of each of them in the herbage is required to be adjusted for their different faecal recovery in relation with the dosed  $C_{32}$ . Both the *n*-alkanes concentration in faeces and the recovery rates of these alkanes in faeces of individual animals were used to estimate the average faecal concentration and intake with data of Mayes et al. (1986), as the full data set was available. The mean *n*-alkanes concentration in faeces and mean faecal recovery rates from a group of animals were used to estimate  $DMI_H$  from Dillon (1993) and Berry et al. (2000). The discrepancy between the estimated and the actual intake for each set of data was calculated as  $[(Actual-Estimated)/Actual]$ . Additionally, the herbage and faecal ratio  $C_{31}/C_{33}$  and the relation ratio  $C_{31}/C_{33}$  in herbage (adjusted for recovery differences) to ratio  $C_{31}/C_{33}$  in faeces for each animal ( $RelH/F$ ) were also estimated for each data set. If the ratio  $C_{31}/C_{33}$  in both herbage and faeces is similar ( $RelH/F=1$ ), the recovery correction compensates the concentration of the *n*-alkanes in faeces and intakes calculated from different *n*-alkanes should be similar.

The same procedure was used to estimate  $DMI_H$  of dairy cows grazing freely on temperate mixed pastures in the two field experiments reported later in this thesis (Chapter 6 and 8). The first study was a long term ‘system’ comparison, which combined three different genotypes of Holstein – Friesian dairy cows farmed at different feeding levels (see Chapter 6 for details, only data from pasture and faeces collected in September 2002 were utilised in the present Chapter). The second experiment was a short-term ‘component’ study that compared the same genotypes grazing swards with different structure (see Chapter 8, data from September and November 2004). The main components of the pasture grazed in both studies were *Lolium perenne* L. and *Trifolium repens* L., with a content of the latter below 20%, and only the early lactation period was analysed. In both experiments, each cow was dosed orally twice a day at each milking with pellets containing 347 mg of the  $C_{32}$ -alkanes for a 10-day period. Faecal samples were taken per rectum from each cow before each milking during the last five days of the dosing period and bulked at the end of the period ( $n=205$  cows for the ‘system’ study and  $n=48$  cows for the ‘component’ study). In addition, pasture samples were taken from the grazed stratum of each paddock grazed during the faecal collection period. Individual faecal and pasture samples were bulked on a weekly basis and analysed for the natural  $C_{31}$  and  $C_{33}$ -alkanes and the dosed  $C_{32}$ -alkane (Mayes et al., 1986b), and then  $DMI_H$

estimated for each cow as described above, utilising the faecal recoveries estimated by Dillon (1993)(see Chapter 2).

On one hand, data from faeces of individual cows from the NZ70 genotype (n=30), half managed at the lowest and half at the highest feed allowance within the range of feeding level utilised in this experiment ( $\blacktriangle$  &  $\triangle$  in Figure 3.1b respectively), were compared in the ‘system’ trial. On the other, data from individual cows of three different genotypes (n=48), half grazing a short sward and the other half a tall sward ( $\blacktriangle$  and  $\triangle$  in Figure 3.2a &  $\bullet$  and  $\circ$  in Figure 3.2b respectively) were compared in the ‘component’ trial. The ratio between the individual faecal concentration of the  $C_{31}$  and  $C_{33}$ -alkanes was calculated for each treatment in both ‘system’ and ‘component’ studies and the means between treatments compared for each experiment. In addition, the relationship between  $C_{31}$  and  $C_{33}$  for the cows in each treatment (one or two dates in the ‘system’ or ‘component’ trial respectively, and in both during the early lactation period), were investigated by fitting different linear models and comparing the intercept and the slope of these models between the lowest and highest feed allowance in the ‘system’ study and between the short and tall swards and dates the cows were sampled in the ‘component’ study, by using the statistical procedures of SAS (SAS, 2002).

Additionally, the faecal concentration of odd and even chain alkanes, the ratio  $C_{31}/C_{33}$  and the RelH/F were calculated with data from the ‘component’ study. The  $DMI_H$  of individual cows was estimated by using the pairs  $[C_{31}+C_{33}]-C_{32}$ ,  $C_{31}-C_{32}$  and  $C_{33}-C_{32}$  and a correction for differences in recovery rate between the odd and even chain alkanes used. Intake was also estimated by using the pairs  $C_{31}-C_{32}$  and  $C_{33}-C_{32}$  without any adjustment for recovery differences. Cows were grouped by their RelH/F being similar to, lower or higher than 1 and the average and standard deviation for the mean faecal concentration of natural and dosed *n*-alkanes, faecal ratio  $C_{31}/C_{33}$ , RelH/F and  $DMI_H$  were estimated for each group of cows.

### 3.3. RESULTS

#### 3.3.1. Accuracy of the ratio $C_{31}+C_{33}/C_{32}$ adjusted by their different faecal recovery to estimate herbage intake

The average discrepancies between measured intakes (Mayes et al., 1986b; Dillon, 1993; Berry et al., 2000) and intakes estimated from the same data using the pair  $[C_{31}+C_{33}]-C_{32}$  adjusted by faecal recovery were small and within a range similar to those obtained using the non-adjusted  $C_{33}$ -alkane alone (Table 3.1).

**Table 3.1: Mean faecal concentration of natural and dosed *n*-alkanes, faecal ratios  $C_{31}/C_{33}$ , and herbage intakes estimated using the sum of the natural  $C_{31}+C_{33}$ -alkanes adjusted by recovery rates from different experiments.**

Reference	Feed	Faecal concentration & ratios					DMI <sub>H</sub>		
		C <sub>32</sub>	C <sub>31</sub> +C <sub>33</sub>	RC <sub>31</sub> /C <sub>33</sub>	Rel H/F	Actual	C <sub>31</sub> +C <sub>33</sub>	DIS (%)	
Mayes <i>et al.</i> , 1986	L G	68.1	79.03	1.13	0.99	0.688	0.682	0.84	
Mayes <i>et al.</i> , 1986	L G+C	75.9	63.15	1.15	1.02	0.470	0.475	-0.94	
Dillon, 1993	DC G	31.9	90.7	1.56	1.03	14.20	14.54	-2.4	
Dillon, 1993	DC G	32.2	89.7	1.56	1.05	14.94	15.27	-2.2	
Dillon, 1993	DC G	18.3	53.7	1.00	0.99	14.20	14.45	-1.8	
Dillon, 1993	DC G	19.4	54.6	0.99	0.99	14.00	13.99	0.1	
Dillon, 1993	DC G	25.8	57.3	1.09	0.98	12.03	11.91	1.0	
Dillon, 1993	DC G	22.0	57.5	1.08	0.96	14.50	14.46	0.3	
Berry <i>et al.</i> , 2000	DC H+C	5.9	20.1	2.46	0.98	10.35	9.78	5.5	
Berry <i>et al.</i> , 2000	DC H	6.6	33.9	2.46	1.01	15.29	15.59	-1.9	
Berry <i>et al.</i> , 2000	DC H&H+C	6.3	27.0	2.46	1.00	12.70	12.76	-0.5	

L: lambs, DC: dairy cows; G: grass; G+C: G plus concentrate; H: mix of herbage; H+C: H plus concentrate; DMI<sub>H</sub>: herbage dry matter intake. Faecal concentration C<sub>32</sub>; C<sub>31</sub>+C<sub>33</sub> (mg 100 g<sup>-1</sup> DM). RC<sub>31</sub>/C<sub>33</sub>: ratio in faeces. Rel. H/F: relation between Ratio C<sub>31</sub>/C<sub>33</sub> in herbage and faeces. DIS: discrepancies (%).

However, discrepancies were not always of the same magnitude and direction. The average ratios  $C_{31}/C_{33}$  in faeces were different between experiments (Table 3.1) but in all similar to the ratio  $C_{31}/C_{33}$  measured in the herbage offered (RelH/F = 1).

### 3.3.2. Change in the faecal concentration of the natural *n*-alkanes $C_{31}$ and $C_{33}$ and the ratio $C_{31}/C_{33}$ . Differences between estimates based on different *n*-alkanes

As expected, there was a strong linear relationship ( $y = 6.39 + 0.59 x$ ;  $r^2 = 0.88$ ;  $n = 205$ ; where 'y' is the concentration of  $C_{33}$  and 'x'  $C_{31}$ ) between the faecal concentration of  $C_{31}$  and  $C_{33}$ -alkanes in cows from the different systems in the 'system' study (Figure 3.1a), except for the system farmed at the highest stocking rate and lowest feeding level ( $y = 8.29 + 0.35 x$ ;  $r^2 = 0.57$ ;  $n = 15$ ) (▲ in Figure 3.1b).

The ratio  $C_{31}/C_{33}$  in the herbage offered to the two treatments of the 'system' study were slightly different, with a mean value of 1.41 and 1.58 for the lowest and highest feed allowance. In addition, the mean faecal ratio  $C_{31}/C_{33}$  from cows grazing these treatments was also different to the mean ratio measured in the pasture they grazed. For instance, the mean faecal ratio  $C_{31}/C_{33}$  of cows that grazed the lowest feed allowance (highest stocking rate) was 1.60 and higher than the pasture grazed (▲ and solid line in Figure 3.1b), and vice versa for cows of the same genotype at the highest feed allowance (mean ratio: 1.43).

Differences between the mean faecal ratios  $C_{31}/C_{33}$  between treatments were also observed; however, at the lowest feed allowance the ratio  $C_{31}/C_{33}$  was higher than at the

highest feed allowance, which was opposite to what was previously reported for the ratio  $C_{31}/C_{33}$  in the pasture grazed by each herd. The higher variability of the faecal ratio  $C_{31}/C_{33}$  for cows that grazed at the lowest feeding allowance was determined by one cow with a much lower value than the mean for the herd; therefore, if this cow was not considered the variation between cows would have been similar (CV%: 2.0). Differences for the linear relationship between the faecal  $C_{31}$  and  $C_{33}$ -alkanes for cows grazing different treatments were significant ( $P < 0.001$ ), with the linear relationship showing different intercept but similar slope between treatments (Figure 3.1b).

**Figure 3.1: Concentration of  $C_{31}$  and  $C_{33}$ -alkanes in faeces from all cows utilised in the experiment (a) and those from similar genotype managed at contrasting feeding allowance (high or low) (b). Data from 'System' study.**

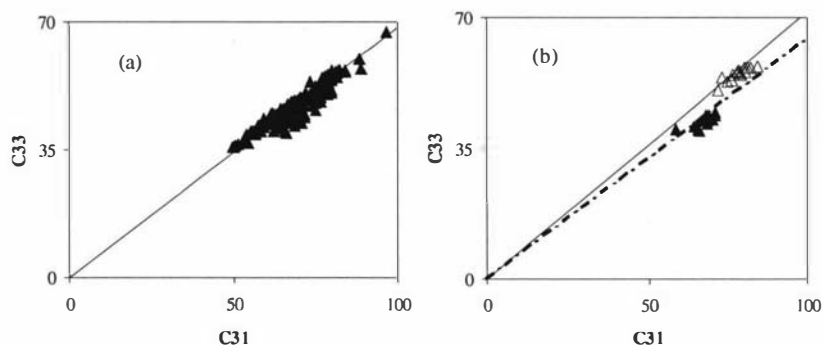


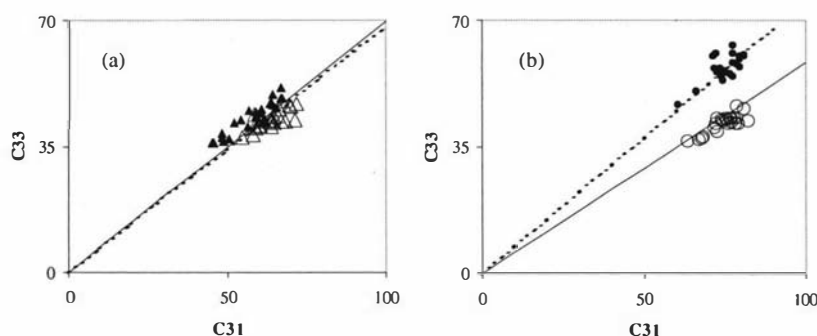
Figure (a): n-alkanes ( $\text{mg } 100\text{g}^{-1} \text{ DM}$ ) in faeces of all cows in the system study. The solid line indicates the mean ratio  $C_{31}/C_{33}$  of the herbage grazed (mean of 11 systems). In Figure (b): n-alkanes ( $\text{mg } 100\text{g}^{-1} \text{ DM}$ ) in faeces of cows managed at the lowest ( $\blacktriangle$ ) and highest ( $\triangle$ ) feed allowance. The mean ratio  $C_{31}/C_{33}$  of the herbage of pastures grazed at the lowest and highest feed allowance (solid line and dotted line respectively) are indicated.

In the 'component' study, the ratio  $C_{31}/C_{33}$  in the herbage offered to the short sward was similar between September and November (1.50 and 1.46 respectively) while for the tall sward was lower in September than in November (1.35 and 1.74 respectively). This suggests a positive effect of management on pasture characteristics. In addition, similarly to what was previously observed, the mean faecal ratio  $C_{31}/C_{33}$  from cows grazing these treatments were different to the mean ratio  $C_{31}/C_{33}$  of the pasture they grazed ( $P < 0.05$ ). The mean faecal ratio of cows in both short and tall swards was below and above the ratio  $C_{31}/C_{33}$  in the pasture during September and November respectively (Figures 3.2a and 3.2b), with a higher variability between these cows within each treatment than that observed in the system study (CV%: 6.5; mean of short and tall swards and both dates).

The linear relationships between the faecal concentrations of  $C_{31}$  and  $C_{33}$  of cows grazing the short and tall swards were not significantly different (considering data from September and November together); however the differences observed for cows grazing the same sward condition on different dates were significantly different ( $P < 0.001$ ).

The linear relationship measured for the faecal concentrations of  $C_{31}$  and  $C_{33}$ -alkanes calculated for all the data in the 'component' study was also estimated ( $y = 15.29 + 0.45x$ ;  $r^2 = 0.33$ ;  $n = 96$ ; where 'y' is the concentration of  $C_{33}$  and 'x'  $C_{31}$ ), the fit of this equation was poor compared with that obtained from the whole dataset for the 'system' study. This probably occurred as a result of the wider range in pasture characteristics that were measured in the 'component' trial (as indicated by the large difference for the pasture ratio  $C_{31}/C_{33}$  between the short and the tall swards and also for the tall swards between dates), compared to the lowest and highest feed allowance in the system study. This is a possible indication that the cows in the 'component' study had a higher opportunity to graze selectively than in the 'system' study.

**Figure 3.2: Concentration of the  $C_{31}$  and  $C_{33}$ -alkanes in faeces of individual cows grazing short (a) or tall (b) swards in September and November 2004. Data from 'Component' study.**



The concentration of *n*-alkanes ( $\text{mg } 100\text{g}^{-1}\text{DM}$ ) in faeces from each cow grazing the 'short' (a) or 'tall' (b) pasture was plotted. In each figure the solid and dotted lines indicate the mean ratio  $C_{31}/C_{33}$  of the herbage grazed. Data from September ( $\blacktriangle$ ) and November ( $\triangle$ ) are presented in both figures. Mean ratio  $C_{31}/C_{33}$  in herbage for September: 1.50 and 1.35 for short and tall swards, in November: 1.46 and 1.74 for short and tall swards. Mean ratio  $C_{31}/C_{33}$  in faeces for September: 1.34 and 1.32 for short and tall swards, in November: 1.51 and 1.80 for short and tall swards.

Considering groups with similar  $\text{RelH/F}$ , the variation ( $\text{CV}\%$ ) in the individual faecal ratio  $C_{31}/C_{33}$  within each group was higher during November than in September and in the tall than in the short sward (Figure 3.2 and Table 3.2). Cows with  $\text{RelH/F}$  equal to 1 had similar  $\text{DMI}_H$  calculated by using either the pair  $[C_{31}+C_{33}]-C_{32}$  or each of the pairs  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$  adjusted by the mean recovery rate in faeces from Dillon's work. Differences between estimates appeared when the faecal ratio  $C_{31}/C_{33}$  was different from the pasture ratio  $C_{31}/C_{33}$  ( $\text{RelH/F} < \text{ or } > 1$ ). The group with  $\text{RelH/F} > 1$  (ratio  $C_{31}/C_{33}$  in herbage higher than in faeces), had the lowest and highest intakes estimated with the adjusted  $C_{31}$  and  $C_{33}$ -alkane respectively, and vice versa for the group with  $\text{RelH/F} < 1$  (Table 3.2). The estimate obtained using the pair  $[C_{31}+C_{33}]-C_{32}$  was between the values estimated using each adjusted natural *n*-alkane alone however not equal to the average of their estimates. The  $\text{DMI}_H$  estimated from the pairs  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$  non-adjusted for

the differences in faecal recovery rate were lower than those obtained when the same pairs were used but differences in recovery not corrected.

### 3.4. DISCUSSION

One of the greatest advantages of the *n*-alkanes methodology is that it can provide an independent estimate of dry matter intake and digestibility for each individual animal in a group grazing freely on the same pasture.

The use of the adjusted pair  $[C_{31}+C_{33}]-C_{32}$  in Equation 3.1 to estimate  $DMI_H$  using data from Mayes et al. (1986), Dillon (1993) and Berry et al. (2000) indicates that the procedure proposed is accurate, with estimates being similar to the ‘actual’ values measured in these works (Table 3.1). Dove and Mayes (1991) pointed out that the incomplete recovery would not affect the estimate of  $DMI_H$ , if both of the *n*-alkanes used have similar faecal recovery rates; however, the differences between ‘actual’ and estimated  $DMI_H$  in the previous studies obtained by using the pair  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$  might be slightly improved if the small differences in recovery between the natural and dosed *n*-alkanes were corrected.

**Table 3.2: Faecal concentration of natural and dosed *n*-alkanes, faecal ratios  $C_{31}/C_{33}$  and herbage intakes of dairy cows. Data from ‘Component’ study.**

		Faecal concentration and ratios					$DMI_H$				
		$C_{32}$	$C_{31}+C_{33}$	R	Rel	$C_{31}+C_{33}$	$AdC_{33}$	$AdC_{31}$	$C_{33}$	$C_{31}$	
Rel	N			$C_{31}/C_{33}$	H/F	(a)	(b)	(c)	(d)	(e)	
September											
~1	9	Mean	21.6	125.9	1.35	0.99	17.02	17.02	17.03	16.45	16.17
~1		SD	3.17	13.9	0.03	0.005	4.08	4.08	4.09	3.92	3.85
>1	28	Mean	21.26	108.3	1.30	1.11	14.53	15.60	13.84	15.08	13.16
>1		SD	3.01	18.16	0.06	0.091	3.15	3.23	3.21	3.11	3.01
<1	11	Mean	21.27	127.7	1.39	0.96	17.39	16.91	17.77	16.35	16.87
<1		SD	3.49	11.20	0.04	0.017	3.53	3.51	3.57	3.53	3.36
November											
~1	4	Mean	18.4	105.97	1.57	1.00	17.07	17.07	17.07	16.49	16.21
~1		SD	2.53	4.12	0.11	0.005	2.85	2.81	2.87	2.72	2.73
>1	4	Mean	20.34	108.24	1.49	1.04	16.61	17.06	16.32	16.48	15.49
>1		SD	3.92	8.40	0.16	0.058	5.70	5.52	5.85	5.30	5.51
<1	40	Mean	19.86	112.2	1.68	0.94	17.30	16.51	17.82	15.97	16.92
<1		SD	2.14	8.41	0.16	0.039	3.16	3.09	3.21	2.98	3.03

$DMI_H$ : herbage dry matter intake. Intake was estimated using  $C_{31}$ ,  $C_{33}$  and  $C_{31}+C_{33}$ , all adjusted by recovery rates (Dillon, 1993) in (a), (b) and (c). Non-adjusted estimates are presented in (d) and (e). ‘n’: number of animals in each group. Faecal concentration  $C_{32}$ ;  $C_{31}+C_{33}$  (mg 100 g<sup>-1</sup>DM);  $RC_{31}/C_{33}$ : ratio *n*-alkanes in faeces; Rel. H/F: relation Ratio  $C_{31}/C_{33}$  in herbage and faeces; SD: standard deviation.

It could be argued that the use of an incorrect recovery rate would increase the error associated with the estimation of  $DMI_H$ . Despite this problem, it is apparent that in complex situations where the cow selects a diet different in the content of *n*-alkanes than the mean pasture offered, an improved estimation of  $DMI_H$  can be obtained with the pair

[C<sub>31</sub>+C<sub>33</sub>]-C<sub>32</sub> just because the relative change in concentration of the two main odd *n*-alkanes in the diet compared with the pasture have been considered. The correction for differences in faecal recovery between the alkanes used would contribute to reduce the difference between 'actual' and 'estimated' DMI<sub>H</sub>.

Higher and less variable faecal recovery rates have been reported for sheep than for cattle (Dove & Mayes, 1991), which explains the small differences obtained when Mayes's data was used (Table 3.1). Additionally, recovery is known to vary between animals (Mayes et al., 1986b; Dillon, 1993) and between experimental conditions (Dillon, 1993; Penning, 2004); thus, in the adjusted estimates, discrepancies will depend on using accurate recovery values. This analysis reinforces the original argument of Mayes et al. (1986) that errors associated with incomplete recoveries cancel out in the equation, with natural and dosed *n*-alkanes having similar faecal recoveries and within this context, the *n*-alkanes method has been extensively tested to estimate intake in ruminants (Mayes et al., 1986b; Vulich et al., 1991; Dillon, 1993; Hameleers & Mayes, 1998a; Unal & Garnsworthy, 1999; Berry et al., 2000; Garcia et al., 2000).

Considering the level of accuracy obtained between 'actual' and 'estimated' DMI<sub>H</sub> in Mayes et al. (1986), Dillon (1993) and Berry et al. (2000), it is evident that there was no reason for these authors to consider to use the pair [C<sub>31</sub>+C<sub>33</sub>]-C<sub>32</sub> and any adjustment for recovery. However, in these indoors experiments the capacity of the animal to select was constrained because feed was restricted and no refusals were collected, in addition, either herbage composition or presentation, in cases when cows were fed ad-libitum, represented a constraint to animal selectivity. The similar ratio C<sub>31</sub>/C<sub>33</sub> between herbage and faeces in the mentioned experiments, as indicated by the value of RelH/F equal to 1 (Table 3.1), suggests similar *n*-alkanes composition in faeces and diet and no effect of selectivity, then the composition of the diet should be considered similar to that of the herbage offered.

The main condition for accuracy in the previous indoor works was the similar ratio in herbage and faeces of the main natural *n*-alkanes C<sub>31</sub> and C<sub>33</sub>. In contrast, large differences between animals should be expected due to selectivity in a grazing situation, with a hand-plucked sample not representing the diet of each cow of a herd grazing the same pasture. This is particularly evident from the analysis of the concentration of both C<sub>33</sub> and C<sub>31</sub> in faeces from cows in the 'system' study (Figure 3.1b). The ratio C<sub>31</sub>/C<sub>33</sub> in faeces of cows from the same genotype farmed at low or high feeding level (high or low stocking rate) were on average higher and lower respectively than the adjusted ratio in the average pasture offered. Since the ratio in the pasture offered to each system was similar, the differences observed indicate that cows at the high feeding level had a higher concentration of both natural *n*-alkanes, probably as a result of an increased diet

digestibility. They increased the concentration of  $C_{33}$  relative to  $C_{31}$ , reducing the ratio  $C_{31}/C_{33}$  in the faeces in relation to the ratio in the pasture grazed, which could be explained by higher leaf content in the herbage consumed because animal selection (see Table 2.2).

The same explanation is valid for the ‘component’ study (Figures 3.2a and 3.2b), where the concentration of  $C_{31}$  and  $C_{33}$ -alkanes in faeces was slightly different between cows and also aligned along the mean value of the ratio  $C_{31}/C_{33}$  in the herbage. However, it is interesting to note that whereas the ratio  $C_{31}/C_{33}$  in the short sward was similar between September and November (Figure 3.2a), in the tall sward this ratio was higher during November indicating a greater change in the composition of *n*-alkanes in the herbage offered (Figure 3.2b); in addition, it is apparent that the cows preferred herbage components or plant-parts with higher  $C_{33}$ -alkane content in September, but they harvested herbage with higher  $C_{31}$ -alkane content in November as indicated by the individual faecal ratios being greater or lower than the herbage ratio of the pasture grazed (Figures 3.2a and 3.2b) probably because there was a change in the proportion of herbage components, and then in the content of *n*-alkanes, in the herbage available between dates. These results confirm the fact that differences in the concentration of *n*-alkanes in the diet compared with the pasture being grazed frequently occur; consequently the  $DMI_H$  obtained would be biased accordingly with the pair of odd and even *n*-alkanes selected in Equation 2.1 whereas an improved estimate would be achieved by using the pair  $[C_{31}+C_{33}]-C_{32}$ .

These results indicate that more accurate estimates of the actual  $DMI_H$  of grazing cows selecting species or plant-parts with different compositions of the two main  $C_{31}$  and  $C_{33}$ -alkanes present in the pasture can be obtained by considering both  $C_{31}$  and  $C_{33}$  simultaneously by using the pair  $[C_{31}+C_{33}]-C_{32}$ , as a result of the reduction in the bias that would occur by considering either the pair of  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$  when the ratio  $C_{31}/C_{33}$  in faeces differs from the ratio of same *n*-alkanes in the mean pasture sample. This indicates that the concentration of *n*-alkanes in the reference herbage sample utilised in Equation 3.1 did not represent the diet of the animals grazing this pasture, which is the main requirement for accuracy with the method. This might have occurred especially when cows grazed the long swards in the ‘component’ study, where the possibility to select would be higher, despite the change in pasture composition as indicated by the increased ratio  $C_{31}/C_{33}$ . In this case a large bias would be expected from using either of the single odd alkanes paired with the even dosed.

To improve this further the solution proposed is to estimate the composition of the diet for each animal (Dove & Moore, 1995) and consequently, the dietary proportions of the *n*-alkanes from which to calculate intake (Mayes & Dove, 2000). However, the constraint

that applied to the hand-plucked pasture sample would also apply to the reference herbage pools needed to determine diet composition.

The alternative of using the pair  $[C_{31}+C_{33}]-C_{32}$  adjusted by differences in faecal recovery rate has the benefit of considering the relative changes of the two main *n*-alkanes of the diet relative to their concentration in the average pasture, and compensating the changes occurring due to differences in their faecal recovery. The intakes estimated through this procedure were accurate, as mentioned, using data from Mayes et al. (1986), Dillon (1993) and Berry et al. (2000) (Table 3.1). The similar intakes obtained by using the adjusted  $[C_{31}+C_{33}]-C_{32}$ , the pair  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$  alone for cows with  $RelH/F=1$  in the 'component' study indicate that when the ratio  $C_{31}/C_{33}$  in herbage and faeces is similar, their effects on the intake value cancel out in Equation 3.1. This means that the digestibility of the diet should be similar to the digestibility of the pasture collected, and the concentration of each natural *n*-alkane in faeces determined by the intake of pasture components. Differences between the estimates obtained using the adjusted  $[C_{31}+C_{33}]-C_{32}$  and the non-adjusted  $C_{33}$  or  $C_{31}$  are mostly explained by the recovery rates considered (Table 3.2).

The intakes estimated for cows with  $RelH/F < \text{or} > 1$  using the pair  $[C_{31}+C_{33}]-C_{32}$  adjusted by the differences in the recovery rate in faeces, take into account the relative change in the faeces of the two main *n*-alkanes present in the pasture relative to the mean pasture being grazed, and then the difference in the relative change of these *n*-alkanes in the diet of each cow as a result of selectivity. It is clear that by adopting this approach the bias that could occur in the estimation of  $DMI_H$  due to selectivity of species or plant parts from the pasture differing mainly in these two *n*-alkanes is reduced. The  $DMI_H$  obtained with the pair  $[C_{31}+C_{33}]-C_{32}$  adjusted by the differences in the recovery rate in faeces would lie between the values of  $DMI_H$  obtained with the pairs  $C_{31}-C_{32}$  and  $C_{33}-C_{32}$ , but not necessarily is the mean of these values as depends on the relative change in the two odd alkanes involved (Table 3.2).

### 3.5. CONCLUSION

The use of the pair  $[C_{31}+C_{33}]-C_{32}$  adjusted by differences in the recovery rate between odd and even *n*-alkanes used in faeces improves the accuracy of the  $DMI_H$  estimated with the *n*-alkane method, particularly in complex grazing situations where selectivity for different pasture components or plant-parts occur, and the *n*-alkanes concentration of the diet of different cows in a group differs from that in the pasture being grazed. However, the main limitation is the impossibility of obtaining a sample of the herbage consumed

for each individual cow. One step forward in order to improve results would be to validate the proposed procedure under controlled feeding conditions.

## CHAPTER 4

# ESTIMATION OF HERBAGE AND SUPPLEMENT INTAKES OF INDIVIDUAL COWS GRAZING IN A GROUP CONTEXT: TESTING OF FIELD PROCEDURES

### 4.1. INTRODUCTION

It was concluded in Chapter 2 that a combination of methods that use the n-alkanes (Mayes et al., 1986b) and the stable carbon isotope discrimination techniques (Jones et al., 1979) can be used successfully to estimate herbage dry matter intake ( $DMI_H$ ) of grazing dairy cows simultaneously fed forage and other supplements like pasture and maize silage (Hameleers & Mayes, 1998b; Garcia *et al.*, 2000). However, the preference of the cow for some pasture components can affect the accuracy of  $DMI_H$  from n-alkanes (see Chapter 3). In addition, if the preferred species is a  $C^4$  summer component like *Paspalum dilatatum* poir. ( $PA_{DI}$ ) with low n-alkanes content and similar stable carbon isotope discrimination signature to maize silage, the accuracy of the estimation of maize silage consumption could be affected.

Two main conditions have been mentioned for an odd-chain n-alkane to be utilised to estimate accurately  $DMI_H$ : (a) preferable high concentration in the herbage; and (b) similar recovery rate in faeces to the even-chain dosed n-alkane (Dove & Mayes, 1991; Dove, 1992; Dove & Mayes, 1996). For these reasons, the pair  $C_{33}$ – $C_{32}$  alkanes has been accurately utilised with dairy cows under different experimental conditions (Dillon, 1993; Unal & Garnsworthy, 1999), however, the pair  $C_{31}$ – $C_{32}$  alkanes also provided the lowest discrepancy in particular grazing situations (Hameleers & Mayes, 1998a).

Both  $C_{31}$  and  $C_{33}$ -alkanes have contiguous chain length with the dosed  $C_{32}$ -alkane thus are suitable to be used as internal markers (Malossini et al., 1996). The use of  $C_{33}$  instead of  $C_{31}$ -alkane is supported by the smaller difference in recovery rate with the dosed  $C_{32}$ -alkane, and despite the fact that its concentration in the herbage is usually lower and more variable than that of the  $C_{31}$ -alkane in temperate pastures (Malossini et al., 1996),

although higher than the value of 5 mg/100 g DM considered as the minimum threshold for using these markers (Laredo *et al.*, 1991).

The n-alkane concentration and pattern in the pasture are known to change with the state of growth and composition of the pasture (Laredo *et al.*, 1991; Dillon, 1993; Hameleers & Mayes, 1998b; Genro *et al.*, 2001), with the content of C<sub>31</sub>-alkane being very low in some preferred species like *Trifolium repens* L. (white clover) while in others such as PADI it is just above the suggested limit. The convenience of using one or the other n-alkane to estimate DMI<sub>H</sub> and their recovery rate in the faeces for diet digestibility (DMD) (Mayes *et al.*, 1986b; Dove & Mayes, 1991; Vulich *et al.*, 1991; Dove, 1992; Dillon, 1993; Hameleers & Mayes, 1998a; Unal & Garnsworthy, 1999; Berry *et al.*, 2000) has been tested in short indoor trials by using relatively small numbers of cows. The actual amount of herbage eaten was measured and compared with the DMI<sub>H</sub> estimated from n-alkanes for each individual animal. Cows in these trials do not have the opportunity to behave selectively as the diet is maintained constant. Furthermore, there is no report available where the accuracy of the estimate was evaluated either during a long period where changes in the diet of the cows occurred or with animals selecting different feed components with different n-alkanes. It is apparent that when cows select from the pasture components or plant-parts with different content of C<sub>31</sub> and C<sub>33</sub>, the ratio of these two n-alkanes in the diet of the cows measured in their faeces will be different from the mean ratio of the pasture grazed, thus the DMI<sub>H</sub> value estimated would be affected by the n-alkane utilised in the calculation (see Chapter 3). The fact that different authors have obtained improved accuracy by using C<sub>31</sub> or C<sub>33</sub>-alkane depending on which was the main n-alkane in the diet suggests that a careful approach should be considered when deciding which n-alkane to utilise to estimate DMI<sub>H</sub> in order to not bias the value obtained.

Although procedures to discriminate feeds and even pasture components in the diet have been extensively described (Dove, 1992; Mayes & Dove, 2000) and successfully used (Hameleers & Mayes, 1998b; Hameleers & Mayes, 1998a), the appropriate n-alkanes to use in the equation to estimate DMI<sub>H</sub> must be selected with care, particularly in long term studies where the composition of the pasture is expected to change during the experiment, and then the DMI<sub>H</sub> from different dates being affected by the content of n-alkanes in the diet on that date and the pair of n-alkanes used in the calculation. Differences in the pattern of n-alkanes between pasture components and the diet of the animals and the difficulty involved in obtaining a sample which represents better the mean diet have been reported as the main weakness of the procedure for grazing animals, particularly because the pasture sample must represent the species and plant-parts actually eaten. This is not an easy problem to solve considering the selective capacity of grazing animals.

The combined use of  $C_{31}$  and  $C_{33}$  as the natural markers in the ratio  $[C_{31}+C_{33}]-C_{32}$  was indicated as a way to balance the effect of cow selectivity in the estimation of  $DMI_H$  by using n-alkanes as the relative change in the ratio  $C_{31}/C_{33}$  between diet and pasture is considered with this approach (see Chapter 3). In addition, it would make estimates obtained on different dates directly comparable as the same procedure was utilised (same pair of odd – even alkanes).

## 4.2. MATERIALS AND METHODS

The data used in this chapter is derived from the long term ‘system’ experiment described in Chapter 5 which started in 2001 and lasted three complete seasons. This study compared the productivity of three different genotypes of Holstein-Friesian dairy cows in different feeding systems managed at increased level of feed per cow per year. Genotypes were two modern high-merit strains of New Zealand (NZ) or Overseas origin and one low-merit NZ strain representing the genetic used in NZ in the 1970’s (See Appendix IV for details). In this study, 11 non-replicated systems representing real farm conditions with 15 or 20 cows each were managed in different farmlets, with different feeding levels (n=205 cows, see details in Chapter 5). Systems were based on pasture and also fed forage (pasture silage) and supplement (maize silage and grain), particularly during mid and late lactation and the dry periods. In this study the  $DMI_H$  of the cows was measured by utilising a combination of methodologies to estimate the composition of the diet when forage and supplements were also fed to grazing dairy cows. For the purpose of this Chapter the intake of cows from different systems were managed together and differences between genotypes or feeding levels not analysed. The theory involving the procedures utilised to estimate intake were described in Chapter 1, and also expanded in the following sections of the present Chapter. A description of the particulars conditions of the ‘system’ study is presented with detail in Chapter 5.

The n-alkane content and  $\delta^{13}C$  value from the pastures grazed and the faeces collected from individual cows in the systems were analysed on seven dates along two consecutive lactating periods and the dry period between them. The individual intakes of herbage and supplement were estimated from these data.

In this chapter the procedure utilised to estimate herbage and supplement intakes are described. Data from the n-alkane composition and  $\delta^{13}C$  from the herbage and main components in pastures, the forage and the supplements fed, and in the faeces of individual cows were analysed. Herbage intakes were estimated using different pairs of natural odd and even chain n-alkanes, the values obtained compared and the implications of the use of different pairs and accuracy, determined. In addition, both the carryover

effect of  $DMI_H$  and the effect of the presence of  $PA_{DI}$  during the summer period and preference for the cows on the estimate of maize silage intake were discussed. This was expected to help understanding how to improve the discrimination between diet components with similar  $\delta^{13}C$  signature.

#### **4.2.1. Description of the procedure utilised to estimate intakes of herbage, forage and supplement**

Herbage dry matter intakes were measured by the n-alkane procedure in combination with the stable carbon isotope discrimination technique (Chapter 1), to all individual cows in the experiment on seven dates during the study (three intake measurement during the early, mid and late lactation periods of two consecutive lactations in season 2002-03 and 2003-04 and the dry period between them; see Chapter 5 for details). Cows were dosed orally twice daily before each milking during a 10 day period, with a capsule containing  $350 \pm 5$  mg of the even chain  $C_{32}$ -alkane. Pasture and faecal samples were obtained during the last five days of the dosing period, after equilibrium of the dosed n-alkane in the digestive tract was considered attained.

Pasture samples of similar size were collected daily from each paddock by taking manually 60-80 snips within the grazing stratum along two transects. Grazing height was identified by close observation of the cows while grazing. Daily samples representing each of the daily grazing strips (sub-paddocks) grazed during the faecal sampling period were collected and sub-samples prepared for analysis. Sub-samples representing each 'sub-paddock' were kept aside for individual n-alkanes determination. One bulk representing the pasture grazed during the faecal collection period was obtained for each system on each date  $DMI_H$  was measured..

Faecal samples (similar volume) were collected twice daily (at each milking) from each individual cow and immediately frozen. One bulk sample per cow was generated at the end of the five-day sampling period. Herbage, forage, supplement and faecal sub-samples were freeze dried, ground (1 mm) and analysed by gas chromatography (Mayes et al., 1986b) to determine the concentration of odd chain  $C_{27}$ ,  $C_{29}$ ,  $C_{31}$ ,  $C_{33}$  and  $C_{35}$ -alkanes and even chain  $C_{32}$  and  $C_{36}$ -alkanes. Additional herbage and faecal sub-samples were analysed by the stable carbon isotope discrimination when maize silage was fed (Jones et al., 1979). In addition, weekly bulked herbage, forage and supplement sub-samples were analysed for chemical composition and dry matter percentage.

#### 4.2.2. Adjusted formula used to estimate pasture dry matter intake

Intakes were estimated using either the pairs  $C_{31}$ – $C_{32}$  or  $C_{33}$ – $C_{32}$  alkanes (Dillon, 1993; Hameleers & Mayes, 1998a; Unal & Garnsworthy, 1999) or  $[C_{31}+C_{33}]$ – $C_{32}$  adjusted by their recovery rate in faeces (see Chapter 2). The equation proposed by Hameleers and Mayes (1998) adjusted for recovery was used (see Appendix I). As it was not possible to estimate the proportion of each feed by using Dove and Moore (1995) when the supplement had low n-alkane concentrations, maize silage consumption was calculated separately by using the stable carbon isotope discrimination technique (Jones et al., 1979; Garcia et al., 2000) after  $DMI_H$  was estimated. An explanatory diagram describing the complete procedure is in Appendix III.

#### 4.2.3. Correction for differences in recovery rate

All n-alkanes are recovered incompletely in faeces due to limited absorption in the small intestine (Penning, 2004). Nevertheless, recovery rates are high and very similar between  $C_{32}$  and  $C_{33}$ -alkanes, although slightly higher in these two than in the  $C_{31}$ -alkane (Dillon, 1993; Penning, 2004). As a result, when the  $C_{31}$ -alkane is used to estimate intake a correction for differences in recovery rate with the dosed  $C_{32}$ -alkane was indicated as a way to improve accuracy (Berry et al., 2000). In this thesis a correction for recovery was also used when  $C_{33}$  and  $C_{31}+C_{33}$  were utilised to estimate  $DMI_H$  for consistency in the procedure utilised.

The recovery rate of the individual n-alkanes is known to change with the changing condition of different experiments (Penning, 2004), thus, it would be desirable to use recovery rates estimated from similar conditions to those under which they will be utilised. However, the total collection of faeces needed for this was not possible under the grazing conditions of the present work; hence, the values used were those proposed by Dillon (1993), 0.753, 0.767, 0.826, 0.861, 0.838 and 0.882 for  $C_{27}$ ,  $C_{29}$ ,  $C_{31}$ ,  $C_{32}$ ,  $C_{33}$  and  $C_{35}$ -alkanes respectively, which were measured in a set of grazing experiments with supplemented and un-supplemented cows. These recovery rates have been used extensively by different authors (Hameleers & Mayes, 1998a; Penning, 2004).

#### 4.2.4. Discrimination of grazed herbage and grass silage in the diet

The pasture/grass silage ratio in the diet of cows was estimated using the programme named 'Eatwhat' (Dove & Moore, 1995), from the n-alkane contents of the herbage, forage and cows' faeces. To utilise this tool, the composition of n-alkanes in the two feeds affected by recovery rate, and the concentration of the same n-alkanes in cows' faeces is needed. Once the proportions of pasture and grass silage eaten was calculated

for each cow, the intake of each component was estimated by considering the concentration of n-alkanes in the two feeds (Hameleers & Mayes, 1998a).

#### 4.2.5. Herbage and maize silage intakes

The n-alkanes methodology cannot be used to estimate the individual intakes of feed with low concentrations of the n-alkanes like maize (Bianchi & Avato, 1984; Garcia *et al.*, 2000); however, this summer grass ( $C_4$  photosynthetic pathway) has a different concentration of the ratio of natural  $^{12}C$  and  $^{13}C$  isotopes ( $\delta^{13}C$ ) compared to the cool season species of the pasture like *Lolium perenne* L. (ryegrass) and white clover ( $C_3$  photosynthetic pathway). Results from the ratio of stable isotopes in the pasture and faeces are expressed as the difference from the ratio of a number of atoms of  $^{13}C$  to the number of atoms of  $^{12}C$  in a standard ( $\delta^{13}C$ ). Thus, by knowing the  $\delta^{13}C$  in the pasture, the proportion of each  $C^3$  and  $C^4$  species ingested by the animal can be estimated from the  $\delta^{13}C$  of the faeces (Jones *et al.*, 1979).

The technique depends upon the fact that  $\delta^{13}C$  is about  $-28$  and  $-11$  for  $C^3$  and  $C^4$  species respectively, so if the  $\delta^{13}C$  of both components of the pasture are known, the  $\delta^{13}C$  of a  $C^3$ – $C^4$  mixture in the faeces of a cow is in direct proportion to the amount of these components in the pasture or diet (Jones *et al.*, 1979). The discriminated components are expressed in organic matter (OM) basis and must be corrected by ash content and expressed in dry matter (DM) basis. Finally, the intake of each component of the diet is calculated by using a combination of the n-alkanes –  $^{13}C$  methods (Garcia *et al.*, 2000) modified as indicated in Chapter 2.

The procedure is simple and accurate (Garcia *et al.*, 2000) providing  $DMI_H$  is accurately estimated. Nevertheless,  $PA_{DI}$  was a summer component in the pastures grazed by the cows and its consumption must be corrected to accurately calculate maize silage and total intakes. It is not possible to distinguish maize silage from  $PA_{DI}$  in the diet because they have a similar  $\delta^{13}C$  value. One alternative is to use the procedure described by Dove and Moore (1995), which was already utilised in this thesis to estimate the amount of pasture silage consumed (see point 3.2.4 in this Chapter). A second possibility is to estimate the proportion of  $PA_{DI}$  by using the stable carbon isotope discrimination technique (Jones *et al.*, 1979) and comparing the  $\delta^{13}C$  value of the pasture sample on each date with the value from the winter pasture considered as baseline, and assuming nil content of  $C_4$  species in the latter. Thus the difference in the mean  $\delta^{13}C$  between summer and winter pasture would represent the increased proportion of  $PA_{DI}$  during the summer period and the difference in the mean  $\delta^{13}C$  between summer pasture and faeces would represent the average proportion of  $PA_{DI}$  in the diet.

This procedure considers that any increase in the  $\delta^{13}\text{C}$  of the faeces might have occurred due to maize consumption; nevertheless, it could be expected that estimate would be biased if  $\text{PA}_{\text{DI}}$  were selected or rejected by the cows. Thus, although the mean estimate for the group would be accurate, it could be expected that individual differences exist due to selectivity. Faecal samples were collected from 18 un-supplemented cows in December 2002 and 2003 and March 2003-2004 and the  $\delta^{13}\text{C}$  determined.

#### 4.2.6. Additional determinations

Samples of ryegrass, white clover and  $\text{PA}_{\text{DI}}$  including green leaves, stems and petioles were collected from the pastures grazed during spring and summer and analysed for n-alkanes. To investigate further how differences in the proportion of ryegrass and white clover in the herbage affect the n-alkanes composition of the samples, plant material from these species were combined in the laboratory to generate sub-samples with different proportions of grass and clover, ranging from 100% ryegrass and 0% white clover to 0% ryegrass and 100% white clover, with a proportional decrement and increment of each species in the sample respectively (six samples with different composition of these two species) and the n-alkanes composition of each proportional sample was determined.

#### 4.2.7. Data analysis

The n-alkanes content and the value of  $\delta^{13}\text{C}$  from each pasture grazed (individual paddocks) and a faecal bulk sample from each individual cow in the system, were utilised to estimate intakes of fresh herbage, forage and supplement in the 'system' study on each date. These data plus that from individual samples of the main pasture components, forage and supplement fed, were analysed by using the statistical procedures of SAS (SAS, 2002).

The mean concentration and standard deviation of each of the natural n-alkane, the sum  $\text{C}_{31}$  and  $\text{C}_{33}$ -alkanes, the total n-alkanes, the ratio  $\text{C}_{31}/\text{C}_{33}$  -alkanes and the  $\delta^{13}\text{C}$  from samples collected each date were calculated. The difference between the concentration of  $\text{C}_{31}$  and  $\text{C}_{33}$ -alkanes in herbage and faeces, and between dates during the experimental period were analysed as a mixed model with date as a fixed effect, and the covariance between dates (for pasture) or animals (for faeces) was accounted for in the analysis of repeated measures with a compound symmetry covariance structure. Differences in the total concentration of  $\text{C}_{31}$  plus  $\text{C}_{33}$ -alkanes, and the ratio  $\text{C}_{31}/\text{C}_{33}$  -alkanes between dates were also investigated.

The intake values estimated by using the pairs  $C_{31}$ – $C_{32}$ ,  $C_{33}$ – $C_{32}$  and  $[C_{31}+C_{33}]$ – $C_{32}$  were compared through a mixed model with the *n*-alkanes used to calculate intake as a fixed effect and individual cows as replicates. The concentrations of  $\delta^{13}C$  in the pastures and in the faeces of supplemented and un-supplemented cows were compared for December and March using a mixed model with sample type (herbage or faeces) as fixed effect and paddock or cows as replicate.

The linear relationships between the  $C_{31}$  and the  $C_{33}$ -alkanes in the faeces were estimated by regression analysis, considering all the data available and individually for each sampling date; in addition, the linear relationship between the  $DMI_H$  calculated with the pair  $C_{31}$ – $C_{32}$  and  $C_{33}$ – $C_{32}$  was also estimated.

### 4.3. RESULTS

#### 4.3.1. Concentrations of the odd chain *n*-alkanes in pasture and supplements

The concentrations of the  $C_{29}$ ,  $C_{31}$  and  $C_{33}$ -alkanes in the pasture were higher than for the rest of *n*-alkanes measured ( $C_{27}$ ,  $C_{32}$ ,  $C_{35}$  and  $C_{36}$ ) (Table 4.1), and varied across dates ( $P<0.001$ ; Table 4.1 and Figure 4.1 a).

**Table 4.1: Mean *n*-alkanes concentration, ratio  $C_{31}/C_{33}$ -alkanes, and  $\delta^{13}C$  value in different feeds utilised in the experiment ('system' study).**

Forage	n	$C_{27}$	$C_{29}$	$C_{31}$	$C_{33}$	$C_{35}$	$C_{32}$	$C_{36}$	$C_{31}, C_{33}$	Total	$C_{31}/C_{33}$ ratio	$\delta^{13}C$ ‰
Pasture	77	3.4	11.1	19.8	11.1	1.2	1.11	0.4	30.9	48.1	1.9	-29.7
SE		0.19	0.33	0.42	0.25	0.03	0.02	0.01	0.4	0.8	0.08	0.11
Pasture silage	13	5.8	14.7	28.1	8.8	0.9	1.1	0.4	36.9	59.9	3.2	-28.9
SE		0.60	0.73	0.82	0.36	0.04	0.05	0.02	0.86	2.04	0.16	2.55
Maize silage	14	1.1	1.3	1.6	1.1	0.3	0.3	0.4	2.7	6.1	1.6	-12.0
SE		0.21	0.15	0.09	0.05	0.02	0.03	0.02	0.14	0.5	0.06	0.03
Maize	16	1.2	1.3	1.4	0.8	0.2	0.3	0.4	2.2	5.5	1.8	-12.0
SE		0.27	0.15	0.08	0.04	0.01	0.03	0.01	0.12	0.55	0.09	0.56
Maize grain	1	2.3	1.5	0.8	0.3	0.0	0.4	0.4	1.1	5.7	2.9	-11.4
<i>Paspalum</i> spp. <sup>1</sup>	1	2.5	4.2	11.8	4.2	0.2	0.8	0.4	15.9	24.1	2.8	-11.9

The mean and standard error (Se) for each feedstuff were estimated from bulked samples collected at different dates. 'n': number of bulked samples.  $C_{27}$ : heptacosane;  $C_{28}$ : octacosane;  $C_{29}$ : nonacosane;  $C_{31}$ : hentriacontane;  $C_{32}$ : dotriacontane;  $C_{33}$ : tritriacontane;  $\delta^{13}C$ : difference from the ratio of the number of atoms of  $^{13}C$  to the number of  $^{12}C$  in a standard carbonate; <sup>1</sup>*Paspalum dilatatum* pair; SE=  $sd/\sqrt{n}$ .

The concentration of the  $C_{31}$ -alkane was the highest, with  $C_{31}+C_{33}$  alkanes accounting for more than 50% of the total *n*-alkanes in herbage and faeces. As a result of the variation in the concentration of the individual odd *n*-alkane across dates, the total concentration also varied ( $P<0.001$ ; Table 4.2 and Figure 4.1a), the highest value being in December (both seasons), with the lowest percentage of  $C_{31}+C_{33}$  alkanes in the herbage in relation with the total content of *n*-alkanes. The content of the odd chain  $C_{29}$  and  $C_{31}$ -alkanes in the

herbage followed a similar pattern with peaks in December 2002 and 2003 and the lowest concentration during the winter. In contrast, the concentration of C<sub>33</sub>-alkane was low in December but showed a peak in June (Figure 4.1a). The relative change of the C<sub>31</sub> and C<sub>33</sub>-alkanes in the herbage is well represented by the change in the ratio of C<sub>31</sub>/C<sub>33</sub> during the period (Table 4.1 and Figure 4.1c); the lowest observed in June then increased during September to achieve the highest value in December, and then declined again until the last observation was made in March 2004 (Table 4.2 and Figure 4.1c).

**Table 4.2: Mean *n*-alkanes concentration, ratio C<sub>31</sub>/C<sub>33</sub>-alkanes and <sup>13</sup>C value in pasture samples from different dates, collected in the 'system' study.**

Forage	C <sub>27</sub>	C <sub>29</sub>	C <sub>31</sub>	C <sub>33</sub>	C <sub>35</sub>	C <sub>32</sub>	C <sub>36</sub>	C <sub>31</sub> ,C <sub>33</sub>	Total	C <sub>31</sub> /C <sub>33</sub> ratio	δ <sup>13</sup> C ‰
mg/100 g DM											
September 2002	2.4	10.2	19.2	12.3	1.3	1.1	0.4	30.5	45.8	1.5	nd
SE	0.08	0.19	0.28	0.15	0.03	0.03	0.01	0.38	0.59	0.02	
December 2002	5.1	14.8	25.7	8.9	0.9	1.1	0.4	34.6	56.8	2.9	-29.6
SE	0.21	0.31	0.44	0.17	0.03	0.02	0.02	0.53	0.93	0.06	0.44
March 2003	1.9	8.1	16.4	9.6	1.1	1.1	0.4	26.0	38.6	1.7	-28.8
SE	0.13	0.23	0.44	0.25	0.03	0.03	0.02	0.66	0.97	0.03	0.69
June 2003	1.8	7.3	16.7	14.1	1.6	1.1	0.4	31.2	43.5	1.2	-31.1
SE	0.06	0.18	0.29	0.28	0.04	0.06	0.03	0.41	0.59	0.02	0.51
September 2003	1.9	8.4	16.1	11.6	1.2	1.1	0.4	27.7	40.8	1.4	nd
SE	0.04	0.17	0.25	0.19	0.02	0.01	0.01	0.36	0.54	0.07	
December 2003	6.5	18.9	21.7	9.1	1.0	1.07	0.4	30.8	58.5	2.4	-29.5
SE	0.49	0.84	0.42	0.14	0.02	0.03	0.01	0.47	1.68	0.06	0.45
March 2004	2.6	9.4	17.6	11.5	1.2	1.2	0.4	28.8	43.6	1.5	-29.4
SE	0.18	0.22	0.36	0.23	0.03	0.03	0.01	0.51	0.81	0.02	0.68

Pasture samples were collected daily pre-grazing and bulked at the end of the week, during the faecal collection period. C<sub>27</sub>: heptacosane; C<sub>28</sub>: octacosane; C<sub>29</sub>: nonacosane; C<sub>31</sub>: hentriacontane; C<sub>32</sub>: dotriacontane; C<sub>33</sub>: tritriacontane; δ<sup>13</sup>C: difference from the ratio of the number of atoms of <sup>13</sup>C to the number of <sup>12</sup>C in a standard carbonate; SE= sd/√n; n=55, for all dates.

Differences in the content of the *n*-alkanes were observed between the principal species in the pasture (Table 4.3) with ryegrass having the highest total *n*-alkane content (44.1 mg 100 g<sup>-1</sup>DM) and white clover the lowest (8.3 mg 100 g<sup>-1</sup>DM). The *n*-alkane content of PA<sub>DI</sub> (24.1 mg 100g<sup>-1</sup>DM) was in between the other two species, in addition, these species had different ratio C<sub>31</sub>/C<sub>33</sub> (Table 4.2) As a result of the different *n*-alkanes composition between species, particularly in the content of the C<sub>31</sub> and C<sub>33</sub>-alkanes, the sub-samples prepared in the laboratory with different proportions of white clover – ryegrass showed that as the proportion of clover in the sample increased, the total concentration of *n*-alkanes in the sub-sample decreased whereas the ratio C<sub>31</sub>/C<sub>33</sub> increased, indicating the effect of the different proportion of components with different composition of *n*-alkanes in a sample of herbage.

Paddocks harvested for grass silage were those grazed during the experimental period. The mean concentration of C<sub>29</sub> and C<sub>31</sub>-alkanes was lower and the concentration of C<sub>33</sub>-alkane was higher in the fresh pasture than in the pasture silage. As a result the ratio

$C_{31}/C_{33}$  was higher in the latter, probably more related to the concentration of n-alkanes of the pasture at the time of ensiling. In addition, the concentration of n-alkanes in maize grain or maize silage was very low (Table 4.1).

**Table 4.3: Mean n-alkane concentration and ratio  $C_{31}/C_{33}$ -alkanes in dominant pastures components and in hand constructed rye grass/white clover proportional samples.**

	$C_{27}$	$C_{29}$	$C_{31}$	$C_{33}$	$C_{35}$	$C_{32}$	$C_{36}$	$C_{31}+C_{33}$	Total	Ratio $C_{31}/C_{33}$
	g/100 g DM									
Paspalum spp. <sup>(1)</sup>	2.5	4.2	11.8	4.2	0.2	0.8	0.4	16.0	24.1	2.8
Trifolium repens L.	1.4	3.8	2.0	0.4	0.2	0.2	0.3	2.4	8.3	5.0
Lolium perenne L.	1.9	9.5	18.9	11.2	1.1	1.1	0.4	30.1	44.1	1.7
Proportional mixtures of white clover – rye grass										
80 – 20	1.7	4.9	5.2	2.1	0.2	0.3	0.4	7.3	14.8	2.5
60 – 40	1.7	5.9	9.8	5.0	0.5	0.6	0.4	14.8	23.9	2.0
40 – 60	1.4	6.4	12.6	8.1	0.8	0.6	0.3	20.7	30.2	1.6
20 – 80	1.8	7.8	15.8	9.7	0.9	1.0	0.4	25.5	37.4	1.6

<sup>(1)</sup>Paspalum dilatatum poir (PA<sub>01</sub>). Plant material used in rye grass – white clover mixtures was similar to that in pasture in spring, and for PA<sub>01</sub> in late summer (n=1).  $C_{27}$ : heptacosane;  $C_{28}$ : octacosane;  $C_{29}$ : nonacosane;  $C_{31}$ : hentriacontane;  $C_{32}$ : dotriacontane;  $C_{33}$ : tritriacontane.

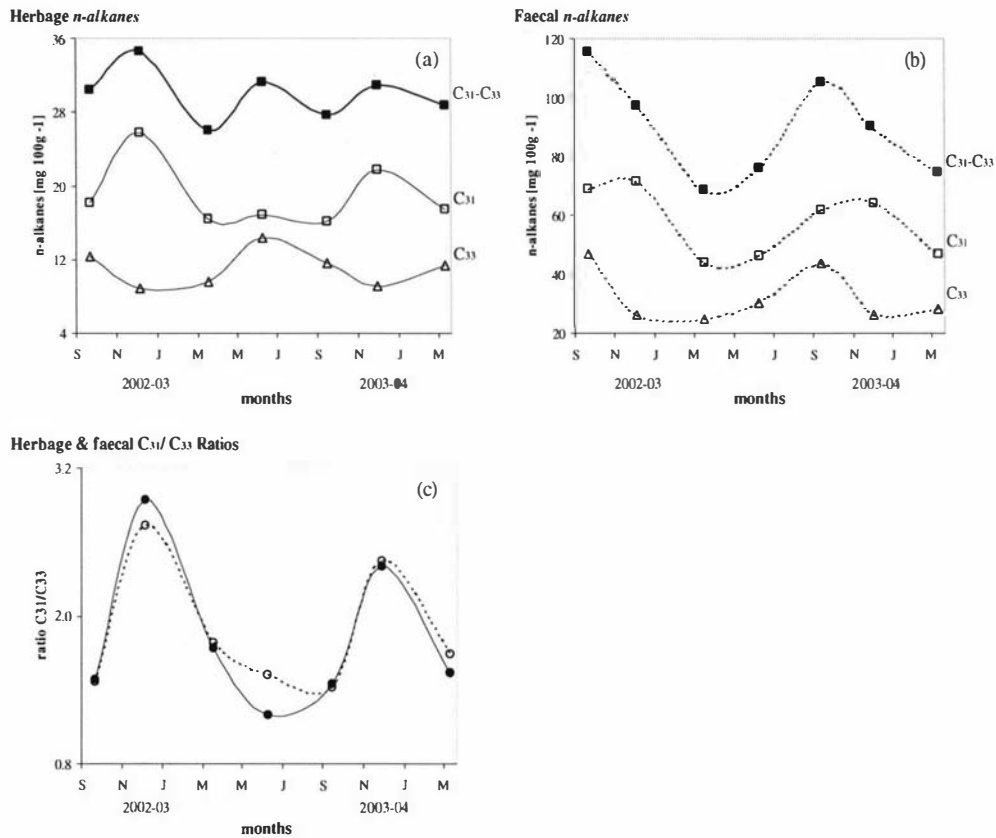
#### 4.3.2. Concentration of the odd chain n-alkanes in the faeces

As expected, the concentrations of all the n-alkanes in faeces were higher than those in the pasture (Figure 4.1b) and also showed significant variation across dates ( $P < 0.001$ ; Figure 4.1b). The content of the  $C_{33}$ -alkane showed a peak in September 2002 and 2003 and then achieved the lowest value in the following December, which was maintained throughout the summer until increasing slightly in March each year. The content of the  $C_{31}$ -alkane in faeces was higher than the content of the  $C_{33}$ -alkane throughout the year, with a similar pattern to that observed in the herbage. The difference between the  $C_{31}$  and  $C_{33}$ -alkanes was larger during the spring – summer period than during the autumn – winter period, with a higher content of the  $C_{31}$ -alkane during the spring than in winter but always above the  $C_{33}$ -alkane values (Figure 4.1b). The sum of the  $C_{31}+C_{33}$  alkanes in faeces also showed significant changes across dates ( $P < 0.001$ ; Figure 4.1b), more pronounced than observed in the pasture. These changes in the  $C_{31}+C_{33}$  alkanes were sustained by a different relative rate of change between these two alkanes, probably associated with changes in the diet as a result of the changes observed in the pastures (Figure 4.1a).

The ratio  $C_{31}/C_{33}$  in faeces showed a similar trend over the study to the ratio of the same n-alkanes in the pasture (Figure 4.1c), however the faecal ratio in June was above the minimum measured in the pasture due to the effect of the pasture silage consumed. Pasture silage had the highest ratio  $C_{31}/C_{33}$ , hence should have affected the mean ratio of the diet. A similar trend, although to a lower extent, was measured in March 2004 when a

high proportion of the cows in the study were also fed pasture silage. An opposite trend was observed in December, particularly in 2002; this time there was no additional feed offered to the cows other than maize, with very low concentration of n-alkanes (Table 4.1), which suggests that other factors affected the composition of the ingesta of the cows promoting a reduction in the ratio  $C_{31}/C_{33}$  relative to the mean value of the pasture (Figure 4.1c).

**Figure 4.1:** Mean concentration of  $C_{31}$ ;  $C_{33}$  and  $C_{31}+C_{33}$  alkanes in pastures (a), faeces (b) and ratio  $C_{31}/C_{33}$  in pasture and faeces (c).



Values are means of all the paddocks and cows in the 'system' study, from dates where intake was estimated.  $[C_{31}+C_{33}]$ -alkanes (■);  $C_{31}$ -alkane (□);  $C_{33}$ -alkane (Δ); Ratio  $C_{31}/C_{33}$  in pasture (○) and in faeces (●).

Although significant, the general linear relationship between the  $C_{31}$  and  $C_{33}$ -alkanes in the faeces was poor when all the data available was considered together ( $y = 11.5 + 0.36x$ ;  $r^2: 0.28$ ;  $p < 0.001$ ; RMSE: 8.17;  $n: 1390$ ; where 'y' is  $C_{33}$  and 'x'  $C_{31}$ ), this probably resulted from the differences between the n-alkanes composition of the pastures grazed on different dates combined with the consumption of pasture silage (Figure 4.2b). This is supported by the fact that the linear relationship estimated for each date improved (Table 4.4). The relative change of the concentration of  $C_{31}$  and  $C_{33}$  in faeces of individual cows

was consistent with the mean concentration of these two n-alkanes in the herbage consumed, and then with the ratio  $C_{31}/C_{33}$  (Figure 4.2b).

**Table 4.4: Linear relationships between the concentration of  $C_{31}$  and  $C_{33}$ -alkanes in faeces. The mean and standard deviation values for the ratio  $C_{31}/C_{33}$ -alkanes in faeces are also presented.**

Dates	Equation	$r^2$	p	RMSE	n	Ratio	
						$C_{31}/C_{33}$	SD
All dates	$y = 11.5 + 0.36 x$	0.28	<0.001	8.17	1390	1.87	0.518
September 2002	$y = 6.44 + 0.59 x$	0.88	<0.001	1.82	200	1.46	0.064
December 2002	$y = 11.7 + 0.20 x$	0.32	<0.001	1.85	204	2.73	0.221
March 2003	$y = 2.35 + 0.51 x$	0.95	<0.001	1.11	189	1.77	0.083
June 2003	$y = 14.5 + 0.34 x$	0.60	<0.001	4.38	203	1.52	0.333
September 2003	$y = 5.74 + 0.61 x$	0.84	<0.001	1.71	200	1.42	0.060
December 2003	$y = 7.63 + 0.29 x$	0.59	<0.001	1.79	205	2.45	0.187
March 2004	$y = 7.96 + 0.42 x$	0.73	<0.001	2.16	189	1.69	0.163

'y' is the concentration of  $C_{33}$ -alkane in faeces and 'x' the concentration of the  $C_{31}$ -alkane. P: probability [significance: \*= $P < 0.05$ ; \*\*= $P < 0.01$ ; \*\*\*= $P < 0.001$ . NS= not significant.];  $r^2$ : coefficient of determination; RMSE: root mean square error; SD: standard deviation; n: sample number.

**Figure 4.2: Relationships between intakes estimated using  $C_{31}$  or  $C_{33}$ -alkanes (a) and the faecal concentration of  $C_{31}$  and  $C_{33}$ -alkanes (b).**

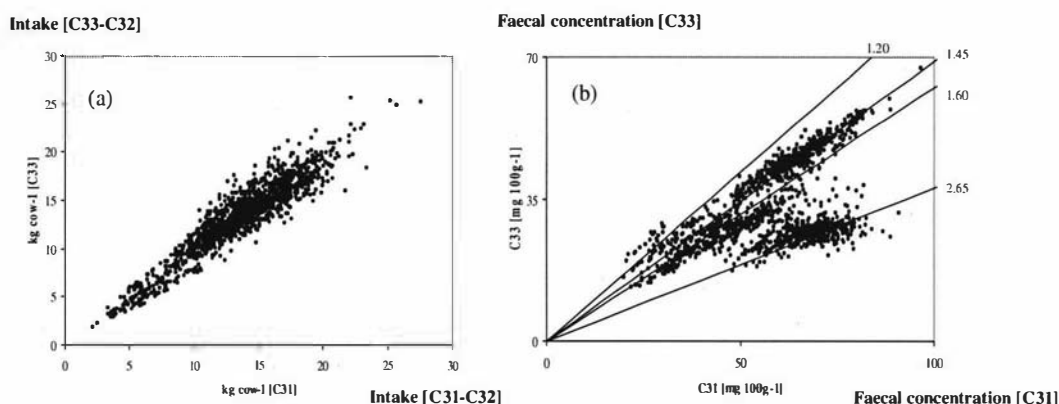


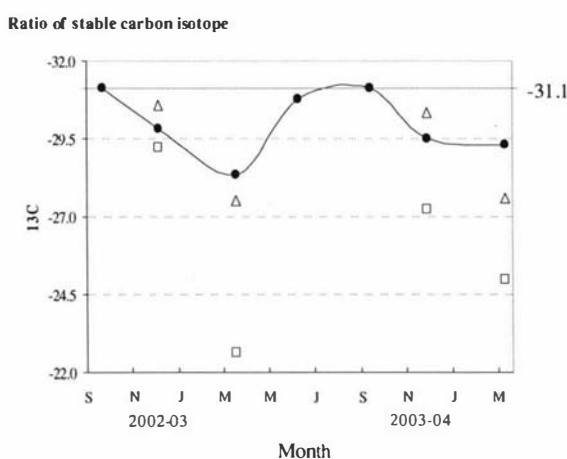
Figure (a) and (b) are individual cows (n = 1390). In Figure (b) solid lines indicate the mean ratio  $C_{31}/C_{33}$  in the pastures grazed [June: 1.20; September: 1.45; March 1.60 & December: 2.65; n=110 in September, March and December and n=55 in June]. A proportion of the cows were fed maize grain or silage in December 2002 & 2003 and March 2003 & 2004; in addition, most of the cows were fed pasture silage in June 2003 and a proportion in March 2004.

### 4.3.3. Concentration of the $\delta^{13}C$ in pastures and faeces

The means and standard error for values of  $\delta^{13}C$  in supplements and pasture estimated for different dates are presented in Table 4.1, Table 4.2 and Figure 4.3. The mean  $\delta^{13}C$  value for maize grain, maize silage and  $PA_{DI}$  were similar and higher (less negative) than the values in the herbage and the forage fed to the cows. The mean  $\delta^{13}C$  of the pasture varied across dates ( $p < 0.001$ ) and showed a rise in value during late spring – summer periods (less negative; Table 4.2 and Figure 4.3).

The mean  $\delta^{13}\text{C}$  in the faeces also varied across dates ( $p < 0.001$ ). The values for un-supplemented and supplemented cows showed the same trend across dates observed in the pasture (Figure 4.3), however, the mean  $\delta^{13}\text{C}$  value in faeces of un-supplemented cows was consistently lower (more negative;  $p < 0.05$ ) during mid lactation and higher (less negative;  $p < 0.05$ ) in late lactation (both seasons), while the cows fed maize grain or maize silage had a higher  $\delta^{13}\text{C}$  value (less negative) on both dates ( $p < 0.001$ ). Significant differences were observed for the  $\delta^{13}\text{C}$  value from un-supplemented and supplemented cows in both mid and late lactation periods.

**Figure 4.3: Mean  $\delta^{13}\text{C}$  values in the pasture and faeces from cows grazing pasture only or from cows grazing and fed maize silage.**



Pasture (●); cows grazing only (Δ); cows grazing and fed maize silage (□). Solid grey line indicates the mean  $\delta^{13}\text{C}$  value for the winter pasture (-31.1). Values were calculated with data from all the paddocks and cows fed maize silage, only 18 cows (6 cows from each strain) feeding only pasture were analysed for  $\delta^{13}\text{C}$  in the faeces.

#### 4.3.4. Dry matter intakes

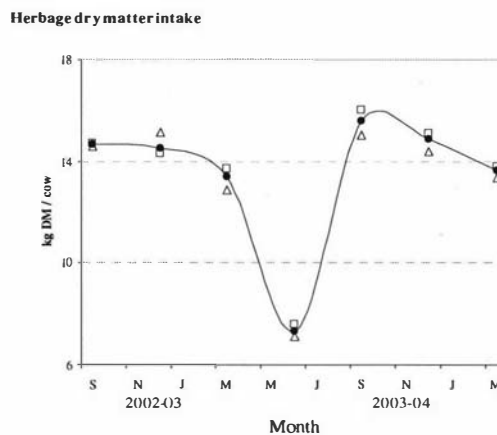
The mean  $\text{DMI}_H$  of all the cows estimated with the pair  $\text{C}_{31}\text{--}\text{C}_{32}$  was slightly higher than that obtained with the pair  $\text{C}_{33}\text{--}\text{C}_{32}$  in most dates, except during December 2002 (Figure 4.4). The mean  $\text{DMI}_H$  obtained using the pair  $[\text{C}_{31} + \text{C}_{33}] \text{--}\text{C}_{32}$  was between those obtained with the other two pairs on all dates. The linear relationship between  $\text{DMI}_H$  calculated with either  $\text{C}_{31}\text{--}\text{C}_{32}$  or  $\text{C}_{33}\text{--}\text{C}_{32}$  ( $y = 0.50 + 0.94 x$ ;  $r^2: 0.88$ ;  $p < 0.001$ ; RMSE: 1.29; n: 1390; where 'y' is intake estimated with  $\text{C}_{33}\text{--}\text{C}_{32}$  and 'x' with  $\text{C}_{31}\text{--}\text{C}_{32}$ ) indicate the high association between the  $\text{DMI}_H$  estimated from one or the other pair of n-alkanes (Figure 4.2a), when high number of cows are involved. However, it should be noted that for any  $\text{DMI}_H$  estimated by the pair  $\text{C}_{31}\text{--}\text{C}_{32}$ , a number of possible corresponding  $\text{DMI}_H$  values estimated with the pair  $\text{C}_{33}\text{--}\text{C}_{32}$  exist, and vice versa (Figure 4.2a). This is attributed to the variability in the ingestion  $\text{C}_{33}$  relative to the ingestion of  $\text{C}_{31}$  between cows, and then due

to the different ratio  $C_{31}/C_{33}$  within animals in a herd and between the value for each cow and the mean pasture (See Chapter 3).

#### 4.4. DISCUSSION

Although the  $C_{31}$ -alkane is known to have a lower recovery rate in faeces than the  $C_{33}$ -alkane (Dillon, 1993), the content of the  $C_{31}$ -alkane in the herbage was higher (Tables 4.1 and 4.2). These two n-alkanes have been used in combination with  $C_{32}$  to accurately estimate intake of pasture by dairy cows (Dillon, 1993; Malossini *et al.*, 1996; Hameleers & Mayes, 1998b; Hameleers & Mayes, 1998a; Unal & Garnsworthy, 1999; Garcia *et al.*, 2000), but the pair  $C_{33}$ - $C_{32}$  is usually preferred (Mayes *et al.*, 1986b) despite the concentration of  $C_{33}$  being lower than  $C_{31}$  in most temperate species (see Chapter 2). It has been recommended that the concentrations of the odd n-alkanes used should be above  $5 \text{ mg } 100 \text{ g}^{-1} \text{ DM}$  for an accurate estimate of  $\text{DMI}_H$  (Laredo *et al.*, 1991; Malossini *et al.*, 1996; Berry *et al.*, 2000), mainly because the potential errors introduced with the analytical procedure at very low concentration of the n-alkane in the herbage (Hameleers & Mayes, 1998a), although the estimates can be correct with lower than  $4 \text{ mg } 100 \text{ g}^{-1} \text{ DM}$  providing a precise analytical determination (Dove H., personal communication).

**Figure 4.4:** Mean herbage dry matter intake estimated by using the pairs  $C_{31}$ - $C_{32}$ ;  $C_{33}$ - $C_{32}$  or  $[C_{31}+C_{33}]$ - $C_{32}$  alkanes. In all cases an adjustment for differences in recovery was utilised.



Values calculated from all the paddocks and cows in the experiment on each date.  $C_{31}$ - $C_{32}$  (□);  $C_{33}$ - $C_{32}$  (Δ) &  $[C_{31}+C_{33}]$ - $C_{32}$  (●).

The absolute concentration of the n-alkanes in the herbage varied (Malossini *et al.*, 1990), as was observed between dates in the 'system' study. In addition, the content of the  $C_{33}$ -alkane in white clover was very low, whereas in  $\text{PA}_{DI}$  it was on the limit proposed by Laredo *et al.* (1991)(Table 4.3). This problem could be reduced by using the

$C_{31}$  instead of the  $C_{33}$ -alkane, considering that the concentration of  $C_{31}$  is 5 and 2.8 times higher in white clover and  $PA_{DI}$  respectively than the concentration of the  $C_{33}$ -alkane.

The concentrations of these two n-alkanes in the pasture also showed complementary peaks. This indicates that by considering the same pair of n-alkanes on different dates to estimate  $DMI_H$ , differences in herbage ingestion associated with changes in pasture components would affect the accuracy of the  $DMI_H$  estimated, particularly if different concentrations of n-alkanes in herbage and diet occurs. This seems to be the case, as the fluctuation of  $C_{31}$  and  $C_{33}$ -alkanes in faeces and pasture were different, and then the ratio  $C_{31}/C_{33}$  in the faeces of individual cows was different to the mean ratio in the pasture grazed.

The quantity and proportion of the pasture in the total diet and their n-alkane concentration corrected by the partial absorption of each n-alkane in the digestive tract, should determine the concentration of the n-alkane in the faeces. Due to the different n-alkane content between pasture silage and maize silage in relation to the pasture, the type of supplement consumed may affect the faecal content of the  $C_{31}+C_{33}$  alkanes. Thus a high proportion of maize grain or maize silage in the diet should dilute the concentration of the n-alkanes from the pasture recovered in faeces, but keep the ratio of  $C_{31}/C_{33}$  unchanged. In contrast, a high proportion of pasture silage in the diet should increase the content of  $C_{31}+C_{33}$  alkanes and affect the ratio of  $C_{31}/C_{33}$  in the faeces, according to the proportion of pasture and pasture silage consumed and the differences in the n-alkane concentration of the two feeds.

Nevertheless, differences between grazing cows would also be expected due to selectivity. For instance, if a cow consumes a higher proportion of a component with a high concentration of  $C_{31}$ -alkane relative to  $C_{33}$ -alkane (ratio  $C_{31}/C_{33}$  higher than the mean in the pasture) and the pair used to estimate intake is  $C_{33}-C_{32}$ , the additional dry matter ingested will not be accurately estimated, simply because the n-alkane associated with the type of herbage ingested is not considered. Thus, a low concentration of the  $C_{33}$ -alkane in the components of the pasture being selected could result in  $DMI_H$  being underestimated with the pair  $C_{33}-C_{32}$ , this could particularly occur during periods where the diet incorporates either white clover or  $PA_{DI}$  as result of their greater availability in the pasture. Then, selective grazing can enhance the content of white clover in the diet of the cows; in addition, the cows showed an increased preference for  $PA_{DI}$  during the autumn.

The change in the ratio  $C_{31}/C_{33}$  in faeces between dates was mainly determined by the greater variability of the  $C_{31}$ -alkane in the pasture between dates, particularly the high content of  $C_{31}$  relative to the  $C_{33}$ -alkane in the pasture for December (Figures 4.1b) as it

is shown by the ratio  $C_{31}/C_{33}$  in the faeces (Figure 4.1c). As indicated, the ratio  $C_{31}/C_{33}$  is not the same between pasture components and plant-parts. In addition, this relationship also changed over the season - probably as a result of the seasonal change between components or in their leaf to stem ratio - which could affect cows' diet (Dillon, 1993; Genro et al., 2001). Despite this, the mean ratio  $C_{31}/C_{33}$  in the faeces across dates (all cows considered on each date,  $n=205$ ) was similar to that observed in the pasture offered. This suggests a similar mean n-alkanes composition between herbage offered and consumed, indicating that the mean proportion of the species components or plant-parts in pasture and diet was similar across dates. This is in agreement with the strip grazing management utilised where most of the herbage available was removed by the cows during each grazing, and suggests that the mean difference between the estimate of  $DMI_H$  for a group of cows by using either of the pairs mentioned would be reduced just because the herbage available was almost totally removed by the cows, even though differences between the  $DMI_H$  estimated for individual cows exist. Differences between animals would be compensated for in a large group of cows, particularly when grazing non-selectively.

Considering that the dose rate of the even chain  $C_{32}$ -alkane during the sampling period was constant, the relationships between the natural and dosed n-alkanes in faeces may be determined by the n-alkane content in the herbage, their ingestion and recovery in faeces. Thus, given that the concentration of the n-alkane in the herbage is high enough to be utilised, the difference between using one or other n-alkane should be defined by the accuracy of the analytical procedures, which could be affected by a low concentration of the n-alkane and the recovery rate. If the herbage ingested has the same concentration of  $C_{31}$  and  $C_{33}$ -alkanes as the pasture and then a similar ratio  $C_{31}/C_{33}$ , the same intake would be estimated by using the pairs  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$  if differences in recovery were corrected; thus differences in intake between cows would be determined by the total amount of n-alkane ingested with the pasture (see Chapter 3). However, differences in the  $DMI_H$  of a cow estimated by using different pair of n-alkanes are supported by the diet having a different ratio  $C_{31}/C_{33}$  than the pasture offered. This occurs because the cows prefer some components of the pasture instead of others, combined with management decisions allowing more selective grazing (Figures 4.2 and 4.4). By considering the ratio  $[C_{31}+C_{33}]-C_{32}$  and a correction for differences in recovery this problem can be solved, as was indicated in Chapter 3. This agrees with the fact that the mean  $DMI_H$  obtained with the pair  $[C_{31}+C_{33}]-C_{32}$  seems to more accurately estimate the intake of individual cows when the mean diet composition differs from the composition of the pasture being grazed, hence the intake estimated lay between those obtained with the other two pairs possible on each date intake was measured, as a result of considering the relative change of  $C_{31}/C_{33}$  in diet and pasture simultaneously (see Chapter 3).

It should be also noted that sward type in both the ‘system’ and ‘component’ experiments described in this thesis differed from the pure perennial ryegrass pasture used by Dillon (1993)(see Chapters 5 and 8). The recovery rates could have introduced a bias in the estimation of the intakes, because of the difference in the type of pasture being grazed. However, values of recovery rate estimated by Berry et al. (2000) for a different type of pasture than that used by Dillon (1993) also confirms the same differences in faecal recovery used by Dillon (see Table 2.3)

Therefore, although it could be argued that the mean  $DMI_H$  from a group of cows would be similar by using either the pair  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$ , this would not be true for individual cows grazing selectively. This problem would increase under lax grazing management. Furthermore, one of the main interests of this method is to detect those animals that have greater  $DMI_H$  by grazing more efficiently. It is apparent that neither the use of the pair  $C_{31}-C_{32}$  nor  $C_{33}-C_{32}$  will give an accurate indication when the ratio  $C_{31}/C_{33}$  in the diet of the cows is different to the ratio in the pasture and the  $DMI_H$  obtained would be biased. The data from the faecal concentrations of the  $C_{31}$  and  $C_{33}$ -alkanes of individual cows, their faecal ratios (Figure 4.2b) and the  $DMI_H$  estimated from them (Figures 4.2a and 4.4) suggest that this is the case under the grazing conditions of the present thesis, in agreement with the analysis presented in Chapter 3. Despite the previous argument, differences between the mean  $DMI_H$  estimated for all the cows in the ‘system’ experiment by using different pairs of odd – even n-alkanes were not significant (Figure 4.4); however,  $DMI_H$  differences appeared for individual cows when different pairs of n-alkanes were involved in the calculation. The difference in the value estimated for the same cow by using these different pairs increased when the faecal ratio  $C_{31}/C_{33}$  was different to the ratio of the same n-alkanes in the pasture, which indicates that the cow has selected a diet different in composition to the mean pasture being grazed (see Chapter 3).

The proportion of the  $C_4$  species in the diet of each cow was estimated from the  $\delta^{13}C$  value from faeces and pasture analysed for the same sampling period. The procedure utilised was already well described and tested; however, as is also based on the determination of  $DMI_H$  by the n-alkanes and involves an increased degree of complexity (see Appendix III), it should be considered that errors at different stages of the procedure could affect the estimation of supplement intake. In addition, due to the similar  $\delta^{13}C$  signatures between  $PA_{DI}$  and maize (grain or silage), the proportion of maize that is determined in the diet of supplemented cows would be biased by the ingestion of  $PA_{DI}$ . Considering the  $\delta^{13}C$  value of the pasture at each date as the reference from which the intake of maize is estimated, the procedure assumes that the preference of  $PA_{DI}$  is similar

between cows and equal in the diet to the mean content determined in the pasture. Thus, the proportion of the C<sub>4</sub> species represents the true value of maize silage intake.

It was observed that PA<sub>DI</sub> was not preferred in December by un-supplemented grazing cows, but preferred in March. If supplemented cows behaved similarly to those un-supplemented, the consumption of maize would be underestimated in December and overestimated in March. Supplemented cows had lower herbage consumption and the standard deviation of the mean  $\delta^{13}\text{C}$  in faeces was smaller than in un-supplemented cows. This could indicate that the  $\delta^{13}\text{C}$  content of PA<sub>DI</sub> in their diets was more likely to be lower than in the pasture during the summer, so maize consumption was probably underestimated. Nevertheless, the similar mean ratio C<sub>31</sub>/C<sub>33</sub> between pasture and faeces indicates that the mean proportions of PA<sub>DI</sub> in pasture and diet was similar, suggesting that the content of maize in the diet was accurately estimated, at least the mean values of each group of cows being supplemented. However, the accuracy of the individual intake of supplement is still in doubt, because of the impossibility of detecting the content of PA<sub>DI</sub> in the diet of the cows by means other than the stable isotope discrimination method, the results of which were confounded with the estimation of maize content.

Thus, a number of problems must be dealt with in order to accurately estimate intake of herbage and supplements in the field: (a) the concentration in the feed and supplement consumed of the natural n-alkane used in the equation should be high (Hameleers & Mayes, 1998a; Berry *et al.*, 2000); (b) the n-alkanes pattern of the components to be discriminated should be different (Dove & Mayes, 1996); (c) the presence of preferred components in the pasture with low n-alkanes content (white clover and PA<sub>DI</sub>) should be considered and the n-alkane to be used in the equation selected accordingly (Hameleers & Mayes, 1998a); and (d) the recovery rate for the natural and dosed n-alkanes selected should be accurately known and differences corrected when the ratio  $[\text{C}_{31}+\text{C}_{33}]-\text{C}_{32}$  is utilised. Furthermore, the proportion of summer grasses such as PA<sub>DI</sub> in pasture and diet and differences in the preference of the cows for these grasses and between dates would affect the estimation of the supplement consumed when the  $\delta^{13}\text{C}$  technique is used. The discrimination of components within the pasture and the diet of the cows by using the procedure proposed by Dove and Moore (1995) appears to be a helpful tool to solve this problem and was successfully used to separate the consumption of fresh pasture from pasture silage in the present thesis; however, when utilised to discriminate PA<sub>DI</sub> from the diet the results were unrealistic (data not presented) and did not agree with the results obtained from the isotope discrimination method, which is known to be very precise (Jones *et al.*, 1979). The reason for this lack of agreement with Dove and Moore's procedure for PA<sub>DI</sub> was attributed to the quality of the herbage sample obtained. This sample must represent exactly those pasture components and plant-parts the animals are

actually consuming; as preference for a component in the pasture is not always similar, this was the main weakness of the procedure described.

The faecal recovery rates (Dillon, 1993; Penning, 2004) and the pair  $[C_{31}+C_{33}]-C_{32}$  utilised were probably appropriate to estimate herbage intake in both the 'system' and 'component' experiments in this thesis, mainly because the changes observed in pasture composition between dates ('system' trial, see Chapter 3 and 5) or as result of management ('component' trial, see Chapter 3 and 8) affected the n-alkanes content of the herbage available to the cows in both experiments. However, it would have been helpful to validate the approach used here by including actual measurements of intake in an indoor study, by using forage obtained from the same sward grazed in the current studies. In addition, the use of a group of control animals being fed pasture components in excess and separately in order to promote their selection, and including both  $PA_{DI}$  and maize silage in the diet to compare discrepancies between the actual intakes obtained of each component and the estimated would have been helpful. An indoor experiment would be useful to provide also estimates of the recovery rates for the n-alkanes used for the experimental conditions and pasture type available in the current studies, and to test some of the assumptions made in this work.

#### 4.5. CONCLUSIONS

Changes in the n-alkane composition of the pastures grazed over the present studies have occurred as a result of natural changes in the relative composition of species during the year, in addition to the capacity of the cows to select a diet with a different ratio  $C_{31}/C_{33}$  than that measured for the mean pasture sample. Considering the requirement to determine  $DMI_H$  of individual cows grazing in a group, that were also fed forage and supplements, the use of  $[C_{31}+C_{33}]-C_{32}$  adjusted for recovery appears to estimate  $DMI_H$  more accurately than either the pair  $C_{31}-C_{32}$  or  $C_{33}-C_{32}$ . In addition, the values of  $DMI_H$  obtained for different dates are comparable despite the relative change of the n-alkane content in the pasture between dates and also in animal preference.

The estimation of maize consumption could be affected by the presence of  $PA_{DI}$  in the pasture, as described. An improved herbage sample, which should represent those species and plant-parts the cow prefers, may improve the capacity to discriminate different components in the diet of grazing dairy cows. This would enhance the determination of the supplement consumed by each cow.

#### 4.6. INTEGRATED CONCLUSIONS

The accuracy of the *n*-alkanes method to estimate  $DMI_H$  for grazing cows depends on the difference in the composition of *n*-alkanes between diet and pasture in relation to the known concentration of a dosed *n*-alkane. The pairs  $C_{33}/C_{32}$  and  $C_{31}/C_{32}$  have been widely used in temperate pastures due to demonstrated accuracy, but when the animals graze selectively, discrepancies between actual and calculated intakes occur. However, an improved estimate for individual animals can be achieved by using the pair  $[C_{31}+C_{33}]-C_{32}$  in the estimate instead of the usually suggested  $C_{33}/C_{32}$  and  $C_{31}/C_{32}$ . The procedure was tested using data from different indoor studies; however one step forward would be to validate it by feeding the cows indoors with a known amount of two different types of herbage with different *n*-alkane content and ratio  $C_{31}/C_{33}$ , in excess of requirements so allowing for cow selection; in addition, a second indoor experiment where cows of the different strains are fed herbage of similar composition collected from the same pasture the cows were grazing in the system study would also bring the possibility of investigating the assumption made about the recovery rate of *n*-alkanes in faeces being similar to those from Dillon (1993) and any possibility of the differences between the genotypes used.

To estimate the consumption of maize the combination of *n*-alkanes and  $\delta^{13}C$  was successfully utilised. The presence of  $PA_{DI}$  in the pastures and diet of the cows, with a similar  $\delta^{13}C$  signature to maize, would introduce an error in the estimation of maize intake. It was assumed that the content of  $PA_{DI}$  in the diet and pasture was similar, so the difference in  $\delta^{13}C$  between pasture and diet was considered to be the true value of maize silage consumption; however, a difference in the preference for this species could bias the estimate of the diet for individual animals. Despite the fact that individual preference for  $PA_{DI}$  could bias the consumption of maize for individual cows, individual bias is considered to be compensated for in the mean herd  $DMI_H$  in strip-grazing management when most of the herbage available is consumed and large numbers of experimental units were utilised.

The main problem of combining the *n*-alkanes –  $\delta^{13}C$  methods is still that the sample of herbage must represent the diet of each animal. The discrimination procedure proposed by Dove and Moore (1995) is probably the best approach to solve this problem, as it brings an opportunity to detect  $PA_{DI}$  in the diet of the cow and avoid its confounding effect in the estimation of maize consumption; however, an improved reference sample must be collected. This could be obtained by a detailed observation of which part-plants and pasture components the cows are actually eating. This is not an easy task, it can be improved by the use of oesophageal fistulated animals; however, their use involves

additional considerations that have been discussed previously (See Chapter 2). However, it is apparent that this problem did not affect the improved estimation of  $DMI_H$  that is achieved by the use of  $[C_{31}+C_{33}]-C_{32}$ . This reduced the negative effects of cow selectivity when the main change in the diet involves a different proportion of the two main natural *n*-alkanes present in the pasture offered. As previously mentioned, the validation of this technique indoors requires further considerations.

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## APPENDIX I

### EQUATION USED AND PROCEDURE DESCRIPTION TO ESTIMATE DRY MATTER INTAKE OF DIFFERENT COMPONENTS OF THE DIET

The equation used to calculate herbage dry matter intake (DMI<sub>H</sub>) in the present thesis is:

$$\text{DMI}_H = \frac{[F_{i+ii}/F_j] \times (D_j \times R_j + I_S \times S_j) - [(I_S \times S_i \times R_i) + (I_S \times S_{ii} \times R_{ii})]}{[(p \times A_i + (1-p) \times B_i)] \times R_i + [(p \times A_{ii} + (1-p) \times B_{ii})] \times R_{ii}} - F_i + F_{ii} / F_j \times [(p \times A_j + (1-p) \times B_j)] \times R_j \times 0.1$$

DMI<sub>H</sub> = Herbage dry matter intake

*j* = C<sub>32</sub>-alkane

*i* = C<sub>31</sub>-alkane

*ii* = C<sub>33</sub>-alkane

D = dosed amount

R = recovery rate

I<sub>S</sub> = maize intake

A, B, S, F = pasture, grass silage, maize and faeces

*p* = proportion in the diet of each herbage origin (pasture or grass silage)

This is a modification of that used by Hameleers and Mayes (1998), derived from Dove and Mayes (1991). This equation considers the proportion of each forage component estimated using Dove and Moore (1995) and the quantity of supplement in the diet estimated by using a modification of the combined *n*-alkane – δ<sup>13</sup>C methods proposed originally by Garcia et al. (2000) as it is detailed in Appendix II.

The difference between this formula and those previously presented formulae is that the C<sub>31</sub> and C<sub>33</sub>-alkanes are considered together in the pair [C<sub>31</sub>+C<sub>33</sub>]-C<sub>32</sub>. In addition, the mean recovery rates of natural and dosed *n*-alkanes from Dillon (1993) were considered to compensate differences in the faecal recovery rate between the *n*-alkanes used.

## APPENDIX II

### MODIFICATION TO THE COMBINED *N*-ALKANES – <sup>13</sup>C METHODS

The procedure utilised by Garcia et al. (2000) to estimate the amount of maize consumed by individual cows in a group-feeding context was utilised. Although this procedure was tested indoors, the accuracy of maize consumption estimate relies on the *in-vitro* digestibility of the pasture sample, which is not a good indicator of the digestibility of the diet ingested by the cow (as was already discussed, see Chapters 2-3) and it was largely recognised as the main limitation of the *n*-alkane determination.

Consequently, errors in the *in-vitro* determination of the herbage sample will affect the estimate of the individual supplement intake in the field. To make an improvement to this procedure, the C<sub>32</sub>-alkane was used to estimate total faecal output, thus maize silage and pasture faecal outputs can be estimated from the faecal ratio of the C<sub>32</sub>-alkane dosed and recovered in faeces instead of using pasture *in-vitro* value as proposed by Garcia et al. (2000). Nevertheless, the *in-vitro* value of the maize sample is still required to complete the procedure and estimate the intake of supplement once faecal output composition is determined.

The benefit of this modification is that total faecal output (TFO) is estimated from the dosed marker affected by recovery (Mayes & Dove, 2000) and considers the C<sub>32</sub>-alkane in the herbage. As the consumption of herbage was already estimated by using the *n*-alkanes method then the content of the C<sub>32</sub>-alkane in the herbage is also known, as well as the proportion of herbage and supplement is estimated by the stable carbon isotope discrimination technique (Jones et al., 1979) and the consumption of maize is calculated by considering the *in-vitro* value of the maize sample.

Total faecal output:

$$\text{TFO} = \frac{(D_j \times R_j) + (\sum A..B_{jj} \times R_j)}{F_j}$$

TFO = Total faecal output

*j* = C<sub>32</sub> -alkane

D = dosed amount

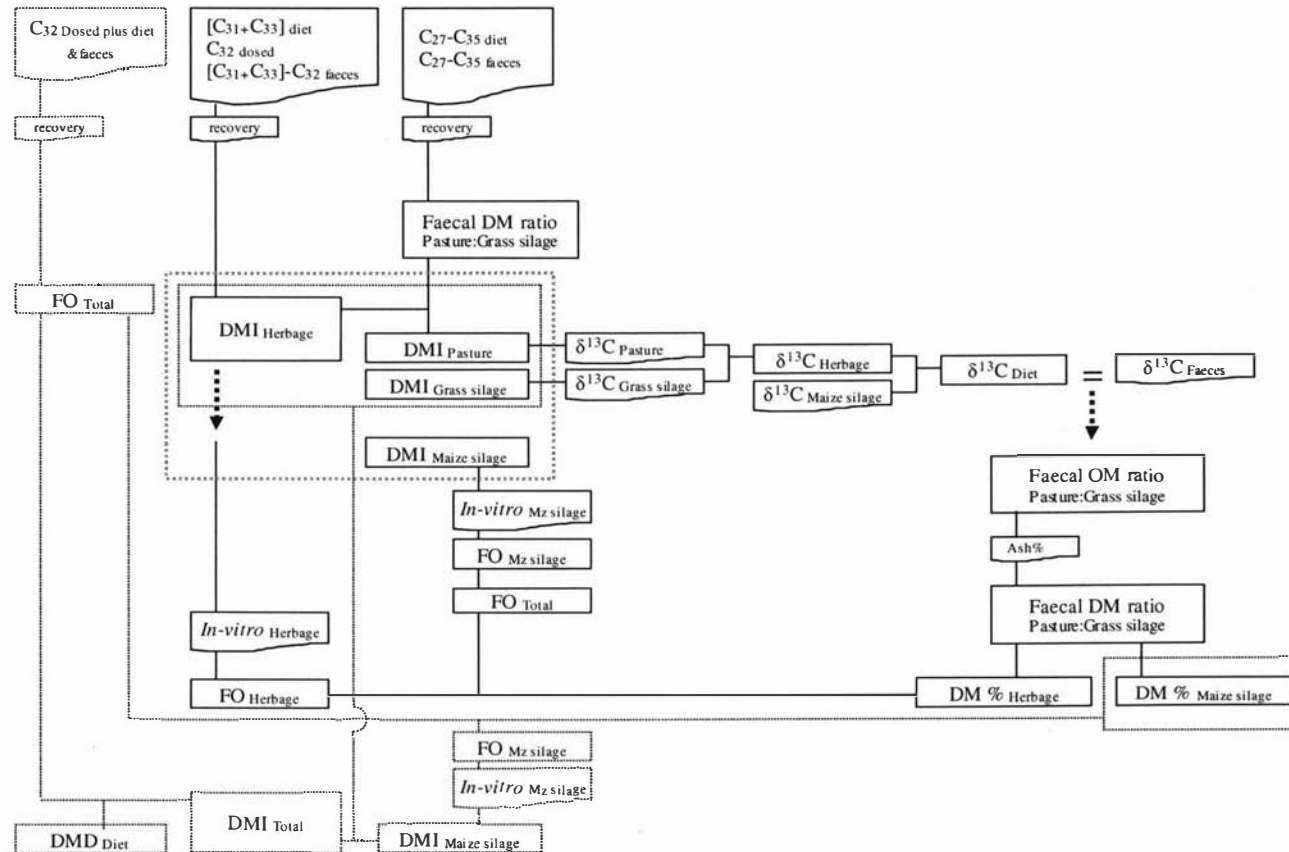
R = recovery rate

A, B, F = pasture, grass silage, faeces

This alternative should reduce the errors compared with using the *in-vitro* value of the pasture sample. It is also assumed that the *in-vitro* value of the supplement would be more uniform than that from the pasture sample. In addition, the estimate obtained still considers the differences in selectivity from different animals, as the digestibility value utilised is not fixed equal in all the animals by using the *in-vitro* value of the pasture sample.

## APPENDIX III

### DETAILED FLUX DIAGRAM INDICATING THE SEQUENCE OF PROCEDURES UTILISED TO ESTIMATE DIET COMPOSITION AND DIGESTIBILITY



The combined n-alkanes -  $\delta^{13}\text{C}$  method was used to estimate the intakes of herbage (pasture plus grass silage) and the proportion of herbage in the faecal dry matter of each individual cow. The  $\text{C}_{32}$ -alkane was used to estimate the total faecal output ( $\text{FO}_T$ ), which combined with the percentage of maize silage in the  $\text{FO}_T$  allowed the estimation of the maize silage faecal output ( $\text{FO}_{Mz}$ ) and, after affected by *in-vitro*, the estimation of the intake of maize silage. From the  $\text{FO}_T$  and the total dry matter intake, diet digestibility was estimated.

## SECTION III

### CHAPTER 5

## PRODUCTIVITY OF PASTURE – BASED SYSTEMS: INTERACTIONS BETWEEN COW GENOTYPE AND MANAGEMENT

### 5.1. INTRODUCTION

High producing cows consume more feed as a result of higher energy demand (Buckley *et al.*, 2000), however the expression of their genetic yield potential may be compromised by inadequate nutrition (Veerkamp *et al.*, 1994) because they are not able to eat enough when grazing pasture (Mayne & Gordon, 1995; Kolver & Muller, 1998). Consequently, only small differences in production are expected between cows of high or low genetic potential for yield when farmed in low-input feeding systems based on pasture (Veerkamp & Emmans, 1995). Because of this, increased concerns have been raised in different parts of the world about whether the advantage of high yielding cows selected to suit high-input systems is maintained in low-input systems (Veerkamp *et al.*, 1994; Holmes, 1995; Veerkamp *et al.*, 1995; Oldham *et al.*, 1996). However, genotype by feeding interactions are not always detected (Holmes, 1995) because they are not evident at a reduced plane of nutrition where the genetic response of the cow cannot be fully expressed (Freeman, 1975); additionally, high producing cows can graze more herbage provided that high daily herbage allowance is offered (Buckley *et al.*, 2000).

It has been determined that high yielding cows developed in New Zealand (NZ) or overseas (OS) have different potentials for yield, with the OS genotypes performing better at a high plane of nutrition on total mixed ration (TMR), unable to achieve their potential on pasture diets (Kolver *et al.*, 2002). However, in a number of recent studies genotype by feeding interactions were measured in pasture-based systems, e.g. for fat and lactose yields and the content of fat in milk (Kennedy *et al.*, 2002; Kennedy *et al.*, 2003;

Linnane *et al.*, 2004). This increased occurrence of genotype by environment interactions occurred because the greater difference between the genotypes utilized, due to genetic improvement.

It is hypothesized that in systems managed on relatively low feed allowance per cow on pasture, genotypes with different potentials for yield would produce similarly, as cows with higher yield potential would be more constrained than cows of low genetic potential for yield and lower energy requirements, when low amount of feed is available. However, production would increase substantially in cows with high genetic potential when the level of feed is increased.

The objective of the present study was to compare the productive performance of three strains of Holstein-Friesian dairy cows with different genetic potentials for yield, when farmed in a range of pasture-based systems that differed in the amount of feed offered per cow, and to explain the effect on milk production in terms of observations on pasture and supplement consumption and grazing behaviour. Attention is focused on milk production and cow performance in this chapter (Chapter 5), on aspects of forage intake in Chapter 6, and in the influence of grazing behaviour on forage intake in Chapter 7.

The different genetic potential of the strains was indicated by the breeding worth (BW) of the cows in each genotype at the start of the experiment (see Appendix IV). Breeding worth is a profit index that estimates the cows' genetic ability to produce, combined with an economic value, per unit of feed required (Holmes *et al.*, 2002); thus cows with high BW will produce more profit per tonne of dry matter (DM) eaten than cows with low BW.

## **5.2. MATERIALS AND METHODS**

### **5.2.1. System description and management**

This trial was located at Dexcel No. 2 dairy farm, Hamilton, NZ and was designed as a long-term system study starting in 2001 and lasting three seasons. Each season started in June each year and continued until the end of May in the following year with cows grazing outside all year round. Genotypes used are described in detail in Appendix IV. Briefly, the genotypes were high breeding worth (BW) Holstein-Friesian of OS (OS90) or NZ origin (NZ90), and a low BW NZ Friesian genotype representing the cow used in NZ during the 1970s (NZ70).

Two year old cows of the different genotypes were allocated to systems before June 2001 balanced by sire, BW, live weight (LW), calving date for the first season and body condition score (BCS). Cows were maintained in the same system during the entire experiment, except those not pregnant at the end of each season that were culled and replaced by two-year-old cows of the same genotype.

Systems were designed to provide a range of feed allowances (FA) from moderate to generous for each genotype, ranging from 4.5 to 7 t DM per cow per year, which were determined before the start of the experiment and achieved through a combination of stocking rate (SR) and forage plus supplements inputs (see Table 5.1). Each system was then identified as in a range of feeding levels from FA1 (the lowest) and FA4 (the highest), different for each genotype (Table 5.1) according with the amount of feed provided per cow during both the lactation and dry periods,

The planned total nominal amount of feed (NTF<sub>0</sub>) allocated to each feeding level at which each system was managed was determined before the start of the experiment in season 2001 – 2002 by considering the expected herbage annual accumulation rate (HAR) for the farm where the experiment was located (17 t ha<sup>-1</sup>), the different mature live weight and yield potential between genotypes. An increased stocking rate (SR) was utilised for the two NZ genotypes to reduce feed allowance per cow for systems with these genotypes farmed at the lowest FA. The mean NTF<sub>0</sub> at which the genotypes were farmed were different in absolute terms but similar relative to the feed requirements of the mature cows of each genotype, considering the expected differences in adult live weight (McNaughton *et al.*, 2002) and yield potential between genotypes according to their different BW and results from previous studies (Kolver *et al.*, 2000; Kolver *et al.*, 2002).

**Table 5.1: Trial design with actual values of stocking rate and nominal (planned) values of feed allowance in each system.**

	Feeding Systems										
	NZ70			NZ90				OS90			
	FA1	FA3	FA4	FA1	FA2	FA3	FA4	FA1	FA2	FA3	FA4
Comparative feed allowance											
Stocking rate (cow ha <sup>-1</sup> )	3.8	3.1	3.1	3.4	3.1	3.1	3.1	3.1	3.1	3.1	3.1
Pasture offered (P <sub>0</sub> )	4.5	5.5	5.5	5.0	5.5	5.5	5.5	5.5	5.5	5.5	5.5
Supplement offered (NS <sub>0</sub> )			0.5			0.5	1.0		0.5	1.0	1.5
Total feed offered <sup>1</sup> (NTF <sub>0</sub> )	4.5	5.5	6.0	5.0	5.5	6.0	6.5	5.5	6.0	6.5	7.0
Total feed offered <sup>2</sup> (NTF <sub>0</sub> )		5.33			5.75				6.25		

P<sub>0</sub>: nominal pasture offered, NS<sub>0</sub>: nominal supplement offered and NTF<sub>0</sub>: nominal total feed offered per cow, all expressed in t cow<sup>-1</sup> year<sup>-1</sup>. SR: stocking rate. Comparative feed allowance (FA) increased from 1 (lowest) to 4 (highest) according to NTF<sub>0</sub><sup>(1)</sup>. NTF<sub>0</sub> was calculated as P<sub>0</sub> plus NS<sub>0</sub>, and P<sub>0</sub> was estimated from the expected annual herbage accumulation rate (HAR, equal to 17 t DM ha<sup>-1</sup>) and the SR at which each system was farmed. <sup>(2)</sup> Mean NTF<sub>0</sub> of all the systems managed with a same genotype.

Comparative FA increased from 1 (lowest) to 4 (highest) according to NTF<sub>O</sub> (identified as FA1 to FA4). Four feeding levels were allocated to the NZ90 and OS90 genotypes but only three for NZ70 where no system was managed at FA2. Hence, there were eleven systems in the experiment managed as self-contained farmlets, with the amount of feed allocated to each FA compounded by a basal amount of pasture (NP<sub>O</sub>) plus a combination of forage and supplement (NS<sub>O</sub>)(Table 5.1). These levels created a range in NTF<sub>O</sub> (expressed in t cow<sup>-1</sup>year<sup>-1</sup>) for each genotype.

Stocking rate was greater at the lowest FA in the NZ70 and NZ90 genotypes (3.8 and 3.4 cows ha<sup>-1</sup> respectively), and higher in all others systems (3.1 cows ha<sup>-1</sup>). The mean NTF<sub>O</sub> at which each genotype was managed (mean of the three or four FA according to genotype) differed between genotypes (5.33, 5.75 and 6.25 t cow<sup>-1</sup>year<sup>-1</sup> in NZ70, NZ90 and OS90 respectively).

Each farmlet or system was managed separately according to a set of common decision rules (Macdonald & Penno, 1998). Similar decision rules were used during all three seasons of the present experiment except for a change in the drying-off rules used in 2003-04, to allow those cows farmed at the highest FA to be milked for longer by reducing the BCS threshold for drying-off (Macdonald *et al.*, 2005).

All cows were first lactating two year old animals in season 2001-02. Systems had two and three year old cows in 2002-03 and two, three and four year old cows in 2003-04. All the systems were spring calving and feed was based on pasture. Calving was concentrated in late winter, with the mean calving date in the first week of August for all three seasons.

A breeding period of 12 weeks was implemented (from late September to mid December), with the first six weeks of artificial insemination and the last six weeks of bull mating (McNaughton *et al.*, 2003).

As previously mentioned, the second level of FA (FA2) was not represented in the NZ70 due to the reduced number of cows available at the start of the experiment, Additionally, in the first season farmlets of the NZ70 strain had 12 cows each, while both the NZ90 and the OS90 had 17 cows. The number of cows increased in 2002-03 and 2003-04 up to 15 cows per farmlet in NZ70 and 20 cows per farmlet in NZ90 and OS90. In total, the area allocated to the trial was 55.8 ha, subdivided into 138 paddocks with 172 cows in 2001-02 and 65.2 ha subdivided into 161 paddocks with 205 cows in the following two seasons.

The experimental area was flat and uniform with pastures of approximately similar age and composition. Paddocks were allocated at random to systems balanced by soil type

and distance to the milking shed before the experiment started, and then remained in the same treatment until the end of the experiment. Within this area, paddocks were allocated at random to each farmlet to ensure that soil type, pasture condition and distance to the dairy shed were balanced between farmlets.

Pasture composition was similar in all the paddocks with a mixture of ryegrass – white clover (white clover below 20%) and a proportion of dallisgrass growing during the summer period (L'Huillier, 1987; Thom, 1991). Paddocks were allocated to each farmlet (treatment) before the start of the experiment and were grazed by the assigned system throughout the experiment.

During the experiment, cows were milked twice a day during the whole lactation, offered a new strip of pasture each morning after the morning milking and returned to the same paddock after the afternoon milking. The mean distance from the paddocks to the milking shed was about 1 km, hence cows had to walk this distance four times each day.

Systems were fed pasture silage when required during the experimental period (subject to pasture silage availability), harvested from the same farmlet. In addition to pasture silage, systems managed at high FA (FA4 in NZ70 and FA3 and FA4 in both NZ90 and OS90 genotypes) were fed maize grain, maize silage or a combination of both. The amount of extra feed offered increased with genotype requirements, thus the lowest amount of supplement was fed to NZ70, intermediate to NZ90 and the highest to OS90. Systems farmed at increased FA were fed a greater amount of extra feed per cow daily and managed in a shorter rotation length. Winter supplementation increased in 2003-04 compared to 2002-03, especially for the OS90 genotype, and the strategy of supplementation also changed in mid and late lactation between these two seasons (see Appendix V-15, V-16 & V-17). For instance, maize grain and maize silage were fed to high FA systems in mid and late lactation in 2002-03 respectively whilst a proportion of the total supplement was also fed during the dry period in 2003-04 and hence the amount fed in mid and late lactation reduced accordingly. Only those systems of each genotype farmed at the highest FA (FA4) were fed maize silage plus maize grain. In mid lactation 2002-03 maize grains were fed alone, in individual buckets during each milking. Subsequently, maize silage alone or with maize grain added was offered, mixed when fed together and brought to each paddock in fibreglass trolley troughs, after the morning milking. When pasture growth rate exceeded the daily requirements of the herd, bales of pasture silage were made from each farmlet. Bales were weighed and kept separately for each farmlet, to be fed back later to the farmlet from where bales had been made. Pasture silage was fed in the paddock on the ground.

### 5.2.2. Pasture measurements

Herbage mass was measured by visual scoring of all the paddocks in each farmlet once weekly (Campbell & Arnold, 1973; Baars & Dyson, 1981). This provided a measure of the amount of feed available in the systems at one time during each week over the season. A team of four observers visually calibrated 11 quadrats weekly (each covering an area of 0.33 m<sup>2</sup>) randomly covering the range of conditions of the sward in two different paddocks, four quadrats in a recently grazed paddock and seven in another area which was about to be grazed. Each observer assessed visually all the quadrats and assigned a 'visual point score' to each (1 visual point score equal to 300 kg DM ha<sup>-1</sup>).

After a first calibration of all the quadrats, the observers walked across every paddock of each farmlet and recorded a single visual score for each paddock. Two observers assessed half of the total area of the farmlets in one week and the other half in the following week in order to balance the effect of the observer between paddocks (and farmlets) over time. After all the paddocks had been scored, the observers returned to the calibration area to score all the quadrats again, then the herbage in each quadrat was cut to ground level and collected, then washed in the laboratory, oven dried (at 60°C for 48 hours) and weighed. The relationship between the average score of the quadrats (all the observers) and the dry matter weight of the material collected in each quadrat was estimated and subsequently used to transform the visual score from each paddock to a value of dry matter per hectare. This allowed a weekly correction of the visual scores obtained each week.

The weekly pasture cover (kg DM ha<sup>-1</sup>) and the actual weekly HAR (kg DM ha<sup>-1</sup>day<sup>-1</sup>) were estimated from the individual visual score of each paddock. The cover for each farmlet was calculated as the average cover of all the paddocks in each farmlet, except those being grazed. The mean change in cover of the non-grazed paddocks, from visual scoring on consecutive weeks, was used to estimate the average HAR during the preceding week assuming a linear accumulation rate. The mean weekly values for cover, HAR, herbage mass (HM) pre-grazing (HM<sub>PRE</sub>) and herbage mass post-grazing (HM<sub>POST</sub>) for each system were used to calculate the monthly estimates. Weekly HAR values were used to estimate the monthly and annual HAR for each system.

Once a week a hand-plucked pasture sample was collected from the grazing stratum of one pre-grazing paddock representing the quality of the pasture consumed on that date; in addition a weekly sample of the supplement fed to each herd that week was also collected from a trough or bale of silage. Weekly samples were bulked monthly within each farmlet, then oven dried (at 60°C for 48 hours) and analysed by Near Infrared Reflectance Spectroscopy (NIRS)(Shenk & Westerhaus, 1994).

### 5.2.3. Pasture and supplement dry matter intakes

The same observers also used the visual scoring system to assess the amount of  $HM_{PRE}$  and  $HM_{POST}$  in the same paddock (different strips each day) on each farmlet three times a week. The difference between the visual scores obtained in these paddocks was transformed to DM values per hectare using the calibrated equation estimated for that week. The difference between  $HM_{PRE}$  and  $HM_{POST}$  for the same paddock was assumed to be the amount of herbage removed by the herd during that grazing. The mean herbage removed by the same herd from different paddocks on three consecutive days weekly, expressed per cow, was considered to be the value of the apparent mean daily dry matter intake that week ( $DMI_V$ ).

The amount of pasture silage and maize silage fed to the different systems were weighed and samples collected for DM determination; the actual total amount of pasture silage and maize silage fed to the cows ( $TS_O$ ) was expressed on DM basis. Average wastage values of 0.2 and 0.1 were considered for grass silage and maize silage respectively during the whole study, to estimate the apparent amount of each supplement consumed. These values were visually estimated from different dates and agreed with on farm estimates. This amount of supplement consumed was added to the  $DMI_V$  value to obtain an estimate of the mean total DM consumed per cow in each system, each season ( $DMI_T$ ).

### 5.2.4. Milk yield, live weight and body condition score

Milk volume per cow was measured once a week on two consecutive milkings (afternoon and the following morning). A 30 ml aliquot from a representative sub-sample of approximately 2.5% of the total volume of each cow's milk was analysed at Dexcel Milk Laboratory to determine fat, protein and lactose concentrations using a calibrated Fossomatic milk-o-scan (FT120; Foss Electric, Hillerod, Denmark). The mean daily yields of milk, fat, protein and lactose were estimated weekly for each cow; the total yield of milk and milksolids (MS) per cow and farmlet were calculated at the end of each season. The lactation length (DIM: days in milk) of each cow was also calculated. Once a week (always on the same day) each cow was weighed with a calibrated Tru-test<sup>TM</sup> electronic scale and body condition scored by one experienced observer after the morning milking (1-10 point scale (Macdonald & Roche, 2004)).

### 5.2.5. Additional calculations

The current annual HAR for each system minus the grass silage conserved was considered the amount of pasture DM offered per hectare to each farmlet [ $P_O = \Sigma$  of monthly average HAR from Table 5.2, considering monthly changes from Figure 5.1,

plus  $TS_0$ ]. This, plus the pasture and maize silage brought into each system ( $TS_0$ ), accounted for the total annual amount of feed offered to each system ( $TF_0$ ), and expressed per cow. The actual value of the ‘comparative stocking rate’ (CSR), expressed as kg of LW per ton of DM available, was calculated for all the systems each season (Penno, 1999).

Pasture utilisation (PU) was calculated as the mean apparent daily pasture consumed per hectare by each herd from pre-post grazing visual scores each week, accumulated for the whole season ( $DMI_V$  multiplied by SR) and expressed as a proportion of  $P_0$  (deducting the amount of grass silage conserved).

The weekly estimates of daily yield were used to estimate total lactation yields. The mean LW and BCS at the start of the season, at one and four weeks after calving, at nadir, at the dry-off date and at the end of the season were estimated for each cow in all the systems from the individual weekly records of LW and BCS. The LW and BCS lost between the first week after calving and nadir was estimated as the mean difference in LW and BCS of all the cows in each system. The length of lactation or DIM was calculated as the interval between calving and drying-off.

#### **5.2.6. Statistical analysis**

Systems were not replicated in this study due to the large amount of resources that this would have required; instead large treatments groups representing real farm conditions were utilised. Nevertheless, this approach would include interactions between cows within farmlets so large that edge effects were avoided (Morley & Spedding, 1968). Considering the importance of the practical implication of these results to NZ dairy farmers, the need to measure the response of the different genotypes to increased levels of feed in the system was more important than the need to replicate each system at the expense of reducing the number of feeding levels (Bransby, 1989).

The experimental site (farm) was considered to be relatively uniform at the start of the experiment. Furthermore paddocks within the farm had similar pasture species composition and were allocated at random to the different systems in a balanced design that accounted for differences in soil type and distance from the milking shed; hence it could be argued that the variations between paddocks and systems at the start of the study were very low.

In contrast, greater variability would be expected between cows. Within strains, cows were allocated at random to systems managed at different FA but balanced by sire, breeding worth, pre-experimental LW and BCS. It was assumed that similar between-cow

variation existed within farmlets and that the use of individual animals as the experimental unit in the statistical analysis would not affect the main conclusions drawn from these results (Linnane et al., 2004).

**Table 5.2: Mean values of daily herbage accumulation rate, pre- and post-grazing herbage mass and herbage composition genotype for each genotype and feed allowance, in each season.**

	Genotype			Feeding Allowance				Significance					
	NZ70	NZ90	OS90	Sed	FA1	FA2	FA3	FA4	SED	GE	FA	GE*FA	
Season 2001 – 2002	HAR (kg DM ha <sup>-1</sup> day <sup>-1</sup> )	47.5	47.0	45.9	1.86	47.5	48.4	45.1	46.2	2.22	NS	NS	NS
	Pre-grazing HM (kg DM ha <sup>-1</sup> )	3465	3463	3499	39.82	3435	3484	3470	3513	47.49	NS	NS	NS
	Post-grazing HM (kg DM ha <sup>-1</sup> )	2120	2068	2131	23.8	2042	2105	2123	2156	28.4	NS	NS	NS
	CP (g kg <sup>-1</sup> DM)	234	233	233	1.01	228	245	233	227	1.19	NS	NS	NS
	Lipids (g kg <sup>-1</sup> DM)	42.0	41.7	41.9	0.32	42.0	42.4	41.5	41.6	0.38	NS	NS	NS
	ADF (g kg <sup>-1</sup> DM)	227	224	224	5.84	223	216	230	231	6.96	NS	NS	NS
	NDF (g kg <sup>-1</sup> DM)	416	418	419	12.1	419	398	425	428	14.4	NS	NS	NS
	ME (MJ kg <sup>-1</sup> DM)	11.2	11.3	11.3	0.15	11.3	11.4	11.1	11.1	0.17	NS	NS	NS
Season 2002 – 2003	HAR (kg DM ha <sup>-1</sup> day <sup>-1</sup> )	54.1	53.1	52.1	1.45	52.3	55.1	51.9	53.0	1.73	NS	NS	NS
	Pre-grazing HM (kg DM ha <sup>-1</sup> )	3175	3122	3113	51.4	3126	3175	3139	3108	61.3	NS	NS	NS
	Post-grazing HM (kg DM ha <sup>-1</sup> )	1840	1823	1858	38.13	1773	1793	1863	1932	45.47	NS	NS	NS
	CP (g kg <sup>-1</sup> DM)	227	229	233	2.84	234	222	227	235	3.39	NS	NS	NS
	Lipids (g kg <sup>-1</sup> DM)	41.3	41.6	41.5	0.39	42.3	41.3	41.4	41.0	0.46	NS	NS	NS
	ADF (g kg <sup>-1</sup> DM)	229	226	227	2.82	219	229	229	231	3.36	NS	NS	NS
	NDF (g kg <sup>-1</sup> DM)	409	410	402	4.16	393	414	413	409	4.96	NS	NS	NS
	ME (MJ kg <sup>-1</sup> DM)	11.3	11.3	11.3	0.11	11.5	11.3	11.3	11.2	0.13	NS	NS	NS
Season 2003 – 2004	HAR (kg DM ha <sup>-1</sup> day <sup>-1</sup> )	52.6	52.9	51.9	1.34	51.1	53.0	52.1	53.7	1.60	NS	NS	NS
	Pre-grazing HM (kg DM ha <sup>-1</sup> )	3106	3069	3133	40.4	3150	3201	3046	3014	48.2	NS	0.08	NS
	Post-grazing HM (kg DM ha <sup>-1</sup> )	1864	1833	1925	60.6	1776	1851	1910	1960	72.3	NS	NS	NS
	CP (g kg <sup>-1</sup> DM)	243	244	249	4.23	240	232	257	252	5.04	NS	*	NS
	Lipids (g kg <sup>-1</sup> DM)	43.7	44.3	44.0	0.42	43.8	43.7	44.5	44.1	0.50	NS	NS	NS
	ADF (g kg <sup>-1</sup> DM)	223	219	218	4.08	222	221	218	219	4.86	NS	NS	NS
	NDF (g kg <sup>-1</sup> DM)	403	399	396	7.81	400	401	398	398	9.31	NS	NS	NS
	ME (MJ ME kg <sup>-1</sup> DM)	11.7	11.8	11.8	0.89	11.6	11.7	11.8	11.7	0.11	NS	NS	NS

HAR: mean daily herbage accumulation rate; HM: herbage mass; CP: crude protein; ADF: acid detergent fibre; NDF: neutral detergent fibre; ME: metabolisable energy. The mean daily HAR for each month was calculated from the weekly visual scores of all the paddocks in each system. Pre- and post-grazing herbage mass was estimated from visual scores. GE: genotype; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.

The year-to-year carryover effects were also important for the outcome of this study as they represented the long-term effect of using different genotypes in a pasture-based system under NZ conditions; hence cows that were not culled at the end of one season were maintained in the same farmlet for the following year. As a result, only the replacement heifers were allocated at random, within strains, to systems at the start of the second and third seasons. Systems differences due to level of feed allowance are expected to increase with seasons as result of the carryover effect of the level of feed allowance on performance per cow.

The monthly values of HAR, mean pasture cover, HM<sub>PRE</sub> and HM<sub>POST</sub> and those representing the mean quality of the pasture in each system for the complete experimental period were analysed as repeated measurements using the MIXED model procedure (SAS, 2002) with compound symmetry for the covariance structure of the data set.

As systems were not replicated, results of total lactation yield of MS per hectare, CSR, the amount of pasture and supplements offered, pasture utilization and  $DMI_V$  were analysed considering a linear effect of FA by using the statistical procedures of SAS (SAS, 2002) as a MIXED model (PROX MIXED) [ $Y_{ijk} = \mu + GE_i + \beta(FA)_j + e_{ijk}$ ], with the additive effect of genotype (GE) and a linear trend of feed allowance (FA).

All the data collected individually for each cow in the systems (yields and milk composition and DIM) were analysed utilising the statistical procedures of SAS (SAS, 2002) as a MIXED model (PROX MIXED) [ $Y_{ijkl} = \mu + GE_i + P_j + FA_k + (GE \times FA) + e_{ijkl}$ ], with the additive effects of genotype (GE), FA, parity (P or age) and genotype by feeding allowance interactions (GE by FA) as fixed effects, and the individual cow within system as a random effect.

Genotypes were compared at the mean nominal FA at which they were farmed, while feeding levels were compared at the mean values for the three genotypes (e.g. FA1 is the average of the values obtained in systems farmed with different genotypes at comparable nominal FA equal to 5 t/cow per year, and FA4 is the average of the values obtained at the highest feeding level, equal to 6.5 t cow<sup>-1</sup>year<sup>-1</sup>). As the experimental design was unbalanced, not all the levels of feed were represented in the three genotypes, hence data for FA2 is just the mean for NZ90 and OS90 (FA2 equal to 5.5 t/cow per year).

## 5.3. RESULTS

### 5.3.1. Grazing conditions and supplement offered

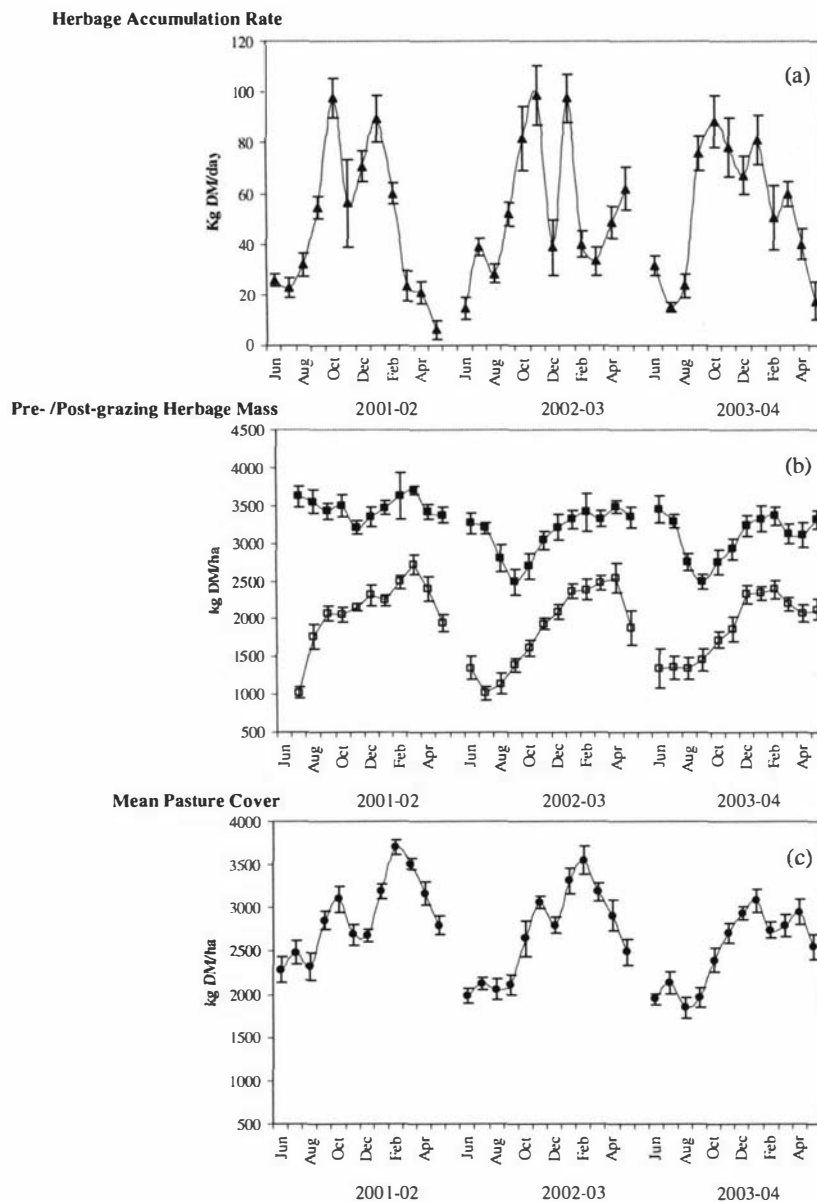
Management differed between systems according to the different FA at which the systems were farmed. Differences involved changes in the area grazed per day, thus in rotation length and instantaneous SR that might be the result of differences in the mean weekly HAR of the farmlets in relation to the amount of extra feed available. The mean values for the conditions of the pasture grazed across strains and feeding levels, and for the supplements fed, are presented in Tables 5.2 and 5.3 respectively.

**Table 5.3: Mean values for composition of the forage and supplements used.**

	n	CP		ADF		NDF		OMD		ME	
		g kg <sup>-1</sup> DM	se	g kg <sup>-1</sup> DM	se	g kg <sup>-1</sup> DM	se	g kg <sup>-1</sup> DM	se	MJ ME kg <sup>-1</sup> DM	se
Pasture silage	22	159	4.8	319	6.4	555	10.8	705	4.9	11.29	0.08
Maize grain	2	78	2.4	29.3	2.2	114	13.2	----	----	13.8	----
Maize silage	14	69	1.2	221	9.3	397	13.9	764	13.4	10.66	0.04
Maize silage + grain	16	75	0.9	167	4.5	327	6.5	813	16.7	----	----

CP: crude protein; ADF: acid detergent fibre; NDF: neutral detergent fibre; OMD: *in-vitro* organic matter digestibility; ME: metabolisable energy. SE: standard error.

**Figure 5.1: Mean monthly values for herbage accumulation rates (a), pre- and post-grazing herbage mass (b) and pasture cover (c) in seasons 2001-02, 2002-03 and 2003-04, across systems managed with three different Holstein-Friesian genotypes.**



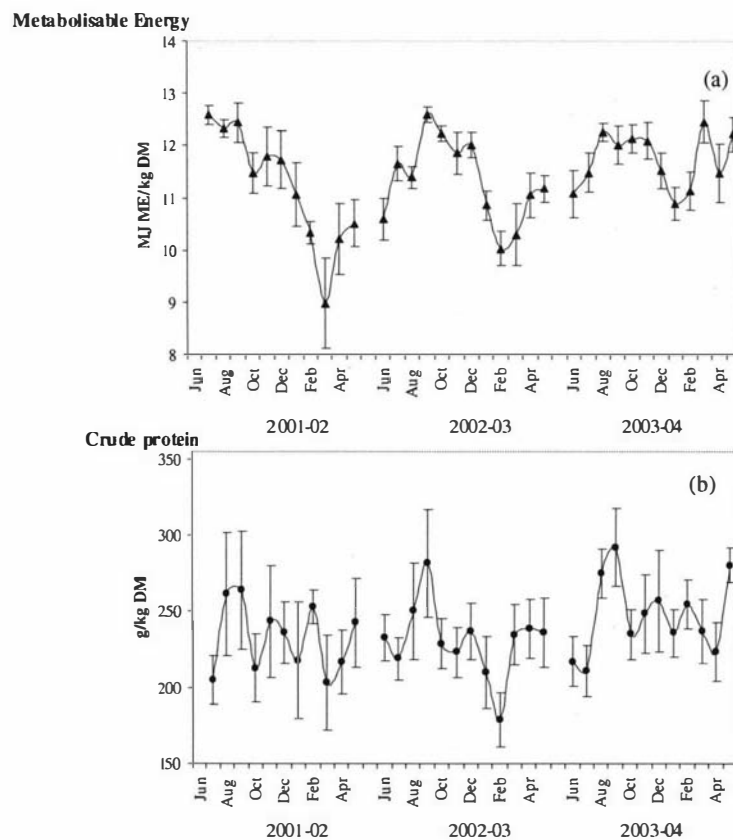
(a) HAR: herbage accumulation rate ( $\blacktriangle$ ) ( $\text{kg DM ha}^{-1}\text{day}^{-1}$ ); (b) Pre-grazing herbage mass ( $\blacksquare$ ) and post-grazing herbage mass ( $\square$ ) ( $\text{kg DM ha}^{-1}$ ) were; (c) Cover ( $\bullet$ ) ( $\text{kg DM ha}^{-1}$ ) (paddocks being grazed were not considered). Bars indicate standard deviation.

The mean annual HAR,  $\text{HM}_{\text{PRE}}$  and  $\text{HM}_{\text{POST}}$  were similar across GE and FA over the three seasons (Table 5.2). Differences in the mean daily HAR for each season were not significant across GE and FA, about  $51 \text{ kg DM ha}^{-1} \text{ day}^{-1}$  and equivalent to  $18.6 \text{ t DM ha}^{-1} \text{ year}^{-1}$  (range from  $46$  to  $54 \text{ kg DM ha}^{-1} \text{ day}^{-1}$  and annual HAR between  $16.8$  to  $19.7 \text{ t DM}$

ha<sup>-1</sup>) with a greater variation across GE and FA during season 2001-02 than in the other two seasons.

The highest daily HAR was observed in spring (75-110 kg DM ha<sup>-1</sup> day<sup>-1</sup>) and mid summer and the lowest was observed during winter (5-20 kg DM ha<sup>-1</sup> day<sup>-1</sup>; Figure 5.1a). A decrease in the daily HAR usually occurred at the end of the spring (greater in 2002-03; Figure 5.1a) and in February (greater in 2001-02), both attributed to dry conditions at these times.

**Figure 5.2: Mean values for the monthly concentration of metabolisable energy (a) and crude protein during seasons 2001-02, 2002-03 and 2003-04, across systems managed with three different Holstein-Friesian genotypes..**



(a) Concentration of metabolisable energy (▲) (MJ ME kg<sup>-1</sup> DM) and (b) crude protein (●) (g kg<sup>-1</sup> DM). Bars indicate standard deviation.

Similar patterns for daily HAR (mean for each month), HM<sub>PRE</sub> and HM<sub>POST</sub> in each season were recorded across GE (Figures 5.1a and 5.1b) and FA (data not presented). The values for HAR and HM<sub>POST</sub> were more stable than HM<sub>PRE</sub> each year (Figures 5.1a and 5.1b). A higher HM<sub>PRE</sub> at the start of the spring was observed in season 2001-02 (3,200 kg DM ha<sup>-1</sup>; Figure 5.1b) than in seasons 2002-03 and 2003-04 (2,500 kg DM ha<sup>-1</sup>). This

was in agreement with the higher mean  $HM_{PRE}$  recorded in 2001-02 (3,500 kg DM ha<sup>-1</sup>) and  $HM_{POST}$  (2,100 kg DM ha<sup>-1</sup>) compared with the trend for lower  $HM_{PRE}$  and  $HM_{POST}$  observed in the following two seasons (Table 5.2 and Figure 5.1b).

The average amount of herbage removed at each grazing was approximately 1,400 kg DM ha<sup>-1</sup> in season 2001-02, and 1,230 during 2002-03 and 2003-04. The lowest amount of herbage removed per hectare was recorded at the end of the summer (late lactation) just before an increased proportion of the cows were dried-off, while the highest amount of herbage removed was recorded during the winter period with dry cows.

The average quality of the pasture grazed was similar in all the systems (Table 5.2) with a mean value of 11.3 MJ ME kg<sup>-1</sup>DM in seasons 2001-02 and 2002-03 and a slightly higher in 2003-04. The lowest energetic value of the herbage was recorded in February each year with the lowest value in February 2002, however quality improved during the autumn-winter period to decrease again from October onwards (Figure 5.2a).

**Table 5.4: Mean values for each genotype across feed allowance and rate of change between feeding allowances for total milksolids yield, comparative stocking rate, actual amount of pasture and supplements offered per cow, pasture utilisation and total intakes per cow for three Holstein-Friesian genotypes managed in three or four feeding systems over three seasons.**

	Genotypes				Feed allowance		Significance		
	NZ70	NZ90	OS90	Sed	Slope	Sed	GE	FA	GE*FA
MS per hectare (kg ha <sup>-1</sup> year <sup>-1</sup> )	1088	1237	1155	49.92	67.10	34.96	0.061	0.096	0.069
MS per cow (kg cow <sup>-1</sup> year <sup>-1</sup> )	333	395	377	8.22	43.43	5.76	***	***	NS
CSR (kg t <sup>-1</sup> DM)	79.12	76.29	77.00	2.19	-7.07	1.53	NS	**	NS
TF <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.92	6.38	6.54	0.13	0.74	0.09	**	***	NS
P <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.60	5.88	6.15	0.39	0.28	0.28	NS	NS	NS
TS <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	0.32	0.50	0.64	0.10	0.40	0.07	*	***	0.051
Pasture utilisation (PU)	0.80	0.79	0.82	0.03	0.00	0.02	NS	NS	NS
DML <sub>v</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	4.68	4.98	5.33	0.11	0.63	0.07	**	***	NS

MS: milksolids; CSR: comparative stocking rate; TF<sub>0</sub>: total feed offered; P<sub>0</sub>: annual pasture offered (DM); TS<sub>0</sub>: total supplement offered including pasture silage; PU: pasture utilization; DML<sub>v</sub>: dry matter intake from visual scoring. The 'slope' indicates the mean rate of change of each variable (considering all the systems) per tonne of 'nominal' feed allowance per cow per annum (NTF<sub>0</sub> from Table 5.1). CSR: Comparative stocking rate (kg LW t<sup>-1</sup>DM of TFO.). GE: genotype; FA: annual feed allowance; GE\*FA: genotype by feed allowance interaction. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = not significant.

The average content of protein in the herbage (CP) was 231 g kg<sup>-1</sup>DM in seasons 2001-02 and 2002-03 and slightly higher in 2003-04 (Table 5.2). Seasonal changes in CP tended to follow the same pattern recorded for ME, especially during 2002-03 (Figure 5.2b).

### 5.3.2. Amount of feed offered and consumed

The TF<sub>0</sub> during the whole experiment (Tables 5.4 and 5.5) was significantly different between genotypes and above that planned for all three strains; it increased significantly

with FA by about 0.74 t DM t<sup>-1</sup>DM of NTF<sub>0</sub>. The range of TF<sub>0</sub> was narrowed than planned during the whole experimental period, however, the differences between TF<sub>0</sub> in all the genotypes were smaller in 2001-02, increased in 2002-03 and increased further in 2003-04, to be closer to the range planned (NTF<sub>0</sub>).

The P<sub>0</sub> to all the genotypes was slightly above that planned due to a higher HAR than expected. Differences in P<sub>0</sub> across genotypes were not significant, although numerically lower for NZ70 and increased with feeding levels. The TS<sub>0</sub> was above that planned for both NZ genotypes but slightly below that planned for OS90 (pasture silage was also considered). Differences in TS<sub>0</sub> across genotypes were, however, in agreement with the design of the experiment.

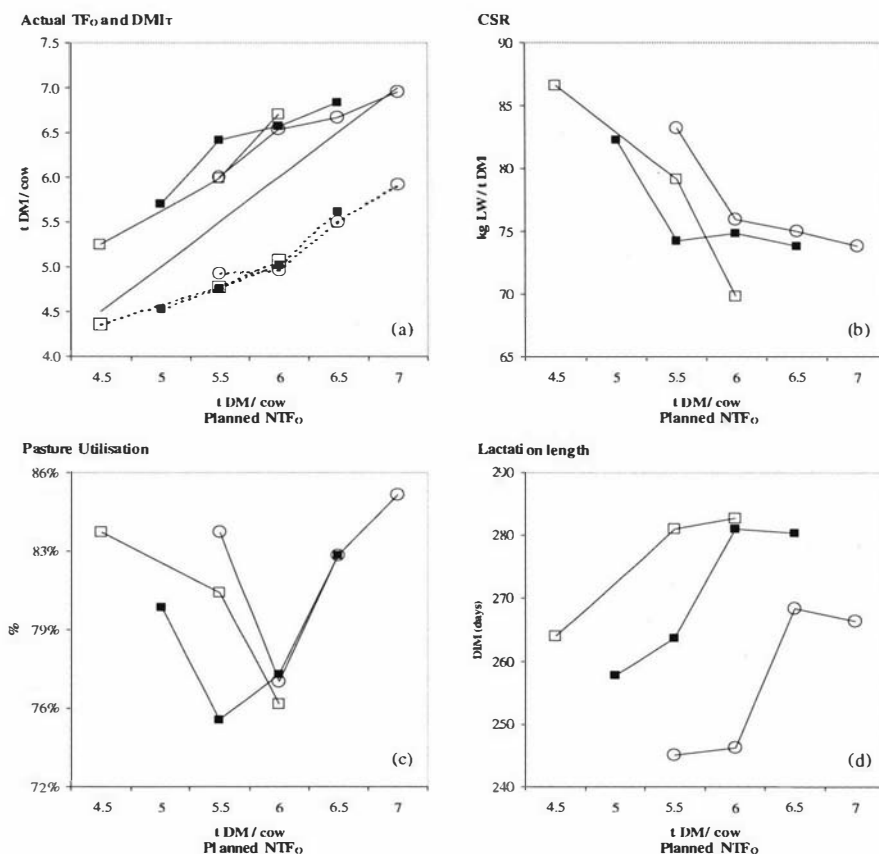
**Table 5.5: Mean values for total milksolids production, comparative stocking rate, actual amount of pasture and supplements offered per cow, pasture utilisation and total intake per cow for each Holstein-Friesian genotype and feed allowance in each season.**

		Feeding Systems											
		NZ70			NZ90				OS90				
		FA1	FA2	FA3	FA1	FA2	FA3	FA4	FA1	FA2	FA3	FA4	
Season 2001 - 2002	SR (cow ha <sup>-1</sup> )	3.8	3.1	3.1	3.4	3.1	3.1	3.1	3.1	3.1	3.1	3.1	
	Annual feeding allowance (t DM cow <sup>-1</sup> year <sup>-1</sup> )	4.5	5.5	6.0	5.0	5.5	6.0	6.5	5.5	6.0	6.5	7.0	
	Comparative feeding allowance	FA1	FA2	FA3	FA1	FA2	FA3	FA4	FA1	FA2	FA3	FA4	
	MS per hectare (kg ha <sup>-1</sup> year <sup>-1</sup> )	996	859	872	1021	945	928	947	857	853	895	889	
	MS per cow (kg cow <sup>-1</sup> year <sup>-1</sup> )	268	289	294	292	315	309	316	286	284	298	296	
	CSR <sub>C</sub> (kg t <sup>-1</sup> DM)	82.8	51.5	71.9	84.2	75.3	76.8	76.1	81.8	75.4	74.9	78.3	
	TF <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.25	5.53	6.16	5.28	6.11	5.98	6.11	5.75	6.23	6.23	6.02	
	P <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	4.80	5.10	5.71	4.91	5.65	5.45	5.53	5.30	5.74	5.55	5.27	
	TS <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> ) [PS included]	0.45	0.42	0.45	0.37	0.46	0.53	0.58	0.45	0.49	0.68	0.75	
	Pasture utilisation (PU)	0.84	0.90	0.81	0.83	0.83	0.84	0.86	0.89	0.79	0.82	0.87	
DML <sub>V</sub> (t cow <sup>-1</sup> year <sup>-1</sup> )	4.38	4.88	4.92	4.35	4.98	4.90	5.14	5.01	4.87	5.08	5.16		
Season 2002 - 2003	MS per hectare (kg ha <sup>-1</sup> year <sup>-1</sup> )	1137	1088	1087	1165	1179	1301	1410	1090	1110	1269	1271	
	MS per cow (kg cow <sup>-1</sup> year <sup>-1</sup> )	306	352	352	353	381	421	456	353	359	411	411	
	CSR <sub>C</sub> (kg t <sup>-1</sup> DM)	82.8	74.2	66.9	77.7	72.7	71.1	68.0	80.9	68.6	72.6	67.2	
	TF <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.34	6.22	6.99	5.94	6.42	6.66	7.26	5.99	6.88	6.66	7.41	
	P <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.30	6.08	6.40	5.81	6.23	6.36	6.19	5.94	6.63	5.85	6.13	
	TS <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> ) [PS included]	0.04	0.14	0.59	0.14	0.19	0.31	1.08	0.05	0.25	0.80	1.27	
	Pasture utilisation (PU)	0.80	0.73	0.72	0.76	0.69	0.72	0.79	0.78	0.69	0.84	0.83	
	DML <sub>V</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	4.28	4.56	5.14	4.47	4.44	4.82	5.86	4.63	4.75	5.60	6.25	
	MS per hectare (kg ha <sup>-1</sup> year <sup>-1</sup> )	1263	1275	1268	1434	1348	1494	1668	1244	1280	1544	1558	
	MS per cow (kg cow <sup>-1</sup> year <sup>-1</sup> )	340	413	411	434	436	483	540	403	414	500	504	
CSR <sub>C</sub> (kg t <sup>-1</sup> DM)	94.2	81.8	70.6	85.1	74.8	76.6	77.3	86.8	83.9	77.5	75.9		
TF <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.17	6.21	6.94	5.87	6.71	7.06	7.11	6.25	6.48	7.11	7.42		
P <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	5.03	5.80	6.41	5.62	6.31	6.34	6.18	5.94	9.05	6.15	6.18		
TS <sub>0</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> ) [PS included]	0.14	0.41	0.53	0.25	0.41	0.72	0.93	0.30	0.43	0.96	1.24		
Pasture utilisation (PU)	0.86	0.79	0.74	0.81	0.73	0.75	0.82	0.83	0.82	0.81	0.85		
DML <sub>V</sub> (t DM cow <sup>-1</sup> year <sup>-1</sup> )	4.43	4.89	5.16	4.74	4.87	5.34	5.87	5.14	5.27	5.84	6.35		

GE: genotype; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]. CSR: Comparative stocking rate (kg LW t<sup>-1</sup> DM of TF<sub>0</sub>). Annual pasture offered (P<sub>0</sub>) was calculated as the Σ of monthly HAR (monthly average HAR from Table 5.2 and monthly changes (Figure 5.1). Pasture silage (PS). Pasture utilization (PU) estimated as pasture DM intake divided by the annual HAR, and pasture DM intake calculated from the weekly pre-post grazing visual scores. Apparent total DM intakes (DML<sub>V</sub>) includes pasture plus supplement, supplement intake estimated as supplement offered (weighed and expressed as dry matter) minus wastage (20% and 10% for grass silage and maize respectively).

No significant differences in pasture utilisation were measured between genotypes, or between FA, although it was slightly higher in the OS90 strain. The lowest total  $DMI_T$  was recorded for NZ70 cows ( $4.68 \text{ t DM cow}^{-1}\text{year}^{-1}$ ) and the highest for the OS90 genotype ( $5.33 \text{ t DM cow}^{-1}\text{year}^{-1}$ ), with  $DMI_T$  for the NZ90 ( $4.98 \text{ t DM cow}^{-1}\text{year}^{-1}$ ) being higher and lower than NZ70 and OS90 respectively.

**Figure 5.3:** Mean values for the seasonal total dry matter offered and consumed per annum (a), comparative stocking rate (b) and proportion of the total amount of feed offered currently utilised (c) of each Holstein-Friesian genotype at the different feed allowances (mean of three seasons).



NZ70 (□), NZ90 (■), OS90 (○). NTF<sub>0</sub>: Nominal (planned) total feed offered (FA); CSR: comparative stocking rate; TFO: actual total feed offered; PU: pasture utilization; DMI<sub>T</sub>: dry matter intake from visual scoring (pasture plus supplement). TFO was calculated as P<sub>0</sub> [as the Σ of monthly herbage accumulation rate (HAR from Table 5.2 and considering monthly changes from Figure 5.1)] plus TS<sub>0</sub>. Pasture utilization (PU) estimated as pasture DM intake % annual HAR (pasture silage conserved was deducted). In (a) TFO (solid lines) and DMI<sub>T</sub> (light dotted lines), line for equal TFO equal to NTF<sub>0</sub> is also shown. In all figures the 'x' axis indicates the value of NTF<sub>0</sub> at which the systems were managed, expressed in tonnes of DM per cow per year.

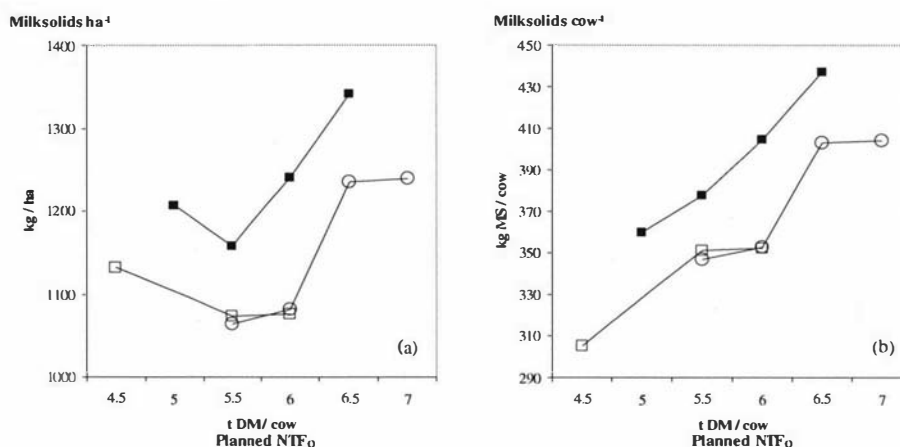
The estimated  $DMI_T$  increased about  $0.63 \text{ t DM t}^{-1}\text{DM}$  of NTF<sub>0</sub> allocated to the systems (Table 5.4). If it is considered that the mean actual increase in TFO was just  $0.74 \text{ t DM t}^{-1}\text{DM}$  of NTF<sub>0</sub>, this indicates that 85% of the feed offered to each cow was consumed. The  $DMI_T$  increased from the first to the third seasons probably as a result of the increased age, yield and size of the cows in the systems. Differences in CSR ( $\text{kg LW t}^{-1}$

DM) across genotypes were not significant; however, the CSR decreased significantly for each genotype as feed allowance increased (Table 5.4 and Figure 5.3b).

### 5.3.3. Systems results

The mean MS yield per hectare in all feeding levels (mean of three seasons) was greatest for NZ90, intermediate for OS90 and lowest for NZ70 (1,237, 1,155 and 1,088 kg MS ha<sup>-1</sup> respectively) and showed a trend to improve with feeding level at a rate of 91 kg MS ha<sup>-1</sup> t<sup>-1</sup>DM of TF<sub>0</sub> per cow (67 kg MS ha<sup>-1</sup>t<sup>-1</sup>DM of NTF<sub>0</sub>; Table 5.4), particularly for NZ90 and OS90 (Figure 5.4a).

**Figure 5.4: Mean values for the seasonal milksolids yield per hectare (a) and per cow (b) of each Holstein-Friesian genotype at the different feed allowances (mean of three seasons).**



NZ70 (□), NZ90 (■), OS90 (○). In all figures the 'x' axis indicates the value of NTF<sub>0</sub> at which the systems were managed, expressed in tonnes of DM per cow per year. NTF<sub>0</sub>: Nominal (planned) total feed offered (feed allowance).

However, strains were managed at a slightly different mean SR, because systems managed with the NZ70 and NZ90 genotypes at the lowest feeding level (FA1) had 3.8 and 3.4 cows ha<sup>-1</sup> respectively while all the other systems were managed at 3.1 cows ha<sup>-1</sup>, so the mean SR at which the genotypes were farmed was slightly different (3.3, 3.2 and 3.1 cows ha<sup>-1</sup> for NZ70, NZ90 and OS90 respectively). The mean MS yield per cow was higher for NZ90, intermediate for OS90 and lower for NZ70 (395, 377 and 333 kg cow<sup>-1</sup> respectively) and also increased with feeding allowance at a rate of 59 kg MS cow<sup>-1</sup>t<sup>-1</sup> DM of TF<sub>0</sub> per cow (43 kg MS cow<sup>-1</sup>t<sup>-1</sup> DM of NTF<sub>0</sub>; Table 5.4). Thus, systems managed with the two NZ genotypes at the higher SR but low feeding level per cow improved yield per hectare (Figure 5.4a) even though they had lower yields per cow (Figure 5.4b).

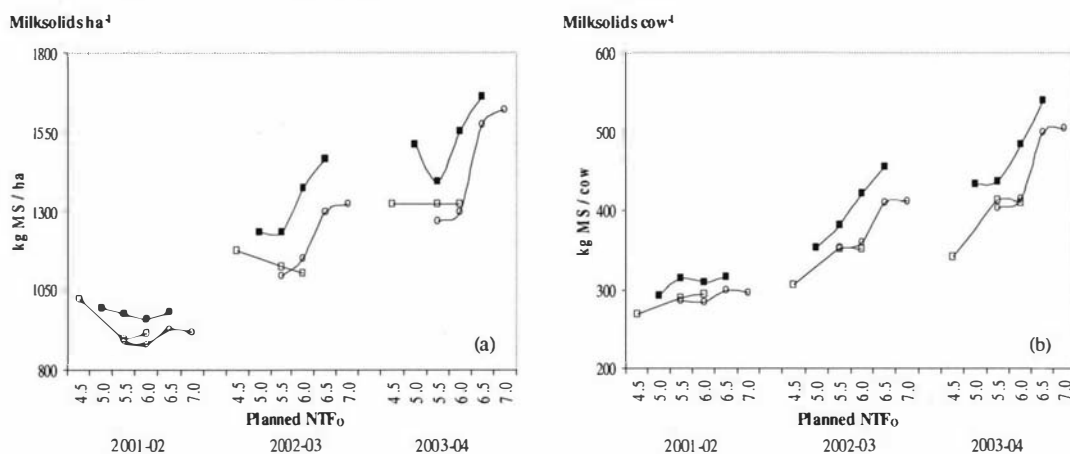
Yield per hectare and per cow increased from the first to the third season (Figures 5.5a and 5.5b) supported by both the increased mean age of the cows in the systems and  $TF_0$ . In the last season of the study yield per hectare for the NZ90 was above that achieved with the OS90 and NZ70 genotypes at comparative FA (see Table 5.1 for the comparative levels of feed between genotypes), in addition, greater for OS90 than NZ70 at highest FA but similar at the lowest FA due to the higher SR of the NZ70 at FA1, despite the differences observed in yield per cow at this level of feed.

### 5.3.4. Milk yield and composition

#### 5.3.4.1. Total lactation yield per cow

During 2001-02 MS yield was higher for NZ90 than for the OS90 and NZ70 genotypes and slightly higher for the OS90 than the NZ70 (Table 5.6). In 2002-03 the OS90 and NZ90 genotypes had higher yields of milk and MS than NZ70; in addition, MS yield was greater for NZ90 than OS90. Similar results were recorded for MS yield in 2003-04; additionally, MS yield increased with FA due to the significant effect of FA on fat and protein yields and DIM (Figure 5.3d). A significant GE by FA interaction was measured for milk yield in both seasons 2002-03 and 2003-04. The OS90 strain showed a larger increase in milk yield as FA increased than the two NZ genotypes (Tables 5.6 and 5.8).

**Figure 5.5: Milksolids yield per hectare (a) and per cow (b) of each Holstein-Friesian genotype at the different feed allowance during seasons 2001-02, 2002-03 and 2003-04.**



NZ70 (□), NZ90 (■), OS90 (○).  $NTF_0$ : Nominal (planned) total feed offered (feed allowance). In all figures the 'x' axis indicates the value of  $NTF_0$  at which the systems were managed, expressed in tonnes of DM per cow per year. Figures (a) and (b) show changes in yield per hectare and per cow with time for each Holstein-Friesian genotype managed at different feed allowance ( $NTF_0$ ). Season 2001-02 only 2 year old cows, season 2002-03 two & three years old and in season 2003-04 two, three & four years old.

In 2001-02 the NZ90 genotype had greater fat and protein yields than NZ70 (Table 4.6) despite slightly shorter lactation lengths. In contrast the higher MS yield of the NZ90 genotype than the OS90 resulted from a greater fat yield and more DIM (Table 5.6 and Figure 5.3d). The NZ70 and OS90 had similar fat yield in 2001-02, but NZ90 and OS90 showed greater fat yield in 2002-03 and 2003-04; in addition, protein yield was also greater for these genotypes than for NZ70. Lactose yield was similar across strains in 2001-02 but it was greater for NZ90 and OS90 than NZ70 in 2002-03. A GE by FA interaction was measured in 2003-04 with a larger increase in lactose yield with increments in FA for the OS90 strain than for the two NZ genotypes, as was recorded for milk yield (Tables 5.6 and 5.8).

**Table 5.6: Mean values for total lactation yields, lactation length, live weight and body condition score for the whole lactation, and liveweight and body condition score lost from calving to nadir for each Holstein-Friesian genotype, age group and feed allowance in each season.**

	Genotype				Age				Comparative feeding allowance					Significance				
	NZ70	NZ90	OS90	Sed	2Y	3Y	4Y	Sed	FA1	FA2	FA3	FA4	SED	GE	A	FA	GE*FA	
Season 2001 – 2002	Milk (L cow <sup>-1</sup> )	3755	3880	3922	107	3852	---	---	---	3638	3932	3921	3919	119	NS	---	0.06	NS
	MS (kg cow <sup>-1</sup> )	284	308	291	7.78	294	---	---	---	282	294	299	302	8.64	**	---	0.06	NS
	DIM (days)	267	258	249	3.42	258	---	---	---	255	252	263	260	3.79	***	---	*	NS
	Fat (kg cow <sup>-1</sup> )	166	175	162	4.79	168	---	---	---	162	166	171	172	5.30	**	---	NS	NS
	Protein (kg cow <sup>-1</sup> )	117	132	129	3.32	126	---	---	---	120	127	128	129	3.67	***	---	*	NS
	Lactose (kg cow <sup>-1</sup> )	185	192	191	5.33	189	---	---	---	178	194	192	193	5.92	NS	---	*	NS
	LW (kg cow <sup>-1</sup> )[1]	422	430	439	7.42	430	---	---	---	427	434	431	429	8.23	0.06	---	NS	NS
	LW lost (kg cow <sup>-1</sup> )[2]	-43	-62	-73	5.00	-59	---	---	---	-55	-59	-60	-65	5.54	***	---	NS	NS
	BCS [1]	4.72	4.43	4.12	0.08	4.42	---	---	---	4.49	4.44	4.39	4.37	0.09	***	---	NS	NS
BCS [2]	-0.76	-1.03	-1.34	0.09	-1.04	---	---	---	-1.03	-1.05	-1.04	-1.07	0.10	***	---	NS	NS	
Season 2002 – 2003	Milk (L cow <sup>-1</sup> )	4213	4952	5011	139	4437	5014	---	116	4162	4516	5054	5168	154	***	***	***	NS
	MS (kg cow <sup>-1</sup> )	325	408	381	10.7	348	395	---	8.90	330	350	397	409	11.9	***	***	***	NS
	DIM (days)	275	270	252	6.05	273	258	---	5.05	247	259	278	280	6.70	**	**	***	NS
	Fat (kg cow <sup>-1</sup> )	189	230	208	6.14	198	220	---	5.13	188	199	223	227	6.86	***	***	***	NS
	Protein (kg cow <sup>-1</sup> )	136	178	173	4.96	150	175	---	4.14	142	151	174	182	5.55	***	***	***	NS
	Lactose (kg cow <sup>-1</sup> )	204	242	243	6.78	217	243	---	5.66	202	220	244	252	7.58	***	***	***	NS
	LW (kg cow <sup>-1</sup> ) [1]	434	450	460	7.08	424	471	---	5.92	439	443	450	460	7.85	**	***	*	NS
	LW lost (kg cow <sup>-1</sup> )	-42	-57	-60	4.71	-55	-51	---	3.93	-59	-53	-48	-53	5.26	**	NS	NS	NS
	BCS [1]	4.85	4.54	4.13	0.06	4.63	4.38	---	0.05	4.48	4.46	4.45	4.64	0.07	***	***	*	0.081
BCS [2]	-1.00	-1.20	-1.50	0.07	-1.60	-0.90	---	0.06	-1.30	-1.40	-1.20	-1.20	0.08	***	***	NS	NS	
Season 2003 – 2004	Milk (L cow <sup>-1</sup> )	4816	5632	5945	163	4821	5637	5935	184	4906	5087	5847	6018	184	***	***	***	*
	MS (kg cow <sup>-1</sup> )	378	471	452	11.8	383	447	472	13.4	389	394	467	485	13.4	***	***	***	NS
	DIM	278	284	267	4.87	291	273	266	5.50	265	257	291	293	5.51	**	***	***	NS
	Fat (kg cow <sup>-1</sup> )	220	267	248	6.91	217	253	266	7.80	221	225	263	272	7.81	***	***	***	NS
	Protein (kg cow <sup>-1</sup> )	157	204	204	5.45	166	194	206	6.14	168	169	204	214	6.15	***	***	***	NS
	Lactose (kg cow <sup>-1</sup> )	233	274	285	8.01	235	271	285	9.09	237	248	280	291	9.11	***	***	***	*
	LW (kg cow <sup>-1</sup> ) [1]	457	483	509	8.12	456	478	516	9.17	475	470	493	494	9.17	***	***	**	0.148
	LW lost (kg cow <sup>-1</sup> )	-64	-78	-92	4.32	-78	-72	-84	4.87	-74	-76	-76	-86	4.88	***	**	*	NS
	BCS [1]	4.49	4.27	3.71	0.09	4.30	4.03	4.14	0.09	4.15	3.93	4.22	4.34	0.10	***	0.04	**	0.054
BCS [2]	-1.13	-1.26	-1.92	0.10	-1.63	-1.24	-1.44	0.11	-1.25	-1.57	-1.48	-1.47	0.11	***	**	*	*	

MS: milksolids; DIM: days in milk; LW: live weight; BCS: body condition score; [1] & [2] LW and BCS mean & lost during lactation. GE: genotype; A: age [2Y: two years old; 3Y: three years old; 4Y: four years old]; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = not significant.

### 5.3.4.2. Daily yield per cow

The NZ90 strain had greater fat yields than both NZ70 and OS90 in 2001-02, whilst similar fat yields were recorded for NZ70 and OS90 (Table 5.7). Fat yield increased in

NZ90 and OS90 in 2002-03 and 2003-04 compared with the previous season. Protein yield was also greater for NZ90 and OS90 than for NZ70 over the whole study. Consequently, the differences in daily MS yield between strains increased from the first to the third season, probably as a result of the higher yield potential of the NZ90 and OS90 and both the significant effect of the age and FA on daily yield.

**Table 5.7: Mean values for daily yields and milk composition for each Holstein-Friesian genotype, age group and feed allowance in each season.**

	Genotype				Age				Comparative feeding allowance					Significance				
	NZ70	NZ90	OS90	Sed	2Y	3Y	4Y	Sed	FA1	FA2	FA3	FA4	SED	GE	A	FA	GE*FA	
Season 2001 – 2002	Milk (L cow <sup>-1</sup> )	14.1	15.1	15.8	0.42	15.0	---	---	---	14.3	15.7	14.9	15.1	0.46	***	---	*	NS
	MS (kg cow <sup>-1</sup> )	1.06	1.19	1.17	0.03	1.14	---	---	---	1.11	1.17	1.14	1.16	0.03	***	---	NS	NS
	Fat (kg cow <sup>-1</sup> )	0.62	0.68	0.65	0.02	0.65	---	---	---	0.64	0.66	0.65	0.66	0.02	**	---	NS	NS
	Protein (kg cow <sup>-1</sup> )	0.44	0.51	0.52	0.01	0.49	---	---	---	0.47	0.51	0.49	0.50	0.01	***	---	0.06	NS
	Lactose (kg cow <sup>-1</sup> )	0.70	0.75	0.77	0.02	0.74	---	---	---	0.70	0.78	0.73	0.74	0.02	**	---	*	NS
	FC (g kg <sup>-1</sup> )	44.5	45.4	41.4	0.08	43.8	---	---	---	44.7	42.4	43.8	44.1	0.09	***	---	0.12	0.103
	PC (g kg <sup>-1</sup> )	31.3	34.2	33.1	0.04	32.8	---	---	---	33.1	32.4	32.9	33.0	0.05	***	---	NS	NS
	LC (g kg <sup>-1</sup> )	41.9	49.5	48.7	0.03	49.1	---	---	---	49.1	49.4	48.9	49.1	0.03	***	---	NS	0.051
	Protein – Fat ratio	0.71	0.76	0.80	0.01	0.76	---	---	---	0.75	0.77	0.75	0.75	0.01	***	---	NS	NS
Season 2002 – 2003	Milk (L cow <sup>-1</sup> )	15.4	18.4	20.1	0.53	16.3	19.6	---	0.44	17.2	17.8	18.3	18.6	0.59	***	***	0.05	*
	MS (kg cow <sup>-1</sup> )	1.19	1.51	1.53	0.03	1.28	1.54	---	0.03	1.36	1.37	1.43	1.47	0.04	***	***	**	NS
	Fat (kg cow <sup>-1</sup> )	0.69	0.86	0.83	0.02	0.72	0.86	---	0.02	0.78	0.78	0.80	0.81	0.02	***	***	NS	NS
	Protein (kg cow <sup>-1</sup> )	0.49	0.66	0.69	0.02	0.55	0.68	---	0.01	0.59	0.59	0.63	0.66	0.02	***	***	***	NS
	Lactose (kg cow <sup>-1</sup> )	0.75	0.90	0.97	0.03	0.80	0.95	---	0.02	0.83	0.88	0.88	0.91	0.03	***	***	0.06	*
	FC (g kg <sup>-1</sup> )	45.4	46.7	41.9	0.09	45.1	44.2	---	0.07	45.5	44.4	44.7	44.1	0.09	***	NS	NS	0.093
	PC (g kg <sup>-1</sup> )	32.7	35.7	34.6	0.05	33.9	34.8	---	0.04	34.2	33.6	34.5	35.1	0.05	***	*	0.15	*
	LC (g kg <sup>-1</sup> )	48.4	48.9	48.4	0.02	48.8	48.4	---	0.02	48.5	48.8	48.4	48.8	0.02	*	*	NS	NS
	Protein – Fat ratio	0.72	0.77	0.83	0.01	0.76	0.79	---	0.01	0.76	0.76	0.78	0.80	0.01	***	**	**	NS
Season 2003 – 2004	Milk (L cow <sup>-1</sup> )	17.5	19.9	22.4	0.49	16.7	20.7	22.4	0.56	18.6	20.2	20.3	20.6	0.56	***	***	***	0.05
	MS (kg cow <sup>-1</sup> )	1.37	1.66	1.71	0.03	1.32	1.64	1.78	0.04	1.48	1.56	1.62	1.66	0.04	***	***	***	NS
	Fat (kg cow <sup>-1</sup> )	0.80	0.94	0.94	0.02	0.75	0.93	1.00	0.02	0.84	0.89	0.91	0.93	0.02	***	***	***	NS
	Protein (kg cow <sup>-1</sup> )	0.57	0.72	0.77	0.01	0.57	0.71	0.78	0.02	0.64	0.67	0.71	0.73	0.02	***	***	***	NS
	Lactose (kg cow <sup>-1</sup> )	0.84	0.97	1.07	0.02	0.81	0.99	1.08	0.03	0.90	0.98	0.97	0.99	0.03	***	***	***	*
	FC (g kg <sup>-1</sup> )	46.2	47.6	42.2	0.09	45.4	45.5	45.1	0.10	45.6	44.7	45.5	45.4	0.10	***	NS	NS	0.072
	PC (g kg <sup>-1</sup> )	32.9	36.3	34.4	0.04	34.6	34.5	34.7	0.04	34.3	33.5	34.9	35.5	0.04	***	NS	***	**
	LC (g kg <sup>-1</sup> )	48.4	48.7	48.0	0.03	48.9	48.1	48.1	0.03	48.3	48.7	48.1	48.4	0.03	***	**	NS	NS
	Protein – Fat ratio	0.72	0.77	0.82	0.01	0.77	0.76	0.78	0.01	0.76	0.76	0.77	0.79	0.01	***	NS	*	NS

MS: milksolids daily yield; FC: milk fat concentration; PC: milk protein concentration; LC: milk lactose concentration; GE: genotype; A: age (2Y: two years old; 3Y: three years old; 4Y: four years old); FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = not significant.

No significant effect of FA was observed on MS yield in 2001-02, however MS yield increased with FA in 2002-03 and 2003-04 due to the positive effect of FA on protein yield in 2002-03 and on both protein and fat in 2003-04 (Table 5.7). It is apparent that FA has a greater effect on protein than fat as indicated by the increased protein/fat ratio measured as FA increased in 2002-03 and 2003-04. The NZ90 and OS90 genotypes produced larger amounts of milk and lactose in season 2001-02, but significant GE by FA interactions were observed in 2002-03 and 2003-04 because of the larger increments in daily milk and lactose as FA increased in the OS90 genotype than for the two NZ strains.

A significant and positive effect of age on total lactation and daily milk, protein, fat and lactose yields was recorded (Tables 5.6 and 5.7). In season 2002-03 and 2003-04, second

and third calving cows had shorter lactations than first lactating animals. The high BCS pre-calving and early calving of first lactating animals appear to be the reasons for their longer lactation length in seasons 2002-03 and 2003-04, which should have affected lactation length for the same cows in the following season. Lactation length of first lactation cows improved from the first to the third season probably as a result of an increased  $TF_0$ , particularly due to a larger increase for systems managed at higher FA (Table 5.5). Another reason for the shorter lactation length in 2001-02 could be an early drying-off due to very slow growing conditions in early autumn 2002 (Figure 5.1a).

Nevertheless, longer lactations lengths were observed in 2002-03 and 2003-04 (Table 5.6), particularly for the NZ90 and OS90 genotypes. This was as a result of the greater amount of  $TF_0$  and its effect on end of season BCS (Table 5.5), particularly in 2003-04 when the drying off decision rules changed for systems managed at high FA.

#### 5.3.4.3. Concentration of milk components

Milk fat content was greater for both NZ genotypes than OS90 during the complete study (Table 5.7). Protein content was greater for the NZ90 and OS90 genotypes than for NZ70 in 2001-02 and showed a significant GE by FA interaction in 2002-03 and 2003-04, with a larger increment of protein content in milk as FA increased for the NZ90 genotypes than for the NZ70 and OS90 strains (Tables 5.7 and 5.8).

**Table 5.8: Significant Holstein-Friesian genotype by feed allowance interactions measured during seasons 2002-03 and 2003-04 (from Tables 5.5 and 5.6).**

	Comparative FA	NZ70			NZ90			OS90			SED	GE	Significance				
		FA1	FA3	FA4	FA1	FA2	FA3	FA4	FA1	FA2			FA3	FA4	A	FA	GE*FA
Season 02-03	Milk (L cow <sup>-1</sup> )	14.72	16.53	15.42	18.16	18.33	17.45	19.47	18.98	19.13	20.98	21.23	1.13	***	***	*	*
	Lactose (kg cow <sup>-1</sup> )	0.71	0.80	0.74	0.89	0.90	0.84	0.96	0.91	0.93	1.02	1.03	0.05	***	***	**	*
	PC (g kg <sup>-1</sup> )	33.1	32.6	33.5	34.4	34.5	36.8	36.9	35.1	34.5	33.9	34.6	0.97	***	*	0.14	*
	BCS End of Season	4.95	5.03	5.56	4.61	4.42	4.76	4.97	4.62	4.40	4.37	4.55	0.25	***	NS	***	*
Season 03-04	Milk yield (L cow <sup>-1</sup> )	4219	5316	5051	5401	5370	5552	6228	5274	5500	6558	6767	330	***	***	***	*
	Milk (L cow <sup>-1</sup> )	15.94	18.59	17.26	19.54	20.11	19.01	20.79	21.34	22.07	22.86	23.97	1.00	***	***	***	*
	Lactose (kg cow <sup>-1</sup> )	205	255	243	264	264	266	304	251	264	315	325	16.2	***	***	***	*
	Lactose (kg cow <sup>-1</sup> )	0.78	0.89	0.83	0.95	0.99	0.91	1.01	1.01	1.06	1.10	1.15	0.05	***	***	***	*
	PC (g kg <sup>-1</sup> )	33.4	32.9	34.2	35.1	34.9	37.6	38.0	34.1	34.2	34.2	34.2	0.74	***	NS	**	**
	BCS Nadir	4.07	4.00	4.26	3.69	3.44	3.95	4.13	3.10	3.07	3.09	3.12	0.19	***	*	***	*
BCS lost	-0.90	-1.30	-1.10	-1.20	-1.60	-1.10	-1.20	-1.80	-1.80	-1.90	-2.10	0.20	***	***	*	*	

PC: milk protein concentration; BCS: body condition score; GE: genotype; A: age [2Y: two years old; 3Y: three years old; 4Y: four years old]; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \*= $P < 0.05$ ; \*\*= $P < 0.01$ ; \*\*\*= $P < 0.001$ . NS= not significant.

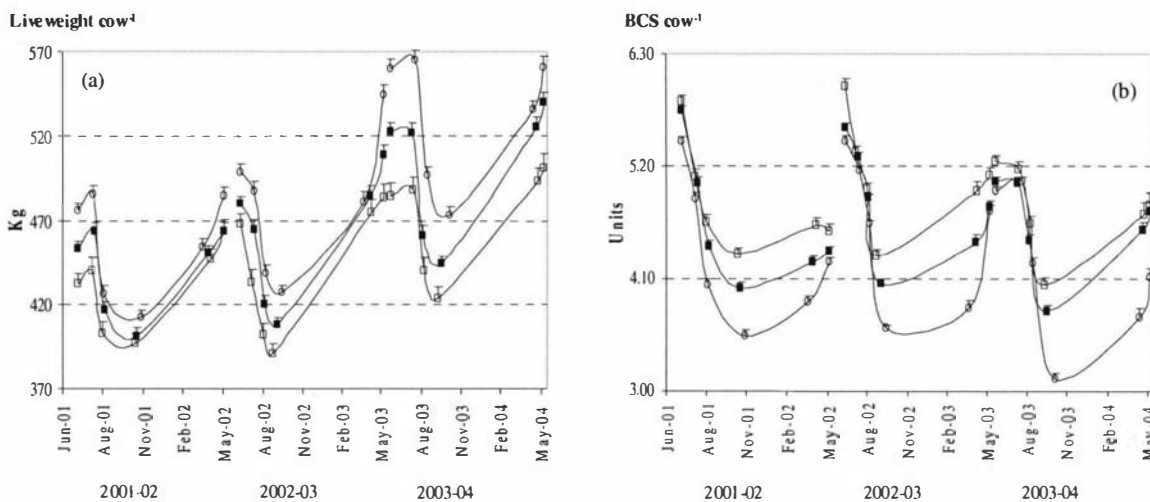
The highest protein/fat ratio was observed for the OS90 genotype, intermediate for NZ90 and the lowest for the NZ70 strain in all three seasons. Milk lactose content was greater for the NZ90 genotype than for both the NZ70 and OS90 strains; differences between

NZ70 and OS90 were not significant although numerically greater for the NZ70 strain. A non-significant effect of FA on the lactose content in milk was observed in all three seasons.

### 5.3.5. Live weight and condition score during the season

The mean actual LW during the lactation and dry periods was below the mature LW expected (McNaughton *et al.*, 2002), particularly for the first season when all the cows were first lactating animals and still growing. In addition, the difference between actual and mature LW was greater for cows of the OS90 and NZ90 strains, with higher adult LW.

**Figure 5.6: Mean live weight (a) and body condition score (b) of three Holstein-Friesian genotypes averaged across feed allowance, at different dates during seasons 2001-02, 2002-03 and 2003-04.**



NZ70 (□), NZ90 (■), OS90 (○). Dates plotted for each season are: start of season (S); calving (C); week 4<sup>th</sup> post-calving (W4); nadir (N); drying-off (D); and end of season (E). Figures (a) and (b) show changes in live weight and body condition score for each Holstein-Friesian genotype across the different levels of feed allowance (NTF<sub>0</sub>) at which they were managed. Season 2001-02 only 2 year old cows, season 2002-03 two & three years old and in season 2003-04 two, three & four years old.

The mean LW and BCS of cows of the three genotypes during the season declined after calving, but increased once the nadir was achieved until the end of the lactation and again from then until the end of each season (May 30<sup>th</sup> each year; see also Figures 5.7a and 5.7b, also Appendix V-13 and V-14). The mean LW of the cows of the three genotypes increased as the mean age of the cows increased, particularly for the OS90 genotype at the end of season 2002-03 and also in 2003-04. However, BCS tended to be lower at the end of season 2003-04 compared to season 2002-03, probably as a result of the extended

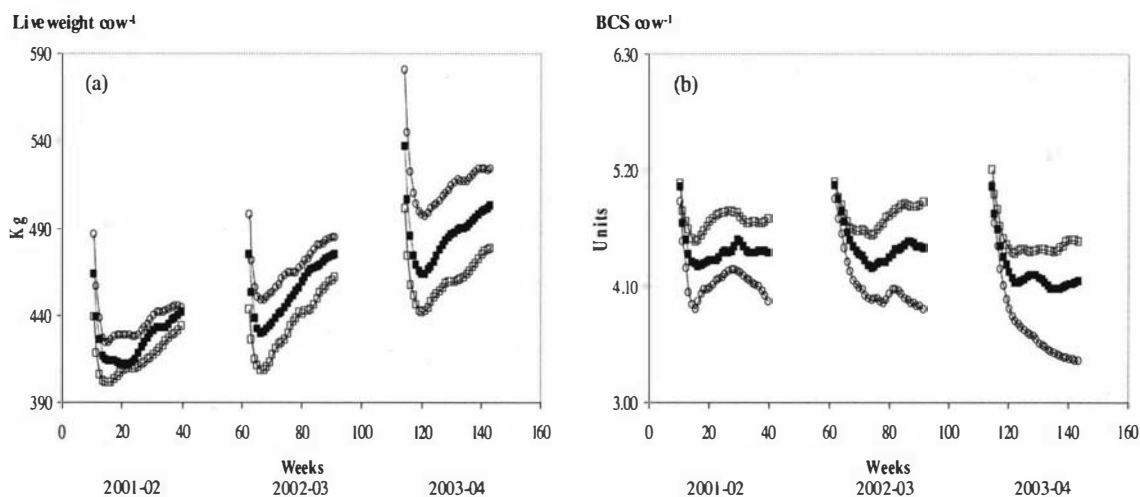
lactation of the three genotypes due to a change in decision rules (Macdonald *et al.*, 2005).

When the mean LW and BCS of the genotypes were compared for cows at similar days post-calving (Figures 5.7a and 5.7b) the decline in LW post-calving and the improvement post-nadir showed a trend similar to that observed in Figure 5.6a, with the differences between strains increasing from season 2001-02 to season 2003-04 (Figure 5.7b). However, the trend for BCS between genotypes was different from season 2001-02 to season 2003-04 and showed that OS90 cows continued losing BCS from calving to week 30 of lactation, while a different response was observed for the two NZ strains. In 2001-02 all the genotypes recovered BCS about the same date post-calving and earlier than observed in the second and third seasons. The OS90 genotype decline at the same rate as NZ cows in 2003-04 while all cows are losing BCS, but then continues losing BCS after NZ cows have reached their nadir (Figure 5.7b).

### 5.3.5.1. Liveweight changes

The average LW during lactation increased from the first to the third season (Table 5.7; Figure 5.6a) due to the significant effects of age and FA in 2002-03 and 2003-04 (Table 5.7).

**Figure 5.7: Mean values for the weekly live weight (a) and body condition score (b) of three Holstein-Friesian genotypes from calving to week 30<sup>th</sup> post-calving during each of three lactations over the experimental period.**



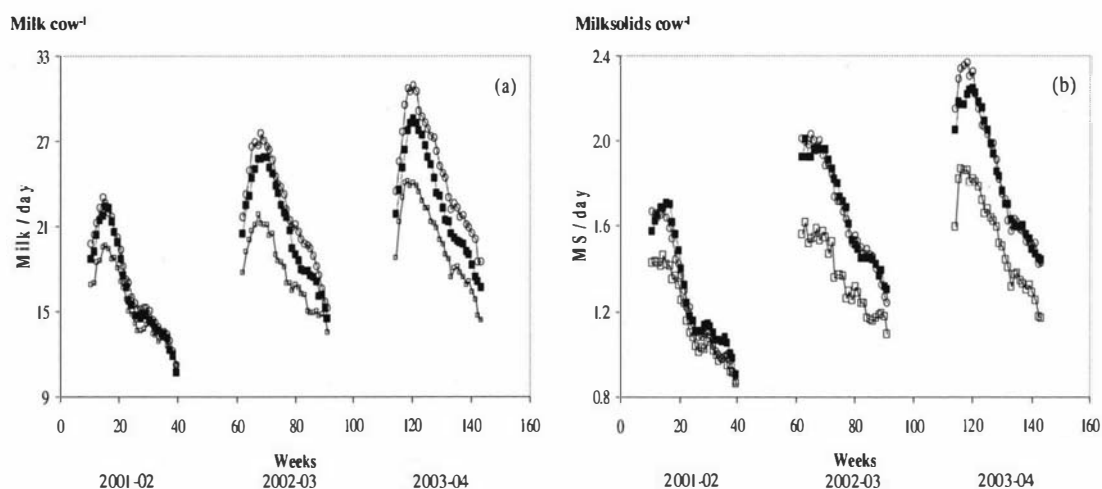
NZ70 (□), NZ90 (■), OS90 (○). Axis 'x' gives weeks from the beginning of the experiment. Figures (a) and (b) show changes in weekly mean live weight and body condition score for each genotype Holstein-Friesian across the different levels of feed allowance (NTF<sub>0</sub>) at which they were managed, plotted over the experimental period. Season 2001-02 only 2 year old cows, season 2002-03 two & three years old and in season 2003-04 two, three & four years old.

The OS90 strain was heavier than NZ90 and NZ70; however differences between NZ90 and OS90 were not significant in 2001-02 and 2002-03. The NZ90 genotype was heavier than the NZ70 strain, although differences were not significant in the first season (Table 5.6). The OS90 cows tended to be heavier at each date during lactation and the dry periods while the NZ70 cows were lighter (Figures 5.7a and 5.8a). Additionally, all the genotypes increased LW as they grew as indicated by the mean LW of genotypes and feeding levels from the first to the third season, showing a positive effect of feeding level and possibly a result of more strategic use of the extra feed available in 2002-03 and 2003-04 (see Appendix V-15, V-16 and V-17). The mean LW lost between calving to nadir was greater for both high yielding genotypes than in NZ70 (Table 5.6); the OS90 lost more LW than NZ90 (although not significantly in 2002-03; see Appendix V-13).

### 5.3.5.2. *Body condition score changes*

The lowest BCS during lactation were observed for OS90 and the highest for NZ70 (Table 5.6; Figures 5.7b and 5.8b), with NZ90 being significantly lower than NZ70 but greater than OS90. Body condition score was greater in first lactation cows, with the lowest BCS measured during the second lactation. No effect of FA on BCS was observed in 2001-02, however BCS improved with FA in 2002-03 and 2003-04.

**Figure 5.8: Mean values for the weekly milk (a) and milksolids yields (b) of three Holstein-Friesian genotypes from calving to week 30<sup>th</sup> post-calving during each of three lactations over the experimental period.**



NZ70 (□), NZ90 (■), OS90 (○). Figures (a) and (b) show changes in the weekly mean milk and milksolids yield for each Holstein-Friesian genotype across the different levels of feed allowance (NTF<sub>0</sub>) at which they were managed, plotted between week 1<sup>st</sup> and 30<sup>th</sup> of lactation (all cows) over season 2001-02, 2002-03 and 2003-04. Season 2001-02 only 2 year old cows, season 2002-03 two & three years old and in season 2003-04 two, three & four years old.

All genotypes lost BCS after calving (Figures 5.6b and 5.7b). The OS90 showed the greatest BCS lost and the NZ70 the lowest. Second lactation cows lost less BCS than first lactation animals in 2002-03 and had a lower mean BCS during lactation (Table 5.6). There was a non-significant effect of FA on BCS lost in 2001-02 and 2003-04; a significant GE by FA interaction on BCS lost was detected in season 2003-04 where the OS90 strain lost more BCS after calving and showed a consistent trend to continue losing BCS during lactation even though FA had increased. In contrast, it was apparent that the loss in BCS in both NZ strains during lactation was more responsive to an imbalance between  $TF_0$  and requirements sometime during the lactation period (Table 5.8).

Differences in BCS between genotypes at the start of the season and at calving were smaller than observed post-calving, although significant (Figure 5.7b; see Appendix V-14). The largest differences between strains occurred at BCS nadir and at the end of the season, particularly in seasons 2002-03 and 2003-04, and between the two NZ genotypes and the OS90 strain. The differences in BCS between genotypes increased after calving until the BCS nadir was achieved, with the OS90 genotype showing the lowest BCS at nadir whereas the NZ70 the highest. Second lactation cows had lower BCS at start of the season and soon after calving but recovered BCS faster at the end of the lactation and achieved similar BCS at drying-off date to the younger and older cows (see Appendix V-14).

A significant effect of FA on BCS at drying-off date was detected in the last two seasons, as well as at the start of season, at calving and during early lactation in 2003-04 (Table 5.8). A long period from calving to BCS nadir was observed for OS90 and the shortest for NZ70 in 2001-02 and 2002-03. This period was shorter for the NZ90 genotype than in the OS90 strain but still longer than observed for the NZ70.

In season 2002-03 a significant GE by FA interaction was observed for BCS at the end of the season (Table 5.8). This occurred because BCS increased as FA increased for the two NZ strains, but not for the OS90 genotype. In season 2003-04, a significant GE by FA interaction was observed for BCS at nadir and the period from calving to nadir. Body condition score at the nadir increased as FA increased in both NZ genotypes but not in OS90. Additionally, the length of the period from calving to nadir increased as FA increased for NZ70 but decreased for NZ90 and OS90, with a faster reduction in the NZ90 genotype.

## 5.4. DISCUSSION

### 5.4.1. Feeding management

The mean values of HAR,  $HM_{PRE}$  and  $HM_{POST}$  were not significantly different across strains and feeding levels (Table 5.2) although differences in  $HM_{PRE}$  and  $HM_{POST}$  between systems were expected due to differences in management between strains. However, large differences were measured over the season due to the seasonal variation in HAR (Figure 5.1a) and its effects on grazing management (Figures 5.1b and 5.1c). In addition, seasonal variation in the ME and CP of the pasture were observed (Figures 5.2a and 5.2b). Despite the lower  $TF_0$  in 2001-02 than in 2002-03 and 2003-04, the pastures grazed by all three genotypes showed higher  $HM_{PRE}$  and  $HM_{POST}$  in 2001-02 (Figure 5.1b). This suggests that the cows were not constrained by pasture management. In addition the mean quality of the pasture grazed in 2001-02 was lower than in the following two seasons as indicated by the high NDF and low CP (Table 5.2), which also indicates that HAR was greater than the rate at which the herbage was harvested in 2001-02.

All three genotypes were managed in systems across ranges of annual feed allowances per cow planned according to their mature energy requirements (Kolver *et al.*, 2002; McNaughton *et al.*, 2002). Thus, the NZ70 genotype was offered the lowest annual feed allowance and the OS90 the highest (5.33, 5.75 and 6.25 t DM cow<sup>-1</sup>year<sup>-1</sup> respectively for NZ70, NZ90 and OS90) (Table 5.1). The mean amount of feed actually offered to the genotypes (Table 5.4) was slightly higher than proposed in the design of the experiment (5.79, 6.28 and 6.44 t DM cow<sup>-1</sup> year<sup>-1</sup> for NZ70, NZ90 and OS90 respectively), by about 9% for both NZ strains and 3% for the OS90.

The mean  $TF_0$  across feed allowances in OS90 was 7% lower than planned in 2001-02, but 7% and 9% higher in 2002-03 and 2003-04 respectively (Table 5.1). As a result, the difference in the mean  $TF_0$  between genotypes was almost similar across seasons (Table 5.5) and smaller between NZ90 and OS90 than between these two genotypes and the NZ70 strain. The difference between actual and planned level of feed for the NZ strains was higher than that observed for the OS90 strain, the range in  $TF_0$  within each genotype concurred with the range planned and increased with FA (Table 5.5), even though at a lower mean rate (0.74 t DM of  $TF_0$  t<sup>-1</sup>DM N $TF_0$ ; Table 5.4).

The mean  $DMI_V$  was about 80% of  $TF_0$  (4.68, 4.98 and 5.33 t DM cow<sup>-1</sup>year<sup>-1</sup> respectively for NZ70, NZ90 and OS90, Table 5.4), in agreement with the expected differences between genotypes and increased with feeding level (Figure 5.3a).

It is possible that for the two NZ strains, the larger  $TF_O$  relative to their energy demands (which are lower in comparison to the demands of the OS90) had a positive effect on the mean yield per cow even though the mean SR was slightly higher. The mean PU was 3% lower for the NZ strains than in OS90, which suggests that under the actual management and at the mean FA at which the genotypes were farmed, the intake achieved by NZ90 cows was close to potential while it was possibly constrained in the OS90 genotype. Moreover, the reduction in the mean PU by the NZ90 suggests a higher substitution effect considering the whole season, although a similar trend was recorded between the NZ90 and OS90 across feed allowances (Figure 5.3c). An indication of the higher substitution of the modern NZ than the OS Holstein-Friesian is the lower difference in intake reported by Kolver *et al.* (2002) between ‘only pasture’ and TMR diets. This suggests that the modern NZ Holstein-Friesian achieve higher proportion of potential intake on pasture alone, which was consistent with the higher substitution rate reported for the modern NZ dairy cows than for OS Holstein-Friesian on pasture-based systems in other recent studies (Linnane *et al.*, 2004; Horan *et al.*, 2006). In contrast, substitution rate was greater for the OS90 genotypes when measured during late lactation of season 2002-03 in the present study (see Chapter 7).

Differences in the substitution of pasture by supplement between genotypes possibly resulted from the previous management affecting the actual yield and energy requirements of the cows when the supplement was fed. Because of this, the response to additional feed would be determined by the ‘relative energy deficit’ of the cow (Faverdin *et al.*, 1991; Penno *et al.*, 2001; Buckley *et al.*, 2005), which would be determined by the actual yield potential of the cows, and also affected by the previous level of feed. It is hypothesised that at similar SR, the lower PU achieved in systems managed at low FA occurred because intake was constrained by grazing only pasture in early lactation hence energy in the milk output would reach equilibrium at a lower yield potential, consequently reducing feed demand (Penno *et al.*, 2001).

Pasture utilisation would have a great effect on the cost per unit of MS produced in pasture-based systems, particularly if the proportion of grazed grass in the diet of the cow is high (Shalloo *et al.*, 2004). Although the differences between strains were small, the extreme difference in PU between systems was about 10% (Figure 5.3c) and equal to 1.86 t DM ha<sup>-1</sup> or 0.6 t DM cow<sup>-1</sup> (3.1 cows ha<sup>-1</sup>). This was equivalent to the amount of supplement brought into systems managed at high FA with the NZ90 strain and about 65% of the mean amount of extra feed offered to the OS90 genotype (mean of FA3 and FA4). Furthermore, feed utilisation was high in systems managed at higher SR in both NZ strains, but also increased with FA in systems farmed with the NZ90 and OS90 genotypes at similar SR. This did not occur with the NZ70 genotype (Figure 5.3c). It

seems that high producing cows fed on pasture that receive strategic inputs of feed to increase total production through a higher lactation peak and slower decline post-peak, increase energy requirements and feeding drive and have a higher motivation to graze. Hence, PU improves despite the system being managed at lower SR.

#### 5.4.2. Systems results

All the genotypes increased yield per hectare from the first to the third season (Figures 5.6a and 5.6b). There was greater mean MS yield per hectare (mean all systems) in the NZ90 genotype in all the seasons; the highest mean yield per hectare was achieved in season 2003-04 with NZ90 cows managed at the highest feed allowance (1,673 kg MS ha<sup>-1</sup> and 540 kg MS cow<sup>-1</sup>), 7% higher than OS90 in the same season.

In the first season of the study MS yield per hectare was most favourable for NZ70, but improved to a greater extent for the OS90 genotype as the study progressed due to a greater yield per cow. For instance, in 2001-02 and 2003-04 the OS90 strain produced -4% and +10% MS ha<sup>-1</sup> respectively compared to NZ70, however MS yield per cow over the whole lactation was +3% and +17% higher for the OS90 (mean all systems). Hence initial differences in yield per hectare occurred due to differences in SR, then yield per hectare increased as yield per cow improved for NZ90 and OS90 cows. In contrast, the increased yield per cow observed for NZ70 cows did not offset the decrease in SR (Figure 5.3b).

In 2003-04 the mean yield per hectare was higher for OS90 than NZ70, but if only systems managed at similar SR and feeding level are considered (5.5 and 6.0 t cow<sup>-1</sup> respectively). Differences across these two genotypes were minimal (1,276 vs. 1,266 kg MS ha<sup>-1</sup> for NZ70 and OS90 respectively) and lower (-12%) than for NZ90 (1,425 kg MS ha<sup>-1</sup>). The NZ90 and OS90 strains were fed mainly pasture at these feeding levels while the NZ70 strain managed at the highest feeding level was fed supplement and had a lower CSR (Table 5.5).

Although for all three genotypes the TF<sub>O</sub> was above that planned (Table 5.4), the difference between actual and planned FA for the OS90 was smaller than that observed for both NZ genotypes, and thus intake may have been affected differently. However, this is not suggested by the actual DMI<sub>V</sub>, greater for OS90, and the small difference in the mean PU (Table 5.4) and pre-/post-grazing conditions between strains (Table 5.2). Yield per hectare for both NZ genotypes was higher at FA1 because of the higher SR utilised (Figure 5.4a). Increases in yield per hectare at higher feed allowances were constrained for the NZ70 because of its lower genetic potential. The TF<sub>O</sub> probably increased to levels higher than required; hence PU was further reduced as FA increased (Figure 5.3c).

Despite the higher requirements of the OS90 cows, when these cows were fed a small amount of supplement (at FA2) PU was initially reduced compared to the systems where only pasture was fed (at FA1). It is suggested that the timing of supplementation affected the seasonal response of systems farmed with both NZ90 and OS90 and as a result cows in systems managed at FA1 and FA2 had similar total lactation yield per cow. In systems managed at FA2, supplements were fed mainly during late lactation with the objective being to improve BCS; therefore intake and yield were probably constrained in early lactation with an effect on total lactation requirements that affected PU at the actual TFO. It is apparent that the NZ90 was affected to a lower extent than the OS90, probably because pasture intake was less affected during early lactation (see Appendix V-7 and Figure 9.1 in Chapter 9)

Pasture intake might have been lower in early lactation in the OS90 than for the NZ90 genotype as suggested by the higher loss in BCS, possibly sustained by a higher rise in yield after calving than in DM intake (Horan et al., 2006); this is particularly evident on pasture diets as a result of constraints to herbage ingestion (Kolver & Muller, 1998; Kolver *et al.*, 2002). However, these cows are metabolically prepared to compensate a reduced energy intake by mobilising body fat reserves, particularly in early lactation; in addition, the total amount of DM consumed during the whole season was higher for these cows suggesting that they probably increase intake at the end of lactation or during the dry period as a compensatory mechanism to recover body reserves at the end of the pregnancy. This response is supported by the trend observed for the LW and BCS of the genotypes during each season (Figures 5.6a, 5.6b, 5.7a and 5.7b), even though these cows were also influenced by the strategy of supplementation used and drying-off decision rules (Macdonald *et al.*, 2005); the OS90 animals clearly showed a trend to partition more energy to milk yield instead of building up body reserves than the NZ genotypes, even at the end of lactation.

#### **5.4.3. Daily and total lactation yield per cow**

The greater daily MS yield of the NZ90 genotype was achieved through improvements in both fat and protein yields compared to NZ70, however, the OS90 genotype had a numerically higher MS yield sustained by a greater yield of protein than the NZ90 strain. Differences in daily yield between the NZ90 and OS90 genotypes increased from the first to the third season as a result of an increased milk yield (Figure 5.8a), and agreed with those reported by Linnane et al. (2004). The improvement observed as the experiment progressed was probably sustained by the increased mean age of the cows in the systems. However, lactation curves also showed a higher rise in milk yield and peak post-calving in OS90 than NZ90 cows that increased with season. Nevertheless, total lactation yield

per cow is the result of daily MS yield and lactation length. In season 2001-02 the NZ70 genotype had the lowest daily yield but the longest lactation length, in contrast the OS90 genotype had the highest daily yield but the shortest lactation length. All three genotypes increased daily yield and lactation length from the first to the third season, but the NZ90 and OS90 cows did so to a greater extent. The largest increase in total lactation yield observed for these genotypes as the study progressed was sustained by a larger increase in daily yield, even though lactation length was also extended. In fact, lactation length was always shorter for the OS90 genotype than for both NZ strains, despite changes in the drying off decision rules.

The main effect of daily yield on total lactation production is evident for cows in their second lactation, as total yield was improved in these cows despite a reduced lactation length. Calving date is the main determinant of lactation length in seasonal calving systems (Macmillan *et al.*, 1984), although delaying drying-off date can also improve lactation length. The main factor determining the drying-off date is the BCS of the cow at the end of lactation (Macdonald & Penno, 1998). As the cow has to achieve a BCS of 5 at calving, the length of the period from drying-off to calving, the BCS at drying-off and the amount of feed available during the period determines the possibility for the cow of achieving this target. Thus, cows that lost more BCS in early lactation and did not recover enough BCS in late lactation had to be dried-off earlier to allow them to recover condition before next calving.

Feeding level had a positive effect on BCS and thus on lactation length, with greater effects on both NZ genotypes, particularly the NZ70. Drying off decision rules were similar in the first two seasons of the study, but they penalised cows farmed at a high feeding level that lost more BCS during lactation, particularly OS90, despite the fact that these cows can recover BCS faster if fed more supplements during the dry period. So drying-off decision rules changed in 2003-04 (Macdonald *et al.*, 2005), lactation length was extended further in systems managed at the highest feeding level and cows were allowed to be dried-off at a lower BCS. Although these changes improved lactation length and the resultant total lactation yield, the increases were larger for the NZ genotypes than for the OS90 strain.

#### **5.4.4. Genotype by feeding level interactions**

Genotype by FA interactions for milk and lactose yield indicates a greater response in daily and total lactation milk and lactose yield for the OS90 than for NZ90 to increased feed allowance. This interaction was not observed for total MS yield, probably because the content of solids in the milk, particularly protein, showed a trend to increase to a

greater extent in the NZ90 than in the OS90 genotype as feeding level increased. The occurrence of GE by FA interaction may increase when the differences in the nutritional plane of the diets or the genotypes used increase (Kolver *et al.*, 2002). In the present study the greater difference between the genotypes used, particularly for yield potential, may have increased the opportunity to detect GE by FA interactions even though all of them were managed in pasture-based systems where the contrast in the plane of nutrition achieved was lower than in Kolver's study. Thus, without the great difference in the genotypes used, it would be expected that the possibility of detecting GE by FA interactions would be reduced considering that the intake of cows of high genetic potential is constrained on pasture, and that there must be a limit to the extent at which body reserves can be mobilised (Veerkamp *et al.*, 1995).

In contrast, the actual LW achieved by the genotypes at the start of the experiment in relation with their adult LW (McNaughton *et al.*, 2002) suggests that the maturity of the different genotypes was achieved in younger cows for NZ70 and older for OS90. Thus, although age was balanced in the systems, actual yield increased as the experiment progressed as a result of the increased proportion of mature cows in the systems from the first to the third season. This suggests that the potential yield of both the NZ90 and to a greater extent the OS90 genotypes would be constrained in the first season of the study, and the possibility to detect GE by FA interactions in the first season was low (Kolver *et al.*, 2000; Kolver *et al.*, 2002). This is in agreement with Linnane's work, probably because the intake capacity of two year old cows is lower - about 80% of the intake of multiparous animals (Jarrige *et al.*, 1986). This explains why an increased number of GE by FA interactions, particularly for yield and BCS, were observed in the second and third seasons of the present study, as it was suggested would occur (Veerkamp *et al.*, 1994; Kolver *et al.*, 2000).

Even though the NZ90 strain yielded more total lactation MS per cow than the OS90 genotype over the three seasons, these GE by FA interactions indicate that the genotypes responded differently to the increased level of feed at which the systems were farmed, with a poor yield of milk achieved by the OS90 genotype at the lowest FA despite the amount of body reserves mobilised.

Differences in the metabolism of the cows probably determined the difference observed between genotypes. For instance, it was suggested that cows would reduce feed intake (or increase production) when more lipid was available for mobilisation (Buckley *et al.*, 2000) and this could be the reason for the higher use of BCS in the OS90 until equal BCS nadir was achieved, as most of the energy available was partitioned to yield (Table 5.8; season 2003-04). However, it seems there is a different mechanism or threshold operating

the level of body reserves mobilisation for the two NZ genotypes, preventing the excessive use of body reserves during lactation. This mechanism would have a different effect on the intake of the cows as is suggested by the fact that yield for the NZ90 cows was less affected at low FA than the OS90 and the cows utilised less body corporal reserves (Table 5.8). Veerkamp *et al.* (1995) concluded that the source of genetic variation in gross energetic efficiency (defined as the amount of yield produced per unit of feed consumed) would be determined by the extent to which body reserves are mobilised and the capacity for feed intake of the cow; therefore differences in partition between these two sources of energy are crucial for the cow to perform and survive in a pasture-based system, where the existence of constraints on herbage intake for cows with high yield genetic potential have been demonstrated elsewhere. If it is considered that a limit to the rate of tissue mobilisation must exist, and it seems that OS90 cows reach this limit whatever the FA at which they were farmed, not only would yield potential be affected but the capacity to sustain other functions like reproduction or the health of the cow would also be compromised (Lucy *et al.*, 1991; Beam & Butler, 1999; Butler, 2000; Kolver, 2001; McNaughton *et al.*, 2003). The effect of selection on the BCS lost post-calving, as an indication of a more negative energy balance of the OS90 cows, could be of great economic value as it is generally related to poorer health and fertility (Veerkamp, 1998).

The calculation of the energy balance of the cows requires knowing the energetic value per unit of LW lost or gained. This in itself involves the problem of dealing with differences in gut fill between cows under different management, where differences in intake are expected. In addition, with the fact that the estimation of the energy provided by the LW lost could actually underestimate the amount of energy diverted to milk production, it is considered less probable for the genotypes to provide a similar amount of energy per unit of LW loss when the evidence available indicates that they are losing LW and BCS at different rates. It is also apparent that changes in the energy in milk output would occur as a consequence of the different selection priorities under which these strains were selected. This is supported by the different strategies in the allocation of energy observed by the genotypes to increase total MS yield (increase in milk volume that occurred for OS90 cows or the concentration of solids in the milk that occurs for the NZ90 genotype). Differences in the energy cost involved in these processes would affect the energy balance of the cows during lactation. In addition, as the main objective for NZ dairy farmers is to increase the amount of MS yield, the energetic efficiency of achieving this through an increase in milk volume should be further investigated. Changes in the partition of the energy available per unit of MS produced between genotypes (AFRC, 1993; Reynolds & Beaver, 1995) could contribute to differences in the daily energy

balance during lactation, in line with the GE by FA interaction observed for the BCS lost and the value achieved at nadir.

#### 5.4.5. Live weight and body condition score

Rearing conditions during the pre-experimental period were similar between strains, however, although heavier, the LW achieved at first calving by OS90 was only 80% of the mature LW of this genotype, compared with 87% and 90% respectively for NZ70 and NZ90. Differences in LW between genotypes in 2002-03 and 2003-04 were smaller than expected, considering the mature LW for the strains [adult LW: 540 kg for both NZ genotypes and 640 kg for OS90 (McNaughton *et al.*, 2002)]. Differences between current and expected LW for the genotypes indicate differences in the maturity of the cows at the start of the experiment that were probably maintained during 2002-03, and suggests that differences in the growing requirements of the cows existed in first two seasons of the present study. This could have contributed to the reduced performance of the OS90 genotype, particularly at the lowest feeding level. However, it is known that the LW of cows fed only pasture all their lives is lower than that which could potentially be achieved on other feeding systems (Holmes, 1995); and it is also probable that a reduction in the mature LW could have an effect on the energy required for maintenance and grazing activity. Thus it could be possible that the differences in the mature LW of the genotypes suggested by McNaughton *et al.* (2002) were higher than is achievable in pasture-based systems.

The mean LW in all three genotypes increased from the first to the third season as a result of the increased mean age of the cows in the systems (Table 5.4). Live weight declined sharply after calving until LW nadir was achieved and then increased at a slower rate during the rest of lactation, then faster again during the dry period.

Cows in all systems were above BCS 5 just before calving, with a trend for higher BCS for the NZ70 strain. Smaller differences in BCS than LW between strains were observed before calving were observed (Figure 5.6a and 5.6b); in addition, NZ70 cows have achieved higher proportion of mature LW at the start of the experiment, as indicated by the reduced increase in LW that occurred later in the study (Figure 5.6a). Differences in BCS relative to LW probably indicate a different composition of the body mass in the genotypes; therefore the energy provided per unit of LW lost post-calving could be different. The decrease in BCS post-calving indicates that the OS90 cows used more energy from body fat reserves to sustain yield after calving than the other two genotypes; in addition, the use of body fat reserves increased in season 2003-04, particularly for

OS90 cows (Figures 5.7b and 5.8b) despite the increased  $TF_0$ , probably due to the increased age of the cows in the systems.

The mean BCS of the cows was improved at the end of the season relative to that observed at BCS nadir and was lower for the OS90 than in both NZ genotypes. This required an earlier drying-off for cows of the OS90 genotype. Considering the LW lost as a measure of the body reserves mobilised, Tamminga *et al.* (1997) suggested that protein is a high proportion of the LW lost. Maximum protein mobilisation occurred four weeks after calving, after this the LW of the cow increased due to the amount of protein and water gained (protein-to-water ratio in body reserves equal to 1:3.4), this may compensate the weight of the body fat still being mobilised, allowing the LW of the cow to increase before the BCS nadir occurs. This is in agreement with the fact that BCS nadir occurred later than LW nadir for all genotypes. As protein deposition and body fat mobilisation occurred simultaneously after the LW nadir (Tamminga *et al.*, 1997), energy from fat reserves is still mobilised even though increments in LW have been observed. Thus the estimation of the energy provided from reserves by the amount of LW lost could actually underestimate the amount of energy diverted to milk production.

Taking into account both LW and BCS changes during lactation (Figures 5.6a and 5.6b), it is suggested that a greater proportion of the total yield was derived from body reserves (Table 5.8) for the OS90 strain. The lower loss of LW and BCS in the NZ90 compared to OS90 cows indicates the higher intake capacity on pasture relative to energetic demand of the former genotype. The capacity to mobilise body reserves to sustain yield in early lactation and the extent to which these reserves are mobilised seems to be genetically controlled, suggesting a different metabolism between these genotypes. Cows that have a fast rise in yield post-calving mobilise more lipids, however it is apparent that the high mobilisation of fat affects DM intake negatively in early lactation (Ingvarlsen *et al.*, 1999; Buckley *et al.*, 2000). In addition, the fact that OS90 cows did not improve BCS substantially while lactating, even at high feed allowance, suggests that a higher partition of stored reserves is needed to sustain the higher yield potential of these cows.

The high decline in BCS after calving indicates that the main use of the body reserves mobilised resulted from the inability of the high producing dairy cow to achieve potential intake on pasture, thus genetic potential yield is constrained by the feed consumed. As BCS nadir tends to occur later for OS90 than for NZ cows, it is possible that the rise of intake post-calving occurs at a slower rate for OS90 cows. However, there must be a limit to the maximum amount of BCS that can be mobilised, and once this limit is achieved yield must be sustained by the feed consumed. As OS90 cows partition most of the energy consumed to milk yield, only small improvements in BCS during lactation would

be expected unless energy intake was above the current requirements for milk energy output and above the metabolic capacity of the cow to increase yield in the short term.

## 5.5. CONCLUSIONS

The NZ90 genotype has greater capacity to produce MS on pasture-based systems than the OS90 strain, despite the greater genetic potential of the latter. In addition, the OS90 cows lose more BCS post-calving and have lower BCS during lactation, which suggests the intake of these cows is limited on pasture to a greater extent than for the two NZ genotypes, particularly in the early lactation period.

It is apparent that the higher BCS mobilised by the OS90 was linked to a faster rise in daily milk yield after calving, which may be greater than the rise in the amount of dry matter consumed from the pasture. It is hypothesised that due to the higher yield potential of OS90 this unbalance is higher than for the NZ90 and thus requires a higher response from body reserves; this would result in a greater concentration of metabolites and precursors in blood with a negative effect on the motivation of the cow to graze and consequently on dry matter intake. Furthermore, a high proportion of the additional energy ingested by the OS90 during late lactation was partitioned to milk production instead of being used to build up body fat reserves. However, considering that intake was constrained in early lactation, the higher intake estimated for the OS90 strain for the whole season and the similar BCS achieved at calving between strains in the following season suggests that an increase in intake would have occurred for OS90 cows during late lactation and the dry period, once BCS was at their lowest.

Total lactation MS yield improved with feeding level in all three genotypes, but to a greater extent in the high potential yielding NZ90 and OS90 genotypes; however, the NZ90 also produced more total lactation MS in systems managed at low feed allowance. The difference in performance between the NZ70 and NZ90 genotypes was mainly determined by the smaller genetic potential yield of the NZ70, which indicates the improvements made in NZ during the last 30 years by selecting cows that well suit the requirements of seasonal pasture-based systems. The NZ90 genotypes combine high yield potential (although lower than OS90) with the ability to meet energy requirements from the grazed pasture.

This chapter focused on the effects of genotype and management contrasts on the productive performance of self-contained farmlot systems and emphasised the importance of herbage intake data in understanding the factors influencing the productive performance and efficiency of grazing dairy cows. The above interpretation of

differences in levels of milk production and changes in body energy reserves depends to some extent on assumptions about the levels of herbage intake achieved by the three genotypes from the grazed pasture. This aspect in particular, regarding the condition of the pastures grazed by the cows and the capacity of the animals to adjust grazing behaviour to pasture conditions, is considered in more detail in the following chapters.

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## CHAPTER 6

# **PRODUCTIVITY OF PASTURE BASED SYSTEMS: FORAGE AND SUPPLEMENT CONSUMPTION, AND COW NUTRITION**

### **6.1. INTRODUCTION**

Results from the ‘system’ study (Chapter 5) showed that, on pasture-based systems, modern high yielding dairy cows selected in New Zealand (NZ) produced slightly more MS than cows of high yield genetic potential selected in the United States (OS). These results were consistent with Kolver et al. (2002).

The fact that OS90 cows lost more live weight (LW) and body condition score (BCS) in early lactation than NZ90 (see Figures 5.6 and 5.7 in Chapter 5) suggests that intake was constrained to a greater extent for OS90 cows grazing pasture, particularly during the early lactation period. As a result, yield improved with feeding level, however, total lactation yield on pasture was always below the potential achieved on a TMR diet (Kolver et al., 2002). It was also observed that the OS90 cows continued to partition body reserves to milk yield throughout the lactation period (see Chapter 5), which is not sustainable in seasonal pasture-based systems where cows have to regain condition before the next calving. This determined a shorter lactation length for this genotype, according with drying off practice (Macdonald & Penno, 1998).

Even though the continued loss of BCS during whole lactation suggest lower dry matter intake relative to energetic requirements for the OS90 genotype than for the two NZ strains, the total dry matter consumed by OS90 cows was higher than in the two NZ genotypes considering the whole season (see Chapter 5). This suggests that intake for the OS90 cows should have increased to a greater extent than for the two NZ genotypes during the dry period, which is consistent with the increased amount of supplement fed and the fact that all the genotypes had achieved similar BCS at calving for all three seasons. Nevertheless, considering the observed differences in milksolids (MS) yield and in the intake per cow for the whole season (see Chapter 5), estimated from the visual

scoring system, the gross efficiency of MS production on pasture-based systems would be lower for the OS90 genotype.

It is hypothesised that the lower loss of BCS and mobilisation of body reserves of the NZ90 cows in early lactation occurs because these cows can improve herbage dry matter intake ( $DMI_H$ ) to a greater extent than their OS90 counterparts when grazing pasture. The greater intake of the NZ90 would be sustained by an improved grazing capacity of these cows that allows them to improve energy status above maintenance requirements and sustain a higher proportion of production from the herbage consumed, hence, mobilising lower amounts of body fat reserves. The improved grazing capacity of these cows would be the result of years of genetic selection for cows that must produce and survive by grazing pasture.

Measurements of herbage intake and supplement consumption were made on all cows in the second and third seasons of the 'system' trial (2002-03 and 2003-04) and are reported in this chapter (Chapter 6). The objective of the present study is to investigate differences in intake capacity of three Holstein-Friesian genotypes managed in different pasture-based systems during the lactation and non-lactation periods; in addition, to analyse the relationships between the intakes achieved and the characteristics of the pasture grazed.

## **6.2. MATERIALS AND METHODS**

### **6.2.1. Systems and management**

A detailed description of the systems and their management was presented in Chapter 5. Briefly, this experiment was located at Dexcel No. 2 dairy farm, Hamilton, New Zealand, and was designed as a long-term system study starting in 2001 and finishing in 2004. The genotypes (GE) compared were three strains of Holstein-Friesian dairy cows with different genetic potential, as indicated by their breeding worth (BW; see Appendix IV for details). They were high BW Holstein-Friesian of OS (OS90) or NZ origin (NZ90) and a low BW NZ Friesian genotype representing the cow used in NZ during the 1970s (NZ70). In this study, eleven non-replicated systems representing real farm conditions with 15 or 20 cows each were utilised ( $n=205$  cows, see details in Chapter 5). Systems were based on pasture and also fed forage (pasture silage) and supplement (maize silage and grain), particularly during mid and late lactation and the dry periods. Genotypes were managed in self-contained feeding units or systems at a range of feed allowances (FA) from 4.5 to 7.0 t DM per cow per year. This was achieved by combining different stocking rates (SR) and inputs of supplements (see Table 5.1 in Chapter 5).

Cows of each genotype were allocated to each system before June 2001 and were maintained in the same system during seasons 2002-03 and 2003-04, when the present study was completed. Non-pregnant cows at the end of each season were culled and replaced by two year old cows of the same genotype, at a similar rate for the different systems, as a result, systems had two and three year old cows in 2002-03 and two, three and four year old cows in 2003-04, and similar age structure. The range of FA at which each genotype was managed was determined by the different energy requirements of the genotypes, determined by differences in live weight yield potential between strains (see Chapter 5). All feeding levels were not represented in the three genotypes (e.g. the NZ70 was not managed at the second lowest FA); furthermore, systems were not managed at exactly the same mean FA.

### 6.2.2. Pasture and animal measurements

Herbage mass (HM) was measured pre- and post-grazing ( $HM_{PRE}$  &  $HM_{POST}$ ) with a Rising Plate Meter (RPM) (Hodgson *et al.*, 1999); sward surface height (SH) of the same paddocks was also measured ( $SH_{PRE}$  &  $SH_{POST}$ ) with a sward stick.

The RPM was calibrated by using the quadrat technique (Hodgson *et al.*, 1999). Eleven square sampling areas (each quadrat = 0.33 m<sup>2</sup>) were located at random in two different paddocks each week during each sampling period, four of the quadrats in a recently grazed paddock and seven in another paddock that was about to be grazed. Fewer numbers of quadrats were measured in the ungrazed area because the residual post grazing herbage mass was homogeneous. The amount of herbage in each sampling area was first measured with the RPM and then cut to ground level and collected. All the samples were washed in the laboratory, oven dried at 60 °C for 48 hours and weighed. A different equation was calculated for each stage of lactation and the dry period from all of the data from the weekly cuts (two seasons). The relationship between the mean RPM values and the weight (DM) of the herbage collected from each quadrat was estimated. These equations were utilised to transform the mean pre- and post-grazing RPM readings into DM values per hectare (Table 6.1).

**Table 6.1: Rising plate meter equations for different stage of lactation and the dry period.**

Period	Equation	r <sup>2</sup>
Early lactation	$y = 646 + 141 x$	0.77
Mid lactation	$y = 1123 + 136 x$	0.84
Late lactation	$y = 863 + 148 x$	0.77
Dry period	$y = 60 + 118 x$	0.88

Equations were estimated for each period from calibration cuts to ground level. 'y' is the estimated herbage mass (HM) and 'x' is the compressed height measured with the instrument.

Measurements were made on three different paddocks each week, during three consecutive weeks, at each stage of lactation plus the dry period. About a hundred readings with the RPM and twenty readings with the sward stick were obtained along each diagonal transects and the mean obtained.

The bulk density ( $B_D$ ) of the pre-grazing pasture was estimated as the  $HM_{PRE}$  per centimetre of height ( $SH_{PRE}$ ). From the difference in height between pre- and post-grazing, the height reduction of the sward ( $H_R$ ) was calculated ( $SH_{POST}$  to  $SH_{PRE}$ ). The  $H_R$  represents the height of the horizon removed by the cows during each grazing. In addition, the  $H_R$  times the  $B_D$  of the pasture is an estimate of the amount of the herbage removed (DM).

Daily hand-plucked pasture samples were collected from the pre-grazing stratum of the same paddocks, which represented the herbage offered to each herd each day. Daily samples were bulked on a weekly basis within each farmlet, oven dried (at 60 °C for 48 hours) and analysed by Near Infrared Reflectance Spectroscopy (NIRS)(Shenk & Westerhaus, 1994).

The individual daily dry matter herbage intake ( $DMI_H$ ); forage (e.g. pasture silage) and supplements fed (maize grain and silage,  $DMI_S$ ); the intake of herbage plus forage and supplements ( $DMI_T$ ); and the digestibility of the diet consumed (DMD) from each cow in the experiment were also estimated once in early, mid and late lactation of seasons 2002-03 and 2003-04 and the dry period between these seasons, using a combination of the of the n-alkanes (Mayes *et al.*, 1986; Dove & Mayes, 1991; Dove, 1992; Dillon, 1993; Dove & Mayes, 1996; Hameleers & Mayes, 1998b; Hameleers & Mayes, 1998a) and stable carbon isotope discrimination technique ( $\delta^{13}C$ )(Jones *et al.*, 1979; Garcia *et al.*, 2000). Details of the procedure used were explained in detail in Chapter 4.

Cows were milked twice a day during the whole lactation period and the mean daily yields of milk, fat, protein and lactose were estimated for each cow once a week as detailed in Chapter 5. In addition, cows were weighed and condition scored weekly (Macdonald & Roche, 2004). The mean LW and BCS of the cows in each system were calculated for early, mid and late lactation in seasons 2002-03 and 2003-04 and for the dry period between seasons.

### 6.2.3. Energy requirements

The mean energy required for maintenance (including an activity allowance) and milk yield during lactation was calculated from the LW, daily milk yield and its solids content,

for each individual cow in each system (estimated for early, mid and late lactation without considering body reserves mobilised or gained).

The energetic expenditure in grazing activity was estimated as an increase of 0.1 in the energy required for maintenance; in addition, the energy expended in walking a mean distance of 4 km from the paddock to the milking shed daily, considering an energy cost equal to  $2 \text{ kJ kg}^{-1} \text{ LW km}^{-1}$  (Agnew & Yan, 2000).

The mean dietary metabolisable energy (ME) required per unit of milk component was also estimated (Reynolds & Beever, 1995), assuming that 0.1 of the glucose utilised in milk synthesis by the cow was derived from amino acids (Dado *et al.*, 1993). The daily energy required for maintenance is usually calculated from the net energy required for maintenance per unit of  $\text{LW}^{0.75}$  ( $\text{NE}_m$ ) based on fasting metabolism data (fasting heat production + fasting urinary energy output) plus an activity allowance. This is affected by the efficiency of utilisation of the metabolisable energy for maintenance [ $k_m = (0.35 q_m) + 0.503$ ,  $q_m = \text{dietary ME/GE}$ , GE being: gross energy (Reynolds & Beever, 1995; Agnew & Yan, 2000)] and  $k_m$  is directly related to  $q_m$ , and both are affected by the quality of the feed consumed. The  $\text{NE}_m$  used in the current energy systems is  $0.35 \text{ MJ (kg}^{0.75})^{-1}$ , but this value would be accurate only for today's low producing cows (Agnew & Yan, 2000); thus a more accurate estimate for a modern high yielding dairy cow was proposed for the high yielding cows of today [ $0.45 \text{ MJ (kg}^{0.75})^{-1}$  in the present study (Yan *et al.*, 1997; Agnew & Yan, 2000)]. In addition, the energetic value of the feed offered was represented by the value  $q_m$  (Reynolds & Beever, 1995), similar for all the cows in each system.

The mean energy in the feed consumed was also estimated. The energy required was discounted from the energy consumed to obtain an estimate of the daily energy balance for the genotypes.

#### 6.2.4. Statistical analysis

Systems were not replicated, instead large treatment groups representing real farm conditions were used (see Chapter 5). Animal and pasture measurement were collected each week of a three-week period, at each stage of lactation and in the dry period. The individual  $\text{DMI}_H$  and  $\text{DMI}_T$ , LW, DMD; LW and BCS and yield of all the cows in the systems (15 – 20 cows, 11 systems) were measured. The pastures allocated to the cows in each system each period were measured pre- and post-grazing (three different paddocks grazed on consecutive days each week), the area allocated per cow and DHA also recorded.

The statistical procedures of SAS (SAS, 2002) were used to analyse the data by using the MIXED model (PROC MIXED) by fitting parallel lines for the genotypes with  $\beta$  representing the slope across FA. The model  $Y_{ij} = \mu + GE_i + \beta (FA)_j + e_{ij}$  was utilised for the main effect and the model  $Y_{ijk} = \mu + GE_i + \beta_i (FA)_j + (GE*FA)_{ij} + e_{ijk}$  for the GE by FA interaction.

A simple linear correlation analysis (PROC CORR) was used to show the degree of relationship between pasture variables; between yields of milk and MS and DMI<sub>H</sub> per unit of LW; and between DMI<sub>H</sub> per unit of LW and DMD with the main pasture variables identifying the condition of the sward grazed in each system. The mean values from each stage of lactation and all the systems were considered. Data from dates where supplement was fed to the cows were not included, hence only grazing situations were considered to estimate these correlations. The linear relationships (PROC REG) between B<sub>D</sub>, NDF and H<sub>R</sub> with the SH<sub>PRE</sub> of the pasture were also estimated using data from each system from the similar data set; in particular the relationship between H<sub>R</sub> and SH<sub>PRE</sub> was explored for each genotype.

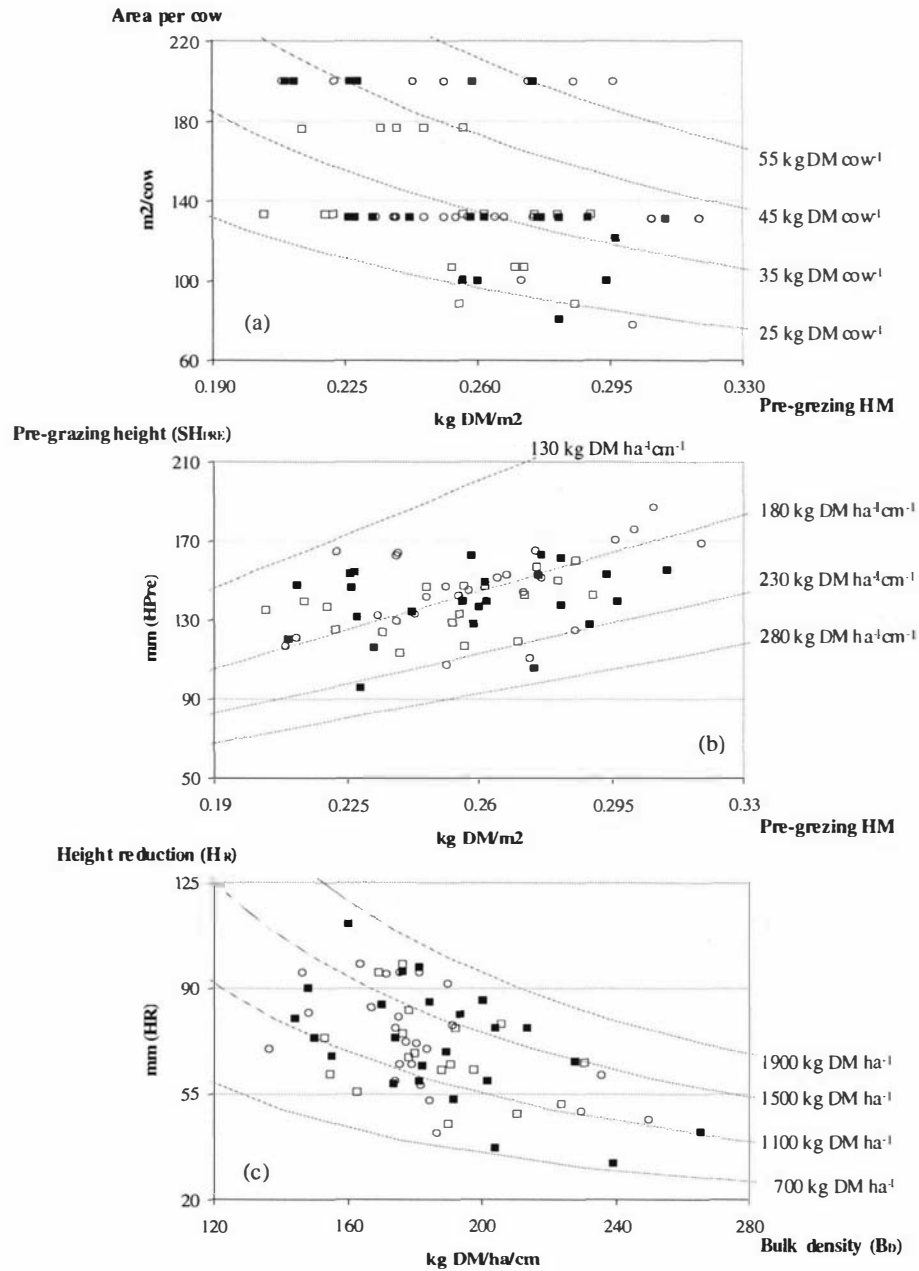
In addition, the mean weekly DMI<sub>H</sub> from three consecutive daily pre- and post-grazing visual scores was estimated for each system on three different weeks (See calculation of the apparent mean daily dry matter intake of herbage DMI<sub>V</sub> in Chapter 5), in coincidence with the use of n-alkane for individual animals (for lactating and non-lactating animals). The linear relationship between these data and the mean DMI<sub>H</sub> estimated from the n-alkane method, during the same period was estimated.

## 6.3. RESULTS

### 6.3.1. Grazing management

The GE were compared at the mean FA of the range at which they were farmed, which differed in absolute values between GE, because they considered the different energy demands of each strain (see Table 5.1 in Chapter 5 for details). In addition, FAs were compared at the mean values for all three genotypes (e.g. FA1 is the average of the values obtained in systems farmed with different genotypes at comparable FA equal to 5 t/cow per year, and FA4 is the average of the values obtained at the highest feeding level, equal to 6.5 t cow<sup>-1</sup>year<sup>-1</sup>). The experimental design was unbalanced and not all the levels of feed were represented in the three genotypes, hence data for FA2 is just the mean for NZ90 and OS90 (FA2 equal to 5.5 t/cow per year).

**Figure 6.1:** Description of grazing management and the main pre-grazing sward conditions in the pastures grazed by three Holstein-Friesian genotypes managed at different feed allowance. Area daily allocated per cow (a), pre-grazing sward surface height at each level of mass (b) and reduction in height pre-/post-grazing for each bulk density (c).



NZ70 (□); NZ90 (■); OS90 (○). Each data point corresponds to the mean grazing condition for each of the systems (genotype by feed allowance) in the experiment during the lactation period of seasons 2002-03 and 2003-04 and the dry period between the two lactations. In (a) the isolines indicate equal daily herbage allowance (kg DM cow<sup>-1</sup>day<sup>-1</sup>); in (b) isolines indicate similar pasture bulk density (kg DM ha<sup>-1</sup>cm<sup>-1</sup>); and in (c) isolines indicate equal amount of pasture removed (kg DM ha<sup>-1</sup>) each grazing for cows that removed herbage according to the difference in pasture height between pre-/post-grazing (H<sub>R</sub>) and its bulk density.

The mean rate of change for each response variable is expressed in units of change in each variable per tonne DM increase in the nominal FA, within the range at which each genotype was farmed. It was determined that the mean actual amount of feed offered in the systems (TF<sub>O</sub>) increased by 0.74 t DM t<sup>-1</sup>DM of nominal total feed offered (NTF<sub>O</sub>; see Chapter 5). As a result, the actual rate of change for each response variable was higher when expressed per tonne of TF<sub>O</sub>.

The daily herbage allowance (DHA) and total allowance (DTA) offered to the cows in the systems changed as lactation progressed according to growing conditions and supplement inputs (see Appendix V-15, V-16 and V-17). Both DHA and DTA increased as FA increased, mainly as result of both more pre-grazing herbage mass (range between 2,000-3,200 kg DM ha<sup>-1</sup>) and increased grazed area (Figure 6.1a).

Systems managed at the lowest FA within each GE were fed only pasture; cows were offered low DHA by grazing a small area with high HM<sub>PRE</sub>. In contrast, in systems managed at the highest FA cows were offered high DHA by grazing a larger area with lower HM<sub>PRE</sub>. Pastures were grazed in a shorter rotation length to avoid a decline in pasture quality, particularly during periods of fast growth rate (Table 6.2).

These systems were fed supplements in mid and late lactation to achieve an even greater DTA. Differences in pasture management resulted in GE by FA interactions for sward conditions, for instance for HM<sub>PRE</sub> and SH<sub>PRE</sub>, which confounded the effects of DHA and sward conditions on DMI<sub>H</sub> and cow performance (see Appendix V).

**Table 6.2: Grazing rotation length and instantaneous stocking rate during early, mid and late lactation (two lactations) and the dry period of three Holstein-Friesian genotypes managed in systems at different feed allowances.**

			NZ70			NZ90				OS90			
			FA1	FA3	FA4	FA1	FA2	FA3	FA4	FA1	FA2	FA3	FA4
Season 2002-2003	Early	RL (days)	20	24	18	23	25	25	16	25	25	16	16
		ISR (cows ha <sup>-1</sup> )	75	75	57	76	76	76	50	76	76	50	50
	Mid	RL (days)	20	20	15	22	21	21	14	21	21	14	14
		ISR (cows ha <sup>-1</sup> )	94	75	57	84	76	76	50	76	76	50	50
	Late	RL (days)	31	30	24	38	32	25	25	32	25	25	25
		ISR (cows ha <sup>-1</sup> )	114	94	75	125	100	76	76	100	76	76	76
Dry	RL (days)	81	73	73	91	81	79	65	81	81	65	65	
	ISR (cows ha <sup>-1</sup> )	300	226	226	301	250	242	200	250	250	200	200	
Season 2003-2004	Early	RL (days)	20	18	24	23	25	16	16	25	25	16	16
		ISR (cows ha <sup>-1</sup> )	75	57	75	76	76	50	50	76	76	50	50
	Mid	RL (days)	20	15	18	20	25	14	16	21	21	14	16
		ISR (cows ha <sup>-1</sup> )	75	57	57	76	76	50	50	76	76	50	50
	Late	RL (days)	31	30	24	30	32	25	25	42	25	25	25
		ISR (cows ha <sup>-1</sup> )	114	94	75	100	100	76	76	134	76	76	76

FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; RL: rotation length; ISR: instantaneous stocking rate. RL: rotation length; ISR: instantaneous stocking rate (cows ha<sup>-1</sup> for 24 h.)

**Table 6.3: Daily herbage allowance per cow, pre- and post-grazing herbage mass and sward height, and composition of the herbage grazed of three Holstein-Friesian genotypes during lactation and dry periods. Mean values across feed allowances for the genotypes and rate of change within the range of feed allowances at which each genotype was farmed.**

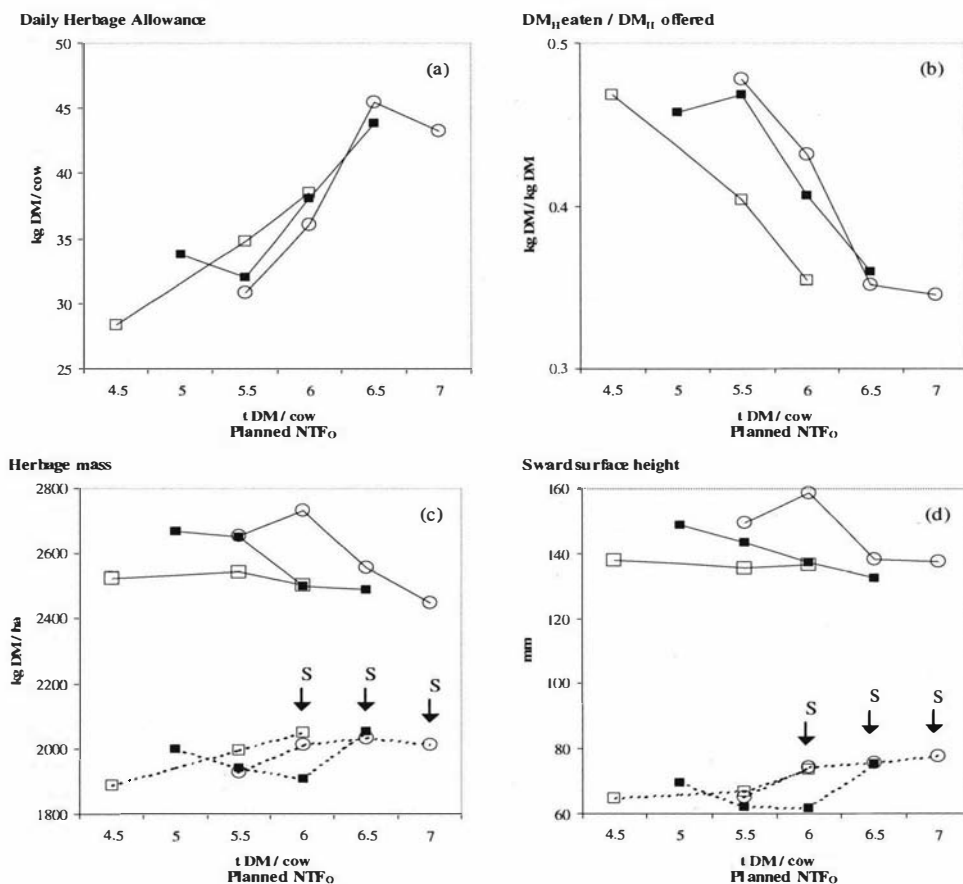
		Genotypes				Feed allowance		Significance		
		NZ70	NZ90	OS90	SED	Slope	SED	GE	FA	GE*FA
Lactation period										
Daily herbage allowance per cow and LW unit										
DHA	kg DM cow <sup>-1</sup>	33.3	36.8	38.9	1.9	7.8	1.4	NS	***	NS
DHA/LW	g DM kg <sup>-1</sup> LW	75.9	79.6	80.8	4.5	14.9	1.58	NS	**	NS
DTA	kg DM cow <sup>-1</sup>	33.7	37.8	40.1	2.0	8.8	1.4	*	***	NS
DTA/LW	g DM kg <sup>-1</sup> LW	76.9	81.6	83.3	4.6	16.9	3.2	NS	***	NS
Pre- and post-grazing sward conditions										
HM <sub>PRE</sub>	kg DM ha <sup>-1</sup>	2531	2572	2598	54	-105	38	NS	*	NS
HM <sub>POST</sub>	kg DM ha <sup>-1</sup>	1973	1983	1995	34	63	24	NS	*	NS
SH <sub>PRE</sub>	mm	137	139	146	4.5	-8.2	3.1	0.195	*	NS
SH <sub>POST</sub>	mm	68	68	73	3.0	5.7	1.1	0.186	*	NS
Bulk <sub>D- PRE</sub>	kg DM ha <sup>-1</sup> cm <sup>-1</sup>	183	188	182	3.6	4.0	2.5	NS	0.148	NS
Composition of the pre-grazing herbage										
NDF	g kg <sup>-1</sup> DM	465.8	464.6	457.4	5.8	7.7	3.7	NS	0.075	NS
ADF	g kg <sup>-1</sup> DM	212.8	216.7	216.4	2.7	3.3	1.7	NS	0.093	NS
CP	g kg <sup>-1</sup> DM	256.6	250.7	256.2	6.8	4.3	2.0	NS	*	NS
OMD	g kg <sup>-1</sup> DM	81.9	82.8	82.1	0.7	-0.22	0.49	NS	NS	NS
Energy	MJ ME kg <sup>-1</sup> DM	11.7	11.9	11.8	0.11	-0.02	0.07	NS	NS	NS
Dry period										
Daily herbage allowance										
DHA	kg DM cow <sup>-1</sup>	7.0	7.8	9.0	0.69	0.38	0.48	NS	NS	NS
DHA/LW	g DM kg <sup>-1</sup> LW	14.4	14.8	15.9	1.26	0.27	0.88	NS	NS	NS
DTA	kg DM cow <sup>-1</sup>	9.4	10.6	11.9	1.02	2.04	0.74	NS	*	NS
DTA/LW	g DM kg <sup>-1</sup> LW	19.4	20.2	21.1	2.11	3.44	1.48	NS	0.05	NS
Pre- and post-grazing sward conditions										
HM <sub>PRE</sub>	kg DM ha <sup>-1</sup>	1793	1924	2001	124	-327	87	NS	**	0.093
HM <sub>POST</sub>	kg DM ha <sup>-1</sup>	684	723	754	43	39	30	NS	NS	NS
SH <sub>PRE</sub>	mm	177	198	195	0.78	-11.6	5.4	0.069	0.072	NS
SH <sub>POST</sub>	mm	52	57	59	0.38	10.8	2.6	NS	**	NS
Composition of the pre-grazing herbage										
NDF	g kg <sup>-1</sup> DM	471	476	475	15.7	0.0	11.0	NS	NS	0.053
ADF	g kg <sup>-1</sup> DM	225	217	219	7.0	-6.0	4.9	NS	NS	NS
CP	g kg <sup>-1</sup> DM	257	251	256	6.2	12.8	4.3	NS	*	NS
OMD	g kg <sup>-1</sup> DM	81.1	81.0	82.1	1.8	2.76	1.29	NS	0.070	0.162
Energy	MJ ME kg <sup>-1</sup> DM	11.4	11.5	11.6	0.27	0.4	0.2	NS	0.078	0.143

DMI<sub>h</sub>: herbage dry matter intake; LW: live weight; DMD: diet dry matter digestibility; HM<sub>PRE</sub>: pre-grazing herbage mass; SH<sub>PRE</sub>: pre-grazing sward height; H<sub>R</sub>: height reduction between pre-/post-grazing of the pasture; B<sub>P</sub>: bulk density of the pre-grazing pasture estimated as HM<sub>PRE</sub> / SH<sub>PRE</sub>; NDF: neutral detergent fiber; ADF: acid detergent fiber; CP: crude protein; OMD: organic matter digestibility. The 'Slope' indicates the mean rate of change of each variable (considering all the systems) per tone of 'nominal' feed allowance per cow per annum (NTF<sub>0</sub> from Table 5.1). GE: genotype; FA: annual feed allowance; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = not significant.

Despite differences in grazing management, mainly determined by DHA, sward characteristics were within a similar range for all three genotypes during the experimental period (Figures 6.1a, 6.1b and 6.1c). This might have provided similar opportunities for all three genotypes to express their capacity to graze. For instance, the HM<sub>PRE</sub> at which the pastures were grazed varied between 2 and 3.1 t ha<sup>-1</sup> and the daily area grazed was between 70 and 190 m<sup>2</sup> cow<sup>-1</sup>, with a wide range of DHA (25-55 kg DM cow<sup>-1</sup>) in the systems (Figure 6.1a). Within the range of HM<sub>PRE</sub> at which the pastures were grazed,

differences in  $SH_{PRE}$  were observed (90-180 mm), also indicating a similar range of  $B_D$  (130-280 kg DM ha<sup>-1</sup>cm<sup>-1</sup>; Figure 6.1b). As a result, the amount of DM removed from the pasture might have been affected by the capacity of the cow to harvest different proportions of the initial  $SH_{PRE}$  in pastures of different  $B_D$  (Figure 6.1c).

**Figure 6.2** Daily herbage allowance (a), grazing efficiency (b), pre- and post-grazing herbage mass (c) and pre- and post-grazing sward height (d) in systems managed with three Holstein-Friesian genotypes in different feeding systems (mean of early, mid and late lactation periods).



NZ70 (□); NZ90 (■); OS90 (○). In (b) grazing efficiency was estimated as the proportion of herbage consumed ( $DM_{it}$ ) of the total herbage offered (DHA) both expressed per unit of LW. In (c) and (d) 'S' indicates systems that were supplemented with maize silage in mid and/or late lactation; however note that some systems were also fed grass silage in late lactation 2003-04 (NZ70: FA2; NZ90: FA1, FA2 and FA3; OS90: FA1). Pre-grazing herbage mass and height in solid lines at the top of figures (c) and (d), and post-grazing in dotted lines at the bottom.

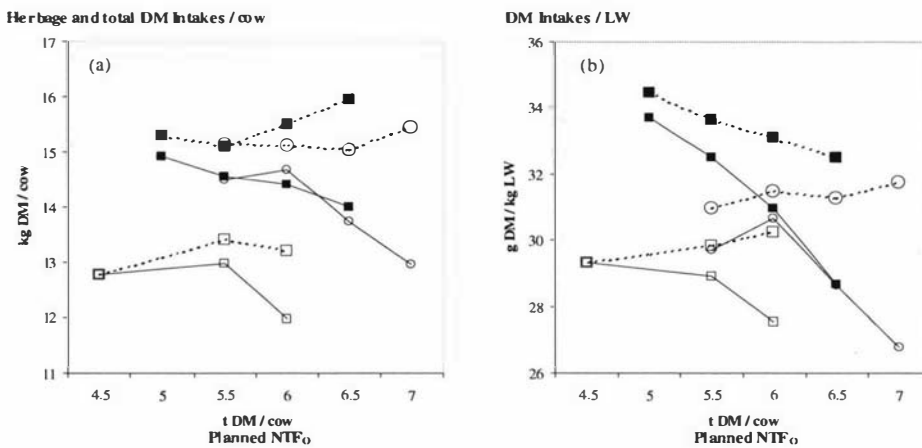
### 6.3.2. Lactation period

The mean DHA for OS90 (39 kg cow<sup>-1</sup>) was slightly greater than for NZ90 (37 kg cow<sup>-1</sup>), and higher in these two genotypes than in NZ70 (33 kg cow<sup>-1</sup>) although not significantly different (Table 6.3). A significant effect of FA on DHA was observed with a significant increase in DHA of 7.8 kg cow<sup>-1</sup> t<sup>-1</sup>DM of  $\text{NTF}_0$  (Figure 6.2a). A significant effect of GE

was observed for DTA, with a trend similar to that observed for DHA, but with larger differences between genotypes due to the supplement fed.

The mean DTA for the OS90 genotype was about 40 kg cow<sup>-1</sup>, slightly greater than NZ90 (38 kg cow<sup>-1</sup>), and these were higher than for NZ70 (34 kg cow<sup>-1</sup>). A significant effect of FA on DTA was also observed with DTA increasing by 8.8 kg cow<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>0</sub> (Table 6.3). There were no significant differences in DHA/LW and DTA/LW across genotypes, probably due to the different LW of the genotypes, with a significant effect of FA on DHA/LW and DTA/LW (Table 6.3).

**Figure 6.3: Daily herbage (solid lines) and total dry matter (dotted lines) intakes per cow (a) and per unit of live weight (b) achieved in systems managed with three Holstein-Friesian genotypes in different feeding systems during lactation.**



NZ70 (□); NZ90 (■); OS90 (○). In (a) daily DMI<sub>H</sub> and in (b) daily DMI<sub>H</sub> per unit of LW were plotted in solid lines; and in (a) daily DMI<sub>T</sub> and in (b) daily DMI<sub>T</sub> per unit of LW were plotted in dotted lines. DMI<sub>H</sub> and DMI<sub>T</sub> are means of early, mid and late lactation for each combination of genotype and feed allowance.

The differences observed in HM<sub>PRE</sub> between GE were not significant (mean of three GE: 2567 kg DM ha<sup>-1</sup>; Table 6.3) and declined significantly as FA increased (105 kg DM ha<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>0</sub>; Figure 6.2c). The differences in HM<sub>POST</sub> across genotypes were not significant (mean of three GE: 1984 kg DM ha<sup>-1</sup>; Table 6.3), which increased as FA increased (63 kg DM ha<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>0</sub>; Figure 6.2d).

There were non-significant differences across GE for SH<sub>POST</sub>, although the OS90 showed a trend for higher SH<sub>POST</sub> than the two NZ genotypes (73 vs. 68 mm respectively), whereas SH<sub>POST</sub> increased significantly as FA increased (6 mm t<sup>-1</sup>DM of NTF<sub>0</sub>; Figure 6.2d). The differences in SH<sub>PRE</sub> across GE were also not significant (Table 6.3), even though the OS90 showed the highest SH<sub>PRE</sub> (146 mm) and NZ70 the lowest (137 mm), in addition SH<sub>PRE</sub> declined significantly as FA increased (8 mm t<sup>-1</sup>DM of NTF<sub>0</sub>; Figure 6.2d).

**Table 6.4: Pearson correlation coefficients (*r*) for the relations between sward height reduction between pre-/post-grazing and sward conditions during lactation.**

Dependent	GE	Mean	CV%	n	Independent	GE	r	P	Mean	CV%	n	
H <sub>R</sub>	NZ70	7.02	22.3	14	SH <sub>PRE</sub>	NZ70	0.69	**	13.6	10.1	14	
						NZ90	0.79	***	14.4	10.2	17	
						OS90	0.71	**	14.3	14.3	16	
						SH <sub>POST</sub>	NZ70	-0.53	*	6.6	17.7	14
							NZ90	-0.39	0.12	6.3	15.4	17
							OS90	-0.03	NS	6.8	20.1	16
						B <sub>D</sub>	NZ70	-0.23	NS	184	11.9	14
							NZ90	-0.54	*	185	14.9	17
							OS90	-0.52	*	181	15.9	16
						NDF	NZ70	-0.18	NS	38.6	14.2	14
							NZ90	-0.47	0.05	36.6	12.5	17
							OS90	-0.05	NS	36.5	12.9	16

Only dates pasture alone was fed (not supplements) were considered. SH<sub>PRE</sub>: pre-grazing sward height; SH<sub>POST</sub>: post-grazing sward height; H<sub>R</sub>: height reduction between pre-/post-grazing of the pasture; B<sub>D</sub>: bulk density of the pre-grazing pasture estimated as HM<sub>PRE</sub> % SH<sub>PRE</sub>; NDF: neutral detergent fiber. GE: genotype; P: probability [significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.]; r: coefficient of correlation; CV%: coefficient of variation; n: sample number.

**Table 6.5: Linear relationships between bulk density and NDF content of the herbage pre-grazing with sward height, and between sward height reduction pre-/post-grazing with sward height pre-grazing.**

Regression equation		r <sup>2</sup>	RMSE	P	n
<b>DMI<sub>H</sub>/LW vs. DHA</b>					
DMI <sub>H</sub> = 31.1 + 0.02 DHA		0.002	3.25	0.77	47
<b>B<sub>D</sub> vs. SH<sub>PRE</sub></b>					
B <sub>D</sub> = 327.9 – 10.29 SH <sub>PRE</sub>		0.43	19.9	<0.001	47
<b>NDF vs. SH<sub>PRE</sub></b>					
NDF = 44.4 – 0.52 SH <sub>PRE</sub>		0.03	4.86	0.24	47
<b>Grazing efficiency vs. DHA</b>					
Only pasture	G <sub>EF</sub> = 0.784 – 0.01 DHA	0.75	0.042	<0.001	47
	NZ70 G <sub>EF</sub> = 0.797 – 0.01 DHA	0.88	0.029	<0.001	14
	NZ90 G <sub>EF</sub> = 0.859 – 0.01 DHA	0.82	0.032	<0.001	17
	OS90 G <sub>EF</sub> = 0.796 – 0.01 DHA	0.80	0.041	<0.001	16
<b>H<sub>R</sub> vs. SH<sub>PRE</sub></b>					
All dates	H <sub>R</sub> = – 2.40 + 0.68 SH <sub>PRE</sub>	0.51	1.17	<0.001	66
Only pasture	H <sub>R</sub> = – 1.98 + 0.68 SH <sub>PRE</sub>	0.52	1.08	<0.001	47
	NZ70 H <sub>R</sub> = – 3.75 + 0.79 SH <sub>PRE</sub>	0.48	1.18	<0.001	14
	NZ90 H <sub>R</sub> = – 3.89 + 0.83 SH <sub>PRE</sub>	0.63	0.96	<0.001	17
	OS90 H <sub>R</sub> = 0.021 + 0.52 SH <sub>PRE</sub>	0.50	1.09	0.002	16
Pasture + supplement	H <sub>R</sub> = – 4.30 + 0.74 SH <sub>PRE</sub>	0.80	0.78	<0.001	19

Linear regressions were estimated for the whole lactation period only for dates that the cows in the systems were grazing only (no supplements were fed), otherwise it is indicated. SH<sub>PRE</sub>: pre-grazing sward height; H<sub>R</sub>: height reduction between pre-/post-grazing of the pasture. P: probability [significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.]; r<sup>2</sup>: coefficient of determination; RMSE: root mean square error; n: sample number.

The differences in pasture B<sub>D</sub> between GE and FA were not significant (mean 185 kg DM ha<sup>-1</sup>cm<sup>-1</sup>); however, they showed a trend to increase with FA (Table 6.3). Similar results were measured for NDF (463 g kg<sup>-1</sup> DM), ADF (215 g kg<sup>-1</sup> DM) and CP (254 g kg<sup>-1</sup> DM) while the ME concentration of the pasture showed a trend to decrease (11.8 ME kg<sup>-1</sup> DM);

Table 6.3). There was a significant increase of CP with FA ( $4.3 \text{ g kg}^{-1} \text{ DM t}^{-1} \text{ DM}$  of  $\text{NTF}_0$ ).

For all three genotypes, but particularly for NZ70,  $H_R$  was positively correlated with  $\text{SH}_{\text{PRE}}$  and negatively with  $\text{SH}_{\text{POST}}$  (Table 6.4). Herbage height reduction was also negatively correlated with  $B_D$ , although this was only significant for NZ90 and OS90, and negatively correlated with NDF, although this was significant only for NZ90.

A significant linear relationship was measured between  $B_D$  and  $\text{SH}_{\text{PRE}}$  (Table 6.5) indicating a decline in  $B_D$  as  $\text{SH}_{\text{PRE}}$  increased, which is in agreement with previous correlations, and with the significant linear relationship between  $H_R$  and  $\text{SH}_{\text{PRE}}$ . This relationship showed a different response of  $H_R$  to increments in  $\text{SH}_{\text{PRE}}$  for systems fed only pasture, where  $H_R$  showed a trend to be greater than in systems fed supplements. Differences between the NZ90 and OS90 genotypes on pasture were also observed, with  $\text{SH}_{\text{PRE}}$  explaining a greater proportion of the variation in  $H_R$  for the NZ90 genotype than in the OS90 strain (Table 6.5). There was a non-significant trend for a decline in NDF content in the herbage offered as  $\text{SH}_{\text{PRE}}$  increased, which suggests that only small changes in quality occurred as  $\text{SH}_{\text{PRE}}$  increased, probably because the sample of herbage collected from the pasture avoided the herbage below the grazing horizon.

The significant linear relationship measured between grazing efficiency ( $G_{\text{EF}}$ ), estimated as  $\text{DMI}_H/\text{LW}$  expressed as a proportion of  $\text{DHA}/\text{LW}$  (Combellas & Hodgson, 1979), and  $\text{DHA}$ , indicate a decline in  $G_{\text{EF}}$  with the increase in  $\text{DHA}$ . In addition, no significant relationship was observed between  $\text{DMI}_H$  and  $\text{DHA}$ .

### 6.3.3. Dry period

The OS90 had the highest  $\text{DHA}$  ( $9.0 \text{ kg cow}^{-1}$ ) and the NZ70 had the lowest ( $7.0 \text{ kg cow}^{-1}$ ; Table 6.3); the effect of FA on  $\text{DHA}$  did not differ between genotypes. The effect of GE on  $\text{DTA}$  was non-significant, although  $\text{DTA}$  tended to be higher for the OS90 ( $11.9 \text{ kg cow}^{-1}$ ) and lower for NZ70 ( $9.4 \text{ kg cow}^{-1}$ ). A significant effect of FA on  $\text{DTA}$  was observed ( $+2.0 \text{ kg cow}^{-1}$  in  $\text{DTA t}^{-1} \text{ DM}$  of  $\text{NTF}_0$ ). Neither the effect of GE nor FA on  $\text{DHA}/\text{LW}$  were significant, however a significant effect of FA on  $\text{DTA}/\text{LW}$  was measured ( $+3.44 \text{ g kg}^{-1} \text{ LW}$  in  $\text{DTA}/\text{LW t}^{-1} \text{ DM}$  of  $\text{NTF}_0$ ; Table 6.3).

The effect of GE on  $\text{HM}_{\text{PRE}}$  was not significant ( $1906 \text{ kg DM ha}^{-1}$ ; Table 6.3) but a significant effect of FA indicated a decline of  $327 \text{ kg DM ha}^{-1}$  in  $\text{HM}_{\text{PRE t}^{-1} \text{ DM}}$  of  $\text{NTF}_0$  (Figure 6.3). No significant effects of GE and FA on  $\text{HM}_{\text{POST}}$  were observed ( $720 \text{ kg DM ha}^{-1}$ ). Both  $\text{SH}_{\text{PRE}}$  ( $190 \text{ mm}$ ) and  $\text{SH}_{\text{POST}}$  ( $56 \text{ mm}$ ) did not differ across GE. In addition,

SH<sub>PRE</sub> showed a significant decline of 12 mm t<sup>-1</sup>DM of NTF<sub>O</sub>, and SH<sub>POST</sub> a significant increase of 11 mm t<sup>-1</sup>DM of NTF<sub>O</sub>.

There was no significant effect of GE on the NDF (474 g kg<sup>-1</sup> DM), ADF (220 g kg<sup>-1</sup> DM), CP (255 g kg<sup>-1</sup> DM), OMD (81.4 g kg<sup>-1</sup>) and the mean energy value (11.5 ME kg<sup>-1</sup> DM; Table 6.3). No significant effect of FA was measured although CP increased significantly by 13 g kg<sup>-1</sup> DM t<sup>-1</sup>DM of NTF<sub>O</sub>.

#### 6.3.4. Intakes and diet digestibility during the lactation period

The mean DMI<sub>H</sub> was higher for NZ90 (14.5 kg DM cow<sup>-1</sup>) than OS90 (13.9 kg DM cow<sup>-1</sup>) and greater for these two strains than for NZ70 (12.6 kg DM cow<sup>-1</sup>; Table 6.6). Herbage intake decreased as FA increased by 0.71 kg DM cow<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>O</sub> (Figure 6.3a), mainly as a result of the increased amount of supplement consumed at the highest feeding levels.

**Table 6.6: Daily intakes and diet digestibility of three Holstein-Friesian genotypes during lactation and dry periods. Mean values across feed allowances for the genotypes and rate of change within the range of feed allowances at which each genotype was farmed.**

		Genotypes				Feed allowance		Significance		
		NZ70	NZ90	OS90	SED	Slope	SED	GE	FA	GE*FA
<b>Lactation period</b>										
DMI <sub>H</sub>	kg DM cow <sup>-1</sup>	12.6	14.5	13.9	0.28	-0.71	0.20	***	**	NS
DMI <sub>T</sub>	kg DM cow <sup>-1</sup>	13.1	15.5	15.2	0.16	0.32	0.11	***	*	NS
DMI <sub>H</sub> /LW	g DM kg <sup>-1</sup> LW	28.8	31.5	28.9	0.71	-1.10	0.25	**	**	0.178
DMI <sub>T</sub> /LW	g DM kg <sup>-1</sup> LW	29.8	33.4	31.4	0.48	-0.05	0.17	***	NS	***
DMD	g kg <sup>-1</sup> DM	779.8	783.0	779.4	3.51	0.63	2.46	NS	NS	NS
<b>Dry period</b>										
DMI <sub>H</sub>	kg DM cow <sup>-1</sup>	5.4	5.8	5.8	0.75	-1.22	0.53	NS	NS	NS
DMI <sub>T</sub>	kg DM cow <sup>-1</sup>	7.8	8.7	8.7	0.55	0.44	0.39	NS	NS	NS
DMI <sub>H</sub> /LW	g DM kg <sup>-1</sup> LW	11.16	11.1	10.2	1.44	-2.69	1.01	NS	*	0.139
DMI <sub>T</sub> /LW	g DM kg <sup>-1</sup> LW	16.10	16.5	15.4	1.03	0.48	0.72	NS	NS	NS
DMD	g kg <sup>-1</sup> DM	733	726	739	16.2	-7.82	11.39	NS	NS	NS

DMI<sub>H</sub>: herbage dry matter intake; DMI<sub>T</sub>: total dry matter intake; LW: live weight; DMD: diet dry matter digestibility. GE: genotype; FA: annual feed allowance; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS: not significant.

The mean DMI<sub>T</sub> was higher for NZ90 (15.5 kg DM cow<sup>-1</sup>) than OS90 (15.2 kg DM cow<sup>-1</sup>), despite the larger amount of supplement that was fed to the latter, with a smaller difference between strains than observed for DMI<sub>H</sub> (Table 6.6). The mean DMI<sub>T</sub> was higher for these two genotypes than for NZ70 (13.1 kg DM cow<sup>-1</sup>). There was a significant effect of FA on DMI<sub>T</sub>, but in contrast to the decline observed for herbage intake, DMI<sub>T</sub> increased by 0.32 kg DM cow<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>O</sub> (Table 6.3; Figure 6.3a).

**Table 6.7: Pearson correlation coefficients (*r*) for the relations between daily herbage intake (LW unit) and diet digestibility, with the condition of the sward during lactation.**

Dependent	GE	Mean	CV%	n	Independent	GE	r	P	Mean	CV%	n
DMI <sub>H</sub> /LW	NZ70	29.9	8.7	14	DHA/LW	NZ70	0.31	0.26	76.1	21.9	14
						NZ90	-0.01	NS	81.7	14.0	17
						OS90	-0.11	NS	80.3	21.8	16
	HM <sub>PRE</sub>	29.9	8.7	14	DHA/LW	NZ70	-0.11	NS	2477	10.9	14
						NZ90	-0.12	NS	2556	11.7	17
						OS90	-0.09	NS	2524	11.1	16
	SH <sub>PRE</sub>	29.9	8.7	14	DHA/LW	NZ70	0.38	0.18	13.6	10.1	14
						NZ90	0.57	*	14.2	10.2	17
						OS90	0.16	NS	14.3	14.3	16
	H <sub>R</sub>	29.9	8.7	14	DHA/LW	NZ70	0.52	0.06	7.02	22.3	14
						NZ90	0.55	*	7.97	19.1	17
						OS90	0.21	NS	7.47	20.0	16
	B <sub>D</sub>	29.9	8.7	14	DHA/LW	NZ70	-0.40	0.15	184	11.9	14
						NZ90	-0.67	**	184	14.9	17
						OS90	-0.25	NS	181	15.9	16
	NDF	29.9	8.7	14	DHA/LW	NZ70	-0.63	*	38.6	14.2	14
						NZ90	-0.58	*	36.6	12.5	17
						OS90	-0.42	0.10	36.5	12.9	16
DMD	29.9	8.7	14	DHA/LW	NZ70	0.81	***	784	3.8	14	
					NZ90	0.64	**	794	4.1	17	
					OS90	0.52	*	785	3.5	16	
DMD	NZ70	784	3.8	14	B <sub>D</sub>	NZ70	-0.69	**	184	11.9	14
						NZ90	-0.83	***	185	14.9	17
						OS90	-0.73	**	181	15.9	16
	NDF	784	3.8	14	B <sub>D</sub>	NZ70	-0.63	*	38.6	14.2	14
						NZ90	-0.68	**	36.6	12.5	17
						OS90	-0.61	*	36.5	12.9	16

Pearson's correlations were estimated only during the lactation period, for dates were supplement was not fed. DMI<sub>H</sub>: herbage dry matter intake; LW: live weight; DMD: diet dry matter digestibility; HM<sub>PRE</sub>: pre-grazing herbage mass; SH<sub>PRE</sub>: pre-grazing sward height; H<sub>R</sub>: height reduction between pre-/post-grazing of the pasture; B<sub>D</sub>: bulk density of the pre-grazing pasture estimated as HM<sub>PRE</sub> / SH<sub>PRE</sub>; NDF: neutral detergent fiber; DMD: diet dry matter digestibility. GE: genotype; P: probability [significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.]; r: coefficient of correlation; CV%: coefficient of variation; n: sample number.

Due to the different mature LWs of the genotypes (McNaughton *et al.*, 2002) and the different LWs achieved by cows managed at different feeding levels (see Chapter 5), a more comparable measure of the intake achieved by each strain is the value of DM consumed expressed relative to the current LW of the cow (Table 6.6). Thus, an increased value for intake per unit of LW indicates the capacity of the cow to consume more dry matter in relation with her maintenance requirements. The mean DMI<sub>H</sub>/LW was higher for the NZ90 (31.5 g DM kg<sup>-1</sup> LW) than for NZ70 and OS90 genotypes (28.9 g DM kg<sup>-1</sup> LW, mean of the two genotypes), and significantly declined by 1.1 g DM kg<sup>-1</sup> LW t<sup>-1</sup>DM of NTF<sub>O</sub>. The DMI<sub>T</sub>/LW was also higher for NZ90 (33.4 g DM kg<sup>-1</sup> LW) than for NZ70 and OS90 (30.6 g DM kg<sup>-1</sup> LW). However, the change in DMI<sub>T</sub> per unit of LW between genotypes across FA showed a significant GE by FA interaction (Figure 6.3b), probably due to the larger increase in DMI<sub>T</sub> of the OS90 relative to LW, as a result of the effect of the supplement fed.

A positive coefficient of correlation ( $r$ ) was observed for the NZ70 genotype between  $DMI_H/LW$  and  $DHA/LW$ , but was not detected for NZ90 and OS90 (Table 6.7). A positive relationship was estimated for the NZ90 genotype between  $DMI_H/LW$  and  $SH_{PRE}$  and between  $DMI_H/LW$  and  $H_R$ , but not for NZ70 and OS90. Also, a negative relationship was observed between  $DMI_H/LW$  and  $NDF$  and between  $DMI_H/LW$  and  $B_D$ , and was particularly significant for the NZ90 genotype. The  $DMI_H/LW$  and  $DMD$  were highly correlated for all three genotypes.

**Table 6.8: Live weight, BCS and the daily yield of three Holstein-Friesian genotypes. Mean values across feed allowances for the genotypes and rate of change within the range of feed allowances at which each genotype was farmed.**

		Genotypes				Feed allowance		Significance		
		NZ70	NZ90	OS90	SED	Slope	SED	GE	FA	GE*FA
<b>Liveweight and BCS</b>										
LW	kg cow <sup>-1</sup>	440	464	484	9.37	11.05	6.56	0.062	0.136	*
BCS	Units	4.61	4.32	3.83	0.09	0.14	0.06	***	0.066	0.055
<b>Daily yield and milk composition</b>										
Milk	Litres	17.3	19.9	21.8	0.45	1.42	0.31	***	***	NS
MS	kg cow <sup>-1</sup>	1.37	1.64	1.64	0.04	0.10	0.02	***	***	NS
Fat	kg cow <sup>-1</sup>	0.79	0.92	0.89	0.02	0.04	0.01	***	*	NS
Protein	kg cow <sup>-1</sup>	0.58	0.72	0.75	0.02	0.06	0.01	***	***	NS
Lactose	kg cow <sup>-1</sup>	0.83	0.96	1.05	0.02	0.07	0.02	***	***	NS
Fat	%	46.6	47.3	41.6	1.18	-1.12	0.83	**	NS	0.132
Protein	%	33.5	36.7	34.7	0.69	0.43	0.48	**	NS	0.062
Lactose	%	48.0	48.3	47.9	0.32	-0.02	0.22	NS	NS	0.160
<b>Daily energy input, output and balance</b>										
En <sub>I</sub>	MJ ME day <sup>-1</sup>	154.3	183.0	178.8	2.19	3.85	1.53	***	*	NS
En <sub>O</sub>	MJ ME day <sup>-1</sup>	94.0	110.8	113.4	2.00	6.82	1.40	***	***	NS
En <sub>M</sub> <sup>(1)</sup>	MJ ME day <sup>-1</sup>	46.3	61.8	63.9	0.92	1.08	0.64	***	0.138	*
En <sub>T</sub> <sup>(1;2)</sup>	MJ ME day <sup>-1</sup>	148.4	182.5	187.6	2.84	8.09	1.99	***	**	NS
En <sub>B</sub> <sup>(1;2)</sup>	MJ ME day <sup>-1</sup>	5.87	0.54	-8.76	2.78	-4.24	1.95	*	0.067	NS

En<sub>I</sub>: energy intake; En<sub>O</sub>: energy in the milk produced; En<sub>M</sub>: daily energy requirements for maintenance; En<sub>T</sub>: daily energy requirements for maintenance plus grazing activity plus yield; En<sub>B</sub>: energy balance. (1) The energy partitioned from and to body reserves or LW change was not considered. (2) This estimate includes an allowance for the daily activity of the animal. GE: genotype; FA: annual feed allowance; GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. SED: standard error of the differences. NS= not significant.

The digestibility of the diet consumed by the NZ90 cows during the whole lactation was slightly higher than the mean for the period (781 g kg<sup>-1</sup> DM) but non-significantly different between strains (Table 6.6). The effect of FA on DMD was not significant, with the DMD of all the genotypes being reduced when the supplement was fed. Diet digestibility was highly correlated with  $B_D$  and  $NDF$  content of the pasture, particularly for the NZ90 strain.

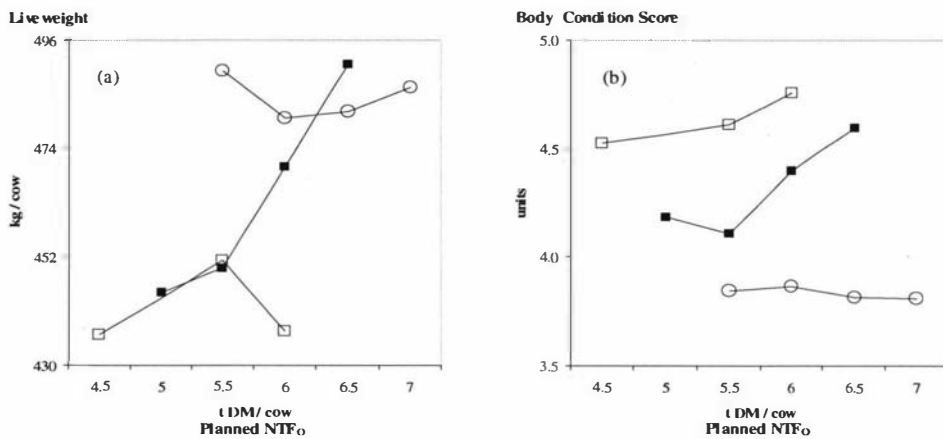
### 6.3.5. Intakes and diet digestibility during the non-lactation period

There was a not significant effect of GE on  $DMI_H$  ( $5.7 \text{ kg DM cow}^{-1}$ ) but a significant effect of FA showed a decline of  $1.2 \text{ kg DM cow}^{-1} \text{ t}^{-1}\text{DM}$  of  $NTF_0$  as a result of the increased amount of supplement eaten as FA increase (Table 6.6). There was no significant effect of GE or FA on  $DMI_T$  ( $8.4 \text{ kg DM cow}^{-1}$ ). The lowest DMD was measured in the NZ90 genotype; however, neither the effect of GE nor FA was significant ( $733 \text{ g kg}^{-1} \text{ DM}$ ).

### 6.3.6. Live weight, body condition score, daily yield and milk composition

During the lactation period the NZ70 cows were the lightest ( $440 \text{ kg cow}^{-1}$ ) and the OS90 the heaviest ( $484 \text{ kg cow}^{-1}$ ), with the LW of the NZ90 genotype intermediate ( $464 \text{ kg cow}^{-1}$ ; Table 6.8). Differences between strains were smaller than expected from their expected mature LW (McNaughton *et al.*, 2002), probably as a result of the young age of the cows in the systems (two to four years old) and loss of LW during lactation (see Chapter 5). Differences in BCS between strains were also observed (Table 6.8). The mean LW of the cows showed a trend to increase with FA, and a significant effect of FA on BCS was measured, except in the OS90 (Figures 6.4a and 6.4b).

Figure 6.4: Mean live weight (a) and body condition score (b) of three Holstein-Friesian genotypes managed in different feeding systems during lactation.

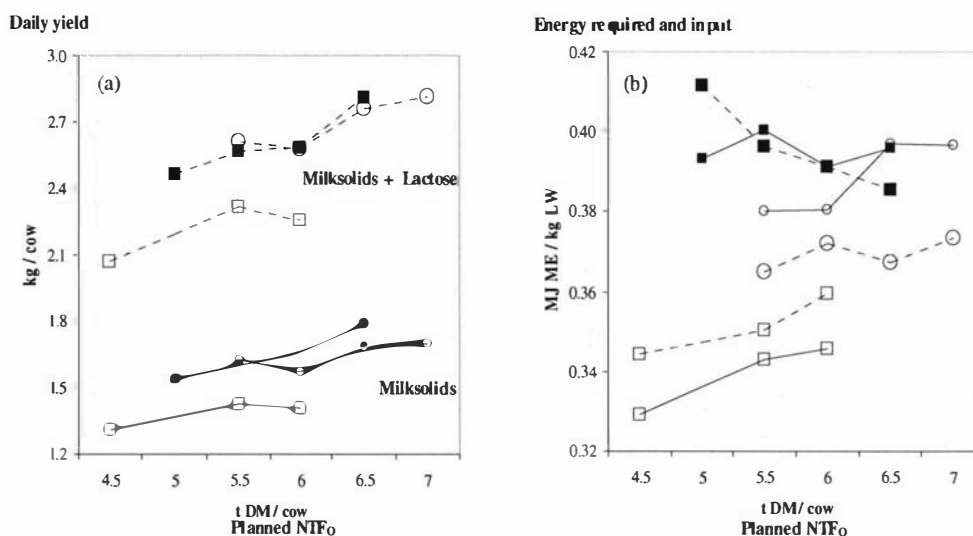


NZ70 (□); NZ90 (■); OS90 (○).

Milk yield was significantly higher for the OS90 ( $21.8 \text{ litres cow}^{-1}$ ) than for NZ90 ( $19.9 \text{ litres cow}^{-1}$ ) and greater for these two genotypes than for NZ70 ( $17.3 \text{ litres cow}^{-1}$ ). A significant effect of FA on milk yield showed an increase of  $1.4 \text{ litres cow}^{-1}\text{day}^{-1} \text{ t}^{-1}\text{DM}$  of  $NTF_0$  (Table 6.8). Even though milk yield was higher for the OS90 genotype, daily MS production was similar between the OS90 and NZ90 strains. There was a significant

effect of FA on MS yield with an increase of 0.10 kg MS cow<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>0</sub> (Table 6.8; Figure 6.5a). In systems fed only pasture, both milk and MS yields were highly correlated with DMI<sub>H</sub>/LW (Table 6.8), although with a lower correlation coefficient (r) for NZ90 and OS90 indicating a greater partition of body fat reserves to yield during lactation.

**Figure 6.5: Mean daily milksolids and milksolids plus lactose production per cow (a), mean daily energy required for maintenance and yield (b) and ingested (c) per unit of live weight, for three Holstein-Friesian genotypes managed in different feeding systems during lactation.**



NZ70 (□); NZ90 (■); OS90 (○). In (a) yield of MS are shown in solid lines and small symbols whereas MS + lactose are shown on top of the same figure in light dotted lines and big symbols. In (b) energy output is shown in solid lines and in (c) energy ingested is shown in light dotted lines, the difference between values from (b) and (c) indicates the energy balance. Both energy input and output were estimated as indicated in the text (AFRC, 1993; Reynolds & Beever, 1995; Agnew & Yan, 2000).

The greater MS yield of the NZ90 and OS90 was the result of a greater fat and protein yield and these genotypes produced more lactose (Table 6.8). Although not significant, fat yield tended to be higher and protein lower for NZ90 than OS90. Lactose yield was significantly higher for OS90 than NZ90. A significant effect of FA on fat, protein and lactose yields was observed. The NZ90 genotype showed similar fat content in milk to that of the NZ70 but higher than OS90 and also higher protein content in milk than both NZ70 and OS90. Similarly, the NZ90 genotype showed higher lactose content in milk although not significantly different from the other two strains. The effect of FA on milk components content was not significant; nevertheless, trends for significant GE by FA interactions were observed, particularly for protein content in milk. It is apparent that while the response of the OS90 genotype to increased feed allowance was to increase milk yield and then lactose and MS yield, the NZ90 strain showed a lower increase in milk yield but greater increase in the content of milk components. As a result, MS yield

was greater for NZ90 than OS90 at high FA, but this difference disappeared when lactose yield was also included with MS (Figure 6.5a).

### 6.3.7. Energy requirements

The highest mean daily energy required for maintenance and milk yield was measured for the OS90 strain (188 MJ ME day<sup>-1</sup>), significantly higher than observed for the NZ90 genotype (183 MJ ME day<sup>-1</sup>) and lowest for the NZ70 strain (148 MJ ME day<sup>-1</sup>). The mean daily requirements of the cows increased as FA increased (8.1 MJ ME day<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>0</sub>) as a result of the improved mean yield and LW of the cows in systems farmed at higher FA (Table 6.8). The higher energy required by the OS90 compared to the NZ90 strain (5.1 MJ ME day<sup>-1</sup>) was mainly sustained by the higher energy required for maintenance and daily milk yield (2.1 and 2.6 MJ ME day<sup>-1</sup> respectively), plus a small difference in the activity allowance. As a result of the higher daily energy required for a similar amount of daily MS and the lower daily energy consumption by the OS90 cows (4.2 MJ ME day<sup>-1</sup>) compared to the NZ90, the mean daily energy balance during lactation was positive for the NZ90 but negative in the OS90 genotype (-8.8 MJ ME day<sup>-1</sup>). These differences were consistent with the corresponding difference in BCS changes during lactation (see Figures 5.6 and 5.7 in Chapter 5).

**Table 6.9: Pearson correlation coefficient (*r*) between daily milk and milksolids yields and herbage dry matter intake during lactation.**

Dependent	GE	Mean	CV%	n	Independent	GE	r	P	Mean	CV%	n
MS yield	NZ70	1.42	19.6	14	DMI <sub>11</sub> /LW	NZ70	0.64	*	29.9	8.7	14
	NZ90	1.70	21.5	17		NZ90	0.50	*	34.3	7.0	17
	OS90	1.69	22.4	16		OS90	0.23	NS	30.7	9.4	16
Milk yield	NZ70	18.2	22.9	14	DMI <sub>11</sub> /LW	NZ70	0.64	*	29.9	8.7	14
	NZ90	21.1	24.2	17		NZ90	0.54	*	34.3	7.0	17
	OS90	22.5	25.2	16		OS90	0.22	NS	30.7	9.4	16

Pearson's correlations were estimated only during the lactation period, for dates when supplement was not fed. DMI<sub>11</sub>: herbage dry matter intake; LW: live weight. GE: genotype; P: probability [significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.]; r: coefficient of correlation; CV%: coefficient of variation; n: sample number.

## 6.4. DISCUSSION

The use of the whole systems as treatments provided a very effective basis for evaluating practical aspects of the management required by the different genotypes utilized. However, there were a number of difficulties in dealing with (a) the differences in mean feeding levels between genotypes, particularly due to the fact that differences in live weight between strains were evident; (b) at the lowest FA at which both NZ strains were managed stocking rate was the highest, higher for NZ70 than NZ90; (c) differences in pasture management occurred due to the increased amount of supplement fed in systems

managed at increased FA and increased substitution effect. As result, there were a substantial number of interactions between pasture characteristics and their effect on  $DMI_H$ , and consequently on body reserves mobilization and daily yield; in addition to this, previous management affected the status of the cow and probably  $DMI_H$  on the dates intake was measured with *n*-alkanes. Although these complications made it difficult to be conclusive about the effects observed and the differences between genotypes and results must be analysed with care to avoid over interpretation, the information presented in this Chapter highlighted those factors affecting the  $DMI_H$  of the strains that should be further investigated.

#### **6.4.1. Relation between intakes estimated by *n*-alkanes or the visual scoring system**

The  $DMI_H$  from the visual scores (See Chapter 5) were estimated with a minimum difference of 0.25 score, equivalent to 75 kg DM ha<sup>-1</sup>. Assuming that the score given on both pre- and post-grazing was correct, this score represents a mean intake difference of about  $\pm 0.5$ –1.5 kg DM cow<sup>-1</sup> for a herd of 15 or 20 cows, grazing 0.1 and 0.4 ha day<sup>-1</sup> respectively (during winter or spring). This agrees with the increased dispersion of  $DMI_H$  obtained with the visual scores as the  $DMI_H$  measured with *n*-alkanes increased (Figure 6.6), considering that  $DMI_H$  and the daily area allocated to each herd was lower during the dry than the lactation period.

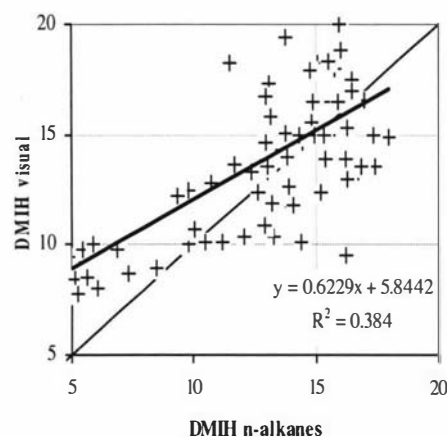
Similar bias could be expected for the visual determination of pre- and post-grazing herbage masses, with a proportionally larger influence on post- than pre-grazing observations. Similar biases would have no effect on the estimation of intake if they have similar direction, but the effect would increase when these biases for pre- and post-grazing have opposite direction, or similar direction but different magnitude. For these reasons, the bias for the  $DMI_H$  introduced from the visual score system was expected to be higher than with the *n*-alkanes method, despite the fact that the latter is affected by the accuracy of the pasture sample collected (See Chapter 3 and 4) and the complexity of the procedure.

It was demonstrated that when the cow graze selectively, the faecal ratio  $C_{31}/C_{33}$  from individual animals is different from the ratio in the pasture, therefore the diet of the cows is not well represented by the pasture sample (Chapter 3). Despite this being the main limitation of *n*-alkanes, the method is accurate enough to detect relative differences in  $DMI_H$  within cows in a group grazing together on the same pasture. Despite the variability observed in the faecal ratio  $C_{31}/C_{33}$  between the cows grazing the same pasture (Chapter 3; Figure 3.2a and 3.2b) the mean faecal ratio  $C_{31}/C_{33}$  for the herd would compensate the differences observed between animals, and be close to the pasture ratio if

the cows graze uniformly the daily area allocated. This would occur just because the reference sample collected would represent better the mean diet of the cows in the herd than the diet of each individual. Moreover, the variability between cows in a herd would diminish when the capacity of the cows to select is reduced and they leave a low and uniform post-grazing residual (See Chapter 2), and when large number of cows are used.

The linear relationship between the means  $DMI_H$  of the ‘systems’ estimated from visual scores with those obtained from the n-alkanes was estimated ( $y = 5.84 + 0.62x$ ;  $r^2 = 0.38$ ;  $n=77$ ; where ‘y’ is the estimate from the visual score and ‘x’ the estimate from the n-alkanes; Figure 6.6).

**Figure 6.6: Linear relationship between the mean herbage dry matter intake estimated from the visual score system and the n-alkane method.**



Each point represents the mean herbage dry matter intake of each system in the experiment (15 or 20 cows), for all the dates n-alkanes were measured.

The values of  $DMI_H$  from the visual score were overestimated during the dry period, when all herds grazed intensively on a small area. It is possible that in these conditions the increased trampling caused an underestimation of the post-grazing residual herbage mass, thereby  $DMI_H$  was overestimated. It is also probable that the higher and more variable post-grazing residuals observed during mid lactation also increased the variability of the visual estimates recorded. Despite the bias observed during the winter period with the visual score system, and even though the relationship is not particularly strong, there is no evidence of consistent bias between  $DMI_H$  estimated by the different procedures.

The mean dry matter intakes from n-alkanes for the lactation and dry periods were used to estimate the total dry matter intake ( $DMI_T$ ) for the whole season, accounting for the length of each period. The mean value for all three strains was  $4.7 \text{ t DM cow}^{-1} \text{ year}^{-1}$ ,

about 6.7 % lower than the mean intake estimated with the visual score system (mean of two seasons, see Chapter 5). Differences between the n–alkanes and visuals were larger for the NZ70 and OS90 genotypes than for the NZ90, and higher with the visual scoring than with n–alkanes, by 9 and 11% respectively.

The differences in  $DMI_T$  estimated by n–alkanes or the score system probably occurred because the values obtained with the n–alkanes for particular dates were assumed to represent the changes that occurred over a long period, whereas in fact diet changed more frequently. Nevertheless, the general agreement between the two methods supports the use of the weekly-calibrated visual scores to estimate the mean amount of herbage removed at each grazing and the dry matter consumed by the herd during the complete season.

#### 6.4.2. Pasture and supplement intakes and diet digestibility

During lactation, cows of the NZ90 and OS90 genotypes had higher  $DMI_H$  and  $DMI_T$  than the lower yielding NZ70 Friesian strain (Table 6.3). The higher intake of cows with high genetic potential for yield has been reported on diets based either on total mixed ration (Kolver *et al.*, 2002) or pasture (Grainger *et al.*, 1985; Buckley *et al.*, 2000; Kolver *et al.*, 2002). Cows selected to produce more yield have higher energy requirements and DMI as indicated by the high genetic correlation between yield and intake (Van Arendonk *et al.*, 1991; Veerkamp, 1998). These cows also mobilise more body reserves post-calving. The combined response of high feed intake and high mobilisation of body reserves for NZ90 and OS90, sustains a higher yield than for the NZ70 strain. However the amount of BCS lost in early lactation (see Chapter 5) and the mean BCS for the whole lactation indicate a more negative energy balance for these strains despite their higher intake (Veerkamp, 1998). The energy balance was probably also more negative for OS90 than for NZ90, as indicated by the different BCS lost between these two genotypes (see Chapter 5), which is in agreement with the mean difference in the energy balance estimated during lactation (Table 6.8). This difference, particularly for the early lactation period, indicates a greater deficit between the increase in milk yield and the increase in nutrient intake immediately after calving for the OS90 (see Chapter 5), because intake was also constrained in the OS90.

The daily  $DMI_H$  was 0.51 kg DM higher for NZ90 than OS90 but the difference in  $DMI_T$  was reduced to 0.27 kg DM due to the greater amount of supplement fed to OS90. Similar increases in  $DMI_T$  were observed between GE as FA increased (0.32 kg DM cow<sup>-1</sup> t<sup>-1</sup>DM of NTF<sub>0</sub>; Table 6.6). Thus, even though intake improved for OS90 by feeding more supplements,  $DMI_T$  was still lower than for NZ90 cows.

It is not known if the higher mobilisation of body reserves for OS90 than NZ90 resulted from intake being further constrained on pasture, or if the reduced intake on pasture is to some extent affected by the high rate of body reserves mobilisation, resulting in a reduced motivation to graze due to metabolic reasons. If a restriction to milk synthesis occurs due to the contrast between diet characteristics and nutrients intake and the potential requirements of a high yielding cow, cows mobilising body reserves at a higher rate would increase the level of metabolites in the circulating energy pool. A link between the increase in the concentration of metabolites associated with body reserves mobilisation (NEFA) and the decrease in intake observed post-calving is recognised (Ingvartsen et al., 1999). Although a genetic control of fat reserves was proposed (Veerkamp *et al.*, 2003), the total amount of the reserves lost appeared to be determined by the availability of feed, the intake capacity of the animal and the energetic costs required to harvest, process and digest the feed. Both NZ genotypes, but particularly the NZ70, used lower amounts of body reserves than the OS90. It was suggested that the amount of body reserves mobilised are controlled genetically and linked to the genetic ability to produce milk, rather than resulting from feed intake and yield not increasing at similar rates (Veerkamp et al., 2003). This results in a dynamic association between yield and LW (Veerkamp, 1998) that may affect the amount of energy required for maintenance as it is apparent that lean animals have higher maintenance requirements per kilogram of LW<sup>0.75</sup> (Agnew & Yan, 2000).

Intake per unit of LW declined from early to late lactation in association with a decrease in daily yield and LW of the cows, affected by mobilisation of body fat. A trend for a higher DMI<sub>H</sub>/LW in NZ90 than OS90 was observed in early lactation but this difference was smaller in late lactation (see Appendix V-9). This indicates that the energy status of the OS90 cows relative to maintenance improved in late lactation, however a substantial improvement in BCS was not observed as it is apparent that these cows continue to partition most of the energy ingested to sustain yield (see Chapter 5).

Considering the whole lactation, the mean DMI<sub>H</sub>/LW was 2.6 g DM kg<sup>-1</sup> LW higher for NZ90 than OS90 (31.5 vs. 28.9 g DM kg<sup>-1</sup> LW; Table 6.6 and Figure 6.3b), in agreement with recent results obtained in Ireland for genotypes with similar genetic potential (Horan et al., 2006), whereas the mean DMI<sub>H</sub>/LW achieved by the OS90 cows was similar to the NZ70 despite differences in milk output and maintenance requirements. This result was unexpected considering the differences in the energetic requirements between genotypes and it also differs from the DMI<sub>H</sub> of 30.0 vs. 32.0 g DM kg<sup>-1</sup> LW reported for animals of small and large body size (Stakelum & Connolly, 1987). However, it is consistent with the higher BCS of the NZ70 during the lactation period (see Chapter 5) and the lower

mean yield of the NZ70 (1.37 vs. 1.64 kg MS cow<sup>-1</sup> for NZ70 and OS90 respectively), which indicates this strain partitioned more energy to body reserves than to yield.

The decrease in the DMI<sub>H</sub>/LW as FA increased for cows of all three genotypes (-1.10 g DM kg<sup>-1</sup> LW t<sup>-1</sup> of NTF<sub>O</sub>, Table 6.6) reflected the decline in daily DMI<sub>H</sub> (-0.71 kg DM cow<sup>-1</sup> day<sup>-1</sup> t<sup>-1</sup> of NTF<sub>O</sub>; Table 6.6). Considering that the mean TF<sub>O</sub> increased at a rate of 0.74 t DM t<sup>-1</sup> of NTF<sub>O</sub> (see Table 5.4 in Chapter 5), the actual decline in DMI<sub>H</sub> was -0.96 kg DM cow<sup>-1</sup> day<sup>-1</sup> t<sup>-1</sup> of TF<sub>O</sub>. This was partially determined by the effect of the supplement fed (Figures 6.3a and 6.3b), because the mean response was calculated for the whole lactation and the supplement was fed in mid and late lactation, only to systems farmed at high FA. The extra feed offered to each system included the extra pasture growth according to a higher measured annual herbage accumulation rate (HAR) than planned (46.6 kg DM day<sup>-1</sup> planned vs. 52 – 54 kg DM day<sup>-1</sup> actual during 2002-03 and 2003-04; Table 5.2 in Chapter 5).

Even though DMI<sub>H</sub> decreased as FA increased, the mean DMI<sub>T</sub> of the genotypes increased 0.32 kg DM cow<sup>-1</sup> day<sup>-1</sup> t<sup>-1</sup> of NTF<sub>O</sub> during lactation (or 0.44 kg DM cow<sup>-1</sup> day<sup>-1</sup> t<sup>-1</sup> of TF<sub>O</sub>); and was affected by the changes in the LW of the cows (Figures 6.3a and 6.3b). Hence, although daily DMI<sub>T</sub> improved, DMI<sub>T</sub>/LW declined for the NZ90 cows as result of the improved LW relative to the OS90 cows. This suggests a probable increased influence of the energetic maintenance requirements for cows that improved condition when managed at high feeding level; nevertheless DMI<sub>T</sub>/LW was still higher for NZ90 than OS90 at the highest feeding level. The increment in LW for NZ90 cows indicates that a proportion of the energy consumed was used to improve body reserves. The same trend was observed for the NZ70 strain despite the relative lower intake per unit of LW achieved, probably as a result of the lower energy requirement for maintenance and the lower partition of the energy consumed to milk yield than in the NZ90 strain. In addition to the differences in intake favourable to the NZ90 genotype, these cows showed a trend for a greater DMD during lactation indicating that the intake of nutrients would have increased further in this strain than for both the NZ70 and OS90.

During the dry period, DMI<sub>H</sub> and DMI<sub>T</sub> were similar for NZ90 and OS90 strains and numerically higher than NZ70 (+0.4), but DMI<sub>T</sub> by the OS90 was still lower than for the two NZ strains. Although this was not significant, it indicates a slightly lower energy status for OS90 cows relative to maintenance and suggests that feed allocation should be increased further for this genotype in the dry period. In contrast to what was observed during lactation, the NZ90 showed the lowest DMD and the OS90 the highest, although the difference was not significant. Nevertheless, due to differences in the DMI<sub>T</sub>/LW between strains, the intake of nutrients per unit of LW still showed the trend to be higher for both NZ genotypes than for OS90.

### 6.4.3. Energy requirements

The daily energy required for maintenance was higher for NZ90 and OS90 than for the NZ70 genotype (+16.6 MJ ME day<sup>-1</sup>; Table 6.8). The latter was lighter than NZ90 and OS90 but also it was assumed that the NZ70 cows have lower fasting metabolism per unit of LW<sup>0.75</sup> (NE<sub>m</sub>), due to the lower yield genetic potential (Agnew & Yan, 2000). In contrast, only a small difference in the energy required for maintenance was observed between NZ90 and OS90 (+2.1 MJ ME day<sup>-1</sup>), which resulted from lower mean LW of NZ90 cows (AFRC, 1993).

The daily energy requirement for production was higher for the NZ90 and OS90 than NZ70 (+18.1 MJ ME day<sup>-1</sup>)(AFRC, 1993; Reynolds & Beever, 1995), and numerically higher for OS90 than NZ90 (+2.6 MJ ME day<sup>-1</sup>) even though yields of MS were similar (Table 6.8). The difference in energy output between NZ90 and OS90 was due to the fact that OS90 produced more milk and lactose than NZ90 but similar MS. Additionally, as feeding level increased MS yield increased for both genotypes, but yields of milk and lactose increased to a greater extent for OS90 than NZ90 (Table 6.8 and Figure 6.5a). Allowing for differences in grazing activity between the genotypes, which also scale with LW, the mean daily total energy requirements (for yield, maintenance and grazing activity) were 5.1 MJ ME day<sup>-1</sup> higher for OS90 than NZ90.

Considering the age of the cows and the mature LW suggested by McNaughton et al. (2002), it is apparent that the cows used a proportion of the energy input for growth. If the energy required for maintenance would increase in growing and lean grazing cows (Agnew & Yan, 2000), fat cows that achieved a higher proportion of their mature LW at an early age would have lower maintenance requirements per unit of LW<sup>0.75</sup>. In addition, the NZ90 genotype showed a trend for a higher DMD (Table 6.6; see also Appendix V-6) particularly in early lactation, when energy output was the highest, but had a lower DMD during the dry period.

Differences in the quality of the feed consumed at different stages of lactation were observed in all three genotypes, but to a larger extent for the NZ90 strain. These differences may have occurred due to the different quality of the pasture offered and promoted by management, however it is hypothesised that the NZ90 have a greater capacity to adapt the energy content of the diet to its metabolic energy demand. This would be based on a greater capacity to select plant-parts of higher energy value than the mean available in the pasture. If the animals ingest herbage of higher ME content, the proportion of metabolisable energy relative to the gross energy of the feed ( $q_m$ ) increases and the efficiency of utilisation of energy consumed for maintenance and lactation ( $k_m$  and  $k_l$ ) may improve (AFRC, 1993; Reynolds & Beever, 1995).

The daily energy required for maintenance and production and the energy consumed expressed per unit of LW indicate a positive energy balance for NZ70 cows and negative balance for the OS90 genotype regardless of the greater FA (Figure 6.5b). The OS90 had a more negative energy balance at high FA because energy output increased more than  $DMI_T/LW$  and thus these cows had to mobilise more body fat. For NZ90 cows, energy intake per unit of LW was higher than energy output at the lowest FA, but was lower at the highest FA, and at intermediate FA the energy status of the cows was in balance. This indicates a higher capacity of the NZ90 cows to sustain energy requirements from pasture than for OS90 cows.

#### 6.4.4. Effects of sward conditions on herbage intake

In the present study DHA increased by  $7.8 \text{ kg DM cow}^{-1} \text{ day}^{-1} \text{ t}^{-1} \text{ DM}$  of  $NTF_O$  (Table 6.3), within a range of  $24\text{-}60 \text{ kg DM cow}^{-1} \text{ day}^{-1}$  (Figure 6.1a) by combining changes in  $HM_{PRE}$  and the area allocated per cow. The range of DHA at which the genotypes were managed determined a similar range in grazing conditions for systems managed with the different strains (Figures 6.1a, 6.1b and 6.1c), which showed a similar range in  $SH_{PRE}$  and  $B_D$ .

Despite the increased DHA (Figure 6.2a), the mean  $DMI_H$  of all three genotypes declined as FA increased, and in contrast  $DMI_T$  increased (Figure 6.3a). The decline in herbage intake of supplemented cows is logical and the decline measured in  $DMI_H$  could be partly attributed to substitution. However, the mean  $DMI_T$  was lower than expected from the cows' genetic potential and the main reason was the low  $DMI_H$  of the cows. Considering that  $DMI_H$  should increase with DHA (Combellas & Hodgson, 1979; Peyraud *et al.*, 1996; Wales *et al.*, 1999; Maher *et al.*, 2003) the decline observed was unexpected. It also occurred during the early lactation period when the supplement was not fed and the requirements of the cows were the highest, hence it is possible that the response of the cows to the increased DHA was confounded by the negative effect of the structure of the sward on  $DMI_H$ .

Daily herbage allowance was on average 2.7 times higher than  $DMI_H$ , thus herbage consumption should not be constrained at this level (Greenhalgh *et al.*, 1966; Combellas & Hodgson, 1979). The suggestion that cows would maximise intake when DHA is twice the potential  $DMI_H$  made by Combellas and Hodgson (1979) would be still valid for today's dairy cows, despite their increased genetic yield potential and requirements, if the sward grazed does not represent any constraint to herbage prehension (see Chapter 8). It is known that the response in  $DMI_H$  of the cows to increments of DHA change with the characteristics and quality of the herbage offered (Hodgson, 1990). For instance, Horan *et al.* (2006) reported a mean response for cows of high genetic potential of a  $0.53 \text{ kg}$

increase in DM intake for an increase of 4.3 kg in DHA ( $\text{kg cow}^{-1}\text{day}^{-1}$ ; above 50 mm), whereas  $\text{DMI}_H$  per cow increased 2.6, 5.1 and 7.1 kg DM  $\text{day}^{-1}$  for NZ70, NZ90 and OS90 respectively, when grazed at similar DHA ( $35 \text{ kg cow}^{-1} \text{ day}^{-1}$ , above ground level) but improved sward condition (see Chapter 8).

At the mean  $\text{SH}_{\text{POST}}$  measured in the present study (70 mm), it is apparent that the cows removed most of the available herbage and the residual herbage mass was that proportion of the herbage offered that was more difficult to graze (about  $1,983 \text{ kg DM ha}^{-1}$  or 76% of  $\text{HM}_{\text{PRE}}$  measured above ground level; Table 6.3). This suggests that a small proportion of the total DHA was available for the cows; in contrast, most of the DHA would have been available in Horan's study (measured above 50 mm height).

In the present study, systems farmed at high FA had pastures that were managed in a shorter rotation length (Table 6.2); had lower  $\text{HM}_{\text{PRE}}$ ; were shorter; and a larger area was allocated daily per cow (Table 6.3; Figures 6.2c and 6.2d). Although the cows would have had more herbage available due to the increased area, they may have removed a lower proportion of the herbage offered due to lower herbage availability (Trudell & White, 1981) although DHA had increased (Hodgson, 1990). As expected the linear relationship between  $G_{\text{EF}}$  and DHA indicates a decline in grazing efficiency as DHA increases; however no relationship was measured between  $\text{DMI}_H/\text{LW}$  and DHA suggesting that even though DHA increased the herbage available did not increase similarly. It could be possible that although the herbage in the strata available would have a high proportion of new leaves due to the shorter interval between grazings,  $B_D$  would be low and then the bites prehended lighter (Allden & Whittaker, 1970; Stobbs, 1973). In addition, it would be expected that the cows would have to graze for longer and spend more energy to achieve a similar  $\text{DMI}_H$ . If the sward grazed is short, it is expected that cows with lower ability to perform small bites would be unable to continue grazing once the herbage available was harvested.

The higher mean  $\text{DMI}_H$  of the NZ90 indicates the greater grazing capacity of this genotype compared to that of the OS90. In addition, considering the lower energy requirements of the NZ70 than OS90, the higher  $\text{DMI}_H/\text{LW}$  of the NZ70 also indicates the greater capacity of this genotype to cover its energy requirements from the grazing pasture. This agrees with the positive correlation between  $\text{DMI}_H/\text{LW}$  with  $\text{SH}_{\text{PRE}}$  and  $H_R$  in both NZ genotypes, particularly in the NZ90 strain (Table 6.7), but this correlation was not observed in OS90 cows although the pastures grazed by this strain were numerically taller than those grazed by the two NZ genotypes (146 vs. 139 mm respectively; Table 6.3).

The fact that  $SH_{POST}$  was numerically higher for the OS90 than in the two NZ genotypes (although not significantly different) could be just the result of the higher  $SH_{PRE}$ , however, it also suggests that this strain avoided grazing very close to ground level. If the  $B_D$  of the horizons harvested was different and the proportion of the initial herbage removed was similar,  $DMI_H$  must be different. The mean  $B_D$  of the residual mass (from  $HM_{POST}$  and  $SH_{POST}$ ; Table 6.3) was higher in the pastures grazed by the two NZ genotypes than by the OS90 genotype. This estimate can be greatly affected by animal treading, particularly if it is considered that  $HM_{POST}$  is an indirect estimate from the compressed height obtained from the RPM; nevertheless, there is no reason to suspect that the bias involved would affect differentially the mean value of  $B_D$  of the residual between genotypes. It is possible that the OS90 strain was more constrained than the two NZ strains by the  $B_D$  of the herbage layer close to ground level and then stopped grazing before the two NZ genotypes because the structure of the pasture might have represented a limit to herbage prehension (Peyraud et al., 1996).

The linear decline in  $B_D$  as  $SH_{PRE}$  increased is logical (Table 6.5), as the proportion of new leaves in the  $HM_{PRE}$  might have increased. This suggests that the cows grazing down the sward would face an increased  $B_D$  and then strength in the herbage, probably with a further increase when the stubble is reached (pseudostem layer). This agrees with the fact that  $HM_{POST}$  and  $SH_{POST}$  declined in systems managed at lower FA, this would have been probably associated with a reduced height of the stubble, as suggested by the improved quality and the decline in  $B_D$  of the herbage offered (Table 6.3). Although the amount of herbage removed ( $H_R$  times  $B_D$ ) would be determined by the  $SH_{PRE}$  due to the linear effect on  $H_R$ , it would also increase for cows with greater ability to graze within the lower strata of the pasture.

The greater capacity of the two NZ strains than OS90 to graze within a stratum of higher  $B_D$  would be determined by the ability to: (1) graze selectively; or (2) to reduce the size of the bite and be able to remove the herbage present in the lower strata of the pasture with increased resistance to severance (Ungar, 1996). The high correlation between  $DMI_H/LW$  and DMD during lactation for all three genotypes indicates the preference for high quality herbage, even though differences between the correlation values between strains suggests that NZ70 cows had higher ability to harvest high digestible herbage than the NZ90 and OS90 strains when available (Table 6.6). In addition, the correlation between  $DMI_H/LW$  and the NDF suggest that intake was more affected by the decline in quality of the herbage for both NZ strains than for OS90, probably because the capacity of the animal to select declines in swards of low quality (Stobbs, 1973). Considering that not only the quality of the pasture decreased as lactation progressed (see Figure 5.2a in Chapter 5) and also the yield of the cows in the three genotypes declined in late lactation

(Figure 5.8 in Chapter 5; see Appendix V-11 and V-12), the effect of herbage quality on intake was confounded by the lower requirements of the cows.

However, it is possible that the OS90 were less constrained by the quality of the herbage than by the increase of its shearing resistance close to ground level or a proportionally higher restriction imposed by any limit to bite depth (O'Reagain & Mentis, 1989). It is hypothesised that the characteristics of the strata close to the ground probably affected the harvesting capacity of the OS90 to a greater extent than for both NZ genotypes. It would seem logical that if the energy expended to harvest a bite and the energy ingested per bite are equal, there will be no benefit to the cow to continue grazing.

Although the general linear relationship between  $H_R$  and  $SH_{PRE}$  shows that all three genotypes increased  $SH_{POST}$  as  $SH_{PRE}$  increased, differences between strains suggest that the proportion of the initial  $SH_{PRE}$  removed for both NZ strains was higher than for the OS90 genotype in tall swards, within the range of pasture conditions at which the systems were managed (Table 6.5). At a  $SH_{PRE}$  of approximately 130 mm, all three strains depleted the sward to about the same  $SH_{POST}$ ; above this height the two NZ strains, and particularly the NZ90, reduced the sward to a lower residual than the OS90 strain; however, below this height the  $H_R$  was larger for the OS90.

The low proportion of the herbage that was removed by the OS90 in the tall sward is probably the result of a lower capacity to perform small bites by this strain; hence, the difficulty toprehend herbage may increase when these cows graze the lower horizons of the pasture where  $B_D$  is higher and herbage quality lower. It is not suggested that a dense layer of herbage at the base of the pasture would act as a final limit for the cows to continue grazing, which probably would result from the interaction between sward conditions and the current balance between energy demand and supply. However, this effect would inhibit grazing in OS90 cows to a greater extent than in the two NZ genotypes. In addition, cows that behave selectively would probably spend more time grazing under these sward conditions but compensate by increasing the quality of the herbage harvested, reducing the effort to process and digest the herbage ingested (see Chapter 7).

## 6.5. CONCLUSIONS

Considering the limitations and confounded effects of the different factors analysed in the present Chapter that may affected the  $DMI_H$  of the genotypes, it is difficult to be conclusive about the cause – effect relationships influencing the differences observed between strains. Despite this difficulty, it was evident from the data analysed that the OS90 and NZ90 genotypes consumed more feed than the NZ70 strain, but the OS90 had

lower  $DMI_H$  than NZ90. In addition, the OS90 had higher daily total energy requirements to produce similar amount of MS than NZ90, which resulted in negative mean daily energy balance during lactation.

The decline in  $DMI_H$  as FA increased observed in all three genotypes with increasing FA was unexpected considering that the systems were managed at increased DHA. However,  $DMI_T$  increased in all three genotypes because of the supplement fed. Although  $DMI_T/LW$  increased with FA for NZ70 and OS90, a decline was measured for the NZ90 genotype. This genotype improved LW more than the other two genotypes as feeding level increased and it also showed a trend for a greater increase in MS yield. The reduction observed in  $DMI_T/LW$  for NZ90 suggests a reduction in the energy status of the cow due to increased maintenance costs, however energy intake per unit of LW was still higher than in the OS90 genotype, and the NZ90 cows had more body fat reserves.

There was a confounded effect of DHA and sward conditions on  $DMI_H$  in the present study. It is apparent that the condition of the sward constrained  $DMI_H$  for all the strains even though DHA increased in systems managed at higher FA. The negative effect of sward structure constrained intake for the OS90 strain to a greater extent than in the two NZ genotypes. It was suggested that the NZ90 genotype has a greater capacity to harvest herbage from the strata close to ground level despite the associated increase in  $B_D$ ; as a result, these cows increased the amount of herbage removed from taller swards. It is also suggested that this improved harvesting capacity is determined by the ability to graze selectively and reduce the size of the bite in the face of constraining sward conditions. As a result, this flexible behaviour would allow them to sustain intake under changing sward conditions.

The following chapter (Chapters 7) concentrates on genotype contrasts in ingestive behaviour and the influence of contrasting management on herbage intake and animal performance.

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## CHAPTER 7

# **PRODUCTIVITY OF PASTURE – BASED SYSTEMS: HERBAGE INTAKE AND GRAZING BEHAVIOUR OF THREE HOLSTEIN – FRIESIAN GENOTYPES MANAGED AT LOW OR HIGH FEED ALLOWANCE IN EARLY AND LATE LACTATION**

### **7.1. INTRODUCTION**

It is known that cows of high genetic merit do not achieve their yield potential on pasture (Kolver & Muller, 1998), and high yielding cows of New Zealand origin perform relatively better than overseas animals on pasture (Kolver et al., 2002). The results from a range of spring calving pasture-based systems farmed with three different strains of Holstein-Friesian dairy cows managed at different feed allowance per cow (see Chapter 5), indicate that the modern high producing NZ cows yielded more MS, utilised less body fat reserves during lactation, consumed more pasture and showed an improved energy status during lactation (see Chapter 6), than the modern high yielding OS Holstein-Friesian dairy cow.

It was apparent that sward structure constrained herbage intake in the OS to a greater extent than in NZ high yielding cows, with NZ cows removing a higher proportion of the initial height of the pasture (see Chapter 6). It is hypothesised that the ability of the cows to perform on pasture is determined by their capacity to continue harvesting bites of a relative high digestible mass, despite facing constraining conditions in the sward.

Under strip grazing management the sward conditions change rapidly during the grazing session as the pasture is grazed down. Changes in sward structure occur at a different rate depending on the initial condition of the sward and the rate at which the herbage is ingested. The objective of the present study was to investigate differences in the grazing behaviour of cows from three strains of Holstein-Friesian dairy cows managed in pasture-based systems at contrasting feeding conditions during early and late lactation.

## 7.2. MATERIALS AND METHODS

Observations were made on all the cows in a sub-set of treatments in the system trial offering the lowest and highest levels of allowance during three-week periods in spring and autumn in season two (2003-03)

### 7.2.1. System management

The details of systems and management, which are the base of the present study, were presented in Chapter 5. Briefly, the genotypes were high breeding worth (BW) Holstein-Friesians of OS (OS90) or NZ origin (NZ90) and a low BW NZ Friesian genotype representing the cow used in NZ during the 1970s (NZ70; see Appendix IV for details). In this study, eleven non-replicated systems representing real farm conditions with 15 or 20 cows each were managed in different farmlets, with different feeding levels (n=205 cows, see details in Chapter 5). Systems were based on pasture and also fed forage (pasture silage) and supplement (maize silage and grain), particularly during mid and late lactation and the dry periods. They were farmed in a range of self-contained feeding units or systems, with a range of feeding levels ranging from 4.5 to 7.0 t DM per cow per year achieved through a combination of stocking rate (SR) and inputs of supplements, pasture silage and maize grain or silage (see Table 5.1 in Chapter 5).

**Table 7.1: Stocking rate and nominal (planned) values of feed allowance in systems managed at low or high annual feed allowance.**

CFA		Feeding Systems					
		NZ70		NZ90		OS90	
		1	4	1	4	1	4
Feeding allowance		Low	High	Low	High	Low	High
Stocking rate	Cow ha <sup>-1</sup>	3.8	3.1	3.4	3.1	3.1	3.1
Pasture allowance	T cow <sup>-1</sup> year <sup>-1</sup>	4.5	5.5	5.0	5.5	5.5	5.5
Supplement allowance	T cow <sup>-1</sup> year <sup>-1</sup>		0.5		1.0		1.5
Total feed allowance <sup>(1)</sup>	T cow <sup>-1</sup> year <sup>-1</sup>	4.5	6.0	5.0	6.5	5.5	7.0
Average feed allowance	T cow <sup>-1</sup> year <sup>-1</sup>		5.25		5.75		6.25

CFA: comparative feed allowance [increased from 1 (lowest) to 4 (highest), see Chapter 5 for details]

The experimental site was located at Dexcel No. 2 dairy farm, Hamilton, New Zealand. The total feed allowance (NTF<sub>0</sub> in Chapter 5) at which each system was managed was determined before the start of the experiment in season 2001 – 2002 by considering the expected herbage annual accumulation rate (HAR) for the farm where the experiment was located (17 t ha<sup>-1</sup>), the different mature live weight and yield potential between genotypes. Hence, the mean NTF<sub>0</sub> at which the genotypes were farmed were different in absolute terms but similar relative to the feed requirements of mature cows of each genotype, considering the expected differences in adult live weight (McNaughton *et al.*,

2002) and yield potential between genotypes according to their different BW and results from previous studies (Kolver et al., 2000; Kolver et al., 2002).

From the range of systems utilised in the main 'system' comparison, only systems managed at the lowest and highest FA ( $L_0 = FA1$  and  $H_1 = FA4$  respectively) of the range at which each genotype was farmed were compared during the early and late lactation periods (three weeks in spring and autumn respectively) in season 2002-03 (Table 7.1). All cows were two and three years old during the experimental period.

### 7.2.2. Pasture and animal measurements

Herbage mass (HM) and sward surface height (SH) were measured pre- and post-grazing ( $HM_{PRE} - HM_{POST}$  and  $SH_{PRE} - SH_{POST}$  respectively) with a Rising Plate Meter (RPM) and a sward stick respectively, as explained in Chapter 6. Equations used to transform the compressed height measurements obtained with the RPM from each paddock into DM values per hectare, were those estimated for the early and late lactation periods (Table 6.1 in Chapter 6). The bulk density ( $B_D$ ) of the pre-grazing pasture was estimated as the  $HM_{PRE}$  per centimetre of height ( $SH_{PRE}$ ). In addition, daily hand plucked pasture samples were collected from the pre-grazing stratum of the same paddocks, and processed as explained (see Chapters 5 and 6).

The individual herbage dry matter intake ( $DMI_H$ ), forage (e.g. pasture silage) and supplements (maize silage) fed ( $DMI_{MS}$ ), and the dry matter digestibility of the diet consumed (DMD) were estimated once in early and late lactation of season 2002-03 for each individual cow in the systems by using a combination of the n-alkanes (Mayes *et al.*, 1986; Dove & Mayes, 1991; Dove, 1992; Dillon, 1993; Dove & Mayes, 1996; Hameleers & Mayes, 1998b; Hameleers & Mayes, 1998a) and the  $^{13}C$  techniques (Jones *et al.*, 1979; Garcia *et al.*, 2000). Details of the procedures used to estimate intakes were explained with detail in Chapter 4.

Cows were milked twice a day during the whole lactation. The yield of milk produced by each cow and its composition was measured once a week as explained in Chapter 5. In addition, cows were weighed and condition scored weekly (Macdonald & Roche, 2004).

### 7.2.3. Grazing behaviour

The grazing behaviour of all the cows in each system was recorded (5 am to 11 pm) on two different days at the start and end of a 10-day period when  $DMI_H$  was measured (in early and late lactation). Each cow was identified with a large number painted on each side and a trained team of twelve assistants watched the cows during the observational

period. The observations stopped once the stock handler entered the paddock to yard the cows for morning and afternoon milking. Visual observations of grazing behaviour started once all the cows were back in the paddock after milking.

Each observer shifted at random between systems every three hours after resting for a similar period. Each observer visually scanned the activity of all the cows in the mob at intervals of 10 minutes and recorded their behaviour as grazing (G), eating supplement from a trough (E) or ruminating (R). A bite was identified as a head movement associated with the severance of a bunch of herbage (Hodgson, 1982). Bite rates ( $B_R$ ) were measured during one-hour periods, twice daily, after the cows returned from the morning and afternoon milkings and were actively grazing. The number of bites each cow performed during 30 seconds, and repeated four times during the following hour.

To calculate total G, E and R times ( $G_T$ ,  $E_T$  and  $R_T$ ) the behaviour observed at 10 minutes intervals was assumed to represent each cow's behaviour within this interval. Grazing was grouped in bout sessions or meals. A bout was considered a consecutive set of visual G longer than 20 minutes (two consecutive G observations). Any discontinuation of G activity was considered an indication of the termination of the bout, thus, once G was reinitiated it was considered the start a new G bout. Bout duration was expressed in minutes and estimated as total grazing time per day ( $G_T$ ) divided by the number of bouts per day.

The mean  $B_R$  was calculated as the mean of morning and evening observations, expressed as bites per minute. In addition, the total number of bites per day was calculated as the mean  $B_R$  times  $G_T$ . The mean  $B_R$  of the cows estimated probably overestimated the actual  $B_R$  over the whole day as it was only measured for one hour after each milking; during this period cows were actively grazing.

As grazing mostly occurs during daylight hours (Orr *et al.*, 2001)  $G_T$  was not expected to be affected greatly by the fact that the activity of the cows was observed for only 18 hours, with no night observations. However it has to be noted that the total number of bites per day calculated as  $G_T \times B_R$  could be biased. As a result of  $G_T$  being underestimated, and the  $I_R$  of the herbage consumed was probably overestimated; nevertheless, rankings between treatments were expected to be maintained.

#### 7.2.4. Statistical analysis

Systems were not replicated, instead large treatment groups representing real farm conditions were utilised (see Chapter 5). The individual  $DMI_H$  and  $DMI_{MS}$  intakes and DMD, grazing behaviour, LW and BCS and yield of the cows in the systems were

analysed using individual cows as experimental units. Pasture measurements pre- and post-grazing from each paddock were taken on consecutive days each week during the experimental periods (each three week period) and analysed using the mean of observations of three different paddocks each week as the experimental unit (week).

The statistical procedures of SAS (SAS, 2002) were used to analyse the animal data by using the MIXED model (PROX MIXED)  $Y_{ijkl} = \mu + GE_i + FA_j + P_k + (GE*FA)_{ij} + e_{ijkl}$  ; with the additive effects of genotype (GE), feed allowance (FA), parity (P; or age) and the genotype by feeding allowance interaction (GE\*FA) as fixed effects, and cow within system as random effect. Pasture data was analysed by using the MIXED model procedure (PROX MIXED)  $Y_{ijk} = \mu + GE_i + FA_j + (GE*FA)_{ij} + e_{ijk}$  ; with the additive effects of genotype (GE) and feed allowance (FA) and the genotype by feed allowance interaction (GE by FA) as fixed effects, and week within system as random effect.

### 7.3. RESULTS

#### 7.3.1. Pasture conditions in early and late lactation

In early lactation, there was no significant effect of GE on DHA (mean values of 36.9 kg DM or 86.5 g DM kg<sup>-1</sup> LW; Table 7.2), but a significant effect of FA on DHA was observed.

**Table 7.2: Pasture pre- and post-grazing pasture conditions of systems farmed with three Holstein-Friesian genotypes managed at low or high annual feed allowance in early and late lactation, with maize silage fed at high feed allowance in late lactation. Mean values for each system.**

	Genotype Feeding allowance	NZ70		NZ90		OS90		SED	Significance		
		Low	High	Low	High	Low	High		GE	FA	G*FA
Early Lactation	DHA (kg DM cow <sup>-1</sup> )	27.9	41.2	34.7	39.8	35.0	42.6	3.23	NS	***	NS
	DHA <sub>LW</sub> (g DM kg <sup>-1</sup> LW)	70	102	84	89	78	96	7.84	NS	**	0.076
	Area (m <sup>2</sup> day <sup>-1</sup> )	133	176	132	192	132	200	4.36	0.06	***	**
	HM <sub>PRE</sub> (kg DM ha <sup>-1</sup> )	2097	2341	2625	2066	2653	2132	200	NS	0.07	*
	SH <sub>PRE</sub> (mm)	123	135	154	124	161	129	12.9	NS	*	0.059
	HM <sub>POST</sub> (kg DM ha <sup>-1</sup> )	1598	1851	1857	1692	1823	1692	113	NS	NS	*
	SH <sub>POST</sub> (mm)	52.6	59.4	61.7	63.6	62.8	57.7	10.4	NS	NS	NS
Late Lactation	DHA (kg DM cow <sup>-1</sup> )	25.7	32.5	26.3	31.7	29.3	31.8	2.37	NS	**	NS
	DHA <sub>LW</sub> (g DM kg <sup>-1</sup> LW)	58	72	59	65	61	64	5.10	NS	*	NS
	Area (m <sup>2</sup> day <sup>-1</sup> )	109	133	106	132	122	133	8.89	NS	**	NS
	HM <sub>PRE</sub> (kg DM ha <sup>-1</sup> )	2368	2436	2469	2401	2426	2411	198	NS	NS	NS
	SH <sub>PRE</sub> (mm)	116	125	115	120	121	126	1.76	NS	NS	NS
	HM <sub>POST</sub> (kg DM ha <sup>-1</sup> )	1834	2068	1833	1978	1882	1970	84	NS	*	NS
	SH <sub>POST</sub> (mm)	58.4	69.6	52.8	65.2	60.3	69.7	0.64	NS	*	NS

DHA: daily herbage allowance; DHA/LW: daily herbage allowance per unit of LW; HM<sub>PRE</sub>: pre-grazing herbage mass; SH<sub>PRE</sub>: pre-grazing sward height; HM<sub>POST</sub>: post-grazing herbage mass; SH<sub>POST</sub>: post-grazing sward height. GE: genotype; FA: feed allowance; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.

This showed lower DHA in systems managed at the lowest than highest feeding allowances (32.5 vs. 41.2 kg DM or 77.3 vs. 95.7 g DM kg<sup>-1</sup> LW). Significant GE by FA interactions were measured for HM<sub>PRE</sub> and HM<sub>POST</sub> (Table 7.2), NZ90 and OS90 had higher HM<sub>PRE</sub> and HM<sub>POST</sub> at the lowest feeding allowance whereas NZ70 showed higher HM<sub>PRE</sub> and HM<sub>POST</sub> at the highest feed allowance. There was no effect of GE on SH<sub>PRE</sub> (137.6 mm) or SH<sub>POST</sub> (59.6 mm). In addition, although SH<sub>PRE</sub> was higher at the lowest than highest feed allowance (146.0 vs. 129.4 mm), SH<sub>POST</sub> was similar at both feed allowances (59.6 mm; Table 7.2).

**Table 7.3: Daily yield of milk and milksolids, live weight and body condition score of three genotypes of Holstein-Friesian managed at low or high annual feed allowance in early and late lactation, with maize silage fed at high feed allowance in late lactation. Mean values for each system.**

	Genotype	NZ70		NZ90		OS90		Significance			
		Low	High	Low	High	Low	High	SED	GE	FA	G*FA
Early Lactation	Milk yield (L cow <sup>-1</sup> )	19.2	20.3	24.6	24.8	24.5	27.0	1.69	***	0.05	NS
	MS (kg cow <sup>-1</sup> )	1.49	1.52	1.85	1.94	1.86	1.88	0.13	***	0.18	NS
	Fat (kg cow <sup>-1</sup> )	0.86	0.84	1.01	1.06	1.01	0.97	0.07	***	NS	NS
	Protein (kg cow <sup>-1</sup> )	0.63	0.68	0.84	0.88	0.85	0.91	0.05	***	*	NS
	Lactose (kg cow <sup>-1</sup> )	0.94	0.99	1.24	1.25	1.22	1.34	0.09	***	0.07	NS
	FC (g kg <sup>-1</sup> )	45.0	41.3	41.4	42.8	40.8	35.5	1.71	**	*	*
	PC (g kg <sup>-1</sup> )	33.1	33.7	34.0	35.4	34.6	35.0	0.83	***	NS	0.056
	LC (g kg <sup>-1</sup> )	49.1	48.7	50.1	50.5	49.8	49.7	0.60	***	NS	NS
	LW (kg cow <sup>-1</sup> )	402	402	410	445	451	445	15.8	***	NS	NS
	BCS	4.66	4.76	4.43	4.59	4.12	3.98	0.16	***	NS	*
Late Lactation	Milk yield (L cow <sup>-1</sup> )	10.1	11.3	12.8	14.9	11.9	13.7	1.84	**	*	NS
	MS (kg cow <sup>-1</sup> )	0.91	0.93	1.16	1.40	1.03	1.14	0.16	***	*	NS
	Fat (kg cow <sup>-1</sup> )	0.56	0.55	0.68	0.80	0.59	0.63	0.09	**	0.09	NS
	Protein (kg cow <sup>-1</sup> )	0.35	0.38	0.48	0.60	0.44	0.50	0.07	***	**	NS
	Lactose (kg cow <sup>-1</sup> )	0.49	0.53	0.61	0.72	0.54	0.65	0.09	**	*	NS
	FC (g kg <sup>-1</sup> )	55.7	48.9	52.2	54.0	50.2	46.0	3.14	0.05	0.09	NS
	PC (g kg <sup>-1</sup> )	35.8	34.2	37.8	39.9	36.8	36.7	1.48	***	NS	NS
	LC (g kg <sup>-1</sup> )	48.8	46.1	47.1	47.9	45.4	47.1	0.89	0.08	NS	**
	LW (kg/cow)	441	454	446	491	484	494	22.8	0.14	*	NS
	BCS	4.48	5.05	4.10	4.58	3.70	3.87	0.20	***	**	0.063

MS: milksolids; FC: fat content in milk; PC: protein content in milk; LC: lactose content in milk. GE: genotype; FA: feed allowance; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.

In late lactation, there was no significant effect of GE on DHA (mean values of 29.6 kg DM or 63.2 g DM kg<sup>-1</sup> LW); HM<sub>PRE</sub> (2,418 kg DM ha<sup>-1</sup>) and SH<sub>PRE</sub> (120.3 mm); and HM<sub>POST</sub> (1,927 kg DM ha<sup>-1</sup>) and SH<sub>POST</sub> (62.6 mm). In addition, the significant effect of FA on DHA showed lower DHA in systems managed at the lowest or highest feed allowance (27.1 vs. 32.0 kg DM or 59.3 vs. 66.8 g DM kg<sup>-1</sup> LW), but the effect of FA on HM<sub>PRE</sub> and SH<sub>PRE</sub> was not significant. Significant effects of FA on HM<sub>POST</sub> and SH<sub>POST</sub> showed lower residual yields plus heights in systems managed at the lowest feed

allowance (1,850 vs. 2,005 kg DM ha<sup>-1</sup> and 57.2 vs. 68.2 mm). The area offered daily to the systems showed similar trends in late and early lactation; differences between genotypes were small and not significant, but with larger areas offered to systems managed at the highest feed allowance.

### 7.3.2. Milk yield

In early lactation, milk and MS yields were greater for NZ90 and OS90 than for NZ70 (25.3 vs. 19.4 litres cow<sup>-1</sup> and 1.9 vs. 1.5 kg MS cow<sup>-1</sup>; Table 7.3). In addition, milk yield was lower in systems managed at the lowest than the highest feed allowance (22.6 vs. 24.1 litres cow<sup>-1</sup>). Differences in MS yield (1.8 kg MS cow<sup>-1</sup>) between lowest or highest feed allowances were not significant, however protein yield was greater at the highest feed allowance (0.77 vs. 0.83 kg cow<sup>-1</sup>). Differences in the content of lactose in milk tended to increase at the highest feed allowance. Milk composition showed a significant GE by FA interaction for fat and protein contents in milk, with fat content increased at the highest feeding allowance for NZ90 but decreased for NZ70 and OS90; additionally, protein content increased at the highest feeding allowance to a greater extent for NZ90 than for NZ70 and OS90.

In late lactation, milk and MS yields were also greater for NZ90 and OS90 than for NZ70 cows (13.4 vs. 10.6 litres cow<sup>-1</sup> and 1.2 vs. 0.9 kg MS cow<sup>-1</sup>; Table 7.3) with a trend for a numerically higher milk yield for NZ90 than OS90 (+8.2%) and significantly greater MS yield (+16.6%) sustained by greater fat (+18.3%) and protein (+14.3%; Table 7.3) yields and a small difference in lactose yield (+10.3%). Superior milk, MS (particularly protein) and lactose yields were observed in systems managed at the H<sub>1</sub> feed allowance (Table 7.3). Higher contents of fat, protein and lactose in milk were observed for NZ90 than for NZ70 and OS90, whereas NZ70 produced milk with higher fat, lower protein and similar lactose content than OS90. A trend for greater fat content in milk at low FA was measured, whereas protein and lactose contents in milk were similar across feeding allowances.

### 7.3.3. Live weight and body condition score

In early lactation, the NZ70 strain was the lightest (402 kg cow<sup>-1</sup>) and OS90 the heaviest (447 kg cow<sup>-1</sup>; Table 7.3). Live weight for the NZ90 strain was intermediate between the other two genotypes and significantly different from them (428 kg cow<sup>-1</sup>). The effect of FA on LW (426 kg cow<sup>-1</sup>) was not significant. Body condition score showed a significant GE by FA interaction, increasing for the two NZ genotypes at increased FA, but decreasing for the OS90 genotype; in addition, the lowest BCS was observed for the

OS90 strain (4.1, mean of lowest and highest) and the greatest for the NZ70 (4.7, mean of lowest and highest; Table 7.3).

In late lactation, LW showed a similar trend across GE to that in early lactation, with similar LW for the two NZ strains at the lowest feeding allowance and also similar between NZ90 and OS90 at highest feeding allowance, hence, significant differences were only observed between NZ70 and OS90 (453 vs. 480 kg cow<sup>-1</sup>; Table 7.3). Body condition score also showed a trend for a significant GE by FA interaction. The NZ70 cows were fatter (4.8) than NZ90 (4.3), whereas the OS90 had the lowest score (3.8). Between early and late lactation, the NZ70 improved while the NZ90 held BCS at highest but not at lowest feed allowance, whereas the BCS of the OS90 declined at both feed allowances.

### 7.3.4. Dry matter intake and diet digestibility

In early lactation the NZ90 and OS90 strains had higher DMI<sub>H</sub> than NZ70 (Table 7.4) with the NZ90 strain showing the highest DMI<sub>H</sub> at the lowest feed allowance, as a result of the improved sward conditions (GE by FA interaction). The same occurred for NZ70 at the highest feed allowance, also due to the improved pasture condition. In contrast, the OS90 showed similar DMI<sub>H</sub> between feed allowances even though the differences between swards were of the same magnitude as those observed for the NZ90 strain.

**Table 7.4: Early lactation. Herbage dry matter intake and grazing behaviour of three genotypes of Holstein-Friesian dairy cows managed at low or high annual feed allowance. Mean value for each system.**

Genotype	NZ70		NZ90		OS90		SED	Significance		
	Low	High	Low	High	Low	High		GE	FA	GE*FA
DMI <sub>H</sub> (kg cow <sup>-1</sup> )	12.5	13.4	15.6	14.5	14.0	14.0	0.49	***	NS	***
DMI <sub>H</sub> (g kg <sup>-1</sup> LW)	31.2	33.3	37.9	32.7	31.0	31.4	1.65	***	*	***
DMD (g/kg)	807	825	842	815	815	802	4.05	***	***	***
G <sub>T</sub> (min/day)	495	478	483	539	487	507	14.4	***	*	***
R <sub>T</sub> (min/day)	202	239	277	237	197	253	13.9	**	***	***
G <sub>T</sub> /R <sub>T</sub>	2.59	2.06	1.78	2.39	2.62	2.21	0.21	*	NS	***
Grazing Bouts	5.61	6.14	7.30	6.77	6.23	6.75	0.32	***	NS	0.068
Bout duration (min/day)	90	80	67	82	82	77	4.85	*	NS	***
B <sub>R</sub> (bites/min)	63.9	70.1	63.2	67.9	65.7	69.0	1.69	NS	NS	NS
Bites total (bites/day) *1000	31.5	33.6	30.5	36.7	31.9	35.1	1.36	NS	***	*
I <sub>R</sub> (g/min)	25.5	28.5	32.3	27.1	29.0	27.8	1.27	*	0.08	***
B <sub>Z</sub> (mg/bite)	402	411	513	402	444	408	22.7	**	***	***

DMI<sub>H</sub>: herbage dry matter intake; LW: live weight; DMD: diet dry matter digestibility; G<sub>T</sub>: grazing time; R<sub>T</sub>: ruminating time; G<sub>T</sub>/R<sub>T</sub>: grazing to ruminating time ratio; B<sub>R</sub>: bite rate; I<sub>R</sub>: herbage intake rate; B<sub>Z</sub>: bite size. GE: genotype; FA: feed allowance; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= not significant.

The DMI<sub>H</sub>/LW showed a similar response to that recorded for DMI<sub>H</sub> (Table 7.4). The amount of nutrients consumed relative to the LW of the cows improved with DMI<sub>H</sub> and to a greater extent in lighter cows. In addition to the improvement in DMI<sub>H</sub>, the NZ90 strain showed a trend for higher DMD with the highest value measured at lowest feed allowance. This response occurred in accordance with the better structure of the sward at the lowest feed allowance. Similar trends were observed for NZ70 and OS90 genotypes with a higher DMD at the better sward condition; however, a significant GE by FA interaction was also measured.

**Table 7.5: Late lactation. Herbage and total dry matter intakes and grazing behaviour of three genotypes of Holstein-Friesian dairy cows managed at low or high annual feed allowance, with maize silage fed at high feed allowance in late lactation. Mean values for each system.**

Genotype	NZ70		NZ90		OS90		SED	G	Significance	
	Low	High	Low	High	Low	High			FA	GE*FA
DMI <sub>H</sub> (kg cow <sup>-1</sup> )	12.4	9.9	13.8	11.2	14.1	10.2	0.53	***	***	***
DMI <sub>MS</sub> (kg cow <sup>-1</sup> )	---	2.09	---	5.47	---	5.46	0.53	***	---	---
DMI <sub>T</sub> (kg cow <sup>-1</sup> )	12.4	11.9	13.8	16.6	14.1	15.6	0.71	***	***	***
DMI <sub>H</sub> /LW (g kg <sup>-1</sup> LW)	28.1	22.0	30.8	23.0	29.4	20.6	1.61	0.07	***	0.097
DMI <sub>MS</sub> /LW (g kg <sup>-1</sup> LW)	---	4.51	---	11.1	---	11.1	1.06	***	---	---
DMI <sub>T</sub> /LW (g kg <sup>-1</sup> LW)	28.1	26.5	30.8	34.1	29.4	31.7	1.32	***	0.10	**
DMD (g/kg)	772	755	764	749	761	756	6.82	NS	***	NS
G <sub>T</sub> (min/day)	519	443	529	420	516	390	18.7	*	***	0.083
E <sub>MST</sub> (min/day)	---	38	---	75	---	68	9.22	***	---	---
F <sub>T</sub> (min/day)	519	481	529	496	516	459	16.8	**	***	0.126
R <sub>T</sub> (min/day)	212	227	222	260	253	267	15.6	***	**	NS
G <sub>T</sub> /R <sub>T</sub>	2.57	2.07	2.59	1.67	2.10	1.53	0.19	***	***	NS
Bout number	4.63	6.64	5.58	6.18	6.77	6.28	0.42	**	***	***
Bout duration (min/day)	119	69	103	71	80	64	7.12	***	***	**
BR (bites/min)	62.6	64.8	66.4	66.6	60.7	67.0	1.82	*	**	**
Bites total (bites/day) *1000	32.4	28.7	35.2	27.9	31.3	26.2	1.46	**	***	0.109
I <sub>R</sub> (g/min)	24.2	22.3	26.5	26.9	27.6	26.2	1.54	***	0.07	NS
MS-I <sub>R</sub> (g/min)	---	69.7	---	89.6	---	94.6	15.9	NS	---	---
BZ (mg/bite)	387	350	404	406	459	393	25.5	***	**	**

DMI<sub>H</sub>: herbage dry matter intake; DMI<sub>MS</sub>: maize silage dry matter intake; DMI<sub>T</sub>: total dry matter intake; LW: live weight; DMD: diet dry matter digestibility; G<sub>T</sub>: grazing time; E<sub>MST</sub>: eating maize silage time; F<sub>T</sub>: feeding time; R<sub>T</sub>: ruminating time; G<sub>T</sub>/R<sub>T</sub>: grazing to ruminating time ratio; BR: bite rate; I<sub>R</sub>: herbage intake rate; MS-IR: maize silage intake rate; B<sub>Z</sub>: bite size. GE: genotype; FA: feed allowance; GE\*FA: genotype by feed allowance interaction. SED: standard error of the differences. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = not significant.

In late lactation, cows in systems managed at the highest feed allowance were supplemented with maize silage. A trend for greater DMI<sub>H</sub> was observed for the two high yielding genotypes compared to NZ70 (Table 7.5), in addition, DMI<sub>H</sub> declined at the highest feed allowance to a greater extent in the OS90 strain than for both NZ genotypes as indicated by the GE by FA interaction, probably associated with the larger amount of maize silage fed to the NZ90 or OS90, 2.6 times higher than in NZ70. The NZ90 strain achieved a higher DMI<sub>H</sub>/LW and differences between NZ70 and OS90 were non-significant. A significant effect of FA was observed with lower DMI<sub>H</sub>/LW for cows consuming supplement and a trend for a larger decline in OS90. This resulted in different

substitution rates of pasture by supplement, the highest observed for NZ70 and the lowest for the NZ90 genotype, with an intermediate substitution rate for OS90 cows. The highest  $DMI_H$  was recorded in the supplemented NZ90 cows and no significant differences in  $DMI_H$  were observed between the NZ70 and OS90 genotypes.

The NZ90 and OS90 strains fed supplements increased  $DMI_T$ . The improvement was greater for NZ90 that achieved 16.6 kg DM cow<sup>-1</sup> and 34.1 g DM kg<sup>-1</sup> LW (Table 7.5) whereas  $DMI_T$  in the OS90 was only 15.6 kg DM cow<sup>-1</sup> and 31.7 g DM kg<sup>-1</sup> LW. In contrast, at the  $H_O$  feed allowance, the  $DMI_T$  of the NZ70 decreased due to the larger substitution rate measured with this genotype. Even though the NZ70 cows showed a trend for a superior DMD (763 g kg<sup>-1</sup>; mean of  $L_O$  and  $H_I$ ; Table 7.5), differences across genotypes were not significant and declined significantly at the H feed allowance (2.3%).

### 7.3.5. Grazing behaviour

#### 7.3.5.1. *Systems fed only pasture in early lactation*

Both NZ90 and OS90 strains spent similar  $G_T$  (498 min), slightly higher than NZ70 (486 min; Table 7.4); however, a significant GE by FA interaction for  $G_T$  showed that  $G_T$  increased at the highest feed allowance for NZ90 to a greater extent than for OS90, whereas a decline was observed for the NZ70 strain at the highest feed allowance (Table 7.4). A significant GE by FA interaction for  $R_T$  showed that  $R_T$  increased in OS90 and NZ70 at highest feed allowance, whereas in NZ90 it declined.

Both the NZ90 and OS90 strains showed a trend to perform an increased number of mean daily total bites than the NZ70 genotype (Table 7.4). The daily number of bites also increased at the H feed allowance, nevertheless, a significant GE by FA interaction for the total number of daily bites showed a larger increment for NZ90 than OS90 between feed allowances (Table 7.4), which was larger for these two strains than for NZ70. The increment in the total number of daily bites was due to a faster  $B_R$  of all three strains managed at  $H_I$  feed allowance; however  $G_T$  was adjusted differently in each genotype.

A significant effect of GE was observed on the number of grazing bouts (meals) performed daily, higher for NZ90 cows (7.0 bouts day<sup>-1</sup>), intermediate for OS90 (6.5 bouts day<sup>-1</sup>) and lower for NZ70 (5.9 bouts day<sup>-1</sup>); however, FA had no significant effect on the number of grazing bouts (Table 7.4). A significant GE by FA interaction was measured for the mean length of each bout; this was reduced for NZ70 but increased in NZ90 cows at the highest feed allowance (Table 7.4). The mean daily  $B_R$  showed non-significant effects of both G and FA (66.6 B min<sup>-1</sup>).

As a measure of the time allocated to G or R, the  $G_T/R_T$  ratio was estimated (Table 7.4). A significant GE by FA interaction showed that the NZ70 and OS90 genotypes reduced the  $G_T/R_T$  ratio when managed at highest feed allowance whereas the opposite occurred for the NZ90. Significant GE by FA interactions for  $I_R$  and bite size ( $B_Z$ ) showed that both variables decreased at the  $H_I$  feed allowance for NZ90 and OS90, whereas they increased for NZ70. However, considering the way  $I_R$  and  $B_Z$  were estimated, these variables are highly auto-correlated.

### 7.3.5.2. *Late lactation, with maize silage fed at the higher feed allowance*

In late lactation, cows were offered pasture only at the lowest feed allowance, but pasture plus maize silage at the highest feed allowance. There were significant effects of GE and FA on  $G_T$ ,  $E_T$ ,  $R_T$ , the  $G_T/R_T$  ratio and the daily number of bites (Table 7.5). Grazing time was longer for both NZ strains (477 min) than for OS90 (456 min), whereas the shortest  $R_T$  was measured for NZ70 (219 min) and the longest for OS90 (258 min), with the  $R_T$  of the NZ90 intermediate (241 min). The  $G_T/R_T$  ratio was higher for NZ70 (2.30), lower for NZ90 (2.13) and lowest for OS90 (1.85). The daily number of bites was higher for NZ90 than NZ70 ( $P=0.13$ ) and the lowest was measured for OS90 (Table 7.5).

Cows grazing only pasture at the lowest feed allowance spent more time  $G_T$  (528 min), less  $R_T$  (229 min) and increased the  $G_T/R_T$  ratio (2.46) than cows fed also supplement at the highest feed allowance (Table 7.5). The latter cows spent less time  $G_T$  (418 min) but spent time  $E_T$  from the troughs (60 min), increased  $R_T$  (251 min) and as result reduced the  $G_T/R_T$  ratio (1.73). However, the NZ70 cows did not increase  $R_T$  because  $DMI_T$  was slightly reduced for these cows when supplement was fed. The total daily number of bites was higher for cows fed only pasture, mainly because of  $G_T$  was higher for these cows, and even though  $B_R$  was slower.

The OS90 genotype performed the highest number of G bouts per day (6.5) and the NZ70 the lowest (5.6); however, the GE by FA interaction was significant because the number of bouts increased for the NZ genotypes when supplement was fed, but not for the OS90 (Table 7.5). In addition, all three genotypes showed a decline in bout duration when supplemented, the effect being greater for both NZ genotypes, particularly for NZ70. Even though the NZ90 genotype had higher  $B_R$  ( $66.5 \text{ B min}^{-1}$ ) than NZ70 and OS90, a significant GE by FA interaction showed that the  $B_R$  of the NZ70 and OS90 genotypes was lower when fed on pasture alone, however, the NZ90 showed similar  $B_R$ , supplemented or not. A significant GE by FA interaction was measured for  $I_R$ , with the lowest value for NZ70 ( $23.3 \text{ g DM min}^{-1}$ ), which declined when supplemented, and higher  $I_R$  for the NZ90 and OS90 genotypes ( $26.8 \text{ g DM min}^{-1}$ ), which increased slightly for NZ90 cows, but declined in the OS90 when supplement was fed.

## 7.4. DISCUSSION

As mentioned in the previous chapter, there were a number of difficulties in dealing with the substantial number of interactions between pasture characteristics and their effect on  $DMI_H$  and grazing behaviour, just because not only these variables were determined by the characteristics of the sward grazed on the dates  $DMI_H$  and grazing behaviour were measured, also because previous management affected body reserves mobilization and daily yield, and hence the energetic status of the cow on these dates and her capacity to respond to changes in pasture availability. This involved a substantial number of interactions between sward characteristics and confounded effects on  $DMI_H$  and grazing behaviour. Although conditions associated with the design of the experiment and the way the different feeding levels were achieved made it difficult to be conclusive about the differences in  $DMI_H$  and grazing behaviour between genotypes, it must be noted that the main objective was to compare  $DMI_H$  and grazing behaviour of the genotypes under comparable feeding conditions, determined by the differences in live weight and yield potential between the strains. Although the genotypes could be compared at similar level of feed, farming conditions for the systems would have been unrealistic. In addition, maintaining these conditions over the whole season or over the experiment would not be possible within the scope of a long-term system study.

Daily herbage allowance is the main determinant of pasture intake (Combellas & Hodgson, 1979; Bryant, 1980; Glassey *et al.*, 1980; Holmes, 1987; Suksombat *et al.*, 1994; Peyraud *et al.*, 1996; Wales *et al.*, 1999; Maher *et al.*, 2003), hence, it has been suggested that cows of high genetic potential require higher DHA due to their higher potential requirements (Buckley *et al.*, 2000; O'Connell *et al.*, 2000; Maher *et al.*, 2003). However, changes in the structure of the sward also affect the capacity of the cow to consume herbage (Allden & Whittaker, 1970; Peyraud *et al.*, 1996) so it is expected that the response of the cow to increments in DHA change with sward conditions, as result of the different availability of the herbage in swards of different structure (Hodgson, 1990). In early lactation, a confounded effect between DHA and  $HM_{PRE}$  or  $SH_{PRE}$  on  $DMI_H$  and grazing behaviour was measured. The fact that the current DHA/LW was more than twice the current  $DMI_H/LW$  (Greenhalgh *et al.*, 1967; Combellas & Hodgson, 1979) suggests that the structure of the herbage, other than DHA, was the main factor affecting  $DMI_H$  and grazing behaviour. In late lactation, the pastures grazed had similar structures at the two FA and the supplement fed was the main factor affecting  $DMI_H$  and grazing behaviour.

Differences in previous management affected the LW and BCS of the cows and thus also the metabolic status of the cow and feeding drive in the measurement period. However, pasture management during the measurement period imposed limitations to intake and

determined the actual energy output of the cows. Thus, to investigate further the effect of the contrasting structures of the swards on  $DMI_H$  and grazing behaviour of the different genotypes in the early lactation period, the swards were re-classified as SHORT or TALL, according to  $HM_{PRE}$  and  $SH_{PRE}$ . The SHORT swards had lower  $HM_{PRE}$  (2,100 kg DM ha<sup>-1</sup>) and  $SH_{PRE}$  (125 mm) than the TALL swards (2,540 kg DM ha<sup>-1</sup> and 150 mm respectively), and both swards had similar bulk density. This procedure did not completely eliminate the effect of DHA on  $DMI_H$ , as small differences in DHA are still evident between the genotypes grazing the SHORT or TALL swards, however its effect is reduced as indicated by the lower  $DMI_H$  measured when the genotypes were offered a higher DHA. As a result of the negative effect of DHA on  $DMI_H$ , grazing efficiency estimated as  $DMI_H$  expressed as proportion of DHA (Combellas & Hodgson, 1979) was below 0.5 in all cases (estimated from Tables 7.2 and 7.4) which suggests that a high proportion of the increment in DHA was not available for the cows, thus  $DMI_H$  was affected more by the structure of the sward than by DHA. In late lactation the structure of the sward was similar across GE and FA ( $HM_{PRE}$ : 2,419 kg DM ha<sup>-1</sup> and  $SH_{PRE}$ : 121 mm) and the effect of supplementation on  $DMI_H$  and  $DMI_T$  was the main factor investigated.

#### **7.4.1. Live weight, body condition score and milk yield in early and late lactation**

As differences in BCS and LW were not significant across feed allowance at calving, the differences measured in early lactation indicate that the OS90 cows used more body reserves post-calving than both NZ genotypes, and NZ90 more than NZ70 (see discussions in Chapters 5 and 6). Between early and late lactation, all the genotypes managed either at the lowest or highest feed allowance regained LW (Table 7.3), however, at the lowest feed allowance all three genotypes continued losing BCS whereas at the highest feed allowance BCS in the NZ70 strain was increased, maintained for NZ90 and decreased slightly for the OS90 genotype (Table 7.3). This indicates that the genotypes showed a different partition of nutrients to milk production or body reserves even during the late lactation period. Changes in milk yield and composition are summarised in Table 7.3 and are known to have affected daily energy output (see Chapter 6). The NZ90 and OS90 genotypes achieved greater yield than NZ70 in agreement with the different yield potential of these genotypes. However, differences between NZ90 and OS90 were non-significant in early lactation despite the greater potential of the OS90 strain (Kolver et al., 2002), and more body reserves were mobilised to sustain yield for OS90 than NZ90. In late lactation daily yield was higher for NZ90 than OS90.

The differences in milk yield in early lactation of the genotypes were consistent with the higher  $DMI_H/LW$  measured for the two NZ genotypes than for OS90 and indicate

differences in the energy status of the cows relative to maintenance. Yield for both NZ genotypes was supported to a greater extent from the energy consumed than by mobilisation of reserves. Differences in yield between NZ70 and NZ90 were due to the greater fat reserve mobilisation and the greater  $DMI_H/LW$  of the NZ90.

In late lactation, the higher  $DMI_T/LW$  of the NZ90 than NZ70 and OS90 and the improved energy status of this strain sustained greater daily yield. The  $DMI_T/LW$  was higher for OS90 than NZ70, however it was apparent that body fat reserves were still mobilised for OS90 cows to support daily yield. At the highest feed allowance the NZ70 cows achieved a higher proportion of their adult LW and mature body fat mass, but had lower MS yield, so the lower energy intake is consistent with the greater fatness of the NZ70 cows (Ingvarsen et al., 1999) and their lower daily energy requirements. In contrast, the supplemented NZ90 cows increased MS yield and both  $DMI_H/LW$  and  $DMI_T/LW$  to a greater extent than the OS90 due to a lower substitution of pasture by maize silage. This is in contrast to what was observed for the complete season (Chapter 5).

At the lowest feed allowance, intake and daily MS yield were numerically higher for NZ90 than OS90 (+5% and 13% respectively) probably due to the effect of the previous nutrition on these cows. It is suggested that once a cow achieves a minimum threshold for body fat mass, the capacity to buffer yield under constraining feeding conditions becomes limited and energy output will reach equilibrium at a level determined by energy intake. The higher threshold of the NZ70 cows is associated with a lower partition to milk yield in cows of low genetic yield potential, however this energy would be available to support other functions (like reproduction) or even to maintain yield during periods of reduced feed availability.

#### **7.4.2. Intake and grazing behaviour under different swards conditions in early lactation**

##### **7.4.2.1. Grazing a SHORTER sward**

Even though in the shorter sward all three genotypes had small  $B_Z$ , slow  $B_R$  and extended  $G_T$  (Table 7.4),  $B_R$  was higher for both NZ90 and OS90 than NZ70 with a positive effect on  $I_R$ . Sward conditions were similar for all three genotypes although DHA was slightly lower for the NZ70 than for the other two strains. This probably explains the slightly lower  $DMI_H/LW$  measured for NZ70 than NZ90 and OS90, and the fact that the NZ70 cows grazed to a lower post-grazing residual. However, the DMD (from n-alkanes) of the NZ70 was higher than that achieved by the OS90. The NZ70 had lower yield (Table 7.3) and assumed lower energy requirements for maintenance (see Chapter 6), hence the

energy status of this genotype might be higher than for NZ90 and OS90 genotypes despite its lower  $DMI_H/LW$ , which is in agreement with the reduced amount of fat reserves mobilised during early lactation.

Both NZ90 and OS90 genotypes avoided grazing deeper into the bottom layer of the pasture to a greater extent than NZ70 as indicated by the high values for  $HM_{POST}$  and  $SH_{POST}$  (Table 7.2). Assuming a similar structure and quality in the vertical plane in the pastures grazed by NZ90 and OS90, the higher DMD of the NZ90 suggests a greater capacity to select high quality plant parts than the OS90. It is apparent that the two NZ strains had similar abilities in this respect, although the NZ70 was probably affected by grazing at lower DHA.

The NZ90 cows grazed for longer at a slightly slower  $I_R$  than the OS90, which had a faster  $B_R$ . The NZ90 had the highest total number of bites per day, the OS90 intermediate and the lowest for the NZ70. The extended  $G_T$  of the NZ90 agreed with previous observations (Thorne *et al.*, 2003). The two NZ genotypes showed a trend for longer G bouts, reduced  $R_T$  and increased  $G_T/R_T$  ratio. Cows increase  $G_T$  not by increasing the number of meals but by increasing bout duration (Gibb *et al.*, 1999). The length of each bout was shorter for OS90 than NZ90, as was suggested by the higher  $I_R$  and probably faster gut fill, which would require a frequent alternation between grazing and rumination activities. It was suggested that a rapid alternation between G and R increases rumen fill and retention times so the digestibility of the diet improves (Greenwood & Demment, 1988).

It is apparent that grazing is a slower process for the NZ genotypes than OS90; in addition, it seems that G is prioritised over R for the NZ strains. For instance, the NZ90 consumed more herbage than OS90 but had a shorter  $R_T$  and higher  $G_T/R_T$  (Table 7.4). It is suggested that the reduction in  $R_T$  might have resulted from a combination of improved diet digestibility and the ingestion of smaller plant particles by the NZ90 cows.

Differences in  $B_Z$  between strains were small in this sward condition (SC), probably because herbage prehension was constrained and the possibility of increasing  $B_Z$  was limited (McGilloway *et al.*, 1999). The faster  $B_R$  of the NZ90 strain in this SC, and to a greater extent in the OS90, suggests these cows attempted to compensate for the increased limitations to  $B_Z$  that would occur while grazing down (Gibb *et al.*, 1999). The short  $SH_{POST}$  possibly affected  $B_R$  for the NZ70 cows, as prehension difficulties are apparent with cows grazing close to ground level. O'Connell *et al.* (2000) indicated that high yielding cows had higher  $B_R$  than their low yielding counterparts, however the increase in  $B_R$  is the apparent response of cows with limitations to compensate by adjusting  $B_Z$  further. Biting rate increased in mature swards where leaf material is less

accessible and is associated with an increased number of bites per day and small  $B_Z$  (Stobbs, 1974; Gibb *et al.*, 1999). Considering that NZ90 and OS90 left a higher post-grazing residual than NZ70, a similar  $B_Z$  in the genotypes suggests that the NZ70 cows can maintain  $B_Z$  despite the shorter  $SH_{POST}$ .

#### 7.4.2.2. *Grazing a TALLER sward*

In this sward,  $DMI_H$  was lower for NZ70 than NZ90 and OS90, which is in agreement with the lower mean daily requirements of NZ70 than the NZ90 and OS90 genotypes (see Chapter 6). Hence, considering the  $DMI_H/LW$  achieved by the NZ70, it is possible that these cows had already achieved satiation and thus had a reduced motivation to graze further. The NZ90 strain had the highest  $DMI_H$  and  $DMI_H/LW$ , whereas  $DMI_H$  was higher for OS90 than NZ70, but the opposite occurred when intake was expressed as  $DMI_H/LW$ , which was greater in NZ70 than for OS90 cows (Table 7.4). Thus in this sward, the energy status of the two NZ genotypes was higher than for OS90 cows, even though the NZ90 and OS90 genotypes had similar milk and MS yields and energy output. The improved energy status of the two NZ genotypes agreed with the higher BCS of these strains, the difference in the amount of energy being partitioned to body reserves being supported by the difference in the energy requirements for maintenance and milk production between them (see Table 6.8 in Chapter 6).

In this sward all three genotypes grazed mainly the top layers of the pasture as indicated by both  $HM_{POST}$  and  $SH_{POST}$ , and improved DMD (from n-alkanes). The value of DMD was higher for both NZ strains than for the OS90 even though  $SH_{POST}$  was taller in paddocks grazed by OS90 (non-significantly), which suggests that this strain avoided grazing deeper into the lower stratum of the pasture. It is known that in a vegetative sward the upper stratum of the canopy is made up of living leaf lamina whereas the proportion of leaf sheaths, pseudostems and dead tissue increase close to ground level (Hodgson, 1985). It is possible that in a tall pasture under strip grazing the contrast in quality and bulk between upper and lower canopy layers were greater due to increased senescent material in the residual stubble (Burlison *et al.*, 1991; McGilloway *et al.*, 1999). Thus, those cows grazing at higher  $I_R$  and depleting the pasture faster would face rapid changes in the sward and would be more affected by the rate of these changes, in addition to the inhibitory condition of the stubble. Hence, although  $B_Z$  should have been progressively restricted by limitations to bite depth in all the genotypes, the effect on the OS90 cows seemed to be greater.

### 7.4.3. Differences between grazing SHORTER or TALLER swards

In all three genotypes,  $I_R$  was faster and  $G_T$  shorter in the TALL sward than in the SHORT sward. In addition, the increase in  $I_R$  was greater for the two NZ genotypes than for OS90. This would be expected to affect gut fill, the number of bouts per day and the mean duration of the bout, and hence improve digestibility if rumen retention time is extended (Greenwood & Demment, 1988). Even though the two NZ strains improved  $I_R$  in the TALL compared to the SHORT swards, NZ90 cows reduced  $B_R$  (7%) and increased  $B_Z$  (27%) whereas NZ70 cows increased  $B_R$  (11%) and  $B_Z$  only slightly (2%). The OS90 had a slightly smaller decrease in  $B_R$  between TALL and SHORT swards than the NZ90 cows (5%) but also increased  $B_Z$  to a smaller extent (9%).

The proportional increase of  $B_Z$  for the NZ90 strain in the TALL sward could be the result of a lower effect of bulk density on bite penetration (Barrett *et al.*, 2001). Sward factors are also known to influence  $I_R$  through their effects on  $B_Z$ , but to a lower extent on  $B_R$  (Mayne *et al.*, 1997); however, it is apparent that  $B_R$  played a more important part in the behavioural adjustment of the two NZ strains to different sward conditions. These changes indicate a greater flexibility for the NZ90 cows to adjust ingestive behaviour to changes in sward structure.

It is apparent that the capacity of the NZ90 cows to improve  $DMI_H$  and  $DMI_H/LW$  was based on their ability to adjust  $B_Z$  and  $G_T$  to the differences between the swards. It is suggested that the larger reduction in  $G_T$  between swards also contributed to reducing energy expenditure for NZ90 (Bergman *et al.*, 2001). Nevertheless, it seems that to improve  $DMI_H$  for the OS90, pasture management strategies should focus on maximizing  $B_Z$  for cows of high genetic yield potential, as suggested by Mayne *et al.* (1997). However, an increase in  $DMI_H$  would require a greater  $R_T$ , which could negatively affect the amount of time available to extend  $G_T$  further. The time a cow spends  $R$  is associated with the effort and energy required to process each unit of herbage consumed and thus increases when both the intake and fibre content of the diet increase, whereas it should decline in animals that perform an efficient rumination. A higher  $R$  efficiency has been reported in solely pasture fed cows of the OS Holstein-Friesian in comparison to a high yielding NZ Holstein-Friesian (Thorne *et al.*, 2003), however this desirable capacity would not be fully exploited by the OS90 cows they have their  $DMI_H$  constrained by sward conditions.

Even though  $G_T$  was similar in all the three genotypes in the TALL sward, the two NZ genotypes performed more bouts of shorter duration, increased  $R_T$  and reduced the  $G_T/R_T$  ratio compared with the SHORT sward. It is apparent that although the OS90 cows increased  $B_Z$  in the TALL sward, they simultaneously extended the length of the bout,

and reduced bout number and  $B_R$ . Hence, although changes in  $B_Z$  and  $G_T$  followed similar trends to those observed in the NZ genotypes, the increased length of the bout in the OS90 had probably an effect on bite quality as indicated by the DMD value.

Despite these changes in behaviour, cows did not graze the TALL sward to a similar post-grazing residual as the SHORT sward, and thus  $G_T$  was not extended. The higher  $DMI_H$  of the NZ genotypes in the TALL sward achieved by performing larger bites would require further energy expenditure in processing and digesting the extra feed consumed, but in contrast the OS90 strain did not increase  $G_T$  even though  $R_T$  was reduced as DMD slightly improved. Although daily  $DMI_H$  was kept similar, this behaviour would reduce energy expenditure and improve energy status further for OS90 cows. Hence, it is suggested that the structure of the stubble at the base of the pasture was probably the main reason for the cows of all strains to stop grazing in the TALL sward (O'Reagain & Mentis, 1989; Rook *et al.*, 1994). However, considering the different energy requirement of the strains and the  $DMI_H$  achieved, it is possible that differences in satiation level might have contributed to a greater extent for both NZ genotypes, whereas it is apparent that the OS90 cows were inhibited to a greater extent by the structure of the sward, possibly because any limit imposed by the structure of the plant results in a greater effect on  $B_Z$  in larger animals (Illius & Gordon, 1987; O'Reagain & Mentis, 1989).

Differences in  $DMI_H$  and grazing behaviour between SHORT and TALL swards agreed with the greater reduction in sward height between pre-/post-grazing in the TALL sward, even though  $SH_{POST}$  was still higher. The different structure between top and bottom layers of the herbage offered could be the reason for the higher residual of the TALL sward (McGilloway *et al.*, 1999). If cows preferentially graze the upper stratum of the pasture, the stubble would accumulate more senescent and dead material, have lower quality and higher bulk density (Wade & Le Du, 1982; O'Reagain & Mentis, 1989). As a result of this process, the contrast between upper and lower strata would increase by the subsequent grazing, so increasing the difficulty for the cow to graze more deeply into the bottom layer (Rook *et al.*, 1994).

Cows performing a reduced number of large and digestible bites increase the reward per bite (Thornley *et al.*, 1994). However as the sward is grazing down,  $B_Z$  and bite quality may decrease. At the same time the effort to harvest each bite would increase (Ungar, 1996), whereas the energy required to process and digest low quality herbage would also increase. It is suggested that the NZ90 cows can reduce the size of the bite to a greater extent than OS90 and are also able to increase the quality of the herbage harvested by selectivity. Although this necessitates a slow rate of intake that would require expending more energy per gram of DM ingested in searching for the best quality plant parts available, it would be compensated by a lower requirement to process and digest the

herbage consumed due to the smaller size and higher digestibility of the particles of herbage ingested (Rook *et al.*, 1994). It was estimated that the energy expended in prehension is lower than that expended in mastication (Illius *et al.*, 1995).

#### 7.4.4. Intake and grazing behaviour under supplementation in late lactation

The NZ90 and OS90 strains had higher  $DMI_H$  and  $DMI_T$  than NZ70, even though larger declines in  $DMI_H$  and  $DMI_H/LW$  were observed in the OS90 strain fed maize silage than for NZ90. However, the NZ90 and OS90 were fed much larger quantities (2.6 times) of maize silage than the NZ70 genotype. As a result, substitution rate was highest for NZ70 ( $1.19 \text{ kg } DMI_H \text{ kg}^{-1} DMI_{MS}$ ), intermediate for OS90 ( $0.71 \text{ kg } DM_H \text{ kg}^{-1} DMI_{MS}$ ) and lower for NZ90 ( $0.48 \text{ kg } DM_H \text{ kg}^{-1} DMI_{MS}$ ). This response was different to that previously reported (Linnane *et al.*, 2004; Buckley *et al.*, 2005; Horan *et al.*, 2006).

The higher substitution of the NZ70 occurred even though the amount of supplement fed was lower than that fed to NZ90 and OS90, probably because of the lower daily requirements and feeding drive of the NZ70 (Meijs & Hoekstra, 1984; Penno *et al.*, 2001). Hence, the supplemented OS90 cows had lower  $DMI_H/LW$  (Table 7.5) than the two NZ strains; however,  $DMI_T/LW$  was higher than NZ70 but still lower than NZ90. In contrast, the supplemented NZ70 cows showed a decrease in  $DMI_T/LW$  because of their larger substitution rate.

When compared with unsupplemented cows, all the genotypes fed maize silage reduced  $G_T$ , increased  $R_T$  and reduced the total number of daily bites and  $B_Z$ , which agrees with previous reports (Linnane *et al.*, 2004). However, the two NZ had longer  $G_T$ , shorter  $R_T$  and showed a trend for more total number of bites when compared with the OS90 genotype. In addition,  $I_R$  was slightly higher for NZ90 than OS90. The number of grazing bouts per day increased in the two NZ strains when supplemented, but decreased in the OS90 genotype. In contrast, on pasture only the OS90 had the highest number of grazing bouts per day and the NZ70 the lowest, however, the NZ70 performed longer bouts while the OS90 performed shorter bouts. Bout duration was shorter for supplemented than unsupplemented cows of all three genotypes, also in agreement with Linnane *et al.* (2004).

The greater decline in  $G_T$  of the supplemented OS90 was in agreement with the increase in  $B_R$  despite the decline in  $B_Z$ . Although all three genotypes increased post-grazing residuals when fed supplement, the taller  $SH_{POST}$  of pastures grazed by OS90 suggests that these cows grazed the upper stratum and were able to reduce  $B_Z$  further in this condition, probably in an attempt to select the highest quality herbage available. A similar response was observed with the NZ70 genotype. If these cows improved the quality of

the herbage harvested when fed supplement, this would reduce  $R_T$ . However, if the supplement increases the rate of passage of the digesta through the digestive tract (Reynolds, 2004) and this affects DMD, it could be possible that the fast turnover in the rumen could offset the improvements made in the diet selected. In contrast, a lower effect of the supplement on  $B_Z$  and  $B_R$  was observed for the NZ90 genotype, even though  $G_T$  and thus probably energy expenditure was also reduced.

Cows of the NZ genotypes fed only pasture showed a higher number of daily bites, particularly the NZ90. It is apparent that the NZ90 strain maintained  $B_Z$  and  $B_R$  within a similar range whether supplemented or not and the only change that determined the  $DMI_H$  achieved was  $G_T$ .

Even though maintaining  $B_Z$  between conditions has an effect on the daily number of bites when fed only pasture, this suggests that by performing small  $B_Z$  the two NZ genotypes improved their behavioural capacity to compensate further because of its effect on the size of the plant particles ingested and a reduced need for rumination. In addition, the DMD of the two NZ genotypes on pasture increased to a greater extent than for the OS90 strain, when compared to the low values obtained for supplemented cows, and corresponds with the large decline in  $R_T$  of the NZ90 strain fed pasture only. Thus, it is hypothesised that the capacity to reduce the size of the particles ingested would enable greater allocation of time to G, which resulted in the increment observed in the  $G_T/R_T$  ratio. It was suggested that simultaneous cropping and chewing might occur in short swards (Laca *et al.*, 1994), so this can be important in improving grazing efficiency when the herbage is less available. This behaviour would have increased relevance when grazing low quality pastures.

## 7.5. CONCLUSIONS

The present experiment provided an opportunity to investigate the response in intake and grazing behaviour of three different Holstein-Friesian genotypes grazing on swards differing in structure. Although the effects of DHA and sward structure on the response of the cows was being confounded because of the differences in management between systems, and the limitations to be conclusive about these results considered, differences in the capacity of the genotypes to adjust grazing behaviour to swards of different structure must be recognised. Although cows of high genetic potential for yield required improved sward conditions to achieve the highest  $DMI_H$  possible on pasture, the advantage of the NZ90 genotype over the OS90 on pasture is sustained by its flexible grazing behaviour.

In early lactation the TALL sward improved  $DMI_H$  for the NZ strains but not for the OS90 while DMD increased for all three genotypes, although to a greater extent in the two NZ. The main factor determining  $DMI_H$  was  $I_R$ , which increased as result of a larger  $B_Z$  and despite reduced  $G_T$  of the cows in the TALL swards. In late lactation, feeding supplement reduced  $DMI_H$  as a result of a reduced  $G_T$  in cows of all three genotypes. Both NZ strains performed a slow grazing process that improved the quality of the diet ingested. This occurred to a lower extent in late lactation, probably due to differences in pasture quality.

If the height of the sward is rapidly depleted, the ratio of leaf to stem quickly changes, and because of selectivity, or due to a higher contrast between upper and lower layers of the pasture, constraining conditions would be anticipated. Thus cows may stop grazing because they are inhibited by the condition of the sward. A constraint to herbage prehension would represent an increase in energy expenditure per bite harvested, which in relation with the energetic value of the herbage ingested, may motivate the cow to continue or to stop grazing. Supplemented cows would increase the inhibitory satiety response and increase substitution rate when the relative energy deficit of the cow is low.

The changes observed in the grazing behaviour of the NZ90 genotypes indicate these cows have the ability to adjust their grazing behaviour to a greater extent than both the NZ70 and OS90 genotypes. As a result, they demonstrated greater flexibility to improve their productive performance under contrasting sward and feeding conditions.

It was also apparent that although the NZ90 cows harvested heavy bites ( $513 \text{ mg bite}^{-1}$ ) in the TALL sward, under constraining sward conditions  $B_Z$  was maintained at about 400 mg/bite and the cows increased the total daily number of bites by extending  $G_T$ , while simultaneously increasing  $B_R$ . It is hypothesised that there is a balance between the energy intake per bite and the cost associated with the processing and digesting of the herbage consumed. Therefore, one possibility to reduce these costs is to harvest small herbage particles to reduce the requirement for rumination, particularly considering that the possibility to select plant parts of high quality declines as the pasture is depleted under strip grazing management.

The results of the systems comparisons provide clear evidence of contrasts between the three genotypes under changing sward conditions and confirm earlier suggestions (Kolver, 2003). They are also consistent with the hypothesis of this thesis, suggesting that selective pressure has created behavioural flexibility for the modern NZ Holstein-Friesian genotype compared to the OS strain developed under more specialised feed lot feeding conditions. However, comparisons of this kind involved a substantial number of interactions and variable time sequences in pasture conditions and also carryover effects

in the response of pasture and animals, making it difficult to draw objective conclusions about the links between management variables and productive responses. A final experiment was designed to provide the opportunity to measure the responses of the genotypes in herbage intake and grazing behaviour and their relation with productive performance under a controlled contrast in sward conditions. This experiment is reported in Chapter 8.

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## APPENDIX IV

### DESCRIPTION OF THE HOLSTEIN-FRIESIAN GENOTYPES UTILIZED IN THE PRESENT THESIS

The animals used in the field experiments described in the present thesis were sourced from Dexcel farms and from a number of commercial dairy farms within New Zealand. Dams selected had to have been herd tested and had at least three generations of pedigree; in addition, there was an avoidance of dominance in any sire lines in the pedigrees of the dams. Genotypes had different genetic potential as indicated by the breeding worth (BW), which is a profit index that estimates the cow's genetic ability to produce combined with an economic value per unit of feed required (Holmes *et al.*, 2002), thus cows with high BW will produce more profit per tonne of dry matter (DM) eaten than cows with low BW.

At the start of the experiment, the BW of the animals was similar and described by the formula:

$$\text{\$value} = \text{\$a} \times \text{kg fat} + \text{\$b} \times \text{kg protein} - \text{\$c} \times \text{milk volume}$$

Thus, the BW of the different genotypes was similar. However, the equation to estimate BW changed during the experiment to include the breeding values (BV) for live weight, somatic cells, days survivability and fertility. Breeding value is the measure of the animal's expected genetic merit for the trait (Holmes *et al.*, 2002; LIC, 2005). As a result, although the BV for fat, protein and milk volume were similar between the modern New Zealand and overseas cows, the new BV included in the equation were different and the estimated BW at the end of the experiment was different between these strains. Changes in the way the genotypes were qualified changed in part because of the results of the experiments in which the present thesis was involved, and because of the requirements to identify those animals that will perform better under the pastoral conditions of NZ dairy industry.

The three strains utilized in the present thesis were:

**NZ70 strain:** this was a 1970s low BW strain of New Zealand Friesian. This strain was representative of the selection and breeding policies in New Zealand in the 1970s. These animals were generated using semen set aside from that era by Livestock Improvement Corporation (LIC). Dams were representative of the cows in the New Zealand population in the 1970s selected for high milk fat yields. Estimated BV for the animals used were +9 (SD=9.7) kg fat; +6 (SD=6.3) kg protein; +450 (SD=253.9) kg milk; +50 (SD=21.0) kg LW; +13 (SD=7.9) days survivability; +2.3 (SD=2.7) % fertility; and -18 (SD=29.3) \$BW. These animals had 7% North American genetics.

NZ90 strain: this was a high BW Holstein-Friesian strain of New Zealand origin. This strain was generated using sires and dams with a low proportion of overseas genes and representative of the 1990s New Zealand selection and breeding policies. Estimated BV for the animals used were +32 (SD=8.0) kg fat; +33 (SD=5.4) kg protein; +913 (SD=220.9) kg milk; +53 (SD=4.4) kg LW; 12 (SD=6.4) days survivability; +0.0 (SD=2.7) % fertility; and +118 (SD=25.0) \$BW. These animals had 24% North American genetics.

OS90 strain: this was a high BW Holstein-Friesian strain of overseas origin. This strain was established by breeding NZ born cows with a high proportion of overseas genetics in their ancestry to sires of overseas origin (principally from North America). Estimated BV for the animals used were +27 (SD=8.5) kg fat; +38 (SD=5.8) kg protein; +1,183 (SD=218.6) kg milk; +82 (SD=14.4) kg LW; -2 (SD=9.6) days survivability; -7.1 (SD=3.5) % fertility; and +82 (SD=24.6) \$BW. These animals had 91% of North American genetics.

## APPENDIX V

### ADDITIONAL SYSTEMS RESULTS

In this appendix daily herbage allowance (DHA), pre- and post-grazing herbage mass and sward height ( $HM_{PRE}$ ,  $HM_{POST}$ ,  $SH_{PRE}$  and  $SH_{POST}$ ) were estimated from weekly measurements (See Chapter 6 for details). In-vivo diet digestibility (DMD), dry matter intake of fresh herbage ( $DMI_H$ ) and supplements (grass silage, maize silage or maize grain) were calculated by combining the n-alkane –  $\delta^{13}C$  methodology, for each individual cow in all the systems. The live weight, BCS and milk yield of each cow was weekly measured and a milk sample (pm-am) collected and analysed, and the yield of components estimated (See Chapter 5 for details) for each cow (only milking cows were considered).

Genotypes and FA were compared at the average FA at which the genotypes were farmed (i.e.: FA1 is the average of the lowest feeding level at which the genotypes were farmed equal to 5 t/cow per year, on the other end FA4 is the average of the highest feeding level equal to 6.5 t/cow per year). The experimental design was unbalanced and not all the levels of feed were represented in the three genotypes hence data for FA2 is the mean of NZ90 and OS90 only (FA2 equal to 5.5 t/cow per year).

All the data collected individually for each paddock grazed (DHA,  $HM_{PRE}$ ,  $HM_{POST}$ ,  $SH_{PRE}$  and  $SH_{POST}$ ) or cow (intake, yields and milk composition) in the systems for each measurement period (early, mid and late lactation periods of seasons 2002-03 and 2003-04, and the dry period between seasons) was analysed utilising the statistical procedures of SAS (SAS, 2002b) as a MIXED model (PROX MIXED) [ $Y_{ijkl} = \mu + GE_i + P_j + FA_k + (GE \times FA)_{e_{ijkl}}$ ], with the additive effects of genotype (GE), FA, parity (P or age) and genotype by feeding allowance interactions (GE by FA) as fixed effects, and paddock or cow within system as a random effect.

**Appendix V- 1: Mean daily herbage allowance in pastures grazed by three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and during the dry period between the two seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE FA	NZ70			NZ90				OS90				Sed	Significance		
		1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>	29.6	34.2	43.3	34.1	34.6	36.5	41.9	33.6	34.9	41.8	42.3	2.94	NS	***	NS
	Mid lactation	28.8	38.7	45.1	35.9	38.2	40.9	54.9	33.2	42.1	54.8	57.1	3.19	***	***	*
	Late lactation	22.5	27.0	34.9	30.1	25.6	30.7	32.0	27.21	30.8	32.5	31.5	1.77	NS	***	***
03-04	Dry period	7.10	7.97	6.05	7.57	8.26	6.78	8.53	8.43	7.92	9.59	9.98	0.56	***	NS	***
	Early lactation <sup>(1)</sup>	27.2	37.7	29.3	40.0	29.8	44.4	45.3	31.5	31.4	50.3	44.5	2.93	***	***	*
	Mid lactation	36.8	42.1	41.3	36.5	34.6	45.7	51.7	36.3	36.6	59.3	48.7	1.41	***	***	***
	Late lactation	25.2	29.1	37.5	26.0	29.4	30.1	37.2	23.3	40.5	34.0	35.30	2.11	*	***	**

Daily herbage allowance (kg DM cow<sup>-1</sup>day<sup>-1</sup>). <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 2: Mean pre-grazing herbage mass in pastures grazed by three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and during the dry period between the two seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE FA	NZ70			NZ90				OS90				Sed	Significance		
		1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>	2219	2566	2463	2585	2622	2762	2097	2547	2650	2088	2116	187	NS	*	*
	Mid lactation	2704	2904	2565	2969	2895	3099	2749	2514	3187	2738	2857	183	NS	*	0.056
	Late lactation	2552	2532	2621	2819	2562	2325	2424	2720	2335	2463	2384	151	NS	*	0.14
03-04	Dry period	2130	1800	1367	2280	2067	1642	1705	2107	1981	1918	1997	122	*	***	**
	Early lactation <sup>(1)</sup>	2043	2141	2199	2273	2260	2235	2265	2388	2383	2515	2226	216	0.09	NS	NS
	Mid lactation	2756	2391	2347	2769	2624	2287	2588	2752	2769	2963	2433	89	***	***	***
	Late lactation	2859	2725	2813	2603	2941	2279	2819	3013	3065	2577	2675	126	**	***	*

Pre-grazing herbage mass (kg DM ha<sup>-1</sup>). (1)Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 3: Mean pre-grazing sward surface height in pastures grazed by cows of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and during the dry period between the two seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE FA	NZ70			NZ90				OS90				Sed	Significance		
		1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>	125	146	146	162	148	152	120	142	151	116	121	13.8	NS	NS	*
	Mid lactation	119	142	117	139	127	155	105	107	169	110	124	12.9	NS	**	**
	Late lactation	133	129	147	138	139	116	134	144	132	141	129	11.9	NS	NS	NS
03-04	Dry-off	190	185	153	214	197	188	194	192	202	192	196	9.9	**	*	*
	Early lactation <sup>(1)</sup>	134	139	136	154	153	148	146	163	162	147	165	16.3	0.08	NS	NS
	Mid lactation	157	113	123	163	139	96	128	165	151	170	133	12.8	**	***	**
	Late lactation	160	143	150	136	152	132	162	176	187	145	152	8.5	***	***	**

Pre-grazing sward height (mm). (1)Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 4: Mean post-grazing herbage mass in pastures grazed by three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and during the dry period between the two seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE		NZ70			NZ90				OS90				Significance			
	FA		1	3	4	1	2	3	4	1	2	3	4	Sed	GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>		1620	1732	1929	1798	1826	1882	1620	1751	1770	1648	1648	102	NS	NS	*
	Mid lactation		2024	2520	2254	2412	2351	2376	2339	2101	2360	2357	2365	124	NS	*	NS
	Late lactation		1880	1880	2157	1969	1880	1940	2091	1979	1802	1926	1930	71	NS	***	0.067
03-04	Dry period		673	698	690	698	718	658	816	773	698	698	847	41	*	**	0.053
	Early lactation <sup>(1)</sup>		1513	1743	1770	1656	1616	1384	1718	1630	1785	1957	1879	167	*	NS	NS
	Mid lactation		2238	2049	2057	2188	2061	1961	2292	2213	2234	2616	2321	102	***	NS	***
	Late lactation		2055	2053	2127	1974	1927	1898	2262	1889	2121	1986	1927	121	NS	NS	0.083

Post-grazing herbage mass (kg DM ha<sup>-1</sup>). <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 5: Mean post-grazing sward surface height in pastures grazed by three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and during the dry period between the two seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE		NZ70			NZ90				OS90				Significance			
	FA		1	3	4	1	2	3	4	1	2	3	4	Sed	GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>		58	49	51	51	53	55	56	47	56	59	56	6.7	NS	NS	NS
	Mid lactation		53	64	66	62	62	69	63	46	77	64	75	9.3	NS	*	NS
	Late lactation		56	66	78	62	54	57	75	67	61	64	77	5.9	NS	***	NS
03-04	Dry period		46	58	55	48	54	53	72	54	51	62	70	6.4	0.11	**	NS
	Early lactation <sup>(1)</sup>		61	78	81	80	63	52	79	68	81	88	95	12.1	0.09	NS	NS
	Mid lactation		82	66	79	79	70	64	91	82	82	98	91	7.6	**	*	*
	Late lactation		78	78	87	83	71	74	89	81	89	80	72	5.8	NS	NS	**

Post-grazing sward height (mm). <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 6: Mean daily “in-vivo” DMD of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and the dry period between seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE	NZ70			NZ90				OS90				Significance			
	FA	1	3	4	1	2	3	4	1	2	3	4	Sed	G	FA	GE*FA
02-03	Early lactation <sup>1</sup>	806	830	825	841	813	816	815	815	788	819	801	6.09	***	***	***
	Mid lactation	762	752	778	755	762	748	767	754	760	731	766	6.83	***	***	**
	Late lactation	772	752	759	764	763	781	752	761	783	756	758	8.16	***	***	***
03-04	Dry-off	726	770	702	716	750	715	724	743	749	740	723	8.35	***	***	***
	Early lactation <sup>1</sup>	797	816	814	822	837	825	833	802	821	813	805	5.84	***	***	*
	Mid lactation	780	756	759	779	776	761	759	786	769	777	763	5.25	0.06	***	**
	Late lactation	748	750	781	769	709	761	785	762	774	758	784	7.19	***	***	***

Diet dry matter digestibility (mg kg<sup>-1</sup>DM). <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 7: Mean daily pasture dry matter intake of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and the dry period between seasons. The genotypes were farmed in systems at different feed allowance per cow per year.**

Season	GE FA	NZ70			NZ90				OS90				Sed	Significance		
		1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>	12.5	15.2	13.3	15.6	14.3	15.1	14.5	14.0	12.8	15.2	14.0	0.69	***	***	**
	Mid lactation	12.5	12.5	11.3	14.4	16.1	14.8	15.4	14.5	15.5	12.0	13.6	0.64	***	***	***
	Late lactation	12.4	12.7	9.9	13.8	15.2	16.2	11.2	14.1	15.6	11.6	10.2	0.79	***	***	***
03-04	Dry period	7.06	5.35	3.56	7.87	5.01	4.99	5.34	5.64	5.54	6.46	5.41	0.39	***	***	***
	Early lactation <sup>(1)</sup>	12.0	14.7	13.3	16.7	17.1	16.7	16.4	16.3	16.0	15.3	13.7	0.86	***	**	**
	Mid lactation	13.3	12.4	12.4	15.3	15.0	14.6	13.7	16.8	14.7	15.1	13.6	0.66	***	***	0.096
	Late lactation	13.9	10.4	11.7	13.7	9.6	9.1	12.8	11.2	13.5	13.2	12.7	0.72	***	***	***

Daily dry matter intake in kg DM cow<sup>-1</sup>day<sup>-1</sup>. <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = non significant. Probability values are indicated when approaching significance.

**Appendix V- 8: Mean daily total dry matter intake of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and the dry period between seasons. The genotypes were farmed in different systems at different feed allowance per cow per year.**

Season	GE FA	NZ70			NZ90				OS90				Sed	Significance		
		1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>(1)</sup>	12.5	15.2	13.4	15.6	14.3	15.1	14.5	13.9	12.8	15.2	14.0	0.71	***	***	**
	Mid lactation	12.5	12.5	14.2	14.4	16.1	14.8	16.2	14.5	15.5	12.0	15.4	0.73	***	***	*
	Late lactation	12.4	12.7	11.9	13.8	15.2	16.3	16.6	14.1	15.6	15.0	15.6	0.90	***	**	*
03-04	Dry period	7.06	9.41	7.12	7.87	8.66	8.87	9.19	8.63	8.67	8.71	8.82	0.39	***	***	***
	Early lactation <sup>(1)</sup>	12.0	14.7	13.3	16.7	17.1	16.7	16.4	16.3	16.0	15.3	13.7	0.86	***	**	**
	Mid lactation	13.3	12.4	12.4	15.3	15.00	14.6	15.2	16.8	14.7	16.3	16.4	0.68	***	*	0.140
	Late lactation	13.9	13.0	14.0	16.0	12.9	15.6	16.8	15.2	16.1	16.5	17.6	0.81	***	***	***

Daily dry matter intake in kg DM cow<sup>-1</sup>day<sup>-1</sup>. <sup>(1)</sup>Only pasture was fed in early lactation. G: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = non significant. Probability values are indicated when approaching significance.

**Appendix V- 9: Mean daily herbage dry matter intake per unit of live weight of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and the dry period between seasons. The genotypes were farmed in different systems at different feed allowance per cow per year.**

Season	GE	NZ70			NZ90				OS90				Sed	Significance		
	FA	1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>1</sup>	31.1	36.3	33.2	37.8	34.3	35.5	32.1	30.7	28.9	35.1	31.3	1.7	***	***	***
	Mid lactation	29.9	28.2	26.7	33.5	35.8	32.9	32.7	30.7	33.9	26.7	29.9	1.2	***	***	**
	Late lactation	28.0	27.4	22.0	30.7	33.6	33.8	22.9	29.2	33.2	24.6	20.5	1.7	***	***	***
03-04	Dry-off	14.6	10.9	7.5	15.4	9.9	9.5	9.5	10.0	9.9	11.4	9.3	0.8	**	***	***
	Early lactation <sup>1</sup>	27.7	33.2	30.9	38.1	38.3	35.7	33.9	33.5	33.3	31.2	27.9	2.0	***	***	***
	Mid lactation	29.9	27.2	28.1	33.8	33.4	29.7	27.3	33.3	29.3	29.9	26.9	1.2	***	***	***
	Late lactation	29.4	21.9	25.3	27.8	20.2	17.8	24.0	20.9	26.0	24.6	24.3	1.6	***	***	***

Daily total dry matter intake per unit of live weight (g kg<sup>-1</sup>LW). <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 10: Mean daily total dry matter intake per unit of live weight of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04, and the dry period between seasons. The genotypes were farmed in different systems at different feed allowance per cow per year.**

Season	GE	NZ70			NZ90				OS90				Sed	Significance		
	FA	1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
02-03	Early lactation <sup>1</sup>	31.1	36.3	33.2	37.8	34.3	35.5	32.1	30.7	28.9	35.1	31.3	1.7	***	***	***
	Mid lactation	29.9	28.2	33.3	33.5	35.8	32.9	34.2	30.7	33.9	26.7	33.9	1.5	***	***	*
	Late lactation	28.0	27.4	26.4	30.7	33.6	33.8	33.9	29.2	33.2	31.8	31.6	1.8	***	**	NS
03-04	Dry period	14.6	18.8	14.7	15.4	17.1	16.8	16.5	15.2	15.5	15.3	15.3	0.8	***	***	***
	Early lactation <sup>1</sup>	27.7	33.2	30.9	38.1	38.3	35.7	33.9	33.5	33.3	31.2	27.9	2.0	***	***	***
	Mid lactation	29.9	27.2	28.1	33.8	33.4	29.7	30.3	33.3	29.3	32.4	32.5	1.3	***	***	***
	Late lactation	29.4	27.2	30.2	32.5	27.0	30.7	31.5	28.4	31.0	30.8	33.7	1.6	***	***	***

Daily total dry matter intake per unit of live weight (g kg<sup>-1</sup>LW). <sup>(1)</sup>Only pasture was fed in early lactation. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values are indicated when approaching significance.

**Appendix V- 11: Mean daily yield per cow and milk composition of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04. The genotypes were farmed in different systems at different feed allowance per cow per year.**

	NZ70			NZ90				OS90				Sed	Significance			
	1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA	
Early	Milk (litres cow <sup>-1</sup> )	19.9	23.3	21.6	25.6	24.6	24.7	25.5	25.6	26.6	27.3	26.4	1.71	***	NS	NS
	MS (kg cow <sup>-1</sup> )	1.59	1.66	1.63	1.88	1.83	1.92	2.01	1.98	1.98	2.00	1.88	0.13	***	NS	NS
	Fat (kg cow <sup>-1</sup> )	0.93	0.89	0.89	1.01	0.99	1.04	1.07	1.08	1.06	1.08	0.99	0.09	***	NS	NS
	Protein (kg cow <sup>-1</sup> )	0.66	0.77	0.73	0.87	0.85	0.88	0.94	0.90	0.92	0.92	0.89	0.06	***	NS	NS
	Lactose (kg cow <sup>-1</sup> )	0.98	1.15	1.06	1.29	1.24	1.23	1.26	1.27	1.32	1.37	1.32	0.09	***	NS	0.144
	FC (g kg <sup>-1</sup> )	47.2	38.4	41.0	39.3	40.6	42.2	42.1	42.0	40.2	39.4	37.4	2.54	NS	NS	*
	PC (g kg <sup>-1</sup> )	33.5	32.9	34.1	33.9	34.6	35.7	36.7	35.2	34.7	33.7	34.1	1.00	***	NS	0.157
LC (g kg <sup>-1</sup> )	49.4	49.5	49.2	50.6	50.4	49.7	49.3	49.7	49.7	49.9	49.9	0.61	*	NS	0.163	
Mid	Milk (litres cow <sup>-1</sup> )	15.5	17.3	16.9	19.2	20.0	17.1	19.5	19.4	18.1	18.1	22.5	1.46	***	**	***
	MS (kg cow <sup>-1</sup> )	1.19	1.26	1.33	1.56	1.56	1.40	1.54	1.50	1.33	1.31	1.63	0.09	***	***	***
	Fat (kg cow <sup>-1</sup> )	0.68	0.72	0.76	0.87	0.89	0.80	0.82	0.81	0.73	0.70	0.86	0.06	***	*	**
	Protein (kg cow <sup>-1</sup> )	0.51	0.55	0.58	0.68	0.67	0.60	0.71	0.69	0.60	0.61	0.77	0.04	***	***	***
	Lactose (kg cow <sup>-1</sup> )	0.72	0.81	0.79	0.90	0.95	0.80	0.93	0.90	0.84	0.85	1.06	0.07	***	**	***
	FC (g kg <sup>-1</sup> )	44.8	41.7	44.7	45.7	44.9	47.1	42.8	42.4	40.8	39.1	38.3	2.27	***	0.06	NS
	PC (g kg <sup>-1</sup> )	33.3	31.9	34.3	35.8	34.2	35.6	36.7	35.4	33.4	33.9	34.6	1.14	***	**	NS
LC (g kg <sup>-1</sup> )	46.8	46.8	46.4	46.7	47.2	46.9	47.9	46.2	46.1	47.1	47.0	0.47	*	0.11	*	
Late	Milk (litres cow <sup>-1</sup> )	10.0	12.6	11.4	12.6	12.4	13.6	15.7	13.4	13.2	16.0	13.9	1.63	***	**	0.077
	MS (kg cow <sup>-1</sup> )	0.91	1.07	0.94	1.15	1.08	1.32	1.49	1.12	1.18	1.32	1.16	0.14	***	***	0.057
	Fat (kg cow <sup>-1</sup> )	0.56	0.65	0.55	0.67	0.63	0.78	0.86	0.63	0.67	0.74	0.65	0.09	***	**	0.0158
	Protein (kg cow <sup>-1</sup> )	0.36	0.43	0.38	0.48	0.45	0.55	0.64	0.49	0.51	0.58	0.51	0.05	***	**	*
	Lactose (kg cow <sup>-1</sup> )	0.49	0.60	0.53	0.60	0.60	0.64	0.76	0.63	0.62	0.77	0.66	0.08	***	**	*
	FC (g kg <sup>-1</sup> )	55.7	52.1	49.4	52.3	51.2	57.7	54.7	48.2	51.2	45.9	46.8	3.55	***	NS	0.056
	PC (g kg <sup>-1</sup> )	36.1	34.3	34.3	37.9	36.3	40.5	40.6	36.7	39.3	36.4	37.1	0.14	***	NS	***
LC (g kg <sup>-1</sup> )	49.1	47.8	46.3	47.8	48.6	47.4	48.5	46.7	46.4	48.3	47.1	0.79	*	NS	***	

Means of all the lactating cows in each system at each stage of lactation are presented. FC: fat content; PC: protein content; LC: lactose content. GE: genotype; FA: annual feed allowance [increased from FA1 (lowest) to FA4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values indicated when approaching significance.

**Appendix V- 12: Mean daily yield per cow and milk composition of three Holstein-Friesian genotypes during early, mid and late lactation of seasons 2002-03 and 2003-04. The genotypes were farmed in different systems at different feed allowance per cow per year.**

	GE	NZ70			NZ90				OS90				Sed	Significance		
	FA	1	3	4	1	2	3	4	1	2	3	4		GE	FA	GE*FA
Early	Milk (litres cow <sup>-1</sup> )	19.1	23.7	23.5	23.9	28.9	25.4	26.9	25.0	28.4	28.6	31.3	1.77	***	***	0.149
	MS (kg cow <sup>-1</sup> )	1.52	1.84	1.86	1.86	2.34	2.11	2.21	1.97	2.00	2.21	2.24	0.14	***	***	0.055
	Fat (kg cow <sup>-1</sup> )	0.89	1.06	1.09	1.06	1.35	1.23	1.24	1.14	1.07	1.24	1.21	0.09	***	***	*
	Protein (kg cow <sup>-1</sup> )	0.62	0.79	0.76	0.80	0.99	0.88	0.98	0.83	0.94	0.97	1.03	0.06	***	***	NS
	Lactose (kg cow <sup>-1</sup> )	0.94	1.15	1.15	1.17	1.43	1.23	1.33	1.20	1.40	1.40	1.55	0.89	***	***	0.075
	FC (g kg <sup>-1</sup> )	47.7	44.7	46.0	45.3	46.6	49.3	45.8	45.6	38.1	43.7	39.3	0.26	***	0.07	*
	PC (g kg <sup>-1</sup> )	32.6	33.3	32.7	33.6	34.4	35.2	36.4	33.2	33.3	33.7	32.9	0.10	***	NS	NS
	LC (g kg <sup>-1</sup> )	49.2	48.8	49.2	48.9	49.6	48.7	49.3	47.9	49.3	48.8	49.3	0.06	NS	*	NS
Mid	Milk (litres cow <sup>-1</sup> )	15.5	20.4	18.1	19.1	19.2	20.5	21.2	21.9	21.0	23.2	25.9	1.48	***	***	**
	MS (kg cow <sup>-1</sup> )	1.25	1.44	1.40	1.45	1.49	1.68	1.70	1.61	1.44	1.59	1.76	0.10	***	**	**
	Fat (kg cow <sup>-1</sup> )	0.73	0.80	0.75	0.78	0.83	0.93	0.91	0.87	0.76	0.84	0.90	0.06	***	0.11	*
	Protein (kg cow <sup>-1</sup> )	0.51	0.64	0.59	0.67	0.66	0.75	0.79	0.74	0.67	0.75	0.86	0.04	***	***	**
	Lactose (kg cow <sup>-1</sup> )	0.74	0.95	0.87	0.90	0.91	0.96	1.01	1.01	0.99	1.10	1.21	0.07	***	***	*
	FC (g kg <sup>-1</sup> )	47.5	40.7	42.0	41.9	43.9	45.8	43.1	39.9	36.9	35.9	35.1	0.24	***	*	*
	PC (g kg <sup>-1</sup> )	33.3	31.8	32.9	35.2	34.3	36.9	37.3	33.7	32.4	32.4	33.2	0.08	***	*	*
	LC (g kg <sup>-1</sup> )	47.7	46.9	48.0	47.3	47.3	47.2	47.8	46.2	47.3	47.4	46.9	0.06	NS	NS	NS
Late	Milk (litres cow <sup>-1</sup> )	14.9	14.4	15.0	14.3	13.6	15.6	17.7	19.3	18.4	20.2	18.5	1.93	***	*	NS
	MS (kg cow <sup>-1</sup> )	1.36	1.25	1.33	1.33	1.29	1.47	1.75	1.55	1.48	1.69	1.48	0.16	***	**	0.054
	Fat (kg cow <sup>-1</sup> )	0.83	0.74	0.79	0.76	0.77	0.83	0.99	0.84	0.80	0.94	0.82	0.10	NS	*	NS
	Protein (kg cow <sup>-1</sup> )	0.53	0.50	0.54	0.57	0.52	0.64	0.76	0.71	0.69	0.75	0.67	0.06	***	***	**
	Lactose (kg cow <sup>-1</sup> )	0.74	0.68	0.71	0.69	0.66	0.74	0.85	0.92	0.88	0.97	0.89	0.09	***	*	NS
	FC (g kg <sup>-1</sup> )	55.9	52.6	53.4	53.5	57.2	54.1	56.3	44.2	43.9	46.4	44.3	0.36	***	NS	NS
	PC (g kg <sup>-1</sup> )	36.0	35.6	36.5	40.2	38.4	41.3	42.8	37.2	37.8	37.3	36.3	0.15	***	*	*
	LC (g kg <sup>-1</sup> )	49.2	47.8	47.4	47.9	48.6	47.6	47.8	47.5	47.8	48.2	48.4	0.10	NS	NS	0.173

Means of all the lactating cows in each system at each stage of lactation are presented. FC: fat content; PC: protein content; LC: lactose content. GE: genotype; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]. GE\*FA: genotype by feed allowance interaction. Significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = non significant. Probability values indicated when approaching significance.

**Appendix V- 13: Mean live weight at different dates during the season and days from calving to live weight nadir for three Holstein-Friesian genotypes, age group and feed allowance in each season of the ‘system’ study. The genotypes were farmed in different systems at different feed allowance per cow per year.**

	Genotype				Age				Feed Allowance					Significance				
	NZ70	NZ90	OS90	Sed	2Y	3Y	4Y	Sed	1	2	3	4	Sed	GE	A	FA	GE*FA	
Season 2001 – 2002	LW Start season	432	453	475	7.63	454	----	----	----	448	456	455	456	8.45	***	----	NS	NS
	LW Calving	440	464	486	8.78	463	----	----	----	456	467	463	467	9.80	***	----	NS	NS
	LW 4 <sup>th</sup> Week PC	403	417	427	8.21	416	----	----	----	411	420	417	414	9.10	*	----	NS	NS
	LW Nadir	397	401	412	7.67	404	----	----	----	401	409	403	401	8.57	0.09	----	NS	NS
	LW Dry off	447	451	454	7.83	451	----	----	----	444	450	454	454	8.68	NS	----	NS	NS
	LW End season	463	463	484	8.75	470	----	----	----	461	476	468	476	9.71	**	----	NS	NS
Days Calving to Nadir	48.8	72.3	78.6	9.42	66.6	----	----	----	62.9	67.3	68.8	67.2	10.4	**	----	NS	NS	
Season 2002 – 2003	LW Start season	468	480	498	7.47	479	485	----	6.24	481	482	477	488	8.35	***	NS	NS	NS
	LW Calving	434	465	487	9.07	441	482	----	7.58	460	461	460	466	10.1	***	***	NS	NS
	LW 4 <sup>th</sup> Week PC	402	420	439	8.01	398	443	----	6.69	414	423	419	425	8.96	***	***	NS	NS
	LW Nadir	391	408	427	7.34	387	431	----	6.13	401	408	412	413	8.21	***	***	NS	NS
	LW Dry off	475	485	481	9.59	456	504	----	8.01	468	461	485	506	10.7	NS	***	***	NS
	LW End season	482	498	520	10.4	481	520	----	8.70	491	486	503	522	11.5	***	***	**	NS
Days Calving to Nadir	48.4	51.3	58.7	9.32	48.5	57.1	----	7.78	54.6	64.1	39.4	53.2	10.3	NS	NS	NS	NS	
Season 2003 – 2004	LW Start season	484	522	560	9.09	503	512	552	10.2	516	512	528	534	10.2	***	***	0.07	NS
	LW Calving	488	522	565	9.20	494	515	565	10.3	510	518	533	539	10.3	***	***	**	NS
	LW 4 <sup>th</sup> Week PC	440	461	496	8.90	438	458	502	10.0	454	462	474	474	10.0	***	***	0.05	NS
	LW Nadir	424	444	473	8.22	417	443	481	9.27	436	442	457	454	9.29	***	***	*	NS
	LW Dry off	493	525	535	9.67	495	513	544	10.9	509	497	533	532	10.9	***	***	**	NS
	LW End season	498	536	553	10.1	504	527	557	11.4	527	520	536	534	11.4	***	***	NS	NS
Days Calving to Nadir	59.3	62.7	72.6	8.50	61.6	63.2	69.8	9.58	58.5	78.6	65.0	57.4	9.60	NS	NS	NS	NS	

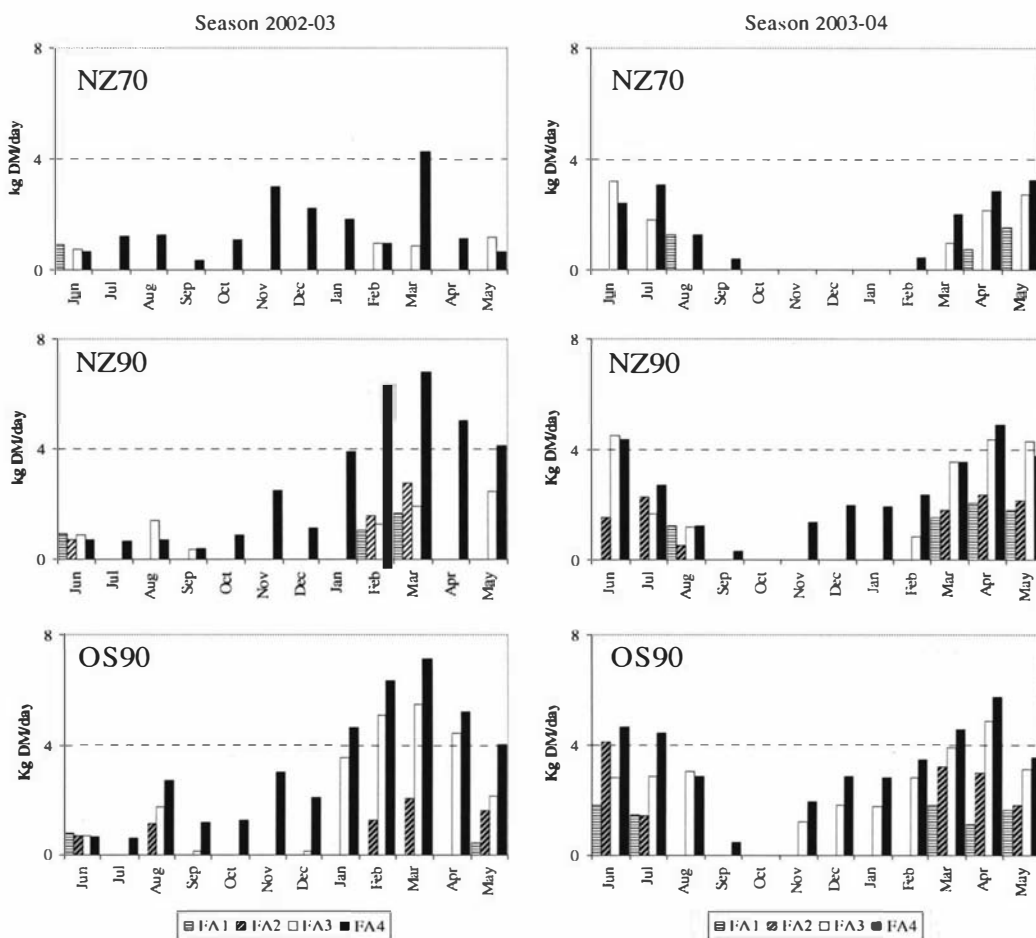
LW: live weight (Kg cow<sup>-1</sup>). GE: genotype; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values indicated when approaching significance.

**Appendix V- 14: Mean body condition score at different dates during the season and days from calving to body condition score nadir for three Holstein-Friesian genotypes, age group and feed allowance in each season of the 'system' study.**

	Genotype				Age				Feed allowance					Significance				
	NZ70	NZ90	OS90	Sed	2Y	3Y	4Y	Sed	1	2	3	4	Sed	GE	A	FA	GE*FA	
Season 2001 – 2002	BCS Start season	5.84	5.74	5.45	0.07	5.67	----	----	----	5.57	5.82	5.61	5.69	0.09	***	----	*	NS
	BCS Calving	5.10	5.04	4.98	0.09	5.01	----	----	----	5.01	5.09	4.97	4.97	0.09	*	----	NS	NS
	BCS 4 <sup>th</sup> Week PC	4.65	4.41	4.04	0.10	4.36	----	----	----	4.39	4.43	4.33	4.33	0.11	***	----	NS	NS
	BCS Nadir	4.33	4.01	3.55	0.09	3.96	----	----	----	3.98	4.04	3.93	3.89	0.10	***	----	NS	NS
	BCS Dry off	4.62	4.27	3.88	0.10	4.25	----	----	----	4.26	4.25	4.25	4.26	0.11	***	----	NS	NS
	BCS End season	4.57	4.36	4.27	0.09	4.40	----	----	----	4.38	4.40	4.38	4.44	0.10	**	----	NS	0.070
	Days Calving to BCS	69.0	96.2	108	18.5	91.2	----	----	----	102	99.1	77.6	85.7	20.5	0.10	----	NS	NS
Season 2002 – 2003	BCS Start season	5.98	5.58	5.44	0.09	6.51	4.83	----	0.08	5.70	5.78	5.53	5.66	0.11	***	***	0.12	NS
	BCS Calving	5.33	5.28	5.16	0.07	5.73	4.78	----	0.06	5.23	5.35	5.21	5.25	0.08	*	***	NS	NS
	BCS 4 <sup>th</sup> Week PC	4.98	4.89	4.64	0.07	5.20	4.47	----	0.06	4.78	4.92	4.79	4.85	0.07	***	***	NS	NS
	BCS Nadir	4.33	4.06	3.61	0.07	4.09	3.91	----	0.06	3.97	3.96	3.98	4.08	0.08	***	**	NS	NS
	BCS Dry off	4.95	4.46	3.81	0.12	4.32	4.46	----	0.10	4.29	4.18	4.43	4.74	0.13	***	NS	***	0.110
	Days Calving to BCS	118	137	165	13.2	175	104	----	11.0	137	168	130	124	14.8	**	***	*	NS
Season 2003 – 2004	BCS Start season	5.24	5.05	4.96	0.08	5.52	4.84	4.89	0.09	5.02	5.01	5.08	5.23	0.09	**	***	*	NS
	BCS Calving	5.17	5.04	5.04	0.09	5.42	4.81	5.03	0.10	4.91	4.99	5.15	5.28	0.10	NS	***	***	NS
	BCS 4 <sup>th</sup> Week PC	4.64	4.47	4.26	0.09	4.73	4.22	4.41	0.11	4.36	4.36	4.52	4.58	0.10	***	***	0.05	NS
	BCS Dry off	4.74	4.58	3.72	0.14	4.34	4.36	4.33	0.15	4.34	3.98	4.40	4.65	0.15	***	NS	***	0.100
	BCS End season	4.83	4.71	3.99	0.15	4.49	4.52	4.51	0.16	4.54	4.39	4.45	4.66	0.16	***	NS	NS	0.107

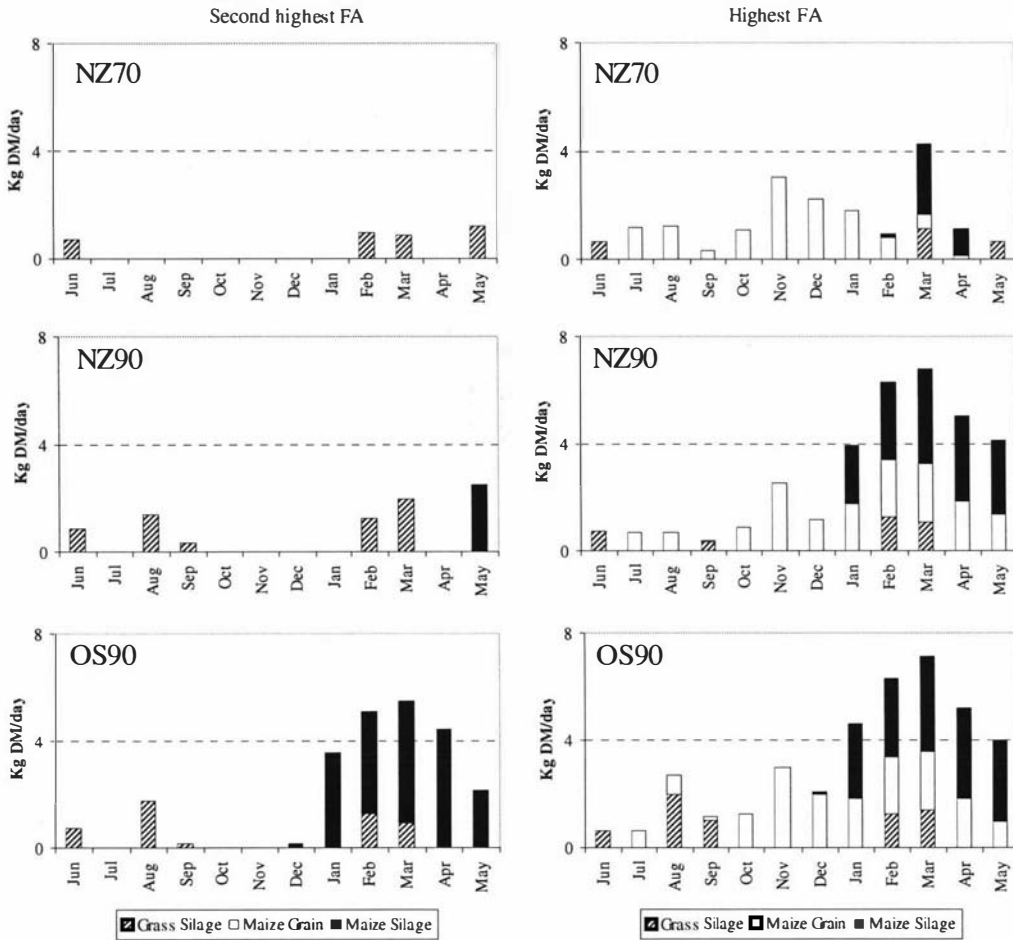
BCS: body condition score. GE: genotype; FA: annual feed allowance [increased from 1 (lowest) to 4 (highest)]; GE\*FA: genotype by feed allowance interaction. Significance: \*=P<0.05; \*\*=P<0.01; \*\*\*=P<0.001. NS= non significant. Probability values indicated when approaching significance.

**Appendix V- 15: Mean daily supplement offered per month (forage, maize silage or maize grain) to systems farmed with three Holstein-Friesian genotypes different feed allowance during season 2002-03 and 2003-04.**



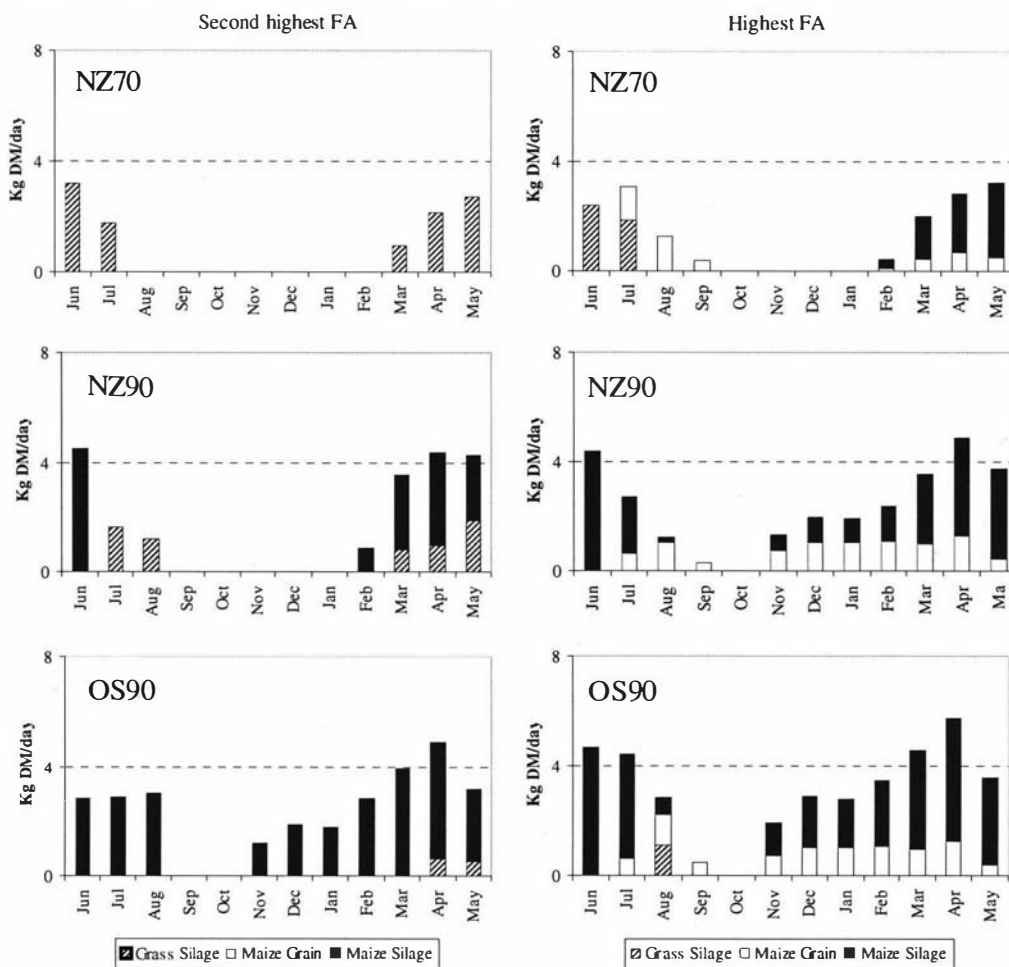
Supplement expressed in  $\text{kg DM cow}^{-1}\text{day}^{-1}$ . Systems farmed at increased feeding allowance (FA) for each genotype (FA1: lowest; FA4: highest). The total amount of supplements fed each month was considered. Supplement lost during feeding was estimated as 20 and 10% respectively for grass silage and maize silage (Grass silage was fed on the soil and maize silage on troughs in the paddocks).

**Appendix V- 16: Mean daily supplement offered per month (forage, maize silage or maize grain) to systems farmed with three different Holstein-Friesian genotypes at the second highest and highest annual feed allowance in season 2002-03.**



Supplement expressed in kg DM cow<sup>-1</sup>day<sup>-1</sup>. Systems farmed at the highest annual feed allowance (FA) for each genotype (FA3: second highest; FA4: highest). The total amount of supplements fed each month was considered. Supplement lost during feeding was estimated as 20 and 10% respectively for grass silage and maize silage (Grass silage was fed on the soil and maize silage on troughs in the paddocks).

**Appendix V- 17: Mean daily supplement offered per month (forage, maize silage or maize grain) to systems farmed with three different Holstein-Friesian genotypes at the second highest and highest annual feed allowance in season 2003-04.**



Supplement expressed in kg DM cow<sup>-1</sup>day<sup>-1</sup>. Systems farmed at the highest annual feed allowance (FA) for each genotype (FA3: second highest; FA4: highest). The total amount of supplements fed each month was considered. Supplement lost during feeding was estimated as 20 and 10% respectively for grass silage and maize silage (Grass silage was fed on the soil and maize silage on troughs in the paddocks).

## SECTION IV

### CHAPTER 8

# HERBAGE INTAKE AND PRODUCTIVE PERFORMANCE OF THREE HOLSTEIN-FRIESIAN GENOTYPES GRAZING UNDER CONTROLLED SWARD CONDITIONS

## 8.1. INTRODUCTION

It was determined that modern Holstein-Friesian cows from overseas (OS) had greater potential for yield than high yielding Holstein-Friesian from New Zealand (NZ) if fed on a total mixed ration (TMR)(Kolver *et al.*, 2002). However results of the grazing systems study (see Chapters 5 to 7) confirmed that the modern New Zealand (NZ) strain produced more milksolids (MS) per cow and per hectare on pasture-based systems than the OS genotype, in agreement with Kolver's results, when both strains were fed on pasture. In this study, the modern NZ cow showed a lower decline of body reserves (BCS) postpartum and during the whole lactation, than the high yielding OS cow (see Chapter 5). Results from this trial showed that daily herbage dry matter intake ( $DMI_H$ ) was higher for the modern NZ than for the OS during the lactation period in agreement with the lower decline in BCS mentioned (see Table 6.6 in Chapter 6 and Figures 5.6b and 5.7b in Chapter 5). This indicates that higher proportion of the yield obtained was sustained by the relatively higher herbage intake measured for the modern NZ (see Chapter 6) and showed that intake per unit of live weight (LW) was affected to a greater extent in OS than NZ cows under constraining sward conditions, probably as a result of lower flexibility in the OS90 genotype to adjust grazing behaviour to the changes in structure that occur in the pasture during the grazing down process (see Chapter 7).

The 'system' comparison provided some opportunities to compare intake and grazing behaviour of three different Holstein-Friesian genotypes under contrasting feeding conditions. However, this comparison involved substantial interactions between pasture

characteristics and their confounded effects on intake as a result of differences in pasture management between the systems, and also the effect of the previous management on performance per cow. This made it difficult to be conclusive about differences observed in relation to the grazing capacity of the genotypes.

The opportunity for cows to compensate for changes in the sward by modifying grazing behaviour may be reduced in short swards where they are unable to eat enough even if the area offered is large, hence grazing would cease once the available leaf material is exhausted. This may occur earlier for those cows grazing at a faster rate. However, cows with higher  $DMI_H$  capacity would continue grazing in the face of limiting sward conditions, and then increasing herbage intake despite the reduced size of the bite harvested. These cows would increase production due to the increased  $DMI_H$ ; hence they would improve performance in pasture-based systems. It is hypothesised that cows would improve the amount of herbage consumed from the pasture if the availability of the herbage grazed improves due to the reduction in sward constraints to bite penetration. In addition, it is expected that because the genetic selection of the modern NZ cows occurred on pasture, this genotype would have developed greater capacity to compensate changes in the structure of the sward that may constrain  $DMI_H$  than OS cows selected for a different type of feeding system, like TMR.

The objective of this study was to investigate the  $DMI_H$  achieved by three different genotypes of Holstein-Friesian dairy cows grazing pastures of different structure at similar daily herbage allowance during the early lactation period. This chapter reports the results of a controlled experiment in which cows of the three different genotypes utilised in the system study (see Section III) were exposed to contrasting sward conditions in a controlled grazing experiment.

## **8.2. MATERIALS AND METHODS**

This experiment was conducted at Ruakura No. 1 Dairy, in Hamilton, New Zealand, from early September to early November 2004, using cows from the same genotypes utilised in the previous study (see Section III and Appendix IV for details).

Sixteen cows of three different genotypes of Holstein-Friesian dairy cows between three and five years old (second to fourth parity), all early lactating animals, were utilised (n=48). Briefly the genotypes were a high breeding worth (BW) Holstein-Friesian of OS (OS90) or NZ (NZ90) origin, and a low BW NZ Friesian genotype representing the cow used in NZ during the 1970s (NZ70), which had different percentage of North American genes according to their ancestry records (7% for NZ70, 24% for NZ90 and 91% for

OS90; see Appendix IV for more details about the characteristics of the genotypes utilised). Cows were allocated to two different sward conditions (SC) determined by their pre-grazing herbage mass and height above ground level ( $HM_{PRE}$  and  $SH_{PRE}$  respectively) and defined as SHORT or TALL swards with a pre-grazing herbage mass of 2,100 and 2,800 kg DM ha<sup>-1</sup> above ground level respectively, at a common daily herbage allowance (DHA), equal to 35 kg DM cow<sup>-1</sup>.

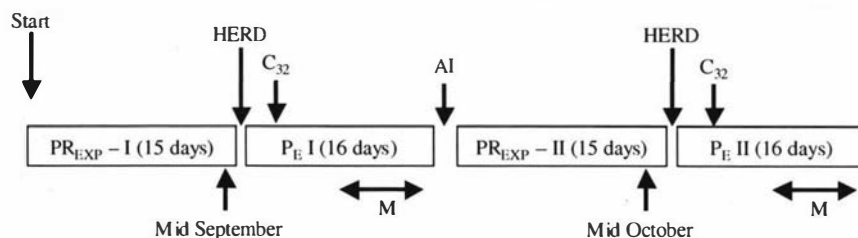
The target DHA was determined considering the mean herbage dry matter intake ( $DMI_H$ ) achieved by cows of high genetic potential from NZ and OS grazing pasture in Kolver et al. (2002), and accounting for a grazing efficiency of 0.50 to maximise intake (Combellas & Hodgson, 1979). To achieve similar DHA in both treatments, a larger daily area per cow was allocated to herds grazing the SHORT sward.

Paddocks used in this experiment were in one area of the farm with similar soil type and pasture composition (L'Huillier, 1987), mainly *Lolium perenne* L. (ryegrass) and *Trifolium repens* L. (white clover). Paddocks in this area were allocated to the treatments ensuring that soil type, pasture condition and distance to the dairy shed were balanced between treatments. The total area and number of paddocks of the farm allocated to the experiment was greater than those finally utilised as some paddocks did not reach the SC required by each treatment. A Rising Plate Meter (RPM) and a sward stick were utilised to measure SC pre- and post-grazing as explained in Chapter 6 and the equation utilised to transform the compressed height measurements obtained with the RPM from each paddock into dry matter values per hectare, was that estimated for early lactation (see Table 6.1 in Chapter 6). Before the start of the experiment, these cows had grazed together as part of a commercial herd, which grazed sequentially all the paddocks allocated to the experiment. Grazing management targeted a mean  $HM_{PRE}$  and  $HM_{POST}$  of 2,500 and 1,500 kg DM ha<sup>-1</sup> respectively and cows were fed 4 kg cow<sup>-1</sup>day<sup>-1</sup> of maize silage.

Eight cows from each genotype were allocated to each of two herds balanced by parity, LW and body condition score (BCS), calving date and pre-experimental MS yield. One herd from each genotype was allocated to the TALL sward and the other to the SHORT sward; hence three herds with cows of different genotypes were managed at each SC during two periods ( $P_E$  -I and  $P_E$  -II, four weeks each), the first in September and the second in November 2004 as is described in Figure 8.1, at a common DHA (35 kg DM cow<sup>-1</sup>day<sup>-1</sup>). Each period consisted of a training period of 15 days ( $PR_{EXP}$ ) where the cows became accustomed to the SC imposed and a sampling period of 16 days ( $SA_{EXP}$ ). Period I and II were separated by a two week period where cows grazed together again and mated to artificial insemination, but were not supplemented. Herds allocated to the

TALL sward in P<sub>E</sub>-I were allocated to the SHORT sward in P<sub>E</sub>-II and vice versa, in a crossover design and each period was considered a replicate for data analysis.

**Figure 8.1: Description of events during each consecutive experimental period during the study.**



HERD: treatments group formation; C32: starting of n-alkane dosage during each experimental period; AI: artificial insemination; PR<sub>EXP</sub>: pre-experimental period; M: measurement period.

After the paddocks had been grazed with the commercial herd during the pre-experimental period, the total herbage mass present in each paddock was measured every two days with a RPM. The mean herbage accumulation rate (HAR) for spring, calculated for the system study (see Chapter 5), was used to estimate the expected grazing date of each paddock according to the SC to which it had been allocated. Paddocks assigned to the TALL or SHORT swards were only grazed if the actual herbage mass was within  $\pm 100 \text{ kg DM ha}^{-1}$  of target by the day before the expected grazing date. Paddocks allocated to the SHORT sward that had more herbage mass than the target were re-allocated to the TALL treatment and grazed once the new target was achieved. A commercial herd grazed all the paddocks allocated to the TALL sward but that achieved more HM than the target for this treatment.

On the day before grazing the appropriate  $HM_{PRE}$  for grazing the SHORT and TALL was measured, the paddocks for each treatment were selected, and the mean grazing area in each treatment calculated. The size of each paddock was 1 ha and the total area grazed by the three herds was  $0.41 \text{ ha day}^{-1}$  for the SHORT sward and  $0.29 \text{ ha day}^{-1}$  for the TALL. The herds spent two and three days in each paddock, with the remaining area being grazed by a herd of dry cows. Within paddocks, one third of the specified area was allocated to each of the genotypes, which were separated by electric fences, with water available. The mass and height of the herbage pre- and post-grazing of the paddocks were measured in each of the strips. Genotypes were allocated at random to each of these strips to minimize the effect of differences in herbage composition between strips and paddocks and also variations in SC within the paddock. Back grazing was avoided by using an electric wire to separate the area actually being grazed from that grazed on the previous day.

Live weight, BCS and milk samples were obtained every week during each  $P_E$  in September and November. Pasture, faecal and blood samples and grazing behaviour were concentrated in the last eight days of each sampling period ( $P_E$ ). Period I was coincident with the start of mating and all cows were treated with a CIDR<sup>®</sup> inserted for the duration of the measurements to avoid oestrus affecting the estimation of  $DMI_H$  and the observations of grazing behaviour. Once the first period of measurements was completed CIDRs<sup>®</sup> were removed and all the cows inseminated two days later, when on heat.

As shown in Figure 8.1, in each  $PR_{EXP}$  period the cows were managed as one mob (within a commercial herd) until they were allocated to each treatment herd (HERD) and the two measurements periods ( $P_{E-I}$  and  $P_{E-II}$ ) of a length of 16 days each started. The start of  $C_{32}$ -alkane dosage indicate the initiation of the n-alkanes procedure used for determination of the herbage intake; in addition, the associated sampling and other measurements (M) occurred during the second part of  $P_{E-I}$  and  $P_{E-II}$  (mainly collection of pasture and faecal samples, milk and blood samples, LW and BCS determination and the observation of grazing behaviour). Cows grazing the SHORT sward in  $P_{E-I}$  were relocated to graze the TALL sward in  $P_{E-II}$  and vice versa.

It was expected that cows grazing the TALL sward would leave greater post-grazing HM than cows grazing the SHORT sward. This would affect  $HM_{PRE}$  and  $SH_{PRE}$  at the next grazing. To reduce this effect all the paddocks allocated to the TALL sward were grazed with a herd of dry cows that followed immediately after the experimental herds. These dry cows achieved a mean  $HM_{POST}$  similar to that obtained in paddocks of the SHORT sward treatment (approximately 1,500 kg DM ha<sup>-1</sup>). Cows were not supplemented during the experiment and were milked twice daily. A new strip of pasture was offered to each herd after the morning milking and cows returned to the same area after the afternoon milking.

### 8.2.1. Pasture and animal measurements

Herbage dry matter intake ( $DMI_H$ ) and in-vivo diet dry matter digestibility (DMD) were estimated for each individual cow of the three genotypes during both experimental periods using the n-alkanes technique (Dove & Mayes, 1991), as was explained in Chapter 4. Intake was expressed per cow and per LW unit ( $DMI_H/LW$ ).

A sample of the herbage grazed was obtained, in both measurement periods, from the pre-grazing stratum of each pasture strip grazed daily. These samples were bulked over each period, oven dried (at 60°C for 48 hours) and analysed by Near Infrared Reflectance

Spectroscopy (NIRS)(Shenk & Westerhaus, 1994) and represented the quality of the herbage offered to the cows grazing each SC.

Grazing behaviour was visually observed during two 24-hour periods, one in each experimental period ( $P_E$  I and  $P_E$  II) to coincide with  $DMI_H$  determination. The grazing behaviour of all the cows while in the paddock was recorded every 10 minutes, as explained in Chapter 7. Behaviours observed were grazing (G), ruminating (R; while lying or standing) and lying or standing not ruminating. Each observation was considered to represent the behaviour of the cow during the previous 10 minute interval, thus the time (T) the cow spent grazing and ruminating during the 24 hour period was calculated as the sum of each particular behaviour over the 24 hour period and the total grazing activity of the cow as the sum of  $G_T$  plus  $R_T$ . Rumination was only observed while the cows were in the paddock and competing with grazing activity, but was not observed for cows on their way to and from the milking shed, in the yard or during each milking. The rate of herbage consumption ( $I_R$ ) was estimated as the relation between  $DMI_H$  and  $G_T$ .

Cows were milked twice a day during the experiment and the amount of milk produced by each cow measured once a week similarly to that explained in Chapter 5. Cows were weighed (LW) and condition scored (BCS) weekly by the same trained observers (Macdonald & Roche, 2004), as in the system study (see Chapter 5). To reduce the bias in the estimation of LW due to differences in the gut fill of cows grazing different sward conditions, the empty body weight was estimated [ $LW_E = LW - 4 DMI_H$ ; (Tamminga *et al.*, 1997)]. If the different sward grazed affects the  $DMI_H$  during early lactation, cows achieving lower  $DMI_H$  would mobilise more body fat reserves to sustain the raise in daily milk yield occurring after calving (see Figure 5.8 in Chapter 5), and as a result, BCS declines (see Figures 5.6 and 5.7 in Chapter 5). Changes in BCS are difficult to measure in such a short period however, it was expected that changes in blood metabolites were more sensible to changes in the differences in cow metabolism expected for cows under different nutritional status. Because this, one blood sample from each cow was taken by tail-vein using a 10 ml evacuated plain grass tube per sample, at the end of each sampling period. After being allowed to clot at room temperature for 60–90 minutes, these blood samples were centrifuged at 2,800 rpm for 15 minutes and aspirated serum was immediately assayed for non-esterified fatty acids (NEFA) and glucose (GLU)(Slein, 1963; Matsubara *et al.*, 1983).

The mean energy required for maintenance ( $En_M$ ; including an activity allowance) and milk yield ( $En_O$ ) was calculated from the  $LW_E$ , daily milk yield and its solids content, of each cow for each measurement period by following the same procedure described in

Chapter 6. The energy balance ( $EN_{BAL}$ ) was estimated from the difference between the mean energy consumed and required.

The breadth of the incisor arcade or size of the jaw ( $J_z$ ), defined as the distance between the outer edges of the canines on the right and left ramus of an animal's jaw (Gordon & Illius, 1988), was also measured for each individual cow with a calliper, once at the end of the experiment. If it is considered a circular shape to estimate bite area, differences in the breadth of the incisor arcade would determine different in bite area for the genotypes.

### 8.2.2. Statistical analysis

Treatments were arranged in a factorial design with three genotypes (GE) and two sward conditions (SC), replicated in time with a crossover arrangement of herds between SC among  $P_E$ . The effect of GE, SC and the GE by SC interaction were analysed using the statistical procedures of SAS (SAS, 2002) as a mixed model (PROC MIXED)  $Y_{ijk} = \mu + GE_i + SC_j + P_{Ek} + (GE \times SC) + e_{ijk}$  with GE, HM,  $P_E$  and the GE by SC interaction as fixed effects and cow within GE as random effects. The breadth of the incisor arcade was analysed by using the model  $Y_i = \mu + GE_i + e_i$  with GE as a fixed effect and cow as random effect. A simple linear correlation analysis (PROC CORR) was used to show the degree of association between  $EN_{BAL}$  with NEFA and GLU.

## 8.3. RESULTS

### 8.3.1. Sward conditions

Both  $HM_{PRE}$  and  $SH_{PRE}$  were similar across genotypes but different between SC (Table 8.1). As expected, the TALL sward had greater post-grazing herbage mass (+276 kg ha<sup>-1</sup>) and was taller (+16.4 mm) than the SHORT sward. Post-grazing residual ( $HM_{POST}$  and  $SH_{POST}$ ) tended to be lower for OS90 than for both NZ genotypes (Table 8.2).

The composition of the herbage offered was similar across genotypes (Table 8.1). The SHORT sward had greater in-vitro DM digestibility (+18 mg kg<sup>-1</sup>) and crude protein (+34 mg kg<sup>-1</sup>), lower soluble carbohydrates (-34 mg kg<sup>-1</sup>) and ADF (-8 mg kg<sup>-1</sup>) and tended to have a slightly greater ME value (+2.6%) and NDF (+1.6), although the latter two differences were not significant (Table 8.1).

The TALL sward was almost 70 mm taller and had about 900 kg DM ha<sup>-1</sup> more herbage accumulated at grazing than the SHORT sward, consequently a smaller area was offered per cow on the TALL sward (118 m<sup>2</sup> cow<sup>-1</sup> day<sup>-1</sup>) than on the SHORT sward (169 m<sup>2</sup> cow<sup>-1</sup> day<sup>-1</sup>).

<sup>1</sup> day<sup>-1</sup>) in order to offer a common DHA (35 kg DM cow<sup>-1</sup>day<sup>-1</sup>). Despite the differences in herbage mass and height, the bulk density of the two swards was similar (133 kg DM ha<sup>-1</sup>cm<sup>-1</sup>).

### 8.3.2. Milk and milksolids production and milk composition

The overall daily milk yield achieved was not as high as expected for three and four year old cows in early lactation. This was attributed to the crossover design of the trial with cows passing through a short period of under-feeding when allocated to the SHORT treatment. It seems that cows of the NZ70 genotype were affected the most on the SHORT sward due to their lower capacity to mobilise body reserves; however, these cows showed the higher increase in MS yield when they grazed the TALL sward, followed by the NZ90 - in contrast, no effect was observed for OS90 cows (Table 8.3).

**Table 8.1: Management and characteristics of the pre-grazing sward conditions of the two types of pastures grazed by different Holstein-Friesian genotypes during the experimental period.**

	SHORT			TALL			SED	Significance		
	NZ70	NZ90	OS90	NZ70	NZ90	OS90		GE	SC	GE*SC
<b>a) Pasture management and pre-grazing sward</b>										
HM <sub>PRE</sub> (kg DM ha <sup>-1</sup> )	2078	2058	2105	2918	3043	2966	41.2	NS	***	0.106
SH <sub>PRE</sub> (mm)	161	162	163	222	224	238	9.7	NS	***	NS
B <sub>D</sub> (kg DM ha <sup>-1</sup> cm <sup>-1</sup> )	131	129	129	132	137	139	4.66	NS	0.06	NS
AREA <sub>OIF</sub> (m <sup>2</sup> cow <sup>-1</sup> day <sup>-1</sup> )	169	171	167	121	116	119	2.22	NS	***	0.117
DHA/LW	78.1	77.4	69.0	75.1	74.3	66.5	----	----	----	----
Grazing efficiency	0.39	0.40	0.41	0.46	0.55	0.61	----	----	----	----
<b>b) Composition of herbage offered</b>										
DMD <sub>IN-VITRO</sub> (mg kg <sup>-1</sup> )	834	827	833	805	813	821	5.97	NS	**	NS
ME (MJ ME kg <sup>-1</sup> )	11.7	11.4	11.4	11.4	11.1	11.1	0.37	NS	NS	NS
Soluble C (g kg <sup>-1</sup> )	119	126	124	151	166	154	14.4	NS	**	NS
CP (g kg <sup>-1</sup> )	263	251	256	218	219	222	14.8	NS	**	NS
NDF (g kg <sup>-1</sup> )	440	449	445	441	438	435	9.87	NS	NS	NS
ADF (g kg <sup>-1</sup> )	215	217	214	223	221	226	1.65	NS	***	0.076

HM<sub>PRE</sub>: pre-grazing herbage mass; SH<sub>PRE</sub>: pre-grazing sward height; B<sub>D</sub>: bulk density; AREA<sub>OIF</sub>: area grazed daily; DHA/LW: daily herbage allowance per unit of actual LW; DMD<sub>IN-VITRO</sub>: in-vitro digestibility; ME: energy content in the herbage; SC: soluble carbohydrates; CP: crude protein; NDF: neutral detergent fibre; ADF: acid detergent fibre; GE: genotype, SC: sward condition, GE\*SC: GE by SC interaction. SED: maximum standard error of the differences of least squares means; NS: non-significant; \*P<0.05; \*\*P<0.01; \*\*\*P<0.001.

The highest yields of milk, protein and lactose, but not fat, were achieved by the OS90 genotype in both sward conditions. A trend for a greater MS yield was observed for the NZ90 strain, particularly the TALL sward. A non-significant effect of GE on MS yield was detected, although the NZ90 (1.89 kg day<sup>-1</sup>) showed a trend to produce more MS than both NZ70 (1.74 kg day<sup>-1</sup>) and OS90 (1.80 kg day<sup>-1</sup>; Table 8.3). The yield of lactose

showed a significant effect of GE, with the lowest lactose yield in NZ70 (1.08 kg day<sup>-1</sup>), intermediate in NZ90 (1.16 kg day<sup>-1</sup>) and the greatest in OS90 (1.30 kg day<sup>-1</sup>).

**Table 8.2: Post-grazing sward conditions of the two types of pastures grazed by different Holstein-Friesian genotypes during the experimental period**

	SHORT			TALL			SED	Significance		
	NZ70	NZ90	OS90	NZ70	NZ90	OS90		GE	SC	GE*SC
HM <sub>POST</sub> (kg DM ha <sup>-1</sup> )	1621	1499	1503	1846	1848	1756	63.0	0.15	***	NS
SH <sub>POST</sub> (mm)	66.8	58.5	60.3	79.2	82.1	73.3	3.3	0.10	***	NS

HM<sub>POST</sub>: post-grazing herbage mass; SH<sub>POST</sub>: post-grazing sward surface height; GE: genotype, SC: sward condition; GE\*SC: GE by SC interaction; SED: maximum standard error of the differences of least squares means; NS: non-significant; \*P<0.05; \*\*P<0.01; \*\*\*P<0.001.

**Table 8.3: Mean daily milk yield and milk components content of three genotypes of Holstein-Friesian dairy cows grazing *short* or *tall* swards in early lactation.**

		SHORT			TALL			SED	Significance		
		NZ70	NZ90	OS90	NZ70	NZ90	OS90		GE	SC	GE*SC
<b>a) Daily yield</b>											
Milk yield	Lt day <sup>-1</sup>	21.7	22.7	25.4	23.3	24.6	28.8	1.24	***	***	*
MS	Kg day <sup>-1</sup>	1.65	1.83	1.79	1.83	1.96	1.80	0.09	NS	**	NS
Fat	Kg day <sup>-1</sup>	0.97	1.07	0.98	1.08	1.12	0.88	0.07	*	NS	*
Protein	Kg day <sup>-1</sup>	0.68	0.76	0.81	0.75	0.84	0.92	0.03	***	***	**
Lactose	Kg day <sup>-1</sup>	1.04	1.12	1.21	1.12	1.20	1.39	0.05	***	***	NS
<b>b) Milk composition</b>											
Fat	g kg <sup>-1</sup>	44.5	47.4	39.1	46.3	46.3	30.4	2.84	***	0.050	***
Protein	g kg <sup>-1</sup>	31.3	33.9	31.7	32.1	34.3	32.0	0.84	**	*	NS
Lactose	g kg <sup>-1</sup>	48.3	49.2	47.8	48.0	48.8	48.3	0.72	NS	NS	*

MS: daily MS yield. GE: genotype, SC: Sward condition, GE\*SC: GE by SC interaction. SED: maximum standard error of the differences of least squares means; NS: non-significant; \*P<0.05; \*\*P<0.01; \*\*\*P<0.001. Dry matter intake, in-vivo digestibility and blood metabolites.

The highest yields of milk (+10%), MS (+6%), protein (+12%) and lactose (+11%) were observed on the TALL sward (Table 8.3); however, there was a significant GE by SC interaction for milk, fat and protein yield. On the TALL sward, milk and protein yield increased more for OS90 than for both NZ strains; whereas fat yield increased for NZ70 more than for NZ90, but declined for OS90. If yields of MS and lactose are considered together the differences previously mentioned for MS and lactose disappear. All three genotypes reduced yield on the SHORT sward. The difference in milk yield between SC was greater for OS90 (-11.8%) than NZ90 (-7.7%) or NZ70 (-6.9%), as were the differences in DMI<sub>H</sub>. The OS90 cows produced less protein (-11.9%) but increased fat yield (+11.4%); in contrast, the NZ90 produced less fat and protein (-4.5% and -9.5% respectively), which were reduced even to a greater extent for NZ70 (-10.2% and -9.3% respectively).

The contents of fat, protein and lactose in milk tended to be higher for the NZ90 genotype (Table 8.3). The protein content in milk was similar between the NZ70 and OS90 genotypes (31.8 g kg<sup>-1</sup>) and lower (-9%) than for the NZ90 (34.1 g kg<sup>-1</sup>). A significant GE by SC interaction for fat and lactose content in milk was measured, with the fat content in milk being higher on the TALL sward for NZ70 but lower for NZ90 and OS90, to a greater extent for OS90 cows; and lactose content was lower for the TALL sward for both NZ genotypes but higher for OS90. The lower fat content of OS90 cows on the TALL sward (30.4 g kg<sup>-1</sup>) compared to the SHORT sward (39.1 g kg<sup>-1</sup>) could be determined by the limited availability of rumen metabolites to increase fat yield to a similar extent than protein yield, combined with the effect of dilution in a higher milk yield. The differences observed for the yield of components between strains on the different sward grazed may be associated to some important metabolic characteristics of the genotypes.

**Table 8.4: Live weight, body condition score, pasture dry matter intake and grazing behaviour of three genotypes of Holstein-Friesian dairy cows grazing short or tall swards in early lactation.**

		SHORT			TALL			Significance			
		NZ70	NZ90	OS90	NZ70	NZ90	OS90	SED	GE	SC	GE*SC
<b>a) Liveweight and body condition score</b>											
LW	kg cow <sup>-1</sup>	448	452	507	466	471	526	10.39	***	***	NS
LW <sub>i</sub>	kg cow <sup>-1</sup>	392	396	449	401	394	441	9.69	***	NS	***
BCS		4.49	4.26	3.75	4.46	4.33	3.79	0.169	***	NS	NS
<b>b) Daily herbage intake and diet digestibility</b>											
DMI <sub>H</sub>	kg DM cow <sup>-1</sup>	13.63	14.06	14.48	16.19	19.20	21.59	0.71	***	***	***
DMI <sub>H</sub> LW	g DM kg <sup>-1</sup> LW	30.4	31.1	28.6	34.8	41.0	40.8	0.13	**	***	***
DMI <sub>H</sub> LWE	g DM kg <sup>-1</sup> LW	34.7	35.5	32.3	40.5	49.2	48.9	0.19	**	***	***
DMD	g kg <sup>-1</sup>	789	791	786	828	832	841	6.71	NS	***	0.121
<b>c) Blood metabolites</b>											
NEFA	mmol l <sup>-1</sup>	0.14	0.13	0.20	0.11	0.09	0.14	0.03	0.061	**	NS
Glucose	mmol l <sup>-1</sup>	3.43	3.21	3.12	3.38	3.36	3.29	0.08	**	0.049	0.049
<b>d) Grazing behaviour</b>											
G <sub>T</sub>	min day <sup>-1</sup>	566	572	517	444	465	473	17.4	NS	***	**
R <sub>T</sub>	min day <sup>-1</sup>	275	279	314	359	365	379	14.9	*	***	NS
G <sub>T</sub> + R <sub>T</sub>	min day <sup>-1</sup>	841	851	831	803	830	852	21.9	NS	NS	*
I <sub>R</sub>	g DM min <sup>-1</sup>	24.5	24.8	28.3	37.3	41.7	46.0	2.00	***	***	NS

LW: actual live weight; LW<sub>E</sub>: empty LW (Tamminga *et al.*, 1997); BCS: body condition score; DMI<sub>H</sub>: daily herbage dry matter intake; DMI<sub>LW</sub>: daily herbage dry matter intake per kg LW; DMD: dry matter digestibility; G<sub>T</sub>: grazing time; R<sub>T</sub>: ruminating time; I<sub>R</sub>: intake rate; GE: genotype; SC: sward condition; GE\*SC: GE by SC interaction; SED: maximum standard error of the differences of least squares means; NS: non-significant; \*P<0.05; \*\*P<0.01; \*\*\*P<0.001.

### 8.3.3. Dry matter intake and diet digestibility

The greatest DMI<sub>H</sub> was obtained by all the three genotypes on the TALL sward (Table 8.4). In addition, DMI<sub>H</sub> was lowest for the NZ70 and highest for the OS90 genotype for both SC, with a larger difference between genotypes for the TALL than on the SHORT

sward. The greatest  $DMI_H/LW$  was also observed for the three genotypes on the TALL sward (+29%; Table 8.4).

There was a significant GE by SC interaction for  $DMI_H$  and  $DMI_H/LW$  (Table 8.4). Daily  $DMI_H$  was similar across GE in the SHORT sward, but with a trend to be highest for OS90 and lowest for NZ70. For the TALL sward the OS90 had the highest  $DMI_H$  while NZ70 the lowest. In contrast, although NZ90 and OS90 had similar  $DMI_H/LW$  on the TALL sward and higher than observed for NZ70, the NZ90 had the highest  $DMI_H/LW$  on the SHORT sward, while the OS90 the lowest (-8.7%).

Diet DMD was similar across GE, but greater on the TALL sward (+6%; Table 8.4). In addition, the OS90 cows showed a superior DMD to both NZ genotypes when grazing the TALL sward, but on SHORT sward the OS90 showed a trend for a lower DMD (although not significant).

### 8.3.4. Live weight, body condition score, blood metabolites and breadth of the incisor arcade

There was a significant difference in LW across genotypes with the OS90 being heaviest and the NZ70 lightest (Table 8.4); however, the differences between the two NZ strains were not significant. When gut fill was discounted (Tamminga *et al.*, 1997), a similar trend was observed but differences between SC were non significant, which suggests that differences in LW between genotypes in the different swards were probably affected by gut fill.

The NZ70 showed higher  $LW_E$  on TALL than SHORT sward whereas the opposite was observed in the OS90 genotype; in contrast, the NZ90 had similar  $LW_E$  in both SC (Table 8.4). However, it is possible that the crossover design of the experiment could have not completely balanced the effect of the different treatments applied in  $P_E$  I during  $P_E$  II, particularly considering the time elapsed between periods and the magnitude of the LW change during the early lactation period (see Chapter 5).

**Table 8.5: Mean breadth of the incisor arcade of three genotypes of Holstein-Friesian dairy cows, three and four years old.**

		NZ70	NZ90	OS90	SED	GE
Jaw size	mm	85.9	88.4	92.4	1.36	***

SED: maximum standard error of the differences of least squares means; \*\*\* $P < 0.001$ .

Differences in BCS were also observed between genotypes during the experimental period (Table 8.4). The OS90 strain presented the lowest BCS while NZ70 the highest and the NZ90 genotype showed an intermediate value for BCS. Differences in BCS between SC were not observed, however a trend for a higher BCS was observed for NZ90 on the TALL sward (+1.2%, non-significantly different from the SHORT sward).

The NZ70 genotype had the lowest concentration of NEFA and the highest concentration of GLU; the opposite was observed for the OS90 (Table 8.4), with NEFA and GLU concentration for the NZ90 between those measured for NZ70 and OS90. In addition, the concentration of NEFA in blood samples was higher on SHORT than TALL sward whereas the opposite was observed for GLU. A significant GE by SC interaction was observed for GLU, which was highest for NZ70 and lowest for OS90 on the SHORT sward, but with no significant differences across GE when measured on the TALL sward. A trend for lower GLU was observed in OS90 than for both NZ genotypes.

The breath of the incisor arcade was different between genotypes (Table 8.5). The OS90 had the highest distance between the canines (92.4 mm), the NZ90 intermediate (88.4) while the NZ70 had the lowest (85.9 mm). This should affect the potential bite area of the genotypes under unconstrained conditions.

**Table 8.6: Mean calculated daily energy ingested and required of three genotypes of Holstein-Friesian dairy cows.**

		SHORT			TALL			Significance			
		NZ70	NZ90	OS90	NZ70	NZ90	OS90	SED	GE	SC	GE*SC
En Ingested	MJ ME day <sup>-1</sup>	169.4	174.8	178.9	210.6	251.2	285.2	9.71	***	***	***
En <sub>M</sub> <sup>(1)</sup>	MJ ME day <sup>-1</sup>	42.2	54.7	60.3	42.3	53.6	58.2	0.90	***	***	***
En <sub>0</sub>	MJ ME day <sup>-1</sup>	115.2	126.1	126.4	126.7	134.3	130.5	5.86	NS	***	NS
En <sub>T</sub> <sup>(1;2)</sup>	MJ ME day <sup>-1</sup>	164.7	189.5	196.2	176.5	196.5	197.9	6.28	***	**	0.146
En <sub>B</sub> <sup>(1;2)</sup>	MJ ME day <sup>-1</sup>	-2.3	-21.4	-24.5	25.9	46.1	74.6	10.24	NS	***	***

En: energy; En<sub>M</sub>: daily energy requirements for maintenance; En<sub>0</sub>: energy in the milk produced; En<sub>T</sub>: daily energy requirements for maintenance plus grazing activity plus yield; En<sub>B</sub>: energy balance. <sup>(1)</sup> The energy partitioned from and to body reserves or LW change was not considered. <sup>(2)</sup> This estimate includes an allowance for the daily activity of the animal. In all estimates empty LW was utilized (Tamminga *et al.*, 1997). GE: genotype; SC: sward condition; GE\*SC: GE by SC interaction; SED: maximum standard error of the differences of least squares means; NS: non-significant; \*P<0.05; \*\*P<0.01; \*\*\*P<0.001.

### 8.3.5. Energy requirements

The En<sub>M</sub> was highest for the OS90 (59.2 MJ EM day<sup>-1</sup>), intermediate for NZ90 (54.2 MJ EM day<sup>-1</sup>) and the lowest for NZ70 (42.3 MJ EM day<sup>-1</sup>; Table 8.6); in addition, En<sub>M</sub> was slightly different across SC (52.4 vs. 51.4 MJ EM day<sup>-1</sup> on SHORT and TALL swards respectively, mean of all three genotypes), mainly as a result of the lighter NZ90 and OS90 cows in the TALL sward, which declined to a greater extent in the OS90 strain.

Differences in the maintenance requirements between the old NZ70 strain and the two modern strains were explained by the different maintenance requirement per unit of  $LW^{0.75}$  (Yan *et al.*, 1997; Agnew & Yan, 2000).

The difference measured for  $En_0$  across genotypes was not significant even though a trend for higher  $En_0$  was observed for NZ90 and OS90 strains (129.3 MJ EM day<sup>-1</sup>, mean of the two genotypes) than NZ70 (120.9 MJ EM day<sup>-1</sup>; Table 8.6), particularly on the SHORT sward, as a result of the greater yield of the NZ90 and OS90 sustained from higher body reserves mobilisation. There was a significant difference in  $En_0$  across SC (122.6 vs. 130.6 MJ EM day<sup>-1</sup> on SHORT and TALL swards respectively; mean of all three genotypes).

The difference in  $EN_{BAL}$  between genotypes was not significant, negative for all three strains grazing the SHORT sward and positive on TALL. A significant GE by SC interaction was observed, with the OS90 having the lowest  $EN_{BAL}$  (more negative) on the SHORT sward and the highest (more positive) for the TALL. This difference between SC was intermediate for NZ90 and the lowest difference was for the NZ70 genotypes (Table 8.6). A higher coefficient of correlation was estimated between  $EN_{BAL}$  and NEFA than between  $EN_{BAL}$  and GLU; in addition differences in the coefficient of correlation between genotypes were also observed (Table 8.7).

**Table 8.7: Pearson correlation coefficients (r) between main blood metabolites and the estimated energy balance.**

Dependent	GE	Mean	SD	n	Independent	GE	r	P	Mean	SD	n
$EN_{BAL}$	All	15.6	51.2	96	NEFA	All	-0.30	**	0.13	0.09	96
	NZ70	10.9	37.6	34		NZ70	-0.31	0.079	0.13	0.09	34
	NZ90	12.0	47.7	31		NZ90	-0.28	0.129	0.10	0.05	31
	OS90	24.3	65.9	31		OS90	-0.41	*	0.16	0.11	31
					GLU	All	0.11	NS	3.29	0.24	96
						NZ70	0.35	*	3.40	0.21	34
						NZ90	-0.03	NS	3.27	0.25	31
						OS90	0.22	0.235	3.19	0.22	31

$EN_{BAL}$ : energy balance; SD: standard deviation; NS: non-significant; \* $P < 0.05$ ; \*\* $P < 0.01$ ; \*\*\* $P < 0.001$ . GE: genotype; P: probability [significance: \* $P < 0.05$ ; \*\* $P < 0.01$ ; \*\*\* $P < 0.001$ . NS= not significant.]; r: coefficient of correlation; n: sample number.

### 8.3.6. Grazing behaviour

All three genotypes spent more time grazing on SHORT than TALL sward, and less time ruminating (Table 8.4). The significant GE by SC interaction observed for  $G_T$  indicated that both NZ genotypes, particularly the NZ90, spent more time grazing on the SHORT sward than the OS90 strain (Table 8.4), whereas the OS90 genotype showed the longest

$G_T$  and NZ70 had the shortest on the TALL sward (although these latter differences were not significant). The OS90 also spent more time ruminating than the two NZ genotypes.

The  $I_R$  estimated from  $DMI_H$  and  $G_T$  showed that all three genotypes achieved the fastest  $I_R$  on the TALL sward; in addition, the fastest  $I_R$  was measured for OS90 while the slowest for NZ70. The  $I_R$  of the NZ90 was similar to that of the NZ70 on the SHORT sward but higher on the TALL, though below that recorded in the OS90 genotype (-12%).

The total time spent in grazing related activities ( $G_T+R_T$ ) did not show any significant effect of GE or SC, however a significant GE by SC interaction showed more total activity for both NZ genotypes on the SHORT sward, particularly for the NZ90 strain. However, on the TALL sward, the OS90 spent more time in grazing related activities than the two NZ genotypes, especially when compared to NZ70.

## 8.4. DISCUSSION

### 8.4.1. Herbage intake, milk yield and plasma metabolites

#### 8.4.1.1. *Herbage intake*

This experiment removed most of the confounding effects of feeding level on DHA and herbage mass, and consequently on  $DMI_H$  and grazing behaviour, that were observed in the 'system' study (see Chapters 6 and 7). However, due to the different adult LW of the genotypes, DHA/LW was still about 12.5% lower for the OS90 cows on both SC than in the two NZ genotypes (Table 8.1), which might account for the lower  $DMI_H/LW$  (-8.7%) of OS90 than NZ90 cows on the SHORT sward (Table 8.4). On the TALL sward similar  $DMI_H/LW$  between NZ90 and OS90 were measured, however, it is possible that OS90 cows could have consumed even more herbage if DHA/LW had been similar for both genotypes. Similar trends were observed when  $LW_E$  was considered.

The  $DMI_H$  and DMD of the three different genotypes were affected differently by the structure of the sward (Table 8.4). On the TALL sward the NZ90 and OS90 strains ate more pasture due to their higher metabolic demand and feeding drive, in agreement with the differences observed for blood concentrations of metabolites and BCS between genotypes. The OS90 genotype consumed more herbage in absolute terms than NZ90, though  $DMI_H/LW$  was similar and comparable to that reported by Kolver et al. (2002) for high yielding cows fed total mixed ration (TMR; about 41 g DM kg<sup>-1</sup> LW), however cows in the present trial were older (three to five years old), lighter (-8% and -12%) and had lower yields of MS (-19% and -31%) than those recorded in Kolver et al. (2002).

The  $DMI_H$  achieved on the TALL sward was greater than that reported in early lactation for cows of the same genotypes managed in the system study (see Chapter 7) and also greater than those reported for high yielding cows grazing pasture in early lactation [18.0 and 18.4 kg DM cow<sup>-1</sup> for comparable NZ and OS cows in first lactation on pasture, at a DHA above 60 kg DM cow<sup>-1</sup> day<sup>-1</sup> (Kolver, 2001); 19.0 kg DM cow<sup>-1</sup> for cows in first to third lactation (Kolver & Muller, 1998); 20.1 kg DM cow<sup>-1</sup> for cows in second lactation (Buckley, 1998 cited by O'Connell et al., 2000); 19.3 and 18.0 kg DM cow<sup>-1</sup> of herbage for supplemented cows in second and third lactation (Buckley *et al.*, 2000)]. The  $DMI_H$  of the NZ70 cows was below that upper limit proposed in the 1980s for grazing dairy cows in early lactation [16.9 kg DM cow<sup>-1</sup> day<sup>-1</sup>; (Meijs & Hoekstra, 1984)]. The difference between this value and that obtained by the modern NZ cow on the TALL sward indicates that the capacity for herbage consumption has improved in the last thirty years as result of the genetic progress for yield.

In contrast, on the SHORT sward the  $DMI_H$  and DMD of the three genotypes were constrained at similar levels. The reduction in  $DMI_H$  between swards was greater for OS90 (-7.11 kg DM cow<sup>-1</sup> day<sup>-1</sup>) than for both NZ genotypes (-5.14 and -2.56 kg DM cow<sup>-1</sup> day<sup>-1</sup> in NZ90 and NZ70 respectively). In addition, the  $DMI_H/LW$  by the OS90 was lower than for the NZ90 (-8.7%) and NZ70 (-6.3%) genotypes. This suggests that on the SHORT sward the OS90 cows had poorer energy status than the two NZ strains, in agreement with the results of the system study (see Chapter 7). The highest NEFA and lowest GLU recorded for the OS90 on the short sward indicate higher mobilisation of fat reserves to sustain yield in this genotype grazing the SHORT sward.

On the TALL sward the NZ90 and OS90 cows achieved similar  $DMI_H/LW$ , however the former strain consumed significantly less pasture (-2.39 kg DM cow<sup>-1</sup> day<sup>-1</sup>). If it is considered that  $DMI_H/LW$  gives an idea of the intake capacity of the cows relative to the maintenance requirement, it could be argued that the NZ90 cows were more efficient than OS90 cows, as they consumed less pasture in absolute terms but achieved similar  $DMI_H/LW$  to the OS90. Furthermore, the NZ90 might have reduced energy expenditure as these cows had reduced total activity. In contrast, even though the OS90 achieved the highest  $DMI_H$ , they still showed the highest NEFA (although lower than observed on the SHORT sward) and lowest GLU (although higher than observed on the SHORT sward), which suggests a different metabolic use of body reserves, despite the fact that on the TALL sward  $DMI_H$  was higher and the estimated energy balance positive (Table 8.6).

#### 8.4.1.2. *Daily yields*

Lactose yield was greater for the OS90 genotype and higher on the TALL than on the SHORT sward, sustained by an increase in milk yield and lactose content at both SC. This response contrasted with that observed for both NZ genotypes where lactose content decreased slightly on the TALL sward (although not significantly), but daily lactose yield also increased on the TALL sward as milk yield increased. The differences in the yields of milk components between genotypes and the way the cow allocates nutrients to milk and its components are controlled metabolically and genetically. Differences between genotypes had probably resulted from the selection policies utilised in NZ compared to those in USA or Europe. The OS90 yielded more MS plus lactose on both the SHORT and TALL swards and also produced more milk yield, therefore the estimated energy output per unit of MS yielded may be higher for the OS90 as was determined in Chapter 6 and indicated by the energy in the milk yielded (Table 8.6).

The greater BCS of the two NZ genotypes is an indication of the difference in utilisation of body fat reserves and metabolism between genotypes (Veerkamp *et al.*, 2003), however it is apparent that this different threshold responded to a different limit in the rate of tissue mobilisation between genotypes. The higher rate of body fat mobilisation for the OS90 may sustain the increased milk yield that occurs post-calving and sustain the faster rise observed in yield than for intake, particularly when fed on pasture. Once body reserves have been depleted to their minimum, the capacity of reserves to buffer fat content in milk will decline, and then any increase in nutrient availability has a faster effect on milk volume and protein yield and content in milk, than on fat yield. A high availability of body fat reserves and a faster rate of fat mobilisation for OS90 than NZ90 would suggest a rapid recovery of the concentration of metabolites in blood as they are being used. If yield is constrained by feeding level and the rate of substrate utilisation is low, the circulating pool of energy and metabolites may be maintained at a high level, which would depress feeding drive and  $DMI_H$  (Weston & Poppi, 1987; Ingvarlsen *et al.*, 1999). The opposite would occur when body fat reserves are low, then if nutrient availability and  $DMI_H$  improve, cows would increase yield and the circulating pool of nutrients and metabolites decrease rapidly, as a result, the feeling of satiation decline and then the motivation to graze increase.

#### 8.4.1.3. *Plasma concentration of blood metabolites*

Metabolic differences between the strains are also supported by the change in the concentrations of plasma GLU and NEFA in blood samples, both metabolites are known to be good indicators of the energetic status of the cow (Clark *et al.*, 2005), in addition

glucose is the main precursor for lactose synthesis in the mammary gland (Holmes *et al.*, 2002). The differences in plasma GLU and NEFA between the genotypes on the SHORT sward are consistent with the differences in  $DMI_H$ , and also in the changes observed in yield and BCS, which indicate a greater energy deficit for the OS90 than for the two NZ genotypes. The difference in yield and BCS between NZ70 and NZ90 indicates that differences in the energetic status between these genotypes also exist. The NZ70 and NZ90 had similar NEFA, however lower GLU was recorded for NZ90 cows suggesting a lower energy balance for them (Clark *et al.*, 2005).

In the TALL sward the energetic status of the NZ90 and OS90 improved. This was probably greater for OS90 than NZ90 as shown by the greater increase in GLU between sward conditions. Both NZ strains had similar GLU and NEFA, hence similar energetic status. The energetic status of the OS90 genotype also improved between SC, as indicated by the plasma concentration of GLU and NEFA. However, GLU was still the lowest and NEFA the highest, despite the fact that  $DMI_H$  was highest for the OS90 genotype. The fact that NEFA was the lowest for the NZ90 strain on both SC probably indicates that a greater proportion of the energy required by this genotype was obtained from the feed consumed, with lower mobilisation of body fat.

#### 8.4.2. Energy requirements and balance

The  $EN_{BAL}$  of individual cows was correlated to a greater extent to the values of NEFA in blood samples than to the values of GLU (Table 8.7), as suggested by Clark *et al.* (2005). The more negative  $EN_{BAL}$  observed in the SHORT sward for the NZ90 and OS90 strains than for the NZ70 is sustained by the greater  $En_M$  and yields, which suggests higher body reserves mobilisation. Even though the NZ90 had greater MS yield, the  $EN_{BAL}$  was slightly less negative for this strain than for OS90, which was mainly explained by the greater  $DMI_H/LW$  of the NZ90 cows.

On the TALL sward, the  $EN_{BAL}$  was positive for all three genotypes but the response in yield of the cows was lower than expected. One possible reason is that  $DMI_H$  was constrained under the pre-experimental conditions and to some extent during the experiment as a result of the crossover design utilised, with greater effect on cows of high genetic yield potential. The evidence available indicates that these genotypes were differently constrained on pasture as is indicated by the different BCS lost during lactation (see Chapters 5, 6 and 7).

Penno *et al.* (2001) suggested that under sub-optimal nutrition, energy output would reach equilibrium at a lower milk target than potential, gradually reducing feed demand. Thus

the effect of an improved sward condition on  $DMI_H$  and energy input was not correlated with a proportional increase in yield, probably because the cows partitioned a high proportion of the extra energy input to increase body reserves instead of increasing milk yield; nevertheless, changes in BCS were not evident during the experiment due to its short duration.

#### **8.4.3. Effects of sward structure on dry matter intake and grazing behaviour**

The amount of herbage consumed by a cow can be described as the product of  $G_T$  and  $I_R$  (Allden & Whittaker, 1970; Hodgson, 1990),  $I_R$  being the product of bite weight and bite rate ( $B_R$ ). Of these,  $I_R$  and  $G_T$  are the main determinants of daily  $DMI_H$  with grazing being extended when  $I_R$  is constrained due to unfavourable sward conditions. This explains the differences in  $DMI_H$  measured between the SHORT and the TALL sward, as the reduced  $G_T$  in the latter indicates that the three genotypes increased  $I_R$  in this sward. Moreover,  $I_R$  was greater for the OS90 genotype in both SC. It is apparent that  $I_R$  increases with milk yield potential due to increased metabolic requirements and feeding drive, but also because the size of the mouth was larger for OS90 cows and so the potential area of the bite (Table 8.5).

Even though the difference in  $DMI_H/LW$  between strains on the SHORT sward agreed with the differences measured in the system study (see Chapter 7), differences in the present experiment could simply reflect the difference in  $DHA/LW$  at which the genotypes were managed and may not demonstrate that real differences in grazing capacity between strains exist. The OS90 strain increased  $DMI_H$  to a greater extent between SC than the two NZ genotypes due to the improved herbage availability of the TALL sward, and then achieved similar  $DMI_H/LW$  despite the fact that  $DHA/LW$  was 10% lower. In order to achieve this, these cows were able to consume 2.4 kg DM more than the NZ90 genotype (12%).

This indicates the higher intake capacity of the OS90 cows when grazing pasture with reduced physical constraints to herbage ingestion. Moreover, the trend for a lower  $HM_{POST}$  and  $SH_{POST}$  measured on the TALL sward indicates that these cows removed a higher proportion of the herbage offered. Grazing efficiency ( $G_{EF}$ ; Table 8.1a), defined as herbage intake expressed as a proportion of the herbage allowance per unit of LW (Combellas & Hodgson, 1979), was similar between strains on the SHORT sward (0.40) and lower than those measured on the TALL sward (0.55). The value of  $G_{EF}$  in the TALL sward agreed with those measured in the system study (see Chapter 7) and support the suggestion that the low  $G_{EF}$  obtained resulted from a decrease of  $DMI_H$  as a consequence of reduced herbage availability, despite the increased  $DHA$  (Hodgson, 1990). The

differences in  $DMI_H$  and  $G_{EF}$  between the NZ90 and OS90 genotypes, but particularly the lower  $DMI_H/LW$  of the OS90 than NZ90, suggest that the OS90 genotype was affected to a greater extent than the two NZ strains in the SHORT sward.

The greater  $DMI_H/LW$  measured in both NZ strains on the SHORT sward suggests these strains, but particularly the NZ90, were able to satisfy a higher proportions of their energy requirements from the herbage consumed, in agreement with the differences observed in blood metabolites and BCS between strains. Thus, the improved performance of the NZ90 strain on pasture is not determined by its higher absolute  $DMI_H$  but by its improved intake relative to energy requirements.

#### 8.4.4. Evidence of differences in grazing ability between strains

Grazing ability was defined as the capacity of the cow to maintain  $I_R$  in the face of limiting sward conditions and to extend  $G_T$  to offset a decline in  $I_R$  (Hodgson, 1985). However, it is unlikely that a cow will continue grazing when (a) her energy requirement was already covered and her motivation to graze reduced due satiation, and (b) the energy gained per bite ingested is lower than the energy expended in harvesting and processing the herbage harvested. Hence, an improved definition should also involve the quality of the herbage ingested per bite (Laca & Demment, 1996).

The larger jaw size of the OS90 genotype (Table 8.5) suggests a larger potential bite area of about 5.7 and 9.1 cm<sup>2</sup> more than NZ90 and NZ70 respectively (assuming a circular shape). Thus  $B_Z$  may be larger if there is no constraint to bite penetration. However, under strip grazing management the capacity to maintain  $B_Z$  would be affected during the grazing down process as the cow will face increases in bulk and decreases in quality of the herbage as it is forced to graze further down into the lower strata of the pasture (Laca *et al.*, 1992). It is apparent that the motivation of the cow to graze within this layer would be determined by its relative energy deficit (Meijs & Hoekstra, 1984; Penno *et al.*, 2001), but also by the physical constraints to herbage prehension. To compensate for a declining  $I_R$  at a declining  $B_Z$ , mainly as a result of prehension constraints, the cows increase  $B_R$  (see Chapter 7).

Differences in quality between SC indicate a greater contrast between the upper and lower strata on the TALL than the SHORT sward, which is in agreement with the higher  $SH_{POST}$  measured in the TALL sward. The fact that pastures grazed with both NZ genotypes had higher post-grazing residuals than those grazed with the OS90, suggests that the two NZ strains did not extend grazing further because at the level of  $DMI_H/LW$  they achieved, they were already satiated. This suggests that although a metabolic signal

probably sets the motivation of the cow to graze, short-term signals associated with the condition of the sward and the difficulty toprehend the herbage may determine the extent of the activity of the cow.

Previous studies indicate that larger animals bite more deeply and are less constrained by the shearing resistance of the herbage (Illius *et al.*, 1995), in agreement with the shorter residual left by the OS90 on the TALL sward. However, both the NZ90 and OS90 genotypes had similar limits on the SHORT sward and grazed to a lower post-grazing residual than of the NZ70 genotype, probably because of the higher metabolic demand and motivation to graze of these two genotypes. The OS90 cows may have the greatest  $B_Z$  while the NZ70 cows the smallest, which agrees with the different  $I_R$  estimated. The fact that the  $I_R$  of the NZ90 in the SHORT sward was similar to NZ70, whereas the potential  $B_Z$  would be different, suggests a slower  $B_R$  for the NZ90 in the SHORT sward, similar to that observed in the system study (see Chapter 7). Under strip-grazing management, the decline in  $B_Z$  would be steeper than the decline in  $I_R$  on a SHORT sward due to the compensating increase in  $B_R$ . However, a cow grazing selectively may reduce  $B_R$  and extend  $G_T$  further, as was observed for the two NZ genotypes (see Chapter 7).

As a consequence of the smaller area allocated to the cows, the rate of depletion of a TALL sward may be faster and then  $I_R$  may decline more rapidly with the faster increase of ingestive constraints as the sward is grazed down (Laca *et al.*, 1994). It is suggested that the contrast between bulk density and quality between the upper and bottom layers of the sward would increase as the mean quality of the sward declines and the constraints to ingestion also increase.

Cows grazing at higher  $I_R$  achieve higher  $DMI_H$  and reduce  $G_T$  and energy expenditure. Ruminating time increased with  $DMI_H$ , nevertheless, the three genotypes showed a similar  $G_T/R_T$  ratio in the TALL sward, indicating a similar proportion of time between activities. The OS90 genotype spent less  $R_T$  per kilogram of dry matter consumed and the opposite was measured for the two NZ genotypes, in agreement with previous studies comparing OS with NZ cows on pasture (Thorne *et al.*, 2003); nevertheless, the opposite response was observed in the SHORT sward in the present study. Cows with higher rumination efficiency showed an improved DMD, particularly on the TALL sward, probably motivated by the improved quality of the herbage offered.

Rumination was reduced further for the two NZ genotypes on the SHORT sward, as is expected from the lower  $DMI_H$ ; as a result the cows had more time available to continue grazing (Table 8.4). The opposite response was observed in the TALL sward. The total

activity ( $G_T+R_T$ ) was lower for the two NZ genotypes grazing the TALL sward, which represented a net saving of time and probably energy expenditure, but the opposite occurred on the SHORT sward.

Illius and Gordon (1987) demonstrated that the ability to obtain adequate energy intake from a SHORT sward is favourable to small animals. They indicated that the relationship between energy intake and body weight is a function of bite dimensions and nutrient density, which explains the preference of the animal to graze the most digestible horizons of the pasture avoiding the lower quality layers. The fact that  $G_T$  was extended on the SHORT sward by both NZ genotypes suggests these cows can get a benefit from each additional bite performed, even though it might be small and despite the extra time required. Cows performing small bites would ingest small particles of herbage; hence, reducing the ruminative requirement even when consuming a low quality diet.

At a higher  $B_z$  chewing per bite increases (Illius *et al.*, 1995) but a decline in  $B_z$  reduces the comminution required, even though an increase in manipulative jaw movements may contribute to slow down the grazing process further. Rook *et al.* (1994) proposed that the small bite size strategy produces small particles directly without the need of additional handling or further rumination, thus in the face of limiting sward conditions this seems to be one possible alternative to compensate increments in grazing activity and energy expenditure. In addition, a decrease in the rate of ingestion caused by a more selective behaviour would increase bite quality where opportunity for diet choice exists.

## 8.5. CONCLUSIONS

The OS90 strain had lower MS yield, similar MS plus lactose and larger milk yields than the NZ90 on both SC. This indicates a different allocation of energy in the milk output, determined genetically. The OS90 strain also had the lowest BCS on both SC suggesting a greater mobilisation of body reserves. Although comparative yields and BCS relative to the  $DMI_H$  achieved could be influenced by the crossover design of the experiment, the concentration of blood metabolites are also indicative of the greater mobilisation of reserves for OS90 than NZ90.

All three genotypes improved  $DMI_H$  on the TALL sward as a result of the increased  $I_R$ , which resulted from decreased limitations to ingestion and increased availability of the herbage offered on this sward. The OS90 can achieve a higher absolute  $DMI_H$  and greater  $G_{EF}$  than NZ90 when highly digestible and readily available herbage is offered. In contrast, the NZ90 demonstrated a higher flexibility to adjust behaviour and achieve high  $DMI_H/LW$  on the constraining SHORT sward, and to improve its energetic status

compared to the OS90 strain. This is in agreement with the lower mobilisation of reserves observed, and the lower concentration of NEFA in blood.

Even though the OS90 had higher  $DMI_H$  than the NZ70 and NZ90 genotypes on both SHORT and TALL swards, the two NZ strains had satisfied a higher proportion of their energy requirements from the herbage consumed in the SHORT sward. This agreed with the results of the system study (see Chapter 7), although the differences observed between strains in the present experiment were also confounded by the difference in DHA/LW at which the genotypes were managed. This indicates the difficulties of creating exactly equal conditions for genotypes differing in LW and milk yield; therefore comparisons between the strains must be interpreted carefully.

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## SECTION V

### CHAPTER 9

## FINAL INTEGRATED DISCUSSION AND CONCLUSIONS

### 9.1. INTRODUCTION

Imported genetic material of the Holstein-Friesian breed from North America has been used in New Zealand (NZ) since the late 1960's. This has diluted the genetic base of the former NZ Friesian strain selected under intensive seasonal pasture-based systems (Harris & Winkelman, 2000), and raised concerns about the extent of the negative influence of these genes on the modern NZ cow (Harris & Kolver, 2001). The North American (NA) genotype is heavier and produces more milk and protein but lower fat yields than the NZ Holstein-Friesian. However, the NA genotype also demonstrated a lower in-calf rate, higher services per conception and lower survival rate under pastoral conditions (Harris & Kolver, 2001).

The modern NZ Holstein-Friesian had lower genetic yield potential than its overseas (OS) counterpart, however, it produced more milksolids (MS), gained more live weight (LW) during lactation and had better reproductive performance on pasture. In contrast, the OS strain improved yield performance to a greater extent when fed with a total mixed ration (TMR)(Kolver et al., 2002). Interactions of this type are difficult to detect under narrow nutritional contrasts (Freeman, 1975; Veerkamp et al., 1995), but have been reported more frequently in recent years, probably as a result of increased genetic differences between the genotypes compared (Kennedy *et al.*, 2002; Kennedy *et al.*, 2003; Linnane *et al.*, 2004).

It was suggested that the reason for the lower performance of the OS Holstein-Friesian on pasture is the lower grazing ability of these cows and their inability to ingest the higher amount of nutrients required to achieve their genetic potential from the pasture, as opposed to a TMR diet (Veerkamp et al., 1995; Kolver et al., 2002; Kolver, 2003). This limitation seems to be more evident in early lactation (Kolver & Muller, 1998), when

energy requirements are high, even when cows graze pastures at high daily herbage allowance (DHA)(Kolver et al., 2002).

In the present thesis, the productive performance of three Holstein-Friesian genotypes was compared under different feeding and grazing managements in two experiments. Three genotypes (GE) were utilized, two modern high breeding worth (BW) Holstein-Friesian strains, of NA (OS90) or NZ (NZ90) origin, and a 1970's NZ Friesian strain (NZ70) of low BW (see Appendix IV for details). In the first experiment the genotypes were farmed at different feed allowance per cow (FA) in a long-term 'system' study. The second experiment was a short-term 'component' study where the same genotypes were compared when grazing different sward conditions at a common DHA.

It was hypothesized that cows of the NZ90 and OS90 genotypes, with high genetic potential for yield, would be more constrained grazing pasture than cows of the NZ70 strain, with lower genetic potential and requirements. Both NZ90 and OS90 genotypes would improve yield to a greater extent than NZ70 when extra feed is utilised, so that the productive performance of the OS90 would then be greater than the NZ90 in agreement with differences in their genetic potential. The two NZ genotypes would perform better on pasture-based systems, due to their greater capacity to achieve higher herbage dry matter intake ( $DMI_H$ ) from grazed pasture.

The first objective of this work was to compare the productive performance of these Holstein-Friesian farmed on pasture-based systems at different FA, secondly, to investigate if differences in performance were sustained by differences in herbage intake and grazing ability when managed on pasture. As the accurate estimation of  $DMI_H$  was central to achieve the previous objectives, it was also important to establish an accurate procedure to measure  $DMI_H$  of grazing dairy cows that were also offered forage and maize supplements, where the composition of the pasture and diet changed regularly.

## 9.2. DISCUSSION

### 9.2.1. Intakes estimates using *n-alkanes*

In the present thesis  $DMI_H$ , forage and maize supplement were accurately estimated by combining the *n-alkanes* –  $\delta^{13}C$  methods (see Chapter 4 for details). The *n-alkanes* combination  $[C_{31}+C_{33}] - C_{32}$  in the equation of Dove & Mayes (1991), adjusted by their different recovery rates in faeces was successfully utilised (see Chapters 3 and 4 for details).

The procedure was tested with data from different indoor studies (see Chapter 3) and demonstrated to be more accurate than the use of individual pairs of n-alkanes (either C<sub>31</sub>–C<sub>32</sub> or C<sub>33</sub>–C<sub>32</sub>) when the cows select components with different n-alkanes composition from the pasture. The difference between the mean DMI<sub>H</sub> of a group of cows grazing the same pasture was small, estimated from different combination of natural n-alkanes relative to the C<sub>32</sub>-alkane dosed; despite the fact that great differences were observed in the DMI<sub>H</sub> estimated for the same cow from different n-alkanes pairs. The pair [C<sub>31</sub>+C<sub>33</sub>]-C<sub>32</sub> gave the most accurate estimate of DMI<sub>H</sub> for individual cows, because it considered the change in the concentrations of both C<sub>31</sub> and C<sub>33</sub>-alkanes in the diet of the cows relative to their concentration in the pasture. Thus, this procedure was used routinely to estimate DMI<sub>H</sub> in the field experiments of the present thesis. The small difference observed for the mean DMI<sub>H</sub> of a group of animals grazing the same pasture under strip grazing, occurred due to the fact that the differences in the relative concentrations of both C<sub>31</sub> and C<sub>33</sub>-alkanes in the diet of different individual cows are compensated when the herd is considered and most of the herbage available is removed.

One step further would be to validate this procedure under controlled indoors conditions, where the capacity to select a diet with a different proportion of plant species or plant-parts, and therefore in n-alkanes composition and ratio C<sub>31</sub>/C<sub>33</sub>, could be investigated by estimating the DMI<sub>H</sub> of the different components with the n-alkanes method and comparing the result obtained with the current intake of each pasture component. In addition, this would provide a test for the method proposed by Dove and Moore (1995) to discriminate between components with different n-alkanes content, particularly when one of the pasture components and the supplement have similar ratios of stable isotopes <sup>13</sup>C and <sup>12</sup>C (Chapter 4). It would also make it possible to test the assumption made about the recovery rate of n-alkanes in faeces being similar between different genotypes.

### 9.2.2. Experimental design

The differences in LW and metabolic requirements between the three Holstein-Friesian genotypes utilised in the present thesis made it difficult to compare them under exactly common experimental conditions. This was particularly evident in the ‘system’ study where they were farmed in systems that offered similar levels of nutrition relative to their genetic potential requirements, but differing in absolute FA. In the ‘component’ study, despite the efforts made to manage all three genotypes under similar DHA, herbage allocation was slightly different when expressed per unit of LW, due to differences in the LW of genotypes. Conclusions about the importance of GE by FA interactions in these studies must therefore be interpreted with caution.

Nevertheless, the results obtained were consistent over years in the ‘system’ study, and between the ‘system’ and ‘component’ trials. Also, the procedure adapted to evaluate interactions, based on a combination of the mean level of intake or production and the slope of the regression of the same variable across levels of FA within genotypes, is considered to be relatively robust. The wide range of levels of FA and of pasture/supplement proportions offered in essentially pasture-based systems also provides basis for confidence in the research outcome and their relevance to practical dairying systems.

### **9.2.3. Performance of different Holstein-Friesian genotypes in pasture-based systems**

Seasonal calving systems synchronise energy ‘demand’ with pasture growth or nutrient ‘supply’. The efficient transformation of pasture into MS ensures a low cost per unit of product; as a result, high pasture utilisation (PU) is usually achieved in systems farmed at higher comparative stocking rate (CSR).

In the present thesis, results from the ‘system’ study indicate that milksolids (MS) yield per hectare and per cow were higher with the NZ90 strain (see Figures 5.4a and 5.4b in Chapter 5). This demonstrates the higher productive performance of this genotype on pasture, within the range of FA at which these systems were farmed (see Table 5.1 in Chapter 5).

The higher productivity of the NZ90 strain was sustained by its higher mean daily MS yield than NZ70 and longer lactation length than OS90 (Table 9.1). The length of the lactation (see Figure 5.3d in Chapter 5) was determined by the body condition score (BCS) at the end of lactation (see Figure 5.6b in Chapter 5), according to the drying-off rules (Macdonald & Penno, 1998). The mean BCS during lactation was lower for the OS90 than for the two NZ strains, despite FA (see Figure 6.4b in Chapter 6), due to a greater loss of BCS in early lactation (see Figures 5.6b and 5.7b in Chapter 5). This loss of body reserves was not recovered while the OS90 cows were lactating; hence, these cows were dried off earlier because their BCS was lower than for the two NZ strains in late lactation. This indicates higher body reserves mobilisation, which occurs as a result of the inability of the OS90 cows to improve  $DMI_H$  on pasture, particularly in early lactation and according to their higher genetic potential for yield, compared to that observed for two NZ genotypes.

**Table 9.1: Summary of results in the ‘system’ study: Planned and actual feeding levels, cow condition and mean productive performance and whole season feed conversion efficiency of three Holstein-Friesian genotypes managed in different feeding systems. Mean values for three seasons (2001-02, 2002-03 and 2003-04) and for season 2003-04 are presented.**

	Genotype	Mean 2001-2004			2003-2004		
		NZ70	NZ90	OS90	NZ70	NZ90	OS90
Stocking rate (SR)	cow ha <sup>-1</sup>	3.27	3.14	3.06	3.27	3.14	3.06
Nominal Total feed offered (NTF <sub>0</sub> ) <sup>1</sup>	t cow <sup>-1</sup> year <sup>-1</sup>	5.33	5.75	6.25	5.33	5.75	6.25
Actual Total feed offered (TF <sub>0</sub> ) <sup>2</sup>	t cow <sup>-1</sup> year <sup>-1</sup>	5.98	6.38	6.54	6.11	6.69	6.81
<b>Apparent feed consumed</b>							
Pasture	t cow <sup>-1</sup> year <sup>-1</sup>	4.44	4.56	4.77	4.52	4.71	4.99
Pasture silage	t cow <sup>-1</sup> year <sup>-1</sup>	0.20	0.23	0.17	0.19	0.18	0.09
Maize silage	t cow <sup>-1</sup> year <sup>-1</sup>	0.04	0.14	0.33	0.07	0.26	0.50
Maize grain	t cow <sup>-1</sup> year <sup>-1</sup>	0.06	0.06	0.06	0.05	0.06	0.07
Total feed consumed	t cow <sup>-1</sup> year <sup>-1</sup>	4.74	4.98	5.33	4.82	5.21	5.65
Liveweight	kg cow <sup>-1</sup>	453	470	490	477	502	534
BCS	units	4.71	4.23	3.98	4.56	4.30	3.69
<b>Annual milksolids yield</b>							
Total MS per hectare	kg ha <sup>-1</sup>	1093	1236	1154	1263	1485	1406
Total MS per cow (whole lactation)	kg cow <sup>-1</sup>	336	395	377	388	473	455
Daily MS per cow	kg cow <sup>-1</sup> day <sup>-1</sup>	1.21	1.45	1.46	1.37	1.66	1.69
Daily MS plus lactose per cow	kg cow <sup>-1</sup> day <sup>-1</sup>	1.97	2.31	2.39	2.19	2.61	2.77
Lactation length	days	276	271	257	283	285	268
<b>Systems efficiencies</b>							
Pasture utilisation efficiency		0.80	0.79	0.82	0.80	0.78	0.83
<b>Feed conversion efficiency</b>							
MS per DM consumed	g MS kg <sup>-1</sup> DM	71	79	70	78	90	83
MS per ME consumed	g MS MJ ME <sup>-1</sup>	6.3	7.0	6.2	7.0	7.9	7.0
MS per LW unit	kg MS kg <sup>-1</sup> LW	0.74	0.83	0.76	0.81	0.94	0.85

SR: stocking rate; <sup>(1)</sup>NTF<sub>0</sub>: Nominal (planned) total feed offered (see Chapter 5 for details). <sup>(2)</sup>TF<sub>0</sub>: Actual feed offered. MS: milksolids. Live weight (LW) and body condition score (BCS) are means for the whole season.

The results from the last season of the ‘system’ study (2003-04) represent better the expected outcome of the commercial systems as in this season they had a higher proportion of mature cows. In addition, the contrast between the actual amounts of feed offered (TF<sub>0</sub>) was larger between systems than in the previous season (see Chapter 5), and also the change in the drying-off decision rules meant that systems managed at high FA were dried-off later (Macdonald *et al.*, 2005). Despite this improvement, the two NZ strains were still milked for longer in 2003-04 (Table 9.1).

Total lactation yield increased from the first to the third season of the study (Figure 5.5a and 5.5b in Chapter 5), as the cows matured (see Tables 5.5, 5.6 and 5.7 in Chapter 5). The shape of the lactation curves also changed as the proportion of mature cows in the systems increased (see Figure 5.8 in Chapter 5). Differences between genotypes were more evident in the last season of the study, where the OS90 achieved the highest production peak but showed a trend for the shortest lactation (or lower persistency). The difference in the shape of the lactation curve between NZ90 and OS90 was larger when estimated for milk than for MS yield, and it is apparent that yields of fat and milk composition are more constrained than milk volume in the grazing OS90.

#### 9.2.4. Genotype by feeding level interactions

The fact that today's modern dairy cows, with high yield genetic potential, have their  $DMI_H$  constrained on pasture compared to a TMR diet (Kolver & Muller, 1998) suggests that the importance of GE by feeding interactions has increased in the last decade as a result of genetic selection, which is more evident with a high contrast between the level of feed at which the genotypes are compared (Kolver et al., 2002). In the present thesis, the high contrast in genetic yield potential and metabolism between the genotypes utilised was probably the reason for the increased number of interactions detected as the study progressed, despite the nutritional contrast being smaller than that utilised by Kolver et al. (2002).

The  $DMI_H$  achieved by cows of high genetic potential was constrained on pasture hence these cows produce similarly to cows with lower genetic potential. However intake increased under improved sward conditions and particularly when more supplement was included in the diet, so yield increased to a greater extent for NZ90 and OS90 than for NZ70. As a result, an increased number of GE by feeding interactions for milk and lactose yields, protein content in the milk and BCS were observed in seasons 2002-03 and 2003-04. This was consistent with the indication that interactions would increase in mature animals due to the increased difference between the genotype's capacity to produce (see Tables 5.6 and 5.7 in Chapter 5)(Veerkamp et al., 1994; Kolver et al., 2000). The lack of interactions observed in 2001-02 was consistent with the results of Linnane et al. (2004) sustained by a lower intake capacity of two years old cows (Jarrige et al., 1986), even though feeding level increased.

The increased productivity of the modern cows is in part sustained by higher mobilisation of body reserves, which play a role in sustaining yield under conditions of low cow nutrition. This however, is not sustainable in the long term (Veerkamp et al., 1995), hence other functions of the cow, for instance reproduction, would be affected under persistent nutritional constraints.

The OS90 strain achieved poor milk yield at low FA despite the higher amount of body reserves mobilised, but they increased milk yield to a greater extent than the two NZ genotypes as FA increased. In contrast, the content of solids in milk, particularly protein, increased to a greater extent as FA increased for NZ90 than OS90 and NZ70 (see Table 5.8 in Chapter 5). The BCS nadir was lowest for the OS90 strain in 2003-04, irrespective of FA, indicates that this genotype satisfied a lower proportion of its energy requirements from the feed, particularly during the early lactation period.

Compared with the NZ70, the NZ90 genotype is heavier and has higher genetic yield potential; hence it needs to consume more feed. It is probable that the NZ90 strain would reduce yield to a greater extent than NZ70 if farmed at an even lower FA. The NZ90 strain partitioned more energy to milk yield from body reserves in early lactation and depleted body reserves further to buffer the sharper rise in yield post-calving than was observed for NZ70 cows (see Figure 5.8 in Chapter 5), even though  $DMI_H$  was higher for NZ90 than NZ70 cows (see Table 6.6 in Chapter 6).

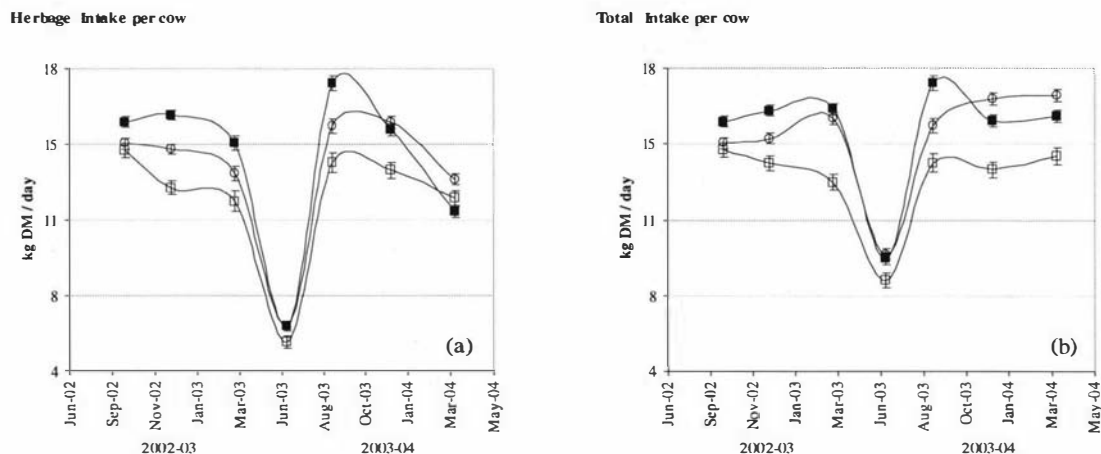
The capacity of the NZ90 strain to adapt grazing behaviour to different sward conditions is higher than for NZ70 and OS90 (Chapters 7 and 8), but higher maintenance requirements were assumed for the NZ90 and OS90 genotypes than the NZ70 cows. Thus, if  $DMI_H$  were constrained, its negative effect would be greater for NZ90 than NZ70 cows. If this constraint occurs during early lactation, both NZ90 and OS90 cows would reduce milk yield to a lower level despite their higher capacity to buffer production from body reserves mobilisation. The NZ90 cows would be less affected considering their lower yield genetic potential and higher  $DMI_H$  than the OS90, hence they would recover condition more rapidly once feed availability is improved.

The fact that the OS90 did not improve BCS at the end of lactation in 2002-03 suggests that these cows continue partitioning most of the energy ingested to milk yield. This would be determined by the difference between nutrient intake and actual energy demand of the cow, and hence also by the relatively low nutrients intake from the grazed pasture in late lactation. If the metabolic capacity of the cow to increase yield in late lactation was constrained by previous feeding, the slow increase in BCS observed would be determined by the intake being constrained relative to cow requirements, rather than by continuing partition of nutrients to milk yield.

#### **9.2.5. Daily dry matter intake**

During the lactation period in the 'system' study, the mean  $DMI_H$  was higher for NZ90, intermediate for OS90 and lower for NZ70 (measured with n-alkanes, see Table 6.6 in Chapter 6) with a trend to decline as lactation progressed for all three genotypes (Figure 9.1a). This decline was more evident in 2003-04, probably because of the increased substitution of pasture by supplement, but also due to the higher  $DMI_H$  of the NZ90 in early lactation. The decline in  $DMI_H$  at the end of the season for all three genotypes, but particularly for OS90 (Figure 9.1a), was compensated by the increment in supplement intake (Table 9.1; Figure 9.1b).

**Figure 9.1: Mean daily herbage dry matter intake (a) and daily total dry matter intake (b) estimated by n-alkanes and the <sup>13</sup>C methods during early, mid and late lactation periods in seasons 2002-03 and 2003-04 and in the dry period between seasons, of three Holstein-Friesian genotypes managed in different feeding systems.**

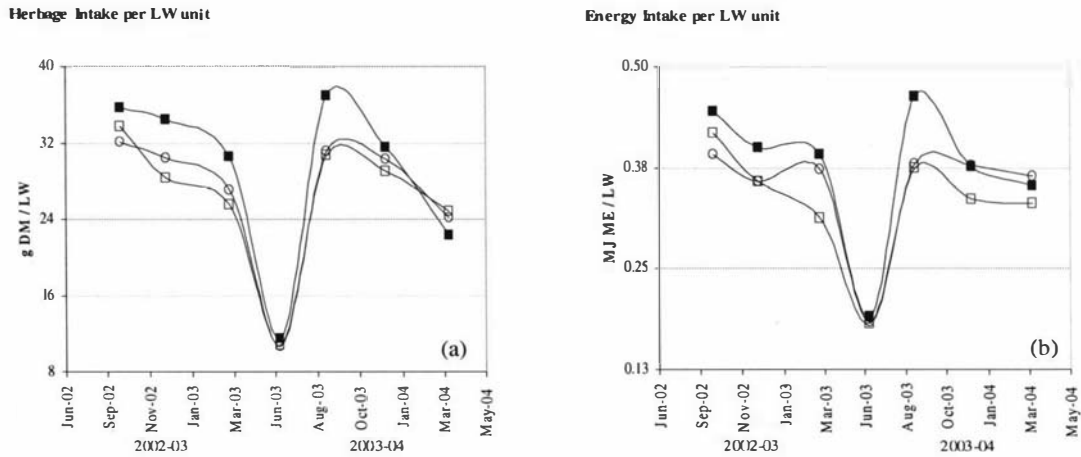


NZ70 (□), NZ90 (■) and OS90 (○). Each value is the mean of the three or four systems at which the NZ70 or NZ90 and OS90 were respectively farmed. Bars indicate standard deviation.

The mean  $DMI_H$  per unit of LW ( $DMI_H/LW$ ) of the NZ90 was higher than those of the OS90 and NZ70 in the ‘system’ study (Figure 9.2a, see Table 6.6 in Chapter 6), as a result of the higher  $DMI_H$  of the NZ90 in most measurement periods. This was consistent with the result obtained on the short sward in the ‘component’ study (see Table 8.4 in Chapter 8). Although total dry matter intake ( $DMI_T$ ) in the OS90 strain was above that recorded for the NZ90 genotype in mid and late lactation 2003-04 (Figure 9.1b), the energy intake per unit of LW was however similar between NZ90 and OS90 (Figure 9.2b). The higher energy intake of NZ90 than OS90 cows in early lactation was consistent with the lower mean BCS lost by the NZ90 strain post-calving (see Figures 5.6 and 5.7 in Chapter 5).

The differences in  $DMI_H/LW$  between NZ70 and OS90 during lactation were not significant; however, the OS90 increased the ME ingested per unit of LW at the end of the lactation due to the supplement fed (Figure 9.2b). As the NZ70 strain was assumed to have the lowest maintenance requirement per unit of LW ( $0.026 \text{ MJ ME kg}^{-1} \text{ LW}$  lower than the NZ90 and OS90 genotypes), a higher proportion of the energy ingested by this strain may be partitioned to milk yield in early lactation, reducing body reserves mobilisation. These results were consistent with the highest BCS during lactation (see Figures 5.6 and 5.7 in Chapter 5). The differences in BCS and LW observed between strains during lactation in the ‘system’ study were consistent with the different concentrations of plasma metabolites measured between strains during early lactation in the ‘component’ study (see Table 8.4 in Chapter 8).

**Figure 9.2: Mean daily herbage intake per unit of live weight (a) and metabolisable energy intake per unit of live weight (b), both estimated by n-alkanes and the <sup>13</sup>C methods during early, mid and late lactation periods in seasons 2002-03 and 2003-04 and in the dry period between seasons, of three Holstein-Friesian genotypes managed in different feeding systems.**



NZ70 (□), NZ90 (■) and OS90 (○). Each value is the mean of the three or four systems at which the NZ70 or NZ90 and OS90 were respectively farmed.

Although it is known that cows with high yield genetic potential are constrained on pasture (Kolver & Muller, 1998), to a greater extent in the OS than in the modern NZ Holstein-Friesian (Kolver et al., 2002; Kolver, 2003), the lowest  $DMI_H/LW$  of the OS90 (see Table 6.6 in Chapter 6) was substantially improved on a non-constraining sward (see Table 8.4 in Chapter 8 where the OS90 cows were capable of harvesting 21.6 kg DM day<sup>-1</sup> of herbage, 2.39 kg DM day<sup>-1</sup> more than the NZ90 cows). This increase in  $DMI_H$  was sufficient to make  $DMI_H/LW$  in the OS90 cows equal to that in the NZ90 cows, despite the heavier LW of the former strain.

### 9.2.6. Factors affecting herbage intake

It is hypothesised that the lower  $DMI_H$  achieved by the OS90 on pasture occurred because (1) metabolic and nutritional constraints interact particularly during the early lactation period, and (2) because physical constraints may limit the rate of energy intake, affecting the benefit obtained by the cow relative to energy expenditure.

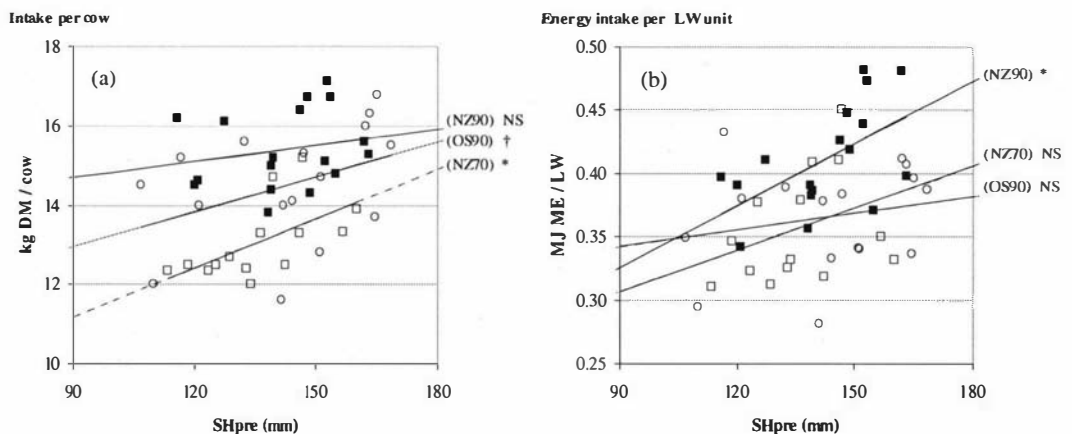
#### 9.2.6.1. Nutritional and metabolic constraints

In addition to the metabolic changes associated with a rapid rise in milk secretion after calving, the cow undergoes physical changes associated with the involution of the uterus immediately post-calving and increases in rumen capacity. These changes would represent an initial physical delay for the cow to achieve potential intake. The magnitude of the energetic constraint would be determined by the initial intake capacity relative to

the potential energy requirement. Under this constraint to DMI, the initial secretion of milk is sustained by a fast rise in reserves mobilisation (Ingvarsten et al., 1999). The capacity of the fat reserves to buffer a period of nutritional deficit would decrease with a decline in the body reserves of the cow. Thus, milk yield would be equilibrated at a lower level according with nutrients intake, depending on both the actual constraints to  $DMI_H$ , and the extent of fat reserves mobilisation. This energetic equilibrium is achieved at a different minimum threshold for body reserves depletion (see Figures 5.6 and 5.7 in Chapter 5) higher for the NZ70, intermediate for the NZ90 and lower for the OS90.

It is proposed that the rate of body reserves utilised post-calving is different between the strains and proportional to the difference between the actual body reserves of the cow and the minimum threshold for each strain, relative to the actual intake of nutrients. Thus, the rate of mobilisation of reserves would be highest immediately post-calving in all three genotypes, and would decline at different rates between genotypes, consistent with the level of reserves at calving and the minimum threshold. It was suggested that the availability of nutrients and their metabolism influence intake immediately after calving, particularly the feedback signals from the oxidation of NEFA in the liver (Ingvarsten & Andersen, 2000). It is possible that the high rate of body reserves mobilisation increases the metabolic feedback signals affecting the satiety of the cow or modifying the sensitivity to inhibitory signals from the pasture, hence, differences in the rate of reserves mobilisation between strains would affect their motivation to graze differently.

**Figure 9.3: Relationships between daily herbage dry matter intake per cow (a) and metabolisable energy intake per kilogram of live weight (b) with pre-grazing sward surface height during the lactation period on dates where cows grazed pasture only (no other feed consumed).**



LW: live weight, SH<sub>PRE</sub>: pre-grazing sward height, DMI<sub>H</sub>: herbage dry matter intake, DMI<sub>H</sub>/LW herbage dry matter intake per unit of LW, ME<sub>H</sub>: metabolisable energy consumed per unit of LW. NZ70 (□), NZ90 (■) and OS90 (○). Solid lines are linear regressions estimated for the whole lactation period. Equations are presented in Table 9.2; significance of the estimated equations are also indicated.

**Table 9.2: Linear relationships between herbage dry matter intake per day and per unit of live weight, and energy intake per unit of live weight and pre-grazing sward height, for three Holstein-Friesian genotypes managed in different feeding systems on pasture during lactation.**

Regression equation		$r^2$	RMSE	P	n
<b>DMI<sub>H</sub> vs. SH<sub>PRE</sub></b>					
NZ70	DMI <sub>H</sub> = 7.45 + 0.0413 SH <sub>PRE</sub>	0.36	0.79	0.02	14
NZ90	DMI <sub>H</sub> = 13.5 + 0.0136 SH <sub>PRE</sub>	0.04	0.99	0.44	17
OS90	DMI <sub>H</sub> = 10.3 + 0.0294 SH <sub>PRE</sub>	0.16	1.40	0.12	16
<b>DMI<sub>H</sub>/LW vs. SH<sub>PRE</sub></b>					
NZ70	DMI <sub>H</sub> /LW = 20.23 + 0.0717 SSH <sub>PRE</sub>	0.14	2.50	0.18	14
NZ90	DMI <sub>H</sub> /LW = 20.67 + 0.0955 SSH <sub>PRE</sub>	0.33	2.02	<0.02	17
OS90	DMI <sub>H</sub> /LW = 27.40 + 0.0233 SSH <sub>PRE</sub>	0.03	2.96	0.54	16
<b>ME<sub>H</sub>/LW vs. SH<sub>PRE</sub></b>					
NZ70	ME <sub>H</sub> /LW = 0.21 + 0.0011 SH <sub>PRE</sub>	0.12	0.042	0.23	14
NZ90	ME <sub>H</sub> /LW = 0.18 + 0.0016 SH <sub>PRE</sub>	0.32	0.036	0.02	17
OS90	ME <sub>H</sub> /LW = 0.30 + 0.0004 SH <sub>PRE</sub>	0.04	0.043	0.43	16

SH<sub>PRE</sub>: pre-grazing sward height (mm), DMI<sub>H</sub>: herbage dry matter intake (kg DM cow<sup>-1</sup>day<sup>-1</sup>), LW: live weight; DMI<sub>H</sub>/LW: herbage dry matter intake per LW unit (g DM kg<sup>-1</sup>LW), ME<sub>H</sub>: metabolisable energy intake per LW unit (MJ ME kg<sup>-1</sup>LW). P: probability [significance: \* = P < 0.05; \*\* = P < 0.01; \*\*\* = P < 0.001. NS = not significant.]; r<sup>2</sup>: coefficient of determination; RMSE: root mean square error; n: sample number. The linear regressions were estimated for the whole lactation period. Only dates with no supplement fed considered.

If conditions of under-nutrition are imposed, the capacity of the OS90 cows to buffer yield would decline sharply as reserves are depleted, and then the energy output reaches equilibrium at a lower yield, consistent with nutrient intake. When these cows are fed on TMR diets these mechanisms would operate similarly but the nutritional status of the cow would be however improved as a result of the higher intake of nutrients. Additionally, energy expenditure would be lower and more energy available to increase actual yield.

#### 9.2.6.2. Differences in grazing ability between Holstein-Friesian genotypes

The correlations between DMI<sub>H</sub>/LW and pre-grazing sward height (SH<sub>PRE</sub>), the reduction in height between pre- and post-grazing measurements (H<sub>R</sub>), and the bulk density (B<sub>D</sub>) of the pastures grazed in the ‘system’ study, were larger for the two NZ strains than for the OS90 genotype (see Table 6.7 in Chapter 6). This suggests that the effect of the structure of the sward on DMI<sub>H</sub> differed between genotypes. However, all three genotypes increased DMI<sub>H</sub>/LW when grazed on tall high quality pastures in the ‘system’ study (Chapter 6), in agreement with the results obtained in the ‘component’ study (see Table 8.4 in Chapter 8), probably because in the taller swards bite penetration is initially less constrained.

It is apparent that OS90 cows preferentially grazed the upper and leafy strata of taller pastures, possibly constrained by a decline in quality and increase in B<sub>D</sub> that might have affected bite penetration. It is possible that the increased contrast between upper and lower layers of mature pasture (Hoogendoorn *et al.*, 1992) would negatively affect the

capacity of the cows to adapt its ingestive behaviour to the fast changes in the sward occurring for cows taking large bites and depleting the sward faster, in particular, the shearing resistance of the herbage to be severed would increase as the pasture is grazed down.

Nevertheless, under the different management utilised in the ‘system’ study, cows that achieved higher bite penetration and depleted the sward further to a lower post-grazing residual, might not necessarily have achieved greater herbage intake. This is because the total amount of herbage removed at each grazing is also affected by the  $B_D$  of the layer removed and the area grazed per cow; in addition, by the physiological state of the cow and its motivation to graze. Furthermore, these results could be also confounded by changes in LW and metabolism during the lactation.

Despite these confounding effects, the results of the ‘system’ study suggest that the two NZ strains have greater flexibility and grazing capacity to perform under a range of different sward conditions, particularly the NZ90 strain with higher daily requirements. In addition, the different responses reported between ‘system’ and ‘component’ studies suggest that differences in the vertical structure of the swards between experiments existed, which constrained to a greater extent the intake of the OS90 cows grazing in the ‘system’ study.

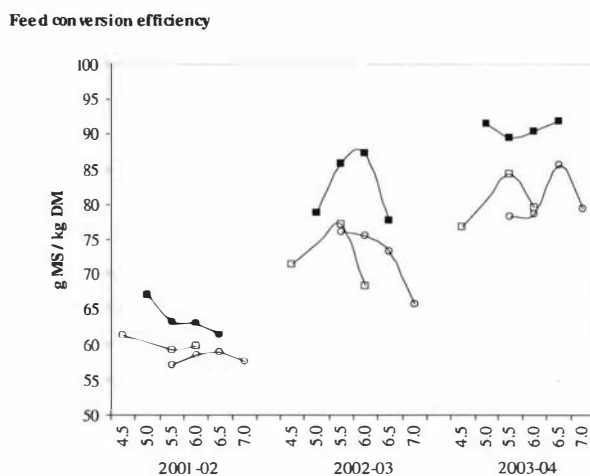
Thus, the  $DMI_H$  of cows fed only pasture was higher for the NZ90 than for the NZ70 and OS90 genotypes during lactation (see Table 6.6 in Chapter 6), with a non-significant decline in  $DMI_H$  as  $SH_{PRE}$  decreased. The opposite was measured for the other two strains (Figure 9.3a and Table 9.4). Thus, within the range of swards grazed in the ‘system’ study, the difference in  $DMI_H$  between the genotypes decreased as sward height increased (Figure 9.3a and Table 9.2); in contrast, the energy ingested per unit of LW increased to a greater extent particularly for NZ90 cows than for both NZ70 and OS90 (Figure 9.3b and Table 9.4). This suggests that although  $DMI_H$  increased, the energetic status of the OS90 cows did not improve to a similar extent to that measured for NZ90, and even compared to the NZ70 strain, because the energy status of the OS90 cows would have been lower than for NZ70 cows due to its higher maintenance requirement, which is consistent with the different mean BCS during lactation between these genotypes.

It is also apparent that the two NZ strains, but particularly the NZ70 genotype, have greater ability to select herbage of higher quality, probably because of an improved capacity toprehend small bites (Gordon & Illius, 1988). Furthermore, the size of the jaw (see Table 8.5 in Chapter 8) would also determine the size of the particles ingested and the requirement for further rumination; in addition, intake rate would be affected (see Tables 7.4 and 7.5 in Chapter 7 and Table 8.4 in Chapter 8). It was demonstrated that the

reason for the higher  $DMI_H$  obtained with NZ90 cows was that these cows adapted their grazing behaviour to a greater extent than OS90 and maintained a higher  $DMI_H/LW$  under different grazing conditions. They were also lighter hence at similar  $DMI_H$  they achieve higher  $DMI_H/LW$  than their OS90 counterparts.

In contrast, the OS90 ingest more dry matter per bite, achieve faster  $DMI_H$  rates and reduce energy expenditure per kilogram of dry matter ingested in swards where the depth of the bite is not constrained. This unconstrained situation is more likely to exist in taller swards that have previously been grazed intensively, where new leaves dominate the top layer and only a shallow layer of pseudostems and dead material is present at the base of the pasture, similar to that obtained in the ‘component’ study (Hoogendoorn et al., 1992).

**Figure 9.4: Mean whole season feed conversion efficiency of three Holstein-Friesian genotypes managed in systems at different feed allowance during seasons 2001-02, 2002-03 and 2003-04.**



NZ70 (□), NZ90 (■) and OS90 (○). Values plotted are systems efficiencies during each season, mean of the three seasons (2001-02, 2002-03 and 2003-04) and for season 2003-04 were presented in Table 9.1. The ‘x’ axis indicates the nominal total feed offered (NTF<sub>0</sub>) at which the systems were managed, expressed in tonnes of DM per cow per year.

It has been suggested that  $DMI_H$  would be maximum when DHA is twice the potential  $DMI_H$  (Combellas & Hodgson, 1979). The evidence from the ‘component’ study agrees with this suggestion, even for cows of high yield genetic potential. The value of  $41 \text{ g kg}^{-1} \text{ LW}$  measured on the tall sward (see Table 8.4 in Chapter 8) is consistent with the values reported for cows feed ‘ad-libitum’ on TMR diets (Kolver & Muller, 1998; Kolver *et al.*, 2002). The value of  $35 \text{ g kg}^{-1} \text{ LW}$  measured for the NZ70 genotype grazing the tall sward can be also considered to be close to potential. Thus, a different DHA should be allocated to each genotype to maximise intake according to the differences in mature LW between strains (Table 9.1)(Buckley et al., 2000). Nevertheless, a high value of DHA does not

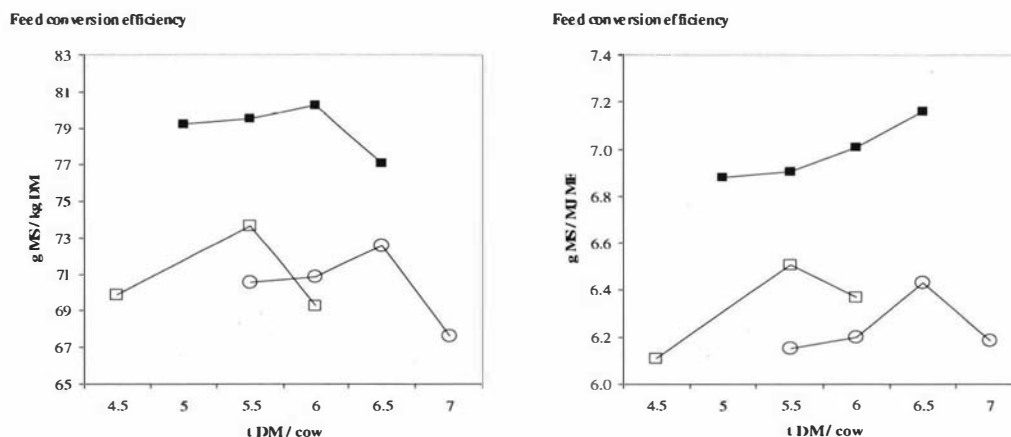
ensure that the highest  $DMI_H$  will be obtained if sward structure constrains bite penetration (see Table 8.4 in Chapter 8).

## 9.2.7. Productive efficiencies

### 9.2.7.1. System efficiencies over the whole lactation

The mean feed conversion efficiency (FCE) in the ‘system’ study ( $73 \text{ g MS kg}^{-1}\text{DM}$ ; Table 9.1) is consistent with previous studies (Penno *et al.*, 1996; Penno *et al.*, 1999). This increased from the first to the third season for all three genotypes because the productive performance of the cows increases with age (Figure 9.4). The mean FCE was greater for NZ90 than NZ70 and OS90 strains (mean 11% - 13% higher respectively in the three seasons of the study and 15% - 8% during 2003-04; Table 9.1 and Figure 9.5a). The efficiency of MS production was also higher for NZ90 when expressed per unit of metabolisable energy ingested (Figure 9.5b) and per unit of LW (Table 9.1).

**Figure 9.5: Mean whole season feed conversion efficiency expressed per unit of dry matter consumed (a) or per unit of metabolisable energy ingested (b) of three Holstein-Friesian genotypes managed in systems at different feed allowance.**



NZ70 (□), NZ90 (■) and OS90 (○). Values plotted are mean of three seasons (2001-03, 2002-03 & 2003-04). The ‘x’ axis indicates the nominal total feed offered (NTF<sub>0</sub>) at which the systems were managed, expressed in tones of DM per cow per year.

It is considered that the extra feed brought into a seasonal pasture-based dairy system causes both an immediate effect on the yield of the cows and a ‘carryover’ effects over the system (Bryant & Trigg, 1982; Thomson & Holmes, 1995; Penno *et al.*, 1998). The decline in FCE observed at the highest FA for all three genotypes in 2002-03 and in NZ70 and OS90 in 2003-04 show that the increase in the amount of feed consumed was proportionally greater than the increment in yield obtained, indicating that the effect of the additional feed was not completely captured by the system. In practice, the extent of

the response would depend on the system's capacity to capture these effects, hence, on management decisions.

### 9.2.7.2. Response to extra feed during different stage of lactation

During the lactation period of the 'system' study the efficiency of MS production was estimated from the data collected from all periods when DMI was measured by n-alkanes (see Tables 6.6 and 6.8 in Chapter 6). The range of mean FCE of the systems was between 100 – 112 g MS kg<sup>-1</sup>DM (Figure 9.6a) with the highest value recorded for OS90 cows (109 g MS kg<sup>-1</sup>DM) and the lowest in NZ70 (104 g MS kg<sup>-1</sup>DM; Table 9.3). The differences in the mean FCE between genotypes were small considering the large differences in yield genetic potential. The two NZ strains showed a steady increase in efficiency as FA increased, but the increase in the OS90 was discontinuous and suggests that the timing of the supplementation could affect the capacity of the cows to transform the extra feed consumed into yield (Figure 9.6a).

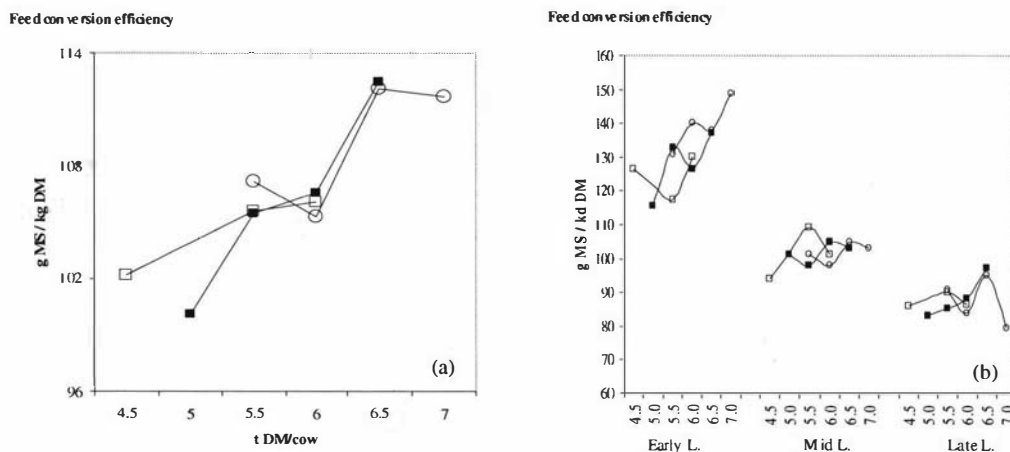
The short-term efficiency in the 'system' study decreased as lactation progressed averaging 131, 102 and 88 g MS kg<sup>-1</sup>DM during the early, mid and late lactation periods respectively (mean seasons 2002-03 and 2003-04 and all FA; Figure 9.6b and Table 9.3), in agreement with Kolver et al. (2002). However, the OS90 showed the highest FCE in early lactation and the NZ70 the lowest. Differences between strains are consistent with the different loss in BCS (see Figures 5.6 and 5.7 in Chapter 5), in contrast with the results from Kolver et al. (2002) and also different from the 'component' study (Table 9.3). The difference between systems or experiments can be attributed to interactions between the use of extra feed, cow condition and metabolic status, depending on previous cow nutrition, and herbage availability when grazing pasture.

**Table 9.3: Mean feed conversion efficiency in early, mid and late lactation in the 'system' study (means of seasons 2002-03 and 2003-04) and in early lactation for the 'component' study (mean of two experimental periods). Dry matter intake measured by the n-alkanes method.**

		Genotype		
		NZ70	NZ90	OS90
<b>'System' study</b>				
Early lactation	g MS kg <sup>-1</sup> DM	124.1	128.1	139.6
Mid lactation	g MS kg <sup>-1</sup> DM	101.6	101.8	101.8
Late lactation	g MS kg <sup>-1</sup> DM	87.7	88.5	87.3
Mean	g MS kg <sup>-1</sup> DM	104.5	106.2	109.6
<b>'Component' study</b>				
Early lactation – Short sward	g MS kg <sup>-1</sup> DM	121.1	130.2	123.6
Early lactation – Tall sward	g MS kg <sup>-1</sup> DM	113.0	102.1	83.4

MS: milksolids. Efficiencies are not adjusted for any change in live weight and body condition score during each period they were estimated.

**Figure 9.6:** Mean feed conversion efficiency during the whole lactation (a) and at each stage of lactation (b) of three Holstein-Friesian genotypes managed in systems at different feed allowance.



NZ70 (□), NZ90 (■) and OS90 (○). In (a) and (b) the 'x' axis indicates the nominal total feed offered (NTF<sub>0</sub>) at which the systems were managed, expressed in tonnes of DM per cow per year.

As expected, FCEs estimated for the lactation period were higher than that for the whole season, however while the OS90 strain showed a trend for the highest efficiency during lactation, the NZ90 had the highest efficiency value for the whole season (Tables 9.1 and 9.3). The efficiency obtained for the whole season would depend on the capacity of the systems to dilute the maintenance requirements of the cows through a greater production. Although the capacity of the cows to partition energy from body reserves increased the efficiency obtained during the lactation period for OS90 cows, the amount of feed required in the non-lactating period to recover the BCS lost during lactation depressed the efficiency value estimated for the whole season. In contrast, the NZ70 required less amount of feed to recover BCS but had lower production performance during lactation.

### 9.2.7.3. *Timing of the extra feeding*

It is known that the energy required by the sharp rise in milk yield post-calving cannot be totally sustained by the energy available from the feed plus body reserves in grazing dairy cows. However, if body reserves are already mobilised at the highest metabolic rate possible and the nutritional constraint important, the cow has a reduced capacity to sustain production and would reduce energy output at a lower milk yield. In addition, if the nutritional constraint persisted, the buffer capacity of the cow to sustain yield from body reserves would decline as reserves are depleted, gradually reducing the feed 'demand' of the cow (Penno et al., 2001). When body reserves are at their minimum, yield may reach equilibrium at a level according with nutrients intake. It is suggested that any nutritional constraint to DMI that the cow suffers immediately post calving would

affect the actual yield of the cow, its relative energy deficit, and the shape of the lactation curve (Friggens *et al.*, 1998). This would affect the immediate and future response of the cow to extra feed, and the capacity of the system to capture carry-over effects.

On pasture, once yield was equilibrated with the actual nutrient intake, any increase in  $DMI_H$  would improve yield according with the metabolic capacity of the cow, however, the immediate response of the cow to extra feed would be low when the relative energy deficit is low (Penno *et al.*, 2001). If yield cannot be improved, any excess of energy ingested would be partitioned to body reserves. Thus, it is apparent that to increase the response to extra feed, the actual potential capacity of the cow must be maintained close to genetic potential. Thus, the immediate response to extra feed would be different according with (1) the type of feed offered, as the increase in yield is relative to the amount of ME supplied (Penno *et al.*, 1998), (2) if most of the additional feed is pasture the response would vary depending on the grazing capacity of the cow utilised and the condition of the sward grazed, (3) the previous feeding of the cow and its effects on the actual yield potential and relative energy deficit; and (4) the physiological state of the cow.

It was demonstrated that  $DMI_H$  could be increased if the cows graze highly digestible and readily available pasture in early lactation (Chapter 8), but this occurred when body reserves were depleted to minimum values and the metabolic constraints to DMI were not fully operating. It is apparent that the different intake of nutrients that occurs immediately after calving between cows fed on pasture or TMR determines the different lactation peak and shape of the lactation between diets. Hence, it may be possible to increase nutrients intake in pasture-based systems by feeding supplements soon after calving. If this occurs, actual yield potential and the shape of the lactation curve would be improved, with a residual effect over the whole lactation if the high level of nutrition required by the cow can be maintained.

Cows maintaining a higher yield potential would have an increased relative energy deficit on constraining sward conditions, then will improve the response to any extra feed given subsequently. This probably would require a continued use of supplement during the whole lactation, or at least until the metabolic constraints associated with body reserves mobilisation are reduced. As yield and requirements would be improved and the relative energy deficit increased, substitution rate would be reduced and PU improved, despite the CSR is lower (see Figure 5.3 in Chapter 5). The requirement for supplement would be greater for the OS90 farmed on a pasture-based system because the higher genetic yield potential of these cows, metabolic constraint to intake post-calving and lower grazing ability.

As most of the energy ingested by OS90 cows plus most of the energy reserves available at calving are partitioned to milk yield during early lactation and kept low until the end of the lactation, the response of the OS90 cows to extra feed would be low if the capacity for an immediate response is affected by previous nutrition. Considering the differences in the metabolic response between genotypes in the present thesis and the apparent interactions between cow metabolism and nutrition, it is possible that systems managed with different genotypes require a different strategy of supplementation during lactation, to correspond with their different energetic requirements and constraints to perform on pasture. If a different timing of supplementation influences the size of the response obtained by the NZ90 and OS90 genotypes, the immediate response plus ‘carryover’ effects would be increased and, as a result, the efficiency of the system enhanced.

#### **9.2.8. Implications of the use of different Holstein-Friesian genotypes in pasture – based systems**

It is clear that the intake of nutrients is one of the main limitations for a cow of high genetic yield potential managed on pasture. The different mobilisation of body reserves and ability to obtain nutrients from the grazed pasture in relation to the maintenance requirement of the genotypes were probably the main reasons that determined the differences observed in productive performance and efficiencies between the strains compared in the present thesis. A better understanding of the metabolic mechanisms sustaining the rate of body reserves mobilisation and links with intake and reproductive performance (not analysed here) would increase the possibility of selecting cows with an improved grazing aptitude.

Considering that about 80% of the herbage produced per hectare can be grazed, the productivity of intensive seasonal pasture-based systems would be determined by (1) the grazing ability and  $DMI_H$  of the cows utilised in these systems, (2) the capacity to increase herbage production, and (3) the proportion of herbage in the total diet of the cows, determined by the availability of low cost supplements. The use of strategic supplementation positively affects cow and system production when the level of feed provided by the pasture is low relative to the feed requirement of the cows. It was suggested that this would occur when the basal feeding level has been high but declining, and insufficient to maintain the current yield of the cows (Penno, 2002).

The mean monthly herbage accumulation rate (HAR) increased from the start of calving (early August) while favourable growing conditions continued until December each season, even though pasture quality showed a slow decrease during the same period (see Figures 5.1a and 5.2a). The fact that a high proportion of the seasonal production of these systems was achieved during the first half of the lactation (see Appendix V-11 and V-12

and Figure 5.8 in Chapter 5), suggest that any improvement in management would greatly affect total season results; in addition, it would increase the opportunity to capture ‘carryover’ effects in late lactation. In contrast, pasture quality showed a slow decline between August and December that would contribute to build up a layer of low quality material at the base of the pasture. This would affect  $DMI_H$  further particularly in OS90 cows due to their lower grazing capacity and flexibility to adapt grazing behaviour to different sward structure. The sharp decline in daily HAR and herbage quality from December onwards suggests that constraints to ingestion would increase further during the summer period, however, the energy requirements of the cows decrease as lactation progresses. This would increase the opportunity for behavioural compensation, provided that enough available herbage is allocated to the cows.

Cows of all three genotypes improved  $DMI_H$  when grazing pastures that did not constrain bite penetration (see Chapter 8), particularly the OS90 that achieved the highest  $DMI_H$  when grazing a taller and leafy sward with low proportion of pseudostems in the base of the pasture at sufficient DHA. Even though the NZ90 strain demonstrated a higher grazing ability and flexibility to perform well under different grazing conditions, metabolic and physical constraints to ingestion can also limit  $DMI_H$  and yield in this strain during early lactation. It is apparent that this does not occur with the NZ70 cows under average grazing conditions on pasture-based systems due to the lower yield genetic potential of this strain.

Thus, it would be possible to increase intake post calving if cow nutrition is improved, as a result the actual yield, energy demand and relative energy deficit of the cow would increase. Hence, it is expected that the immediate response to extra feed would also increase, particularly for OS90 cows due to their higher yield genetic potential. As these cows partition most of the energy available to milk yield during lactation, it is expected that ‘carryover’ effects would be lower compared to NZ90 cows. A similar difference would be expected between the two NZ genotypes.

Furthermore, there is a possibility to increase  $DMI_H$  to a higher level, even for cows of the OS90 strain grazing pasture alone during the early lactation period, by reducing pasture constraints to herbage ingestion. This would occur even though it is probable that with no supplementary feed both the OS90, and to a lower extent the NZ90 strain, would equilibrate actual production to a proportionally lower level than expected, according to their genetic yield predicted. In contrast, the NZ70 strain seems to have buffered this constraint to a greater extent by mobilising body reserves; however, as result of their lower genetic yield potential, lower maintenance requirement and relatively higher  $DMI_H$  capacity, these cows would show a comparatively lower reduction in production than the two high-merit genotypes under a nutritional constraint in early lactation.

It is evident that, to increase FCE with the OS90 genotype, the higher maintenance requirements of these cows have to be diluted by a higher yield. This would be partially achieved by extending lactation length, whereas an opportunity to increase daily yield would exist by targeting a higher yield at lactation peak, by using supplements. This strategy would require a higher level of supplementation than used at the highest FA in the 'system' study, fed over most of the lactation and dry periods as occurred on a TMR diet (Kolver *et al.*, 2002). As a result, daily yield and requirements would be increased and substitution rate reduced. This strategy could also be used for the NZ90 strain.

Hence it is possible to visualize a range of dairy systems, corresponding with the environmental limitations to grow more herbage, the possibility of reducing fluctuations in pasture growth rate between seasons, and the availability of cost-effective supplements. At one extreme the NZ70 cow would fit better a 'forage only' low-input but intensive pasture-based system. On the other extreme, the OS90 would enable productive performance to increase with more extra feed in a 'high-input' intensive pasture plus supplement system, where extra feed would be routinely utilised and the main limitation would be reproductive performance. If high input systems are economically feasible they should not necessarily be seasonal. This would decrease the pressure on reproductive performance, although not eliminate the problem, while cow survival would be still an issue. In addition, it is expected that systems that use a high percentage of supplement in the total diet of the cows would be economically more affected by changes in the price of the supplement. In the middle, the NZ90 genotype would cover most pasture-based situations considering the average conditions of NZ dairy farms (Silva-Villacorta *et al.*, 2005; Hedley *et al.*, 2006). The increased productivity of these systems would be consistent with the capacity to increase the amount of herbage produced at farm level and a more strategic use of extra feed.

It is also possible to think about systems that integrate genotypes with opposite characteristics. For instance, considering the contrasts in grazing capacity and requirements between OS90 and NZ70, and the improvement obtained in the structure of the pasture by grazing close to ground level (Hoogendoorn *et al.*, 1992), a flexible grazing system that combines a 'leader' group of supplemented OS90 cows with a 'follower' group of NZ70 cows (Mayne *et al.*, 1988), at least during the spring period, would improve sward condition and  $DMI_H/LW$  of the OS90 cows and simultaneously achieve high level of pasture utilisation. Although in principle this option seems to be complex in relation to the grazing management required, and its applicability is probably restricted to the early lactation period, it is a valid example of an improved combination of feed availability and cow genotype to improve system efficiency.

### 9.3. CONCLUSIONS

In this context, the two NZ strains were better adapted to seasonal pasture-based systems than their OS90 counterparts. The NZ90 achieved higher productive performance and efficiency than NZ70 by combining larger body reserves mobilisation and  $DMI_H$  capacity on pasture, however total lactation yield for the high-merit genotypes was lower than that reported on TMR diets. This probably occurred because despite the increased FA at which the genotypes were farmed, all the systems were managed on pasture during the early lactation period. This suggests that both production and efficiency can be further improved by strategic supplementation. In contrast, yield performance for NZ70 cows was probably close to potential within the range of FA utilised.

The higher productive performance of the NZ90 genotype was determined by a lower body reserves mobilisation relative to yield performance, sustained by the improved capacity of these cows to maintain  $DMI_H/LW$  under a range of different sward conditions. In contrast, under similar sward conditions, the  $DMI_H$  and to a greater extent  $DMI_H/LW$  of the OS90 were constrained, when compared with the NZ90. Although body reserves mobilisation was higher for NZ90 than NZ70, the former strain showed higher  $DMI_H/LW$  than the NZ70 counterparts.

The  $DMI_H$  achieved by the OS90 cows was below that of the NZ90 because of the lower capacity of the former genotype to accommodate grazing behaviour to swards of different structure, nevertheless the OS90 cows grazed effectively on leafy and taller swards that had been previously grazed intensively. In this sward, bite penetration was not constrained and most of the herbage offered was available, hence the OS90 cows achieved greater  $DMI_H$  than the two NZ however  $DMI_H/LW$  was similar to the NZ90 strain, indicating a similar energy status above maintenance between the OS90 and NZ90 strains.

If the OS90 cow cannot reduce the size of the bite to the same extent as the two NZ genotypes, it would be more difficult for this strain to increase  $DMI_H/LW$  in short swards. In addition, grazing a taller sward requires a further reduction in bite area for the OS90 to compensate for the increased shearing resistance of the herbage while grazing down. Hence, even though OS90 cows are able to maximise bite size when bite penetration is not constrained, it seems that they are not so well able to reduce bite size in order to graze short swards in which bite penetration is restricted.

Differences between genotypes in energy expenditure during grazing may exist. Grazing time increases when herbage availability decreases, but at similar  $DMI_H$  ruminating would also be extended if low quality material were ingested, hence the compensation

capacity of the cow improves when the digestibility of the diet increases and the rumination requirement is reduced. It is usually considered that cows grazing at a faster rate reduce energy expenditure per kilogram of DMI compared to cows grazing at slower rates, but a cow selecting highly digestible bites would increase the benefit per unit of dry matter ingested, with a positive effect on the subsequent cost of rumination and digestion. In addition, by performing small bites the size of the particles ingested is reduced, which would affect rumination independently of the quality of the bite prehended.

These results quantify the progress made in NZ during the last thirty years by genetic improvement. However they also show that the NZ90 strain should be farmed at higher levels of feed than the former NZ70 to achieve this higher yield performance. Furthermore, it is proposed that yield performance in the NZ90 strain could be improved even further by strategic supplementation.

The OS90 strain has demonstrated its inability to perform satisfactorily on pasture-based systems, despite the fact that its productive performance improves with feeding level. It is apparent that to express their higher genetic yield potential these cows should have higher proportions of supplement in their diet. The fact that  $DMI_H$  in the OS90 can be improved when these cows graze unconstrained sward conditions, indicate that fine-tuning management would increase yield performance of these cows even on pasture. Considering the different metabolism and grazing capability of the genotypes, the different strains utilised in the present thesis would probably need different supplementation strategies to take advantage of their different genetic yield potential when managed on pasture-based systems.

It seems that years of genetic selection for yield under feed-lot systems has produced a highly productive and heavy cow, which has higher maintenance requirements and lower flexibility to perform under different grazing managements, particularly at constrained feeding allowance where herbage availability is reduced, because of its lower grazing ability. The reduced flexibility of these cows is partially determined by the large size of the mouth, scaling with LW, which limits their ability to take the small bites required to prehend the herbage in short swards or to maintain the size of the bite when grazing the bulky and low quality herbage of the lower strata of the pasture when they graze taller swards.

The NZ90 cows are also heavier than NZ70; hence the prehension apparatus is also larger, however, it seems that NZ90 cows have developed other metabolic and behavioural characteristics associated to a pasture-based diet, and associated consequences, as a result these cows are more able to adapt their grazing behaviour under a range of different feeding conditions than both NZ70 and OS90 genotypes. The

identification of the mechanisms associated with this improved capacity of the NZ90 strain to perform on pasture requires further attention. This would probably lead to further improvements in the grazing ability and performance of the NZ90 on pasture-based systems, with particular emphasis on critical pasture and management requirements.

#### 9.4. REFERENCES

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## APPENDIX VI

### PUBLICATIONS

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