

Effect of nutritional management during the first 42 days post calving on lactational performance, uterine health, and subsequent reproductive performance post partum

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STRUCTURE OF REPORT

This report begins with an Executive Summary which provides the background to the research, details of the work undertaken, key findings, and practical implications.

The main body of the report describes the study.

The report finishes with a summary of key presentations and publications which have arisen from this work.

TABLE OF CONTENTS

1
5
6 6
6
11
12
12
3
6
6
17
22
26
29
33
33
34
34
35
37
38
38
39
11
ю 16

	Farmers/Industry Meetings and Press Releases	46
	Visitors to Hillsborough	46
AI	PPENDIX 1:	48
AI	PPENDIX 2:	50

EXECUTIVE SUMMARY

- The high milk production potential of the modern dairy cow frequently results in excessive and prolonged periods of negative energy balance (NEB) during early lactation. The most common approach to reduce NEB is to increase energy intake by increasing the proportion of concentrates being offered. However, offering diets containing high levels of concentrates can lead to rumen acidosis, impaired rumen function and reduced intakes, with the latter exacerbating NEB.
- Introducing concentrates into the diet of freshly calved cows at a slower rate is likely to improve rumen function. Furthermore, dietary crude protein (CP) content is likely to be reduced through offering a lower concentrate diet in early lactation. Offering a lower protein diet (14.5%) has been shown to reduce milk output with no adverse effect on dry matter intake, and thus improve cow energy status (Law *et al.*, 2009).
- The objective of this study was to compare two very different strategies by which to introduce concentrates into the diet of dairy cows in early lactation, namely a rapid build-up (RBU) or a delayed build-up (DBU) strategy.
- The study involved 60 multiparous Holstein Friesian dairy cows. From calving onwards, cows were offered a basal diet (via feeder wagon) containing 35% concentrate and 65% forage on a dry matter (DM) basis (150 g crude protein / kg DM and 12.0 MJ metabolisable energy (ME) / kg DM). Animals allocated to RBU were offered 2.0 kg of lactational concentrate on day-one postcalving via an out-of-parlour feeder (OPF) with 0.5 kg increments during the subsequent 10-day period, giving a maximum concentrate allocation of 7.0 kg / day. Animals allocated to DBU received no additional concentrate until day 28 postcalving. From day 28 to day 42 postcalving, these animals were offered a step-wise increase of 0.5 kg / day until they reached a maximum of 7.0 kg / day via OPF. Once these concentrate feed levels had been achieved, diets were designed to have a crude protein and metabolisable energy content of 180 g / kg DM and

12.4 MJ / kg DM, respectively. Cows remained on these two dietary treatments until day 150 of lactation.

• Concentrate build-up strategy had a significant effect on forage dry matter intake. A delayed concentrate build-up strategy significantly increased forage intake in early lactation due to reduced concentrate levels in the diet. However, after maximum concentrate allocation (day-42 postcalving) animals on the DBU treatment continued to have a significantly higher forage intake than animals on the RBU treatment until week 17 of lactation (Figure A).



Figure A: Total forage intake (kg DM / d) of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation.

Milk production was significantly lower for cows on the DBU treatment during weeks 3, 4, 5 & 6 of lactation compared to those allocated to the RBU treatment.
 After week 6, dietary protein levels on both treatments were approximately 18%

and from this point onwards there was no significant difference in milk yields between the two treatments. There was no significant effect of concentrate build-up strategy on milk composition during the experiment.

- Concentrate build-up strategy had no significant effect on liveweight, body condition score or energy balance parameters; however, animals allocated to a DBU treatment tended to have an improved daily energy balance during the first 150 days of lactation.
- Animals allocated to the DBU treatment had significantly lower plasma urea levels during the first 84 days of lactation, a reflection of the lower dietary protein levels in early lactation. Reducing the dietary concentrate level also reduced the overall starch concentration in the diet which significantly reduced plasma glucose levels.
- In early lactation, 44.8% of animals on the RBU treatment were treated for a dilated abomasum, compared to 17.9% on the DBU treatment. In this experiment, the term "dilated abomasum" was used to describe animals which had excess gas in the abomasum, causing movement up the flank, but which did not display any other typical symptoms of a displaced abomasum such as dehydration, diarrhea, ketosis etc. This finding suggests that animals allocated to the DBU treatment had improved rumen function, as was also evident through improved forage intakes.
- Concentrate build-up strategy had no significant effect on the incidence of mastitis or metritis.
- Concentrate build-up strategy had no significant effect on any of the conventional reproductive variables.
- There were no significant treatment effects on the interval to the onset of luteal activity, although there was a numerical difference between treatments (33.0 vs. 44.8 days for RBU and DBU treatments, respectively). This may have been due to the different dietary starch concentrations between treatments. High starch levels will favour elevated plasma insulin concentrations which will promote

steriodogenesis, enhancing ovarian follicle development and reducing the interval to onset of luteal activity.

- Animals on the RBU treatment had a significantly longer inter-ovulatory interval than animals allocated to the DBU treatment. Furthermore, the average inter-ovulatory interval of animals on the RBU treatment was longer than what is generally deemed optimal for good fertility.
- In summary, adopting a delayed concentrate build-up strategy in early lactation improved forage intake and had no detrimental effect on milk production. Delaying concentrate build-up also improved rumen function and tended to improve the energy status of the cow, but had no major effects on fertility performance.
- This study has examined an area that is currently of huge interest to the Northen Ireland dairy sector. This is an area where additional research may be required to provide further information on the responses identified within this study.

INTRODUCTION

Intensive genetic selection has resulted in a dramatic increase in the milk production potential of the modern Holstein-Friesian dairy cow. However, the observed increase in milk production in early lactation has not been matched by a proportionate increase in energy intake (Veerkamp et al., 1995; Ingvartsen et al., 1999). The resulting energy deficit, better known as negative energy balance (NEB), is manifest in the mobilisation of body tissue reserves. Severe and prolonged periods of NEB can predispose the dairy cow to metabolic disorders, immunosuppression, reproductive failure, and behavioural abnormalities, all of which contribute to a decline in the cow's general well-being (Nielsen, 1999). Reducing NEB in early lactation is difficult in cows of a high genetic merit for milk production. The most common approach to reduce NEB is to increase nutrient intake by increasing the nutrient density of the diet. This can be achieved by improving forage quality (high digestible grass silage/inclusion of maize silage), or by increasing concentrate feed levels. Although the latter approach has been widely adopted, offering high concentrate diets, especially in early lactation, can lead to subacute ruminal acidosis (SARA), or in extreme cases to rumen acidosis. Acidosis occurs when the rumen pH falls below 5.6 (normal pH is 6.5) and results in impaired rumen function. In acute cases, normal rumen movement is reduced and fibre digestion decreases, subsequently depressing appetite and milk production. If this is left untreated, the changes in rumen pH alters rumen flora, with acid-producing bacteria becoming dominant. These bacteria produce more acid, which is absorbed through the rumen wall, causing metabolic acidosis. Introducing concentrates to the diet of freshly calved cows at a slow rate is likely to improve rumen function. In addition, there is some anecdotal evidence that a slow build-up in concentrate feed levels post-calving may reduce the extent of NEB experienced by the cow by delaying peak milk yield until closer to the time of maximum dry matter intake.

To address this issue a study was conducted to compare two very different strategies by which to increase concentrate feed levels in early lactation, either a rapid build-up (RBU) or a delayed build-up (DBU) strategy. The DBU strategy was designed to slow the rate of increase in milk production in early lactation. Previous work conducted at this institute highlighted that reducing milk production in early lactation can improve the energy status of the cow and reduce body reserve mobilisation (Law et al., 2009). In

the latter experiment, a reduction in milk production was achieved by decreasing the crude protein content of the diet by lowering the concentrate crude protein level. Dietary crude protein supply is a key driver of milk production as the cow has a limited capacity to rely on body protein reserves to maintain a high level of milk production (Oldham, 1984). In the current study, dietary protein levels were reduced by reducing concentrate inclusion levels in the diet. One potential benefit of the latter approach is higher forage intakes in early lactation, which may improve rumen stability and total dry matter intake.

MATERIALS AND METHODS

Animals and Housing

This experiment involved 60 multiparous Holstein Friesian dairy animals (mean parity, 3.2), calving between 7th September and 20th December 2009 (mean calving date, 20th October). Three weeks prior to calving (close-up dry period), animals were housed as a single group in a free stall house with slatted flooring. Following calving, animals were placed in a single lactating cow group and were kept as a single group in a free stall house with concrete and slatted flooring. The cubicle to cow ratio was $\geq 1:1$ at all times, thus meeting the recommendations of FAWC (1997). Cubicles (2.20m x 1.25m) were fitted with rubber mats and bedded with sawdust thrice weekly. Concrete passageway floors were scraped a minimum of four times daily by an automated system. Lights were left on in the cow house at all times.

Experimental Design, Diets and Feeding

In the close-up pre-calving period (-21 d prior to expected calving date until calving), animals were offered a mixed ration consisting of 50% grass silage, 20% maize silage, and 30% chopped straw (DM basis), with 120 g of dry cow mineral and 50 g of Calcined Magnesite added to the mixed ration per cow per day. In addition, additional concentrates were offered via an out-of-parlour feeder (OPF); 2.0 kg of concentrate in

week -3 precalving, 3.0 kg in week -2, and 4.0 kg in week -1 precalving. The dry cow concentrate composition is detailed in Table 1.

Postcalving, all animals were offered ad libitum access to a basal ration containing 35% concentrate and 65% forage. The forage component consisted of 60% grass silage and 40% maize silage (chemical composition in Table 2). An additional 1.0 kg of concentrate was offered in the parlor / day; 0.5 kg a.m. and 0.5 kg p.m. Cows were also allocated 200 g of Alkacarb (sodium bicarbonate) and 250 g of barley straw / head / day in the basal ration. This basal diet was designed to supply 12.0 MJ metabolisable energy / kg DM, 150 g crude protein / kg DM and 160 g starch / kg DM. Two concentrate build-up strategies were initiated at calving; a rapid build-up (RBU) of concentrates, best representing commercial practice, and a delayed build-up (DBU) of concentrates. Concentrate build-up was acheived via an OPF. Animals allocated to RBU were offered 2.0 kg of lactational concentrate (Table 1) on day-1 postcalving, with concentrates increased by 0.5 kg increments during the subsequent 10-day period, so as to achieve a maximum concentrate allocation of 7.0 kg / day. Animals allocated to DBU received no additional concentrate until day 28 postcalving. From day 28 to day 42 postcalving, concentrate levels for these animals were increased incrementally by 0.5 kg / day until they reached a maximum of 7.0 kg / day, via OPF (Figure 1). In order to prevent mineral deficiencies, 50 g / cow / day of a dairy mineral supplement was added during the first 28 days post-calving. The chemical composition of RBU and DBU is detailed in Table 3, with the energy and protein concentrations of individual ingredients based on published values (AFRC, 1993). The complete diet (basal diet plus 7.0 kg concentrate through the OPF) was designed to provide 175 g crude protein / kg dry matter, 12.0 MJ metabolisable energy / kg dry matter and 200 g starch / kg dry matter. Animals were assigned to postcalving treatments according to parity, previous lactation milk yield, calving date, calving interval, PTA fat, PTA protein and BCS and live weight at day -21 precalving. Animals remained on these diets until d 150 of lactation.



Figure 1: Schematic of concentrate allocation within the rapid (RBU) and delayed build-up (DBU) treatments.

Fresh diets were prepared daily using a mixer wagon and offered between 1000 and 1100h via feed boxes placed on a computer-linked load cell system. Access to feed boxes was controlled by an electronic identification system (Calan gate; American Calan Inc., Northwood, NH, USA), enabling the DM intake of individual animals to be recorded continuously. Daily intakes were used to calculate an average daily intake for each week of lactation. Postcalving, diet allocation had a target excess of 7.0 percent which was removed thrice weekly. On a daily basis throughout the study animals that did not consume 20 kg of diet (fresh weight) were automatically identified by the feed system and these animals were assessed for digestive, metabolic and other health problems. Out-of-parlor feeder intakes were checked twice daily; once before 11.00 am and once in late afternoon.

	Inclusion rate (g/kg)				
Constituent ¹	Dry cow	Basal ration	Out-of-parlour feeder		
Barley (milled)	375	200	200		
Maize (milled)	-	215	215		
Citrus pulp	-	104	104		
Soya hulls	200	105	105		
Soya bean meal (Hi-Pro)	-	171	183		
Rape meal	400	135	135		
Megalac ²	-	16	16		
Dairy cow minerals	-	29	17		
Molaferm ³	25	25	25		
ME (MJ/kg DM)	12.6	13.0	13.2		
CP (g/kg DM)	232	217	225		
Starch (g/kg DM)	230	280	280		

Ingredient composition and chemical composition of the concentrate feedstuffs offered during the dry period and postcalving. Table 1:

¹ ME, metabolisable energy ; CP, crude protein ² Volac Ltd. Orwell, Hertfordshire, UK ³ United Molasses, Belfast, NI, UK

	Grass silage	Maize silage
Oven DM (g/kg)	268	283
VCDM ¹ (g/kg)	274	291
pH	3.8	3.9
Composition of VCDM (g/kg)		
Crude protein	154	97
Ammonia nitrogen (g/kg total N)	78	110
Ethanol	3.3	-
Propanol	0.15	-
Lactic acid	33	-
Acetic acid	4.0	-
Propionic acid	0.23	-
n-butyric acid	0.24	-
Acid detergent fibre	317	259
Neutral detergent fibre	531	496
Ash	81	42
Starch	-	206
Gross energy (MJ/kg DM)	19.9	21.9

Table 2: Chemical composition of grass silage and maize silage offered during the experiment.

¹VCDM, volatile corrected dry matter

		Basal ration	l	C	Complete die	et
	Conc. ¹	Grass silage	Maize silage	Conc. ¹	Grass silage	Maize silage
Proportion of diet (DM basis)	350	390	260	600	240	160
DM content (g/kg)	870	268	283	870	268	283
$CP (g/kg DM)^2$	217	154	97	225	154	97
Starch (g/kg DM)	280	-	206	280	-	206
ME $(MJ/kg DM)^2$	13.0	11.5	11.1	13.1	11.5	11.1
Total ME (MJ/kg DM) ²		11.9			12.4	
Total CP (g/kg DM)		161			187	
Starch (g/kg DM)		152			201	

 Table 3:
 Crude protein, starch and metabolisable energy content on main components of the basal ration and the complete diets, and of the whole diet

¹ Conc., concentrate

² CP, crude protein; ME, metabolisable energy

Measurements

While daily samples were analysed for oven dry matter (85°C), weekly samples of grass silage and maize silage were analysed by near infrared reflectance spectroscopy to estimate ME content (Park *et al.*, 1998). Fresh samples of maize silage and grass silage were also analyzed once every 4 weeks for gross energy (Porter, 1992) and pH. The same samples were analysed for nitrogen and ammonia-nitrogen (ammonia-N) concentrations as described by Steen (1989), and for lactic acid, VFA's, ethanol, and propanol concentrations as described by Porter and Murray (2001). A daily sub-sample was taken from the oven dry matter residues of grass silage and maize silage and bulked for each 4-week period and analyzed for concentrations of NDF (neutral detergent fibre), ADF (acid detergent fibre), and ash as described by Cushnahan and Gordon (1995). Twice-weekly, samples of maize silage were dried at 60°C and then bulked for each 2-week period and analyzed for starch using a Megazyme kit (McCleary et al., 1994). Each batch of concentrates produced was sampled, bulked for each 2-week period, with bulked samples

analyzed for oven dry matter content and nitrogen. Every 28 days, one dried sample for each concentrate was analyzed for ADF, NDF and ash concentrations as described by Cushnahan and Gordon (1995). A separate concentrate sample was dried at 60°C and similarly bulked and analyzed for starch as detailed above (Table 1). Volatility coefficients (Porter and Murray, 2001) were used with the oven dry matter contents of the grass silage and maize silage to produce volatile-corrected dry matter (VCDM) values.

Measurements (animals)

Cows were milked twice daily, between 0530 and 0700, and 1530 and 1700, through a 50 point rotary parlor. Milk yield was recorded automatically at each milking for each individual animal and a mean daily milk yield was calculated for each animal on a weekly basis. Milk samples were taken for fat, protein, lactose, and SCC analysis weekly from two consecutive collections (a.m. and p.m.) from each animal, with a.m. and p.m. samples analyzed separately using an infrared milk analyzer. Each sample had a preservative tablet added (Lactab Mark III, Thompson and Cooper Ltd, Runcorn, UK) and was stored at 4°C until analyzed. A weighted milk composition was subsequently calculated for each weekly sampling occasion.

Milk samples for progesterone analysis were collected on Mondays, Wednesdays and Fridays (all AM) from calving until 100 days into the breeding period or until confirmation of pregnancy. Milk samples were taken aseptically and to each was added a preservative tablet (Lactab Mark III, Thompson and Cooper Ltd, Runcorn, UK). Samples were stored at 4°C until analysis. Milk progesterone concentration was determined using a competitive enzyme–linked immuno-sorbent assay (ELISA) kit (Ridgeway Science Ltd, Gloucestershire, UK). The assay is based on the method of Sauer *et al.* (1986).

Progesterone parameter definitions

A full description of the methodology used to interpret and analyze the progesterone data was provided by McCoy et al. (2006). Briefly, the onset of luteal activity (OLA) is indicated by the first of at least two consecutive progesterone concentrations ≥ 3 ng / ml in whole milk. When measuring this estimate the sampling routine introduces an overestimate of 1.17 days, on average, which was subtracted from the calculated value. The luteal phase (LP) of an individual oestrous cycle is defined as the period between the first progesterone concentration ≥ 3 ng / ml and the last consecutive milk progesterone concentration \geq 3 ng / ml in whole milk. The sampling routine underestimates the interval by, on average, approximately 2.33 days (i.e. 1.17 + 1.17). Therefore, the corrected interval was reported as calculated interval + 2.33 days. The inter-ovulatory interval (IOI) is defined as the period between the first progesterone rise (above 3ng/ml) of one cycle to the first progesterone rise (above 3ng/ml) in the next cycle. No inherent sampling bias exists with this parameter. The inter-luteal interval (ILI) is defined as the period between the demise of one corpus luteum and the rise of the next, and is the interval from the first milk progesterone concentration < 3 ml to the last consecutive milk progesterone < 3ng / ml in whole milk. No inherent sampling bias exists with this parameter.

Abnormal progesterone patterns

Progesterone data was assessed according to Lamming and Darwash (1998) to characterise abnormal progesterone profiles. A delayed ovulation type I (DOV I) was defined as progesterone concentration < 3 ng / ml in whole milk for \ge 45 d (prolonged ovulation) and a delayed ovulation type II (DOV II) was defined as progesterone concentrations < 3 ng / ml in whole milk for \ge 12 d after the OLA (prolonged inter-luteal interval). A persistent corpus luteum type I (PCL I) was defined as progesterone concentrations \ge 3 ng / ml for \ge 19 d on the first luteal phase (delayed luteolysis of the corpus luteum during the first oestrous cycle) and a persistent corpus luteum type II (PCL II) was defined as progesterone to the first oestrous cycle) and a persistent corpus luteum type II (PCL II) was defined as progesterone concentrations \ge 3 ng / ml for \ge 19 d on subsequent luteal phases (delayed luteolysis of the corpus luteum during subsequent oestrous cycles).

Pregnancy was confirmed via an ultrasound scan carried out by a veterinarian at approximately day-30 post insemination and elevated milk progesterone concentrations. All inseminations were carried out approximately 12 hours after an observed oestrus by a trained technician following the commencement of breeding (30th November) and a 42 day voluntary waiting period. All fertility events were recorded.

Body weight was measured weekly from 21 days prior to the predicted calving date until calving and twice daily from calving until the end of the study. Body condition score was measured weekly on a scale from 1 to 5 (Edmonson et al., 1989).

From calving until day 84 of lactation, all animals were blood sampled weekly before being offered fresh food. Blood was taken from the coccygeal vein into uncoated, heparin-coated, and fluoride oxalate coated tubes (BD, Oxford, UK). Plasma was recovered by centrifugation from fluoride oxalate tubes for analysis of glucose and nonesterified fatty acids (NEFA), and from heparinised tubes for analysis of total protein, albumin, urea, and β -hydroxybutyrate (BHBA). All analyses were carried out on a clinical analyzer (AU640, Olympus UK Ltd, Middlesex, UK). Plasma concentrations of total protein, albumin, glucose, and urea were determined using Olympus kits (Olympus Life Science Research Euorpa, Munich, Germany). Reagents for total protein, albumin, glucose, and urea were ready for use and placed appropriately into the analyzer. In the analysis of total protein, cupric ions in an alkaline solution react with proteins and polypeptides containing at least two peptide bonds to produce violet colored complex. The absorbance of the complex at 540/660 nm is directly proportional to the concentration of protein in the sample (Young, 2000). For albumin, a coloured complex is formed when bromocresol green (BCG) reacts with albumin. The absorbance of the albumin-BCG complex is measured bichromatically (600/800 nm) and is proportional to the concentration of albumin in the sample (Young, 2000). Glucose is phosphorylated by hexokinase in the presence of ATP and Mg^{2+} to produce glucose-6-phosphate and ADP. Glucose-6-phosphate dehydrogenase specifically oxidizes glucose-6-phosphate to gluconate-6-phosphate with the concurrent reduction of NAD⁺ to NADH. The increase in absorbance at 340 nm is proportional to the glucose concentration in the sample (Young,

2000). Urea is hydrolysed in the presence of water and urease to produce ammonia and carbon dioxide. The ammonia produced in the first reaction combines with 2-oxoglutarate and NADH in the presence of glutamate-byhydrogenase to yield glutamate and NAD⁺. The decrease in NADH absorbance per unit time is proportional to the concentration of urea (Young, 2000). NEFA concentrations were determined using a standard Wako reagent kit NEFA-HR(2) (Wako Chemicals GmbH, Neuss, Germany). Reagants were prepared according to Kreb et al. (2000). NEFA is converted to Acyl-CoA, AMP, and pyrophosphoric acid by the action of Acyl-CoA synthetase, under coexistence with coenzyme A and adenosine 5-triphosphate disodium salt (ATP). Obtained Acyl-CoA is oxidized and yields 2,3-trans-Enoyl-CoA and hydrogen peroxide by the action of Acyl-CoA oxidase (ACOD). In the presence of peroxidase, the hydrogen peroxide formed yields a blue purple pigment by quantitative oxidation condensation with 3-Methyl-N-Ethyl-N-(β-Hydroxyethyl)-Aniline (MEHA) and 4-aminoantipyrine (4-AA). Nonesterified fatty acids (NEFA) concentration is obtained by measuring absorbance of the blue purple color. β -hydroxybutyrate concentration of plasma was determined according to McMurray et al. (1984).

Serum was recovered by centrifugation of uncoated tubes for analysis of IGF-1. Serum IGF-1 concentrations were determined by RIA from weeks 1-8, 10 and 12 of lactation for all animals on the study. IGF-1 assays were balanced for dietary treatment and parity, with control samples included in each assay. IGF-1 binding proteins were removed by acid-ethanol cryo-precipitation (Wylie et al., 1997) prior to analysis. The primary antibody (AFP 4892898) was rabbit anti-human somatomedin C and was used at a final dilution of 1:400,000.

A clinical examination of the vagina mucus was performed and scored according to Williams *et al.* (2005). Briefly, the cow's vulva was thoroughly cleaned with a disinfected towel and then dried using a paper towel. Then a clean, lubricated, gloved hand was inserted through the vulva into the vaginal area. In each cow, the lateral, dorsal and ventral walls of the vagina and the external cervical os were palpated, and the mucus contents of the vagina withdrawn manually for examination. The vaginal mucus was

assessed by one person for colour, proportion and volume of pus, and a character score assigned as follows: (0) clear or translucent mucus; (1) mucus containing flecks of white or off-white pus; (2) <50 mL exudate containing \leq 50% white or off-white mucopurulent material; and (3) >50 mL exudate containing purulent material, usually white or yellow, but occasionally sanguineous. The vaginal mucus was also assessed by odour, and given a score 0 for normal odour or a score of 1 if a fetid odour was detected. At day 21 post calving, if more than 50 ml of the exudate contained purulent material, usually white or yellow, but occasionally a fetid odour, antibiotics were administered through the vagina and cervical canal into the lumen of the uterus, guided by palpation per rectum (Metricure, Intervet/Schering-Plough Animal Health, Walton Manor, Walton, Milton Keynes, MK7 7AJ). To aid the analysis of vaginal mucous scores data, the scores were translated into one integer; 0 = 0, 0; 1 = 1, 0; 2 = 2, 0; 3 = 3, 0; 4 = 2, 1; 5 = 3, 1.

All additional animal health measurements were recorded and compiled in a database.

Calculation of Energy Balance

The average daily energy balance (DEB) for each animal was calculated for each week of lactation using the equations described by Thomas (2004) {DEB = ME intake – ME requirement $[-10 + (ME_{preg} + ME_{maintmilk} * BW^{0.75})] + [(0.0013*BW)/K_m)]$; K_m, efficiency of energy use for maintenance (0.35 x ME/GE + 0.503)}. Milk yields, DMI, milk compositions, body weight, and feed composition data were all used in the calculations. Missing values (less than 2% of all values) were estimated from data for the week prior to, and the week following, the week of which observations were missing.

Statistical Analysis

Data were analyzed by a repeated measures approach using the Residual Maximum Likelihood (REML) procedure in GenStat (Payne et al. 2007). The model fitted fixed effects concentrate build-up strategy, parity and week of lactation for each parameter.

Average liveweight and condition score for weeks -1 and 1 of lactation were used as covariates. The model included all 2-way and 3-way interactions among these variables.

A logistic regression model was used to analyse the following data; pregnancy rate to first service, pregnancy rate to first and second service, 100 day in calf rate, intervention, delayed ovulation types I and II, persistent corpus luteum types I and II and whether or not there was more than one abnormal progesterone profile observed.

RESULTS

Chemical Composition of the Silages, Concentrates and Diets

The chemical composition of concentrates offered during the dry period and lactation (within basal ration and via OPF) are presented in Table 1. Concentrates had similar ME, NDF, ADF, and ash contents but differed in crude protein and starch contents. The chemical composition of silages, as fed, is presented in Table 2. The grass silage offered postcalving was well preserved having a pH and concentrations of ammonia-N and butyrate of 3.8, 78 g / kg total nitrogen, and 0.24 g/kg of DM, respectively. The maize silage offered postcalving had a dry matter content of 283 g / kg, a crude protein concentration of 97 g / kg DM, and a starch concentration of 206 g / kg DM, typical of average quality maize forage. Table 3 details the mean composition of the TMR as fed.

Effect of Concentrate Build-Up Strategy on Intake and Production Parameters (1-150 d)

During the postcalving period (1-150 d), animals allocated to DBU treatment had higher forage intakes (P < 0.001) and lower concentrate intakes (P = 0.004) than animals allocated to the RBU treatment (Table 4). Concentrate build-up strategy had no significant (P < 0.05) effect on total dry matter intake, milk yield, milk energy, concentrations of fat, protein and lactose, or yields of fat, protein and fat plus protein.

All the above parameters were significantly (P < 0.001) affected by stage of lactation. There was a significant treatment by stage of lactation interaction for total DMI (P < 0.001; Figure 2; SED, 0.62), forage intake (P < 0.001; Figure 3; SED, 0.39), concentrate intake (P < 0.001), milk yield (P = 0.001; Figure 4; SED, 1.36), milk protein concentration (P < 0.001), milk protein yield (P < 0.001), and milk fat plus protein yield (P = 0.016).



Figure 2: Total dry matter intake (kg / d) of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation. Treatment and treatment*week of lactation effects: treatment, P = 0.095 (SED, 0.53); week of lactation * treatment, P < 0.001(SED, 0.623).



Figure 3: Total forage intake (kg DM / d) of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation. Treatment and treatment*week of lactation effects: treatment, P < 0.001 (SED, 0.34); week of lactation * treatment, P < 0.001 (SED, 3939).



Figure 4: Milk yield (kg / d) of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation. Treatment and treatment*week of lactation effects: treatment, P = 0.514 (SED, 0.156); week of lactation * treatment, P = 0.001 (SED, 1.359).

	Concentrate Build-Up Strategy				P-value	
-	Rapid Build-Up	Delayed Build-Up	SED	Treatment	Time	Interaction
Dry matter intake (kg/d)						
Forage	9.0	10.4	0.34	< 0.001	< 0.001	< 0.001
Concentrate	11.5	11.0	0.19	0.004	< 0.001	< 0.001
Total	20.5	21.4	0.53	0.095	< 0.001	< 0.001
Milk yield (kg/d)	38.9	37.9	1.61	0.514	< 0.001	0.001
Milk constituents (g/kg)						
Fat	40.5	41.6	1.43	0.416	< 0.001	0.149
Protein	34.2	33.3	0.71	0.224	< 0.001	< 0.001
Lactose	45.4	45.2	0.33	0.567	< 0.001	0.719
Milk constituent yield (kg/d)						
Fat	1.57	1.57	0.079	0.958	< 0.001	0.379
Protein	1.31	1.25	0.052	0.209	< 0.001	< 0.001
Fat + protein	2.88	2.81	0.124	0.558	< 0.001	0.016
Milk energy output (MJ/d)	120	118	5.1	0.632	< 0.001	0.058

Table 4:Effects of concentrate build-up strategy on dry matter intake, milk yield, milk constituents and constituent yields (fat
and protein) of during the first 150 days of lactation

¹ SED, standard error of the difference;

Effect of Concentrate Build-Up Strategy on Liveweight, Body Condition Score and Energy status (1-150 d)

During the first 150 days of lactation, concentrate build-up strategy had no significant effect (P > 0.05) on liveweight, body condition score, ME requirement, ME intake, daily energy balance (P = 0.065) or cumulative energy balance (Table 5).

All of the above parameters were significantly (P < 0.001) affected by stage of lactation. There was a significant treatment * stage of lactation interaction for ME intake (P < 0.001), ME requirement (P = 0.030), and cumulative energy balance (P = 0.001; Figure 5; SED, 1.36). The treatment * week of lactation interaction for daily energy balance approached significance (P = 0.053; SED, 7.93; Figure 6).

Over the 305 day lactation, concentrate build-up strategy had no significant effect on milk yield, milk composition (fat, protein and lactose), liveweight and condition score (Table 6).

	Concentrate Bu	Concentrate Build-Up Strategy			P-value	
	Rapid Build-Up	Delayed Build-Up	SED^2	Treatment	Time	Interaction
Liveweight (kg)	638	627	7.0	0.100	< 0.001	0.055
BCS ¹	2.33	2.27	0.052	0.295	< 0.001	0.911
ME requirement (MJ/d) ¹	273	268	9.249	0.566	< 0.001	0.030
ME intake $(MJ/d)^2$	252	262	6.33	0.154	< 0.001	< 0.001
Daily energy status (MJ/d)	-21.3	-6.3	7.93	0.065	< 0.001	0.053
Cumulative energy status (MJ)	-2334	-1382	571	0.136	< 0.001	< 0.001

Effects of concentrate build-up strategy on liveweight, body condition score and body energy status of animals during the first 150 days of lactation Table 5:

¹ BCS, body condition score; ME, metabolisable energy ² SED, standard error of the difference;

	Concentrate Build-Up Strategy			P-value		
	Rapid Build-Up	Delayed Build-Up	SED	Treatment	Time	Interaction
Milk yield (kg/d)	35.1	35.1	1.34	0.863	< 0.001	0.057
Milk constituents (g/kg)						
Fat	38.0	39.7	1.25	0.343	< 0.001	0.081
Protein	33.9	33.6	0.71	0.188	< 0.001	0.149
Lactose	45.3	44.9	0.39	0.348	< 0.001	0.448
Liveweight (kg)	637	632	8.9	0.675	< 0.001	0.873
Condition score	2.43	2.37	0.063	0.294	< 0.001	0.220

Table 6:Effects of concentrate build-up strategy on milk yield, milk constituents (fat, protein and lactose) milk constituent yields
(fat and protein) and milk energy output of animals from day 1 to 305 of lactation

¹ SED, standard error of the difference;



Figure 5: Cumulative energy balance (MJ ME) of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation. Treatment and treatment*week of lactation effects: treatment, P = 0.136 (SED, 571); week of lactation * treatment, P < 0.001 (SED, 505).



Figure 6: Energy balance (MJ ME / d) of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation. Treatment and treatment*week of lactation effects: treatment, P = 0.065 (SED, 7.93); week of lactation * treatment, P = 0.053 (SED, 9.735).

Effect of Concentrate Build-Up Strategy on Blood Parameters (1-28d, 1-84 d)

In the first 28 days of lactation, animals on the DBU strategy had lower plasma urea (P < 0.001) concentrations and higher plasma BHBA (P = 0.005) concentrations compared to animals on the control treatment (Table 7). Concentrate build-up strategy had no significant effect (P > 0.05) on plasma concentrations of total protein, albumin, globulin, glucose, or NEFA concentrations.

During the first 84 days of lactation, concentrate build-up strategy had a significant effect on plasma urea (P = 0.041), glucose (P = 0.014) and BHBA (P = 0.011) concentrations. Animals allocated to the DBU strategy had lower plasma urea and glucose concentrations and higher plasma BHBA concentrations than animals allocated the control treatment (Table 7).

During both time periods there was a significant treatment by stage of lactation interaction for plasma urea; 1-28 days (P = 0.025) and 1-84 days (P = 0.029; Figure 7).



Figure 7: Plasma urea concentrations of animals allocated to one of two concentrate build-up strategies (Rapid vs. Delayed) in early lactation. Treatment and treatment*week of lactation effects: treatment, P = 0.041 (SED, 0.156); week of lactation * treatment, P = 0.29 (SED, 0.272).

	Concentrate	Build-Up Strategy			P-value	
Blood parameter	Control	Delayed Build-Up	SED	Treatment	Time	Interaction
Day 1 – 28 of lactation						
Urea (mmol/l)	4.04	3.45	0.205	< 0.001	< 0.001	0.025
Total protein (g/l)	57.6	58.1	3.96	0.651	0.183	0.333
Albumin (g/l)	24.8	24.4	1.71	0.966	0.390	0.408
Globulin (g/l)	32.8	33.7	2.52	0.507	< 0.001	0.346
NEFA $(meq/l)^1$	0.50	0.51	0.058	0.724	0.071	0.951
BHB (mmol/l) ¹	0.81	1.06	0.098	0.005	0.001	0.220
Glucose (mmol/l)	3.14	3.01	0.081	0.099	< 0.001	0.497
IGF-1 $(ng/ml)^{1,2}$	61.2	58.9	3.53	0.436	0.853	0.953
Day 1 – 84 of lactation						
Urea (mmol/l)	4.06	3.74	0.156	0.041	< 0.001	0.029
Total protein (g/l)	56.7	56.1	2.515	0.533	0.003	0.214
Albumin (g/l)	23.3	22.9	1.16	0.583	0.065	0.588
Globulin (g/l)	33.9	33.5	2.014	0.565	< 0.001	0.204
NEFA $(meq/l)^1$	0.36	0.38	0.0378	0.500	< 0.001	0.677
BHB (mmol/l) ¹	0.72	0.85	0.0522	0.011	< 0.001	0.051
Glucose (mmol/l)	3.38	3.25	0.053	0.014	< 0.001	0.686
IGF-1 (ng/ml) ^{1,3}	62.7	60.6	2.60	0.552	0.298	0.879

Table 7: Effects of concentrate build-up strategy on blood constituents of animals during days 1 to 28 and 1 to 84 of lactation

¹NEFA, non-esterified fatty acid; BHB, β -hydroxybutyrate; IGF-1, insulin-like growth factor-1; CP, crude protein; DM, dry matter ²IGF-1 samples (n=24, five time points), and all other parameters (n=90, 16 time points)

Effect of Concentrate Build-Up Strategy on Uterine Health, Digestive Health and Mastitis

Concentrate build-up strategy had a significant (P = 0.026) effect on the proportion of cows that were treated for a dilated abomasum. In animals allocated to a RBU strategy, 44.8% were treated for a dilated abomasum compared to 17.9% of animals allocated to the DBU strategy (Table 8). Concentrate build-up strategy had no significant (P > 0.05) effect on the incidence of mastitis. Similarly, there was no significant effect of concentrate build-up strategy on the incidence of metritis at day 21 post calving. Concentrate build-up strategy had no significant (P > 0.05) effect on vaginal mucous scores at days 14, 21, and 28 postcalving.

Effect of Concentrate Build-Up Strategy on Fertility Parameters

Concentrate build-up strategy had no significant (P < 0.05) effect on days to first service, conception rate to first and second service, 100-day incalf rate, in-calf rate at the end of a 6 month breeding period, or the interval from calving to conception (Table 9). With regard to luteal activity, concentrate build-up strategy had a significant effect on the inter-ovulatory interval. Cows allocated to a RBU treatment had a significantly (P = 0.044) higher (31.6 vs. 22.0; SED, 4.63) average inter-ovulatory interval than cows allocated to a DBU treatment. Concentrate build-up strategy had no significant (P > 0.05) effect on the onset of luteal activity, the luteal phase or on the inter-luteal interval (Table 10). Furthermore, concentrate build-up strategy had no significant (P > 0.05) effect on the occurrence of abnormal cyclic activity; delayed ovulation types I and II or persistent corpus luteum types I and II (Table 10). A high percentage of animals (81.5%) had more than 1 atypical ovarian cycle post calving; however, there was no significant (P > 0.05) effect of concentrate build-up strategy on the occurrence of this.

Table 8: Effect of concentrate build-up strategy on the incidence of right dilated abomasums, mastitis, uterine infection and on uterine mucous scores

	Concentrate B	uild-Up Strategy	Olleretie	CED ¹	Develop
	Rapid Build-Up	Delayed Build-Up	- Odds ratio	SED	P-value
Right dilated abomasum (% of cows)	44.8	17.9	0.27	-	0.026
Mastitis (% of cows)	55.2	42.9	0.61	-	0.352
Uterine infection at day 21 post calving (% of cows)	31.0	32.1	1.05	-	0.928
Mucous score at day 14 post calving ²	2.82	3.16	-	0.460	0.464
Mucous score at day 21 post calving ²	1.74	2.05	-	0.490	0.524
Mucous score at day 28 post calving ²	0.88	1.18	-	0.414	0.471

¹ SED, standard error of the difference ² Mucous score converted to 0-5 to aid the analysis of the data (0 = 0,0; 1 = 1,0; 2 = 2,0; 3 = 3,0; 4 = 2,1; 5 = 3,1)

	Concentrate B	uild-Up Strategy		\mathbf{CED}^1	Davahaa
	Rapid Build-Up	Delayed Build-Up	- Odds ratio	SED	P-value
Days to first service	67.0	75.9	-	7.49	0.239
Conception rate to 1st insemination (%)	40.7	36.4	0.83	-	0.754
Conception rate to 1st and 2nd inseminations (%)	59.3	59.1	0.99	-	0.990
In-calf rate at day 100 of the breeding period (%)	62.9	63.9	1.03	-	0.960
In-calf rate at the end of the breeding period (%)	74.1	86.4	2.22	-	0.281
Interval from calving to conception (d)	93.9	110.9	15.05	-	0.266

Table 9: Effect of concentrate build-up strategy on fertility performance

¹ SED, standard error of the difference

	Concentrate B	CED		
	Rapid Build-Up	Delayed Build-Up	SED	P-value
Onset of luteal activity (d)	33.0	44.8	7.24	0.136
Average luteal phase (d)	15.3	13.8	1.59	0.334
Average inter-ovulatory interval (d)	31.6	22.0	4.63	0.044
Average inter-luteal interval (d)	17.6	12.6	3.35	0.140
Delayed ovulation type I^1 (% of cycles)	20.7	25.9	7.96	0.898
Delayed ovulation type II^1 (% of cycles)	84.8	79.2	3.95	0.571
Persistent corpus luteum type I^1 (% of cycles)	27.2	11.1	7.17	0.115
Persistent corpus luteum type II ¹ (% of cycles)	12.5	14.5	3.87	0.718
Animals with 1 or > atypical cycles (%)	82.1	80.8	7.28	0.897

Table 10:Effect of concentrate build-up strategy on ovarian luteal activity based on 3x weekly analysis of milk progesterone.

¹ SED, standard error of the difference
Effects of Energy Balance on Mucus Score and fertility

Average daily energy balance (week 1-3, week 1-6 and week 1-20) had no significant (P > 0.05) effect on mucous score at days 14, 21 or 28 of lactation, average mucous score, or the recovery of metritis from days 14 to 28 of lactation.

When assessing fertility data, average daily energy balance (week 1-3, week 1-6 and week 1-20) had no significant (P > 0.05) effect on conception rate to first service, conception rate to first and second service, overall conception rate, 100 day in-calf rate, occurrence of a delayed ovulation type I, occurrence of a delayed ovulation type II, occurrence of a persistent corpus luteum type I, the occurrence of a persistent corpus luteum type I, and the occurrence of more than 1 atypical ovarian cycle.

Furthermore, average daily energy balance (week 1-3, week 1-6 and week 1-20) had no significant (P > 0.05) effect on normally distributed fertility data such as interval to the onset of luteal activity, luteal phase duration (cycles 1 - 3), inter-tuteal interval (cycles 1 and 2), inter ovulatory interval (cycles 1 and 2), average inter luteal interval, average inter ovulatory interval, days to conception, and interval to 1^{st} service.

Effect of Mucous Score on Fertility Parameters

Binomial regression of fertility data indicated a negative relationship between the average mucous score and conception rate to 1^{st} service (P = 0.042; r^2 , 0.07) and overall conception rate (P = 0.034; r^2 , 0.10). Concentrate build-up strategy had no significant effect (P > 0.05) on the intercept or trajectory of these relationships. The fitted equation for the relationship between the average mucous score and conception rate to 1^{st} service was Y = 0.383 - 0.505 (X) and for the relationship between the average mucous score and conception rate score and overall conception rate was Y = 0.859 - 0.302 (X)

Regression analysis of data revealed a positive association between the average mucous score and the interval from calving to the onset of luteal activity (P = 0.017; r^2 , 0.101) and the interval from calving to conception (P = 0.047; r^2 , 0.103). Concentrate build-up strategy had no significant effect (P > 0.05) on the intercept or trajectory of these relationships. The fitted equations were "Y = 28.9 + 5.98 (X)" and "Y = 84.4 + 11.78 (X)" for the relationship between average mucous score and the interval from calving to the onset of luteal activity, and the average mucous score and the interval from calving to conception, respectively.

DISCUSSION

The current study was designed to address the issues associated with high concentrate feed levels in early lactation and also to develop strategies to reduce the extent of NEB experienced by the high-yielding dairy cow in early lactation. Reducing NEB in early lactation is difficult; however, the approach most often advocated is to increase the nutrient density of the diet by feeding high levels of concentrate. However, this experiment focused on an alternative approach, namely introducing concentrates to the diet of fresh calved cows at a slow rate. This is aimed at reducing the extent of NEB experienced by the cow by delaying the increase in milk yield so as to be more in line with the increase in dry matter intakes. This approach is also likely to improve rumen function.

Chemical Composition of the Silages, Concentrates and Diets

The target dietary protein content for the basal diet was 150 g / kg DM while actual crude protein levels were slightly higher at 161 g / kg DM, with this reflecting the variability of the forages offered. Similarly, the target of 180 g CP / kg DM for the complete diet was exceeded with an average value 187 g CP / kg DM, again reflecting the variability of forages offered. The target starch content of the basal ration was 165 g / kg DM.

However, due to the low starch levels in the maize silage the actual basal diet starch content was 152 g/kg DM.

Effect of Concentrate Build-Up Strategy on Intake and Production Parameters

As expected, a delayed concentrate build-up strategy significantly increased forage intake in early lactation due to reduced concentrate levels in the diet. However, after maximum concentrate allocation had been achieved (day 42 postcalving) animals on the DBU treatment sustained significantly higher forage intakes than animals on the RBU treatment until week-17 of lactation. This would suggest that improved rumen conditions in animals allocated the DBU strategy facilitated higher forage intakes once concentrate levels were increased. Higher forage intakes in early lactation will improve buffering capacity due to higher fibre intakes stimulating chewing behaviour and subsequently saliva production (Mertens, 1997), which will in turn maintain pH at higher levels. For cows allocated to RBU treatment, the interaction between high concentrate levels and forage may reduce fibre digestion (Bargo et al., 2003). High dietary concentrate levels may cause a reduction in ruminal pH and subsequently have a negative effect on the activity of cellulolytic bacteria, reducing forage digestion and therefore forage dry matter intake. Furthermore, there is evidence to suggest that the substitution rate (the change in forage DM intake per unit of additional concentrate) is greater in early lactation (greater than 0.9 in a study by Ingvartsen et al. (2001)), and this would result in a further depression of forage intake in early lactation. In the current study, the substitution rate for cows allocated the RBU treatment was 0.57 at week-4 of lactation (maximum concentrate intake). However, for cows allocated the DBU treatment, the substitution rate at week-7 of lactation (maximum concentrate intake) was 0.21.

One of the primary objectives of this experiment was to reduce the extent of negative energy balance in early lactation by reducing the increase in milk production in early lactation and delaying peak milk yield to better reflect actual dry matter intakes. This was achieved by reducing the total dietary CP content of the diet in early lactation. Dietary CP supply is a key driver of milk production as the cow has a limited capacity to rely on

body protein resources to maintain a high level of milk production (Oldham, 1984). Previous research conducted by this group indicates that reducing the dietary CP concentration from 173 to 144 g / kg DM suppresses milk production, with no negative effect on DM intake, and as such, improved energy balance (Law et al., 2009). Results from the current study suggest that reducing concentrate levels, and subsequently overall dietary CP content, reduced the acceleration in milk yield in early lactation and delayed peak milk yield. In the current study milk production was significantly lower in cows on the DBU treatment in weeks 3, 4, 5 & 6 of lactation compared to those allocated to the RBU treatment. After week six, dietary protein levels were approximately 18% on both treatments and from this point onwards there was no significant difference in milk yields between the two treatments. The results of the current study illustrate that cows responded in terms of milk production when the dietary crude protein content was increased from 161 to 187 g / kg DM. This is in contrast to work by Cunningham et al. (1996) and Leonardi et al. (2003) who observed no improvement in milk yield when dietary crude protein levels were increased from 165 to 185 g / kg DM and from 161 to 189 g / kg DM, respectively. However, diets offered in these latter studies were different from those offered in the current study.

A significant treatment by stage of lactation interaction suggests that during the first 140 days of lactation the milk yield trajectories were different between treatments. Figure 4 illustrates that cows allocated to the DBU treatment had a much flatter lactation curve than animals on the RBU treatment. Interestingly, when 305 day lactational data was analysed, this interaction was no longer significant suggesting that difference in milk yield were diluted as stage of lactation progressed.

During the first 150 days of lactation there was no significant effect of treatment on milk composition. Due to the significantly higher forage intake during the first 16 weeks of lactation, animals allocated to the DBU may have been expected to have had significantly higher milk fat concentrations. A higher forage proportion in the diet will increase the ratio of acetate:propionate in rumen fluid which should theoretically milk fat concentrations (Bauman and Griinari, 2000).

Effect of Concentrate Build-Up Strategy on Blood Parameters

During the first 28 and 84 days of lactation, animals allocated to the RBU treatment had significantly higher plasma urea concentrations, indicating increased levels of ammonia detoxification in the liver. The balance between effective rumen degradable protein (eRDP) and fermentable carbohydrates is important in dictating the quantity of ammonia that is utilised for microbial growth (Kenny *et al.*, 2001). Diets with high dietary CP concentrations will not only increase the quantity of ammonia available, but also increase the risk of a rumen imbalance between eRDP and fermentable carbohydrate, which will decrease the proportion of ammonia that diffuses into the blood system and undergoes detoxification in the liver to produce urea. Urea is also produced by the liver from the deamination and metabolism of amino acids used for gluconeogenesis, and amino acids originate from digestible undegradable protein, microbial protein and protein catabolism of body reserves (Butler, 1998).

During the first 84 days of lactation, animals allocated to the RBU treatment had significantly higher plasma glucose levels. This will largely be due to the difference in starch concentrations in early lactation between the DBU and RBU treatments. Animals allocated to the DBU treatment received 152 g starch / kg DM compared to 201 g starch / kg DM for animals on the RBU treatment. Starch undergoes fermentation in the rumen to produce volatile fatty acids and higher dietary starch levels will decrease the acetate to propionate ratio in the rumen. As 40–70% of plasma glucose is derived from propionate (Veenhuizen et al., 1988), higher propionate levels will support increased plasma glucose production, thus possibly explaining the difference observed in the current study.

Effect of Concentrate Build-Up Strategy on Health

In early lactation, 44.8% of animals on the RBU treatment, compared to 17.9% on the DBU treatment, were treated for a dilated abomasum. The abomasum is normally positioned at the ventral end, or floor of the abdomen. However, if the abomasum stops contracting an accumulation of gas (abomasal fermentation) can result in the abomasum moving up the abdominal flank towards the dorsal end, eventually causing a displacement. The term "dilated abomasum" was used in this experiment because the animals didn't displayed other typical symptoms of a displaced abomasum such as dehydration, diarrhea, ketosis etc. Animals that didn't consume 20 kg (fresh) of the lactating diet in the previous 24 hour period were automatically identified and checked for ill health. Frequently, these animals had excess gas in the abomasum and these animals were treated accordingly to improve rumen and abomasal motility. It is hypothesised that if these animals were not treated at this stage, a proportion would develop a displaced abomasum. Therefore it can be surmised that a delayed build-up of concentrates improved rumen function which was apparent in higher dry matter intakes of forage and a lower incidence of dilated abomasum.

Effect of Concentrate Build-Up Strategy on Fertility

Concentrate build-up strategy had no significant effect on conventional reproductive variables such as conception rate to first service, Conception rate to 1st and 2nd inseminations, In-calf rate at 100 days of the breeding period etc. (Table 9). In the current study the average conception rate to first service rate (38.6%) was comparable to values reported by Mayne *et al.* (2002) and Royal *et al.* (2000); 37.1 % and 39.7 respectively.

There were no significant treatment effects on the interval to the onset of luteal activity in the current study. However, there was a substantial numerical difference between treatments (33.0 vs. 44.8 days in animals allocated to RBU and DBU treatments respectively). As previously mentioned, during early lactation the dietary starch was substantially different between these treatments. High starch levels will favour elevated

plasma insulin concentrations which will promote steriodogenesis, enhancing ovarian follicle development and reducing the interval to onset of luteal activity. This effect has been shown previously. For example, Van Knegsel *et al.* (2007) illustrated that multiparous cows offered a high-starch diet tended to have an earlier resumption of cyclicity compared to those offered diets containing lower dietary starch concentrations. Furthermore, Gong *et al.* (2002) illustrated that a high-starch diet increased the proportion of animals that ovulated by day 50 postcalving. Interestingly, the mean interval from calving to the onset of luteal activity (38.9 days) in the current study was longer then previously reported; 27.9 days by Royal *et al.* (2000) and 32.2 days by Law *et al.* (2009).

There was a significant effect of concentrate build-up strategy on the inter-ovulatory interval (IOI). Animals allocated to the DBU treatment had a significantly lower IOI than animals allocated to the RBU treatment (22.0 vs. 31.6 days, respectively). An extended IOI is caused by the occurrence of a persistent corpus luteum and / or a delayed ovulation. Interestingly, Royal *et al.* (2000) stated that an IOI outside the range 19-23 days was indicative of reduced reproductive function. In the current study the duration of the IOI appeared to have little effect of subsequent fertility.

The analysis of thrice weekly progesterone concentrations highlighted that the incidence of a delayed ovulation type II (DOV II, 82%) was dramatically greater than previously published values. For example, Lamming and Darwash (1998) presented a DOV II of 13% and Royal *et al.* (2000) quoted a value of 16%. Delayed ovulation may be caused by a delay to, or failure of, the surge in circulating concentrations of luteinizing hormone (LH) which causes thinning and subsequent rupture of the follicle wall, leading to ovulation. It may also occur if there is incomplete luteolysis of the corpus luteum from the previous cycle (Sartori *et al.*, 2004).

CONCLUSIONS

Adopting a delayed concentrate build-up strategy in early lactation improved forage intake in early lactation and had no detrimental effect on overall production performance. Adopting a delayed build-up strategy also improved rumen health denoted by the significantly lower proportion of animals treated for a "dilated abomasum" compared to animals on a rapid build-up of concentrates. However, no effect of concentrate build-up strategy on fertility was identified in the current trial. Future research encorporating higher animal numbers, possibly as part of an on-farm programme, may allow fertility trends to be identified.

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TECHNOLOGY TRANSFER ASSOCIATED WITH PROJECT

Publications in Conference Proceedings

Law, R. A. McGettrick, S. and Ferris, C. P. (2011) Effect of concentrate build-up strategy in early lactation on production performance, health and fertility of high yielding dairy cows. In: *Proceedings of the British Society of Animal Science*, Page 5. (Appendix 1)

Farmers/Industry Meetings and Press Releases

Law, R.A., and Young, F.J., Recent developments in supplementation strategies for highyielding dairy cows during the winter. (2010) AgriSearch Dairy Seminar 'Improving the sustainability of Dairy Farming within Northern Ireland', page 20. (Appendix 2)

Press Release: Effect of delaying concentrate build-up in early lactation on production, health and fertility of the high-yielding dairy cow, United News, September 2010

Visitors to Hillsborough

1)	Presentations to industry	
•	Tom Phillips, Pasture to Profit	28 th May 2010
•	Dairy Co Extension Officers	29 th June 2010
•	AgriSearch Dairy Committee	27 th August 2010
•	John Roche, DairyNZ	31 st August 2010
•	Julie Lee, DairyNZ	8 th October 2010
•	United Feeds Technical Team	19 th January 2011
•	AgriSearch Dairy Committee	28 th March 2011

•	REDNEX, Slovakia	11 th May 2011
•	AB Vista, Birmingham	25 th May 2011
•	United dairy farmers	21 st June 2011
•	Fane Valley Feeds	4 th August 2011
•	McLarnons Feeds	11 th August 2011
•	Dr Kevin McDonald, Dairy NZ	31 st August 2011
•	Drs Joe Jacobs and Chris Korte, Victoria, Australia	1 st September 2011
•	Dr Richard Dewhurst and Christine McCartney, Teagasc	1 st September 2011

2) Presentations to farmer groups at Hillsborough

•	Blacklion discussion group	6 th May 2009
•	Greenmount and farmer group	18 th November 2009
•	Navan Farmers	3 rd February 2010
•	Chilean Dairy Farmers	14 th May 2010
•	Scottish Dairy Farm Managers	9 th June 2010
•	Vision Farmers Group	22 nd July 2010
•	DairyCo Farmers Group	30 th September 2010
•	Antrim Dairy Farmers	26 th October 2010
•	Cookstown Dairy Farmers	9 th March 2011
•	Narberth Grassland Society	6 th June 2011
•	Student field group, University of Kiel, Germany	13 th June 2011
•	North Wales dairy farmers	20 th June 2011
•	Meath Dairy Farmers	16 th August 2011

APPENDIX 1:

Effect of concentrate build-up strategy in early lactation on production performance, health and fertility of high-yielding dairy cows

Introduction The high milk production potential of the modern dairy cow frequently results in excessive and prolonged periods of negative energy balance (NEB) during early lactation. The most common approach to reduce NEB is to increase energy intake by increasing the proportion of concentrates being offered. However, offering diets containing high levels of concentrates can lead to rumen acidosis, impaired rumen function and reduced intakes, the latter exacerbating NEB. Introducing concentrates into the diet of fresh calved cows at a slower rate is likely to improve rumen function. Furthermore, diet crude protein (CP) content is likely to be reduced through offering a lower concentrate diet in early lactation. Offering a lower protein diet has been shown to reduce milk output, have no effect on dry matter intake, and thus improve cow energy status (Law *et al.*, 2009). The objective of this study was to compare two very different strategies by which to introduce concentrates into the diet in early lactation, namely a rapid build-up (RBU) or a delayed build-up (DBU) strategy.

Materials and methods From calving onwards, sixty autumn-calving Holstein Friesian cows (mean parity 3.1) were offered a basal diet (via feeder wagon) containing 35% concentrate and 65% forage on a dry matter (DM) basis (150 g CP/kg DM and 12.0 MJ metabolisable energy (ME)/kg DM). Cows were allocated to one of two post-calving concentrate allocation strategies (via out-of-parlour feeders), namely a rapid build up of concentrates (RBU) or a delayed build up of concentrates (DBU). With the rapid build-up treatment cows were offered 2.0 kg concentrate/cow/day on the day of calving, and this was then built up incrementally (0.5 kg/day) to a maximum of 7.0 kg/cow/day at day 10 post calving. Cows allocated the delayed build-up treatment received no additional concentrate via out-of-parlour feeders until day 28 of lactation, and thereafter received incremental concentrate levels (0.5 kg/day) to a maximum of 7.0 kg /cow/day at day 42 post calving. Once these concentrate feed levels had been achieved, diets were designed to have a CP and ME content of 180 g/kg DM and 12.4 MJ/kg DM respectively. Cows remained on these two dietary treatments until day 150 of lactation. Data were analysed using the residual maximum likelihood procedure via Genstat.

Results Total dry matter intake was unaffected by concentrate build-up strategy (Table 1). However, forage intake was significantly higher for cows allocated to DBU treatment (P<0.001; Figure 1), while concentrate intakes were lower (P<0.01). Neither milk yield nor milk composition was affected by concentrate build-up strategy (P>0.05). Despite the lack of treatment effects on milk production, cows on DBU returned to positive energy balance earlier (week 7 post-calving) than those on RBU (week 19 post-calving). From weeks 3-7 post-calving, cows allocated to DBU produced 3.5kg less milk/day than those allocated to RBU (P<0.001). Reproductive performance was unaffected by treatment (P>0.05).

	Rapid Build-Up	Delayed Build- Up	SED	P value
Dry matter intake (kg / day)	20.5	21.4	0.53	NS
Forage intake (kg DM/ day)	9.0	10.4	0.34	***
Concentrate intake (kg DM /	11.5	11.0	0.19	**
day)				
Milk yield (kg / day)	38.9	37.9	1.61	NS
Milk fat (g / kg)	40.5	41.6	1.43	NS
Milk protein (g / kg)	34.2	33.3	0.71	NS
Energy balance (MJ / day)	-21.3	-6.3	7.91	P=0.06

Table 1: Effect of concentrate allocation strategy on DM intake, milk production and energy balance (day 1-150 of lactation)



Figure 1: Forage intake during the first 140 days of lactation

Conclusions Adopting a delayed concentrate build-up strategy in early lactation improved forage intake while having no detrimental effect on production performance. This resulted in a trend towards improved energy status of the cows on this treatment, although reproductive performance was unaffected.

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APPENDIX 2:

Recent developments in supplementation strategies for high-yielding dairy cows during the winter

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Introduction

Intensive genetic selection has resulted in a dramatic increase in the milk production potential of the modern Holstein-Friesian dairy cow. However, this increase in milk production potential has been accompanied until recently by a decline in functional traits including health, fertility and longevity. In addition, the observed increase in milk production in early lactation has not been matched by a proportionate increase in energy intake (Veerkamp *et al.*, 1995; Ingvartsen *et al.*, 1999). The resulting energy deficit, better known as negative energy balance (NEB; Figure 1), is manifest in the mobilisation of body tissue reserves. A positive correlation has been identified between milk production potential and loss of body condition during early lactation (Ruegg and Milton, 1995). Severe and prolonged periods of NEB can predispose the dairy cow to metabolic disorders, immunosuppression, reproductive failure, and behavioural abnormalities, all of which contribute to a decline in the cow's general well-being (Nielsen, 1999). Thus, minimising the extent and duration of NEB experienced by dairy cows would appear to be an important objective within dairy systems.

Reducing NEB in early lactation is difficult for cows with a high genetic merit for milk production. Nevertheless, the approach most often advocated to reduce NEB is to increase nutrient intake. This can be achieved by either increasing total dry matter intake or by increasing the nutrient density of the diet, although both approaches normally go hand-in-hand. While improved housing and feed barrier management may provide some scope by which to improve total food intake, improving diet quality (nutrient density of diet) is the approach which is likely to be most effective. The nutrient density of the diet can be improved by improving forage quality (high digestible grass silage/inclusion of maize silage), or by increasing concentrate feed levels. The latter approach has been widely adopted in Northern Ireland. This is highlighted in CAFRE benchmarking data which indicates that the average concentrate input of benchmarked dairy herds in Northern Ireland increased from 1.1 to 2.1 tonnes/cow/year between 1997/1998 and 2008/2009.



Figure 1 Relationship between energy intake and energy requirement during the first 25 weeks of lactation.

However, the move to dairy systems involving higher concentrate feed levels has raised a number of challenges and questions, such as: optimum strategies by which to increase concentrate inclusion levels in early lactation; at what part of lactation is the response to additional concentrates maximised; what is the optimal strategy by which to include concentrates in the diet; can the protein content of high concentrate diets be reduced; and can concentrate composition be used to modify energy balance and hormone levels to improve overall fertility. This paper will set out to address these issues, based primarily on the findings of recent research programmes undertaken at AFBI Hillsborough.

Strategies to increase concentrate feed level post-calving

With concentrate feed levels now much higher than in the past, optimum strategies by which to build up concentrate levels in the diet in early lactation need to be identified. This issue is highlighted in that within a short space of time, cows frequently move from a dry cow diet based on a medium quality silage and low concentrate supplementation to a lactating cow diet based on high quality forage and high levels of starchy concentrates. Furthermore, offering high concentrate diets can lead to rumen acidosis. Acidosis occurs when the rumen pH falls below 5.6 (normal pH is 6.5 to 7.0), resulting in impaired rumen function. In acute cases, normal rumen movement stops, and this will decrease fibre digestion and subsequently depress appetite and milk production. If this is left untreated, the changes in rumen pH alter rumen flora, with acid-producing bacteria becoming dominant. These bacteria produce more acid, which is absorbed through the rumen wall, causing metabolic acidosis, and this has the potential to cause death.

Introducing concentrates to the diet of fresh calved cows at a slow rate is likely to improve rumen function. In addition, there is some anecdotal evidence that a slow buildup in concentrate feed levels post-calving may reduce the extent of NEB experienced by the cow by delaying peak milk yield until closer to the time when maximum dry matter intakes have been achieved.

To address this issue a study was conducted to compare two very different strategies by which to increase concentrate feed levels in early lactation, namely a rapid build-up (RBU) or a delayed build-up (DBU) strategy. The slow build-up strategy was designed to slow the rate of increase in milk production in early lactation. Previous research at Hillsborough has highlighted that reducing milk production in early lactation can actually improve the energy status of the cow and reduce body reserve mobilisation. As discussed later in this paper, reduced milk production can be achieved by decreasing the protein content of the diet. Dietary protein supply is a key driver of milk production as the cow has a limited capacity to rely on body protein reserves to maintain a high level of milk production (Oldham, 1984). Reduced dietary protein levels can be achieved by reducing the protein content of the concentrate or by reducing concentrate inclusion levels in the

diet. One potential benefit of the latter approach is higher forage intakes in early lactation, which may in turn improve rumen stability and total dry matter intake.

This study involved 60 winter-calving Holstein-Friesian dairy cows (multiparous), all of which were offered a basal diet containing 35% concentrate and 65% forage (grass silage and maize silage) on a dry matter basis (15% dietary protein content) post-calving. Cows were allocated to one of two post-calving feeding strategies, a rapid build up of concentrates (RBU) or a delayed build up of concentrates (DBU). Out-of-parlour feeders were used to allocate concentrates according to the two feeding strategies (RBU and DBU). With the rapid build-up treatment, concentrates offered through the out-of-parlour feeder were increased during the first 10 days of lactation (0-7 kg/cow/day). With the delayed build-up treatment cows were offered no concentrate via the out-of-parlour feeders until day 28 of lactation, and thereafter had their concentrates built up to 7 kg/cow/day over a 14-day period, so that maximum concentrate allocation the protein content of the diet was 18% on a dry matter basis (Figure 2). Cows remained on these two dietary treatments until day 150 of lactation.



Figure 2 Schematic of concentrate allocation within the rapid (RBU) and delayed build-up (DBU) treatments.

Total dry matter intake was unaffected by concentrate build-up strategy (Table 1). However, forage intake was significantly higher (Figure 3) for cows allocated to DBU compared to those allocated to RBU, while concentrate intake was lower. Neither milk yield nor milk composition was affected by concentrate build-up strategy (Table 1). However, from weeks 3 to 7 post-calving, cows allocated to DBU produced 3.5 kg less milk per day than those allocated to RBU.

Table 1The effect of concentrate allocation strategy (rapid build-up vs delayedbuild-up) on dry matter intake and milk production (mean for first 150 days of lactation),and on dairy cow fertility

	Concentrate bu	Concentrate build-up strategy		
	Rapid build-up	Delayed build-up	Significance ¹	
	(RBU)	(DBU)		
Production				

Dry matter intake (kg/day)	20.5	21.4	NS
Forage intake (kg DM / day)	9.0	10.4	***
Concentrate intake (kg DM / day)	11.5	11.0	**
Milk yield (kg/day)	38.9	37.9	NS
Milk constituents			
Milk fat (g/kg)	40.5	41.6	NS
Milk protein (g/kg)	34.2	33.3	NS
Energy Balance (MJ / day)	-21.3	-6.3	NS
Fertility			
Pregnancy to 1 st service (%)	41	36	NS
100 days in-calf rate (%)	63	64	NS
Overall pregnancy rate (%)	74	86	NS

¹ NS, P>0.05; *, P<0.05; **, P<0.01; ***, P<0.001



Figure 3 Forage intake during the first 140 days of lactation

Despite the lack of treatment effects on milk production, cows on DBU returned to positive energy balance earlier (week 7 post-calving) than those on RBU (week 19 post-calving). This effect can be attributed to improvements in dry matter intake throughout the experimental period and a lower milk yield during weeks 3-7 of lactation. Nevertheless, concentrate build-up strategy had no significant effect on any of the fertility parameters measured within this study (Table 1). There was, however, a numerically higher number of cows pregnant at the end of this breeding period with the delayed build-up feeding strategy.

Adopting a delayed concentrate build-up strategy in early lactation improved forage intake in early lactation, as well as overall intake, and had no detrimental effect on production performance. This resulted in an improved energy status of the cows on this treatment, and this should contribute to improved health and fertility. However, no significant effects on fertility were observed within this study.

How does the response to additional concentrates differ depending on when they are offered?

The previous study examined strategies for increasing concentrate feed levels in early lactation. However, there is relatively little information available on the response of dairy cows to increasing levels of concentrate supplementation during early to mid lactation. While historical data have indicated that the response to the inclusion of additional concentrates decreases with stage of lactation (Broster and Broster, 1983), no similar work has been undertaken using high-yielding dairy cows.

This issue was examined in a recent study at Hillsborough involving 80 winter-calving Holstein-Friesian dairy cows (40 primiparous and 40 multiparous). Post-calving all animals were offered a basal diet of grass silage, maize silage and concentrate (25, 25 and 50% of the diet on a dry matter basis, respectively) plus 1.0 kg concentrate/cow/day in the milking parlour. This diet was formulated to have a protein content of 180 g/kg dry matter. Cows were allocated to one of four treatments, namely a control treatment in which cows were offered the basal diet only, or treatments in which an additional 4.0 kg concentrate/cow/day was included into the diet at 2, 6 or 10 weeks post-calving.

During the first 29 weeks of lactation offering an additional 4.0 kg concentrate/cow/day from weeks 2, 6 or 10 post-calving had no effect on milk yield or milk composition, but resulted in a significant increase in total dry matter intake (Table 2). However, there was a trend for a greater milk yield response when supplementation commenced at week 2 (P=0.051) post-calving, compared to weeks 6 (P=0.135) or 10 (P=0.944) (Figure 4). The milk production response was then examined during each 4-week period after extra concentrates were introduced in the diet i.e. at weeks 1-4, 5-8, 9-12 etc. post inclusion. When analysed on this basis, concentrate inclusion at week 2 post-calving results in a significant increase in milk output (compared to the control treatment) during weeks 1-4 (P=0.014), 5-8 (P=0.008), and 9-12 (P=0.015) weeks post inclusion. However, when additional concentrates were offered at week 6 and 10 post-calving, no significant milk yield response was observed during any of the 4-week intervals examined.

	Treatment				
	Control	2	6	10	Significance ¹
Dry matter intake (kg/day)	17.1	19.7	19.1	19.1	**
Milk yield (kg/day)	32.1	33.8	33.5	33.2	NS
Milk constituents (g/kg)					
Fat	37.6	37.9	38.8	39.4	NS
Protein	32.5	32.6	32.7	32.4	NS
Lactose	50.3	49.5	50.0	49.7	NS
Fat + protein yield (kg/day)	2.22	2.38	2.36	2.36	NS
Liveweight (kg)	566	566	573	571	NS
Energy balance (MJ/day)	-58.4	-44.2	-34.8	-30.3	NS

Table 2Effects of offering additional concentrates at 2, 6 and 10 weeks post-calving,on dry matter intake and milk production (day 1 to 203 of lactation)

¹NS, P>0.05; **, P<0.01



Figure 4 Effects of offering additional concentrate from weeks 2, 6 and 10 postcalving on average daily milk yield (kg/day)

The decreasing response to additional concentrates offered in late lactation appears to be reflected in changes in body tissue reserves. For example, there was a tendency for an improved energy balance (Table 2) in cows receiving additional concentrate later in lactation. This indicates that the responses of dairy cows to supplementation changes according to the stage of lactation when additional concentrates are offered. In early lactation, the additional nutrients consumed tended to be partitioned mainly towards extra milk, whereas after peak milk yield the additional nutrients consumed tended to be partitioned primarily to body tissue reserves.

Complete diet feeding vs out-of-parlour feeders: has the debate moved on?

Historically dairy cows within the UK were offered concentrates twice daily in the milking parlour. However, concentrate feed levels on many farms have increased considerably above the 8-10 kg/cow/day normally considered as the maximum for twice daily in-parlour feeding. Out-of-parlour feeders and complete diet mixer wagons have become increasingly popular methods of achieving higher concentrate intakes. One of

the benefits claimed for both of these systems is the improved synchrony in the supply of dietary fermentable energy and nitrogen, with concentrates being consumed "little and often" throughout the day. Mixer wagons also offer an opportunity to incorporate by-product ingredients, which are often cheaper than cereals, into the ration.

However, there has been considerable debate in recent years as to the relative merits of complete diet feeding compared to the same quantity of concentrate being offered via out-of-parlour feeders. In a review of 14 studies involving comparisons of complete diet feeding and separate feeding of concentrates and silage, Ferris *et al.* (1998) concluded that improvements in milk yield were generally obtained when the proportion of concentrate in the diet was 0.6 or higher. However, many of these studies did not include high-yielding cows or grass silage-based diets. To address this issue a series of three studies were conducted at AFBI Hillsborough (Gordon *et al.*, 1995; Yan *et al.*, 1998) in which the performance of cows offered concentrates, either through an out-of-parlour feeder or in the form of a TMR, was examined.

The results of these studies illustrated that only small improvements in animal performance and efficiency were achieved when concentrates were offered as a complete diet as opposed to being offered separately from the forage through out-of-parlour feeders (Table 3). In these three experiments the average concentrate proportion in the diet was 0.61 and the forage proportion of the diet consisted of good quality grass silage. In contrast to previous work, the results of this series of studies suggest that offering concentrates in the form of a complete diet, as opposed to being fed separately from the forage, actually decreased total dry matter intake by 3%. However, milk yield was increased by 6%, and milk fat concentrations reduced by 4% when cows were offered concentrates as a complete diet rather than separately from the forage.

Table 3Responses to complete-diet feeding vs computerised out-of-parlourfeeding of a concentrate supplement; weighted overall mean responses across threeexperiments (Ferris *et al.*, 1998)

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	DM intake (kg/day)	DM intake (kg/day)	DM intake (kg/day)	yield (kg/day)	(g/kg)	protein (g/kg)	fat plus protein (kg/day)
Weighted mean	7.5	11.5	19.0	32.7	39.8	33.9	2.40
Response to complete diet	-0.9	0.3	-0.6	1.9	-1.7	-0.1	0.05
Proportional response	-0.11	0.03	-0.03	0.06	-0.04	0.01	0.02

However, a valid criticism of these studies was the fact that with the out-of-parlour feeding treatment, the silage component of the diet was mixed using a mixer wagon prior to feeding, and fresh silage was offered daily. In practice when silage and concentrates are offered separately, the silage is likely to be offered in whole blocks or part blocks along a feed barrier, with fresh allocation gauged to last 3 to 4 days. To address the practical shortcomings associated with the earlier studies, two separate experiments were conducted to examine cow performance associated with two winter feeding systems, namely daily complete diet feeding vs separate feeding of the forage and concentrate components. The latter system was specifically designed to incorporate twice weekly feeding of whole blocks of silage (easy-feed system). In addition, an assessment of labour requirements associated with each of the two feeding systems was made.

Experiments 1 and 2 involved 64 and 86 Holstein-Friesian dairy cows, respectively, with the duration of these studies being 144 and 146 days, respectively. Concentrate feed levels in these studies were approximately 8.4 and 10.2 kg/cow/day, respectively. The forage component of the diet consisted of 33% maize silage and 66% grass silage (dry matter basis) in Experiment 1 and 30% maize silage and 70% grass silage (dry matter basis) in Experiment 2. The complete diet was prepared daily using a complete diet mixer wagon. The easy-feed system involved whole blocks of grass silage and maize silage being placed along a movable easy-feed barrier on two occasions per week. The design of the feed barrier allows cows to push the barrier out whilst eating their way

through the forage. The concentrate portion of the ration was offered using out-ofparlour feeders.

In each of the two experiments, feeding system had no effect on food intake, milk production, milk composition or condition score of cows at the end of the winter period (Table 4), and this may reflect the fact that concentrate proportions in the diet on Experiments 1 and 2 were proportionally 0.48 and 0.54, respectively. In terms of labour, it took approximately 156 minutes per week to feed silage within the easy-feed system and approximately 210 minutes per week to feed with the complete diet system. This equates to 78 minutes twice weekly with the easy feed system and 30 minutes every day with the complete diet system.

	Treatr		
-	Daily complete diet	Twice weekly easy-feed	Significance
Experiment 1			
Dry matter intake (kg/day)	17.6	17.0	NS
Milk yield (kg/day)	28.4	29.6	NS
Milk fat (g/kg)	39.4	38.5	NS
Milk protein (g/kg)	33.5	34.1	NS
Milk fat plus protein (kg/day)	2.1	2.1	NS
Final live weight (kg)	539	532	NS
Final body condition score	2.4	2.3	NS
Experiment 2			
Dry matter intake (kg/day)	18.7	18.5	NS
Milk yield (kg/day)	30.0	30.6	NS
Milk fat (g/kg)	41.8	40.2	NS
Milk protein (g/kg)	33.9	33.9	NS
Milk fat plus protein (kg/day)	2.2	2.2	NS
Final live weight (kg)	561	556	NS
Final body condition score	2.5	2.5	NS

Table 4Effect of feeding system on average cow performance over the winterperiod in two separate experiments (Ferris *et al.*, 2006)

¹NS, P>0.05

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The results of this study demonstrate that a simple feeding system, in which the forage part of the diet is offered twice weekly, can result in similar performance to that of a more complex system involving daily feeding. Also, it is clear that mixed forages can be offered as alternate blocks at a feed barrier with good performance and reduced labour. However, this is heavily dependent on maintaining the integrity of the block when placing them at the feed barrier.

Can we reduce the protein content of the concentrates offered (at high concentrate levels)?

Dairy cows require protein for milk production, maintenance, growth, pregnancy and immune function, while protein supply is a key driver of food intake and milk production. These protein requirements are met directly from dietary protein, rumen microbial protein, and from the mobilisation of body tissue reserves. Within Northern Ireland the overall protein content of dairy cow diets is normally approximately 18% (dry matter basis). However, there is currently considerable interest in the use of lower protein diets for dairy cows. The reasons for this are as follows:

- Protein ingredients tend to be the most expensive component of concentrate feeds.
- 2) Offering lower protein diets has been suggested as one option by which to improve cow fertility. As protein supply is a key driver of milk production, reducing the dietary protein content may provide a tool by which to 'reduce milk yield', while maintaining feed intake. This in theory should improve energy status, and as such, also improve fertility.
- 3) Dietary protein is used inefficiently by dairy cows, with approximately 70% of the protein (nitrogen) consumed ending up in manure. Nitrogen excretion is of significant environmental concern at present due to nitrogen losses to waterways (via leaching), which contributes to aquatic eutrophication. Nitrogen lost to the atmosphere as ammonia (via volatilisation), and nitrous oxide (via denitrification), contribute to terrestrial eutrophication and global warming, respectively. Research from AFBI Hillsborough has illustrated that nitrogen excretion in manure is highly correlated with dietary nitrogen intake (Yan *et al.*, 2006).

Thus while there may be clear benefits of offering lower protein diets, this strategy will only be acceptable if it can be achieved without a significant reduction in milk output, and without having a detrimental effect on cow health. To address this issue, a single lactation study was undertaken at AFBI Hillsborough involving 90 high-yielding Holstein-Friesian dairy cows (45 primiparous and 45 multiparous). These were allocated to diets containing one of three dietary protein concentrations (173, 144, or 114 g protein/kg dry matter) from calving until day 150 of lactation. All cows were offered a complete diet containing 55% concentrate and 45% forage (dry matter basis). The forage offered was a mixture of grass silage (27%) and maize silage (18%).

During the first 150 days of lactation, an increase in dietary protein concentration resulted in an increase in dry matter intake and milk yield (Table 5). Animals offered 114 g protein/kg dry matter produced milk with a higher milk fat concentration than animals offered diets containing 144 or 173 g protein/kg dry matter. There was no effect of diet on milk protein concentrations. In addition, the increase in dry matter intake at higher protein levels was not sufficient to supply the additional ME requirement associated with the higher milk output. Thus animals on the high protein diet experienced a more severe NEB than animals offered the medium and low protein diets.

The results presented so far relate to the first 150 days of lactation, and these have demonstrated that milk production is substantially reduced when cows are offered a diet with a protein content of 144 g/kg dry matter. However, it is known that the protein requirements of dairy cows decrease in later lactation. To allow this to be examined in more detail, half of the cows on the high protein diet were moved onto a medium protein diet after day 150 of lactation, while the remaining cows continued to be offered the high protein diet.

During mid to late lactation (day 151 to 305), decreasing the dietary protein concentration from 173 to 144 g/kg dry matter had no significant effect on milk yield or dry matter intake. This highlights that the efficiency of use of dietary nitrogen can be improved by feeding diets with lower protein concentrations (144 g protein/kg dry matter) without detrimental effects on production (Figure 5).

	Dietary protein concentration (g/kg dry matter)			
	114	144	173	Significance ¹
Production				
Dry matter intake (kg/day)	16.5	18.0	18.6	***
Milk yield (kg/day)	25.4	31.8	35.4	***
Milk composition (g/kg)				
Fat	42.0	38.3	38.1	*
Protein	31.4	32.3	32.4	NS
Lactose	48.2	48.1	47.8	NS
Energy balance (MJ/day)	12.8	0.24	-11.1	**
Nitrogen efficiency (Milk N/Dietary N)	0.31	0.30	0.26	***
Fertility				
Pregnancy to 1 st service (%)	35	30	28	NS
Pregnancy to 1 st and 2 nd service (%)	55	63	52	NS
100 day in-calf rate (%)	83	67	62	NS
Overall pregnancy rate (%)	100	93	87	NS

Table 5Effects of dietary crude protein concentration on dry matter intake, milkyield and milk composition (fat, protein and lactose) during the first 150 days oflactation, and on fertility

¹NS, P>0.05; *, P<0.05; **, P<0.01; ***, P<0.001



Figure 5 Effect of lowering the dietary protein content at day 151 of lactation (from 173 to 144 g protein/kg/dry matter) on milk production

Dietary protein concentration had no significant effect on any of the reproductive parameters assessed. However, animals receiving 114 g protein/kg dry matter tended to have a higher 100-day in-calf rate compared to animals receiving 144 and 173 g protein/kg dry matter (Table 5). Within a six month breeding period, 100% of cows became pregnant when allocated a diet containing 114 g protein/kg dry matter during the first 150 days of lactation, compared to 92.9% and 86.7% of cows receiving diets containing 144 and 173 g protein/kg dry matter respectively.

A protein content of 114 g/kg dry matter was inadequate. While the efficiency of nitrogen utilisation was improved with a protein content of 144 g/kg dry matter, there were substantial decreases in production. However, there is scope to reduce dietary protein levels to 144 g/kg dry matter after mid lactation without a loss in performance, but with the benefits of reduced feed costs.

Can the efficiency of nitrogen utilisation be improved by supplementing with specific amino acids?

The results of the previous study have demonstrated that diets containing 144 g protein/kg dry matter are inadequate in terms of maintaining high levels of cow performance. However, there is evidence that this decline in performance may not be due to an overall deficit of protein in the diet, but rather due to a shortage of one or two specific amino acids. The two amino acids that are normally most likely to be limiting in dairy cow diets are lysine and methionine. The optimal ratio of lysine to methionine in dairy cow diets has been identified as 3:1. While the majority of UK rations generally supply sufficient quantities of lysine, in most cases methionine is undersupplied. Methionine supplementation can be achieved using ruminally protected methionine.

To address this issue, 48 Holstein-Friesian dairy cows were allocated to one of two diets; control (180 g protein/kg dry matter) and low protein + methionine (150 g protein/kg dry matter with an additional supplement of 40 g/head/day of protected methionine). Diets consisted of 40% forage (16% maize silage, 24% grass silage) and 60% concentrates on a dry matter basis. Cows entered the experiment at calving and remained on the study until day 210 of lactation.

Offering a low protein diet supplemented with methionine was found to have no effect on either dry matter intake or milk yield compared to a control diet (Table 6). However, milk urea nitrogen was significantly lower in animals offered a low protein diet supplemented with methionine compared to a control diet, suggesting that milk was being used as an excretory route for excess nitrogen with the control diet. In addition, the efficiency of nitrogen utilisation was significantly higher when animals were offered the low protein + methionine treatment compared to the control treatment, a reflection of the lower nitrogen intakes associated with this treatment.
	Treatment		
	Control	Low protein + methionine	Significance ¹
Dry matter intake (kg/day)	19.7	19.4	NS
Milk yield (kg/day)	32.9	32.6	NS
Milk Urea (mg/kg)	164	134	***
Nitrogen utilisation efficiency			
Nitrogen intake (g/day)	569	466	***
Milk nitrogen output (g/day)	180	180	NS
Milk nitrogen/nitrogen intake (g/g)	0.31	0.38	***

Table 6Effect of supplementing a low protein diet with methionine on cowperformance and the efficiency of nitrogen utilisation

¹NS, P>0.05; ***, P<0.001

These results indicate that there may be scope to reduce dietary protein levels without having a detrimental effect on cow performance, provided the diet is supplemented with appropriate limiting amino acids. This has the potential to improve the efficiency of nitrogen utilisation, and as such reduce nitrogen excretion by animals in faeces and urine.

Can dietary protein levels be used as a tool to manage the energy status of individual cows?

As already discussed, severe and prolonged periods of NEB are associated with poor reproductive performance in high-yielding dairy cows (Jorritsma *et al.*, 2003) and has been associated with a delay in the onset of luteal activity (Jolly *et al.*, 1995), an extended interval to first service (Butler *et al.*, 1981) and decreased conception rates (Domecq *et al.*, 1997). Thus it has often been suggested that reproductive performance could be improved by reducing the extent of NEB experienced by dairy cows.

A key outcome of a recent AFBI study was that when total dietary protein content was reduced from 180 to 150 g/kg dry matter of the total diet, total milk output was reduced,

while dry matter intake was unaffected. The overall effect was an improvement in the energy status of the cow (Law *et al.*, 2009). While there was a significant reduction in milk yield within this study, this was associated with a reduced protein diet being offered for a 150-day period. However, it is possible that a lower protein diet could be offered for a shorter period of time to improve the energy status of the cow, without having a long term detrimental effect on cow performance.

To address this issue a study was undertaken in which a Control diet containing 180 g protein/kg dry matter was compared with an "Individual Cow" management treatment. In the latter treatment the energy balance of individual cows was calculated weekly and manipulated to reach a predefined energy balance trajectory. After calving, all cows on this individual cow management treatment were initially offered a diet containing 170 g protein/kg dry matter, plus 5.22 kg dry matter of a concentrate containing 170 g/kg dry matter in the parlour. Commencing 3 weeks after calving, the protein content of the parlour concentrate was altered, if required, in an attempt to maintain energy balance along a predefined trajectory. If the extent of NEB was greater than planned, the protein content of this concentrate was reduced to 140 g/kg dry matter until the energy balance was back on the predefined trajectory. Alternatively, if the extent of NEB was less than the predisposed trajectory, the protein content of the parlour concentrate was increased to 220 g/kg dry matter. This was continued until day 210 of lactation.

Cows on the individual cow treatment had a higher dry matter intake than those on the control treatment (Table 7). None of milk yield, milk fat or milk protein concentrations were affected by dietary treatment. Due to the increase in dry matter intake combined with the absence of a milk yield response, animals on the individual cow treatment had a significantly improved daily energy balance.

	Treatment		
	Control	Individual cow	Significance ¹
Production			
Dry matter intake (kg/day)	19.7	21.0	***
Milk yield (kg/day)	32.8	32.7	NS
Milk constituents			
Fat (g/kg)	37.7	39.4	NS
Protein (g/kg)	33.8	33.9	NS
Urea (mg/kg)	164	174	***
Mean liveweight (kg)	563	557	NS
DEB (MJ/day)	18.4	31.7	***
Reproduction			
Onset of cyclicity (day)	31.5	26.1	NS
Pregnancy to 1 st and 2 nd service (%)	61.9	60.0	NS
100 days in-calf rate (%)	71.4	79.2	NS
Overall pregnancy rate (%)	85.7	95.8	NS

Table 7The effect of managing individual cows to achieve a preset energy
balance, compared to group management, on milk production and energy balance
between 1 and 210 days of lactation

¹NS, P>0.05; ***, P<0.001

However, despite this improvement in energy balance, feeding strategy had no significant effect on any of the reproductive parameters examined (Table 7), although there was a trend for a higher conception rate with the individual cow management treatment.

Adjusting the protein content of the diet shows promise as a strategy by which the energy balance of dairy cows can be manipulated. Furthermore, low protein diets can be offered for relatively short periods of time during lactation without having a long term detrimental effect on cow performance. Although energy balance was improved in this experiment, no clear fertility benefits were identified.

Can we improve reproductive performance with specifically formulated concentrates?

It is considered unlikely that the decline in reproductive performance has a direct 'genetic' origin as conception rates in non-lactating Holstein-Friesian heifers have remained high (at 70-80%) during a period when milk production has increased by 218% (Beam and Butler, 1999). While a number of studies presented within this paper have examined the impact of nutritional strategies on dairy cow energy balance, and subsequent reproductive performance, there is increasing interest in the use of specific nutrients to target various aspects of the reproductive system. For example, diets high in starch have been shown to increase circulating concentrations of insulin (van Knegsel et al., 2007), thus promoting the resumption of ovarian activity and subsequent cyclicity (Gong et al., 2002), while diets high in dietary fat have been shown to be beneficial to embryo growth rate and subsequent survival (Fouladi-Nashta et al., 2007). Indeed the results of a recent study by Gong et al. (2002) indicate that the interval to the onset of cyclicity was reduced when cows were offered a diet high in starch during the first 50 days of lactation. However, work by Fouladi-Nashta et al. (2005) has shown that cows on a high starch diet (resulting in high circulating insulin levels) produced a significantly high number of poor quality oocytes (resulting in lower conception rates). The latter authors state that high plasma insulin levels are detrimental to oocyte quality and suggest that once cycling has commenced a high fat diet is beneficial to blastocyst growth rate in lactating dairy cows (Fouladi-Nashta et al., 2007).

This issue was examined in a study at Hillsborough involving 48 dairy cows, with cows offered either a standard TMR containing 180 g protein/kg dry matter, or a "fertility improver" diet. With the latter, a high starch diet was offered during the first 50 days of lactation to encourage the commencement of cyclicity, followed by a low-starch/high-fat diet (supplemented with 750 g of protected fat per day, Megalac) between day 51 and day 120 of lactation. This latter diet was offered to avoid the detrimental effects of high insulin levels on oocyte quality (Fouladi-Nashta *et al.*, 2005).

The diets offered had no effect on food intake, milk production or milk composition. In addition treatment had no effect on the interval to the onset of cyclicity, although, cows offered the fertility improver ration tended to cycle earlier than those offered the control diet (Table 8).

	Treatment		
	Control	Fertility improver	Significance ¹
Onset of cyclicity (days)	31.5	21.7	NS
Pregnancy to 1 st and 2 nd service (%)	61.9	56.0	NS
100 day in-calf rate (%)	71.4	68.0	NS
Overall pregnancy rate (%)	85.7	88.0	NS

Table 8The effect of offering a fertility improver ration on conception rate and
oestrous cycle characteristics

¹NS, P>0.05; ***, P<0.001

Although the concept of offering high starch diets in early lactation (to reduce the interval from calving to commencement of cyclicity), followed by a high fat diet (to improve embryo quality) appears to be based on sound scientific principles, the results of the current experiment provide no evidence of an increase in overall conception rate with the fertility improver ration. It is possible that this lack of response was related to the relatively low numbers of cows on each of the diets, and that a different result may have been obtained if much larger group sizes had been used.

Future research

Feed costs, especially concentrates, represent approximately 70% of variable costs on Northern Irish dairy farms. During the last decade annual concentrate inputs have increased by approximately 1.0 tonne/cow, while the increase in milk output has been approximately 1000 kg/cow. This poor response highlights a high degree of inefficiency in concentrate use on dairy farms. Future research is required into the development of

robust concentrate allocation strategies for dairy cows which optimise forage use, and ensure nutrient requirements are met across the range of cows within herds.

As technology advances, farms with automated weighing and milk quality monitoring systems will be able to more accurately predict cow energy status. However, farmers often fail to make use of much of this information and research is required to develop algorithms to best allow the farmer to utilise these data within precision management dairy system.

In dairy systems involving high concentrate inputs, the importance of grass silage quality on whole farm profitability may be greatly underestimated. Grass silage remains the key forage within most winter milk production systems, but quality remains extremely variable. Research is required to evaluate the importance of multi-cut silage systems within a whole farm systems context. Furthermore, the relationships between silage quality/concentrate use and whole farm profitability need to be established. This could be achieved by linking CAFRE Benchmarking data with silage quality/feed input data.

The transition period (4 weeks pre-calving to 4 weeks post-calving) is the most traumatic period in the annual cycle of the dairy cow. During this time the cow experiences severe physiological, hormonal, nutritional and metabolic stress. This is due in part to a rapid increase in milk production, and the inability of the cow to consume sufficient feed to meet her metabolic requirements. Previous research has shown that most infectious diseases and metabolic disorders occur during the transition period (Drackley, 1999), with a negative effect on cow performance (Wallace *et al.*, 1996) and reproductive performance (Borsberry and Dobson, 1989) during the following lactation. Developing feeding strategies to improve the transition from the dry period to lactation is critical.

Conclusions

• Delaying concentrate build-up in early lactation improved forage intake, but had no significant effect on milk production. This delayed concentrate build-up

regime improved the energy status of the cow, but did not significantly affect fertility.

- The introduction of additional concentrates into the diet in early lactation resulted in a large milk yield response. However, introducing additional concentrates into the diet later in lactation resulted in a poorer milk yield response, with a large part of the additional energy consumed being partitioned to body tissue reserves.
- Cow performance was unaffected when the concentrate and forage components of the diet were offered separately (the latter twice weekly at a moveable feed barrier), rather than being offered as a mixed ration.
- Reducing the dietary protein content of the diet from 173 g/kg dry matter to either 144 or 114 g/kg dry matter improved the efficiency of nitrogen utilisation but resulted in a substantial reduction in cow performance. However, there is scope to reduce dietary protein levels to 144 g/kg dry matter after mid lactation without a loss in performance, but with benefits of reduced feed costs.
- The use of rumen protected methionine may offer an opportunity to reduce dietary protein levels and improve N utilisation efficiency, without loss in performance.
- Dietary protein content can be used as a tool to manage energy balance. Cows whose energy status was managed to achieve a predefined energy balance had improved dry matter intake and energy status, although this had no effect on reproductive performance.
- Offering a fertility improver ration (high starch in early lactation and high fat in late lactation) tended to reduce the interval from calving to commencement of cyclicity, but resulted in no improvement in overall reproductive performance.

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