Effect of milking frequency and nutrition in early lactation on milk production and body condition in grazing dairy cows

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ABSTRACT

Multiparous Holstein-Friesian cows (n = 120), grazing pasture and milked twice-daily (2X) from calving until ~35 days in milk, were allocated to one of four treatments. Treatments consisted of 2X or once-daily milking (1X), and UnRes (15 kg dry matter (DM)/cow/day) or Res (9 kg DM/cow/day) pasture intake for three weeks. Following treatment, all animals were milked 2X and offered pasture (residuals of 1,600kg DM/hectare) for 20 weeks. During the treatment period (Weeks 1-3), yields of milk, fat, protein and lactose decreased with reduced milking frequency and feeding level. Interactions existed for milk, protein and lactose yields and the decrease due to 1X milking was greater in the UnRes than Res cows. During the post-treatment periods (Weeks 4-12 and Weeks 13-23) no interactions were present. Restricted cows produced less milk, fat, protein and lactose compared with UnRes cows during Weeks 4-12, but no difference existed during Weeks 13-23. During Weeks 4-12, cows milked 1X produced less milk, fat, protein and lactose than those milked 2X and these differences remained during Weeks 13-23. During the treatment period, body condition score was less in Res than UnRes cows, however there was no effect of milking frequency on body condition score. Neither feeding level nor milking frequency altered post-partum anoestrus interval.

Keywords: milking frequency; nutrition; dairy cow.

INTRODUCTION

In pasture-based dairy systems, once-daily (1X) milking is a management strategy often used during early lactation to reduce the heavy workload associated with seasonal calving or to alleviate nutritional stress when pasture quality or quantity is reduced (Clark *et al.*, 2006). The immediate and longer-term effects of restricted pasture in early lactation have been well researched (Broster, 1974; Bryant & Trigg, 1974; Roche, 2007; Burke *et al.*, 2010); however, experiments investigating the combination of reduced milking frequency (MF) and altered nutritional regime or feeding level (FL) in early lactating grazing dairy cows are limited. Previous experiments have been either very short term (Auldist & Prosser, 1998; two days), long term (O'Brien *et al.*, 2005; 20 weeks), conducted at dry off (Lacy-Hulbert *et al.*, 1999) or used high-producing dairy cows in a confinement system (Remond *et al.*, 2002). Nevertheless, these studies consistently indicate an immediate milk production loss when cows are switched to 1X milking of 13% to 32%. However, the interaction between FL and MF, and the duration and severity of the carry–over effect when cows are returned to normalised intakes and twice-daily (2X) milking remains unclear (Auldist & Prosser, 1998; Remond *et al.*, 2002; Phyn *et al.*, 2010). In addition, while some studies indicate a positive effect of 1X milking on energy balance and fertility (Auldist & Prosser, 1998; Rhodes *et al.*, 1998; O'Brien *et al.*, 2005), the longer term effects of reduced MF and FL on body condition score (BCS) are yet to be detemined. Thus, the objective of this experiment was to determine the effects of 1X milking at two FL on immediate and longer-term milk production, BCS and post-partum anoestrus interval in early lactating, grazing dairy cows.

MATERIALS AND METHODS

Animals and treatments

Experimental work was conducted at the Westpac Taranaki Agricultural Research Station, Hawera between July 2009 and April 2010. All procedures involving animals were approved by the Ruakura Animal Ethics Committee, Hamilton, New Zealand.

Multiparous Holstein-Friesian and Holstein-Friesian x Jersey cross cows (n = 120) were grazed as one herd and offered a generous pasture allowance with post-grazing residuals of >1,600 kg DM/ha for the first 34 ± 6 (mean ± standard deviation) days in milk (DIM). Cows were then allocated to one of four treatments, which were balanced for DIM, in a 2 x 2 factorial arrangement. Treatments consisted of two MF (1X or 2X) and two FL (Unrestricted (UnRes) or Restricted (Res)) for three weeks. At the start of the treatment period, pasture allowances were incrementally increased or decreased over a three day period to achieve an average dry matter intake (DMI) of 15 and 9 kg DM/cow/d in the UnRes and Res treatments, respectively.

To reduce variability due to DIM, two experimental cohorts were used. Cows that calved from 17 July to 7 August (n = 66) were included in Cohort 1 and cows that calved from 8 August to 1 September (n = 54) were included in Cohort 2. Both cohorts were managed identically. Following the treatment period, all animals were grazed together (residuals of >1600 kg DM/ha), and were milked 2X for 20 weeks.

Pasture measurements and analysis

Pre- and post-grazing pasture mass was estimated on three days each week using calibrated visual assessment during the treatment periods, and on one day each week for the remainder of the experiment by the same person (Roche, 2007). Average daily DMI (kg DM/cow/d) was estimated as the product of the difference between the pre- and post-grazing mass and area grazed, divided by the number of cows.

Representative pasture samples were collected on five days each week by hand-clipping pasture to the predicted grazing height from paddocks immediately before grazing. Samples were bulked weekly and dried at either 100°C for DM determination or 60°C for analysis of nutrient composition (Roche, 2007). Metabolisable energy (ME) content of the pasture was estimated as described by Roche (2007) and energy intakes (MJ ME/cow/d) were calculated as the product of pasture DMI and ME.

Milk and animal measurements

Individual milk yields were recorded daily (GEA, Oelde, Germany). Milk composition (Milkoscan; Foss Electric, Denmark) and BCS (1-10 scale where 1 = Emaciated and 10 = Obese; Roche *et al*., 2004) were determined weekly for two weeks before, until 10 weeks post-treatment initiation, and fortnightly thereafter.

Blood samples were collected on one day each week after the morning milking from a subset of cows (n = 18/treatment) for the two weeks prior to treatment until the planned start of mating on 10 October. Plasma was harvested and stored at -20°C for subsequent analysis of progesterone (P_4); (Siemens coat-a-count RIA, USA). The post-partum anoestrus interval was estimated as the number of days between calving date and the date of the first plasma sample in which P_4 content was greater than 1 ng/mL.

Statistical analysis

Pre-treatment measurements were used as a covariate and means of daily data for each week and time period (Weeks 1-3, 4-12 and 13-23) were calculated for each cow. These data were then analysed using mixed models fitted with REML in GenStat (Payne *et al*., 2009) including cohort, FL, MF and the interaction of FL and MF as fixed effects and cow as a random effect. Differences were considered significant at $P < 0.05$, and a trend declared at $P < 0.10$.

RESULTS AND DISCUSSION

During the treatment period (Week 1-3), pasture allowance, DMI, and energy intake for the UnRes cows were 31.4 ± 3.9 kg DM/cow/d, 14.3 ± 3.6 kg DM/cow/d, and 173 ± 44 MJ ME/cow/d, respectively. In comparison, the same variables for the Res cows were 14.4 kg ± 5.6 DM/cow/d, 8.3 ± 2.9 kg DM/cow/d, and 100 ± 35 MJ ME/cow/d, respectively. This represented a 42% decrease in dry matter and energy intake for the Res cows. During the post-treatment periods, (Weeks 4-12 and 13-23) the average pasture allowance was 39.2 ± 6.1 kg DM/cow/d, DMI was 14.1 ± 4.6 kg DM/cow/d and energy intakes were 170 ± 56 MJ ME/cow/d.

Table 1 presents milk production during the three week treatment and 20 week post-treatment periods. During the treatment period (Week 1-3), yields of milk, fat, protein and lactose were less in the Res and 1X cows compared with the UnRes and 2X cows, respectively. Furthermore, there was an interaction between FL and MF for milk, protein and lactose yield. Decreases in milk, protein, and lactose yields due to 1X milking were greater in UnRes than Res cows (21 vs. 14%, 21 vs. 12%, and 24 vs. 16%, respectively). These data support those of Auldist and Prosser (1998), who reported a greater production loss when well-fed grazing cows were switched to 1X milking (20%) than when cows on a resticted allowance were swithed to 1X milking (14%). Remond *et al*. (2002) also reported a trend for a greater decrease in milk production due to 1X milking in cows fed a high energy ration compared with those fed a low energy ration (32 vs. 23%, respectively). Phyn *et al*. (2010) suggested the interaction between MF and FL may be due to well-fed cows producing more milk and consequently reaching maximum udder capacity earlier. Thus if MF is reduced, their udders are subjected to longer periods of negative feedback mechanisms, evoking a greater negative effect on milk production (Davis *et al.,* 1999; Stelwagen *et al*., 2001).

In the present study, cows milked 1X with a restricted FL produced the least milk and milk components during the treatment period (Table 1 and Figure 1a), indicating a separate and additive negative effect of MF and FL on milk production.

This confirms previous results (Guinard-Flament *et al*., 2007), where it was postulated that the milk yield loss with reduced MF was additional to any effects of underfeeding, a MF and FL acted via independent physiological pathways to decrease mammary blood flow, lactose and milk production. Feed restriction reduces energy intake and available nutrients, resulting in reduced total cardiac output and decreased mammary blood flow, whereas reduced MF decreased mammary blood flow via local rather than systemic regulation (Davis *et al.,* 1999; Stelwagen, 2001; Guinard-Flament *et al*., 2007; Wall & McFadden, 2007).

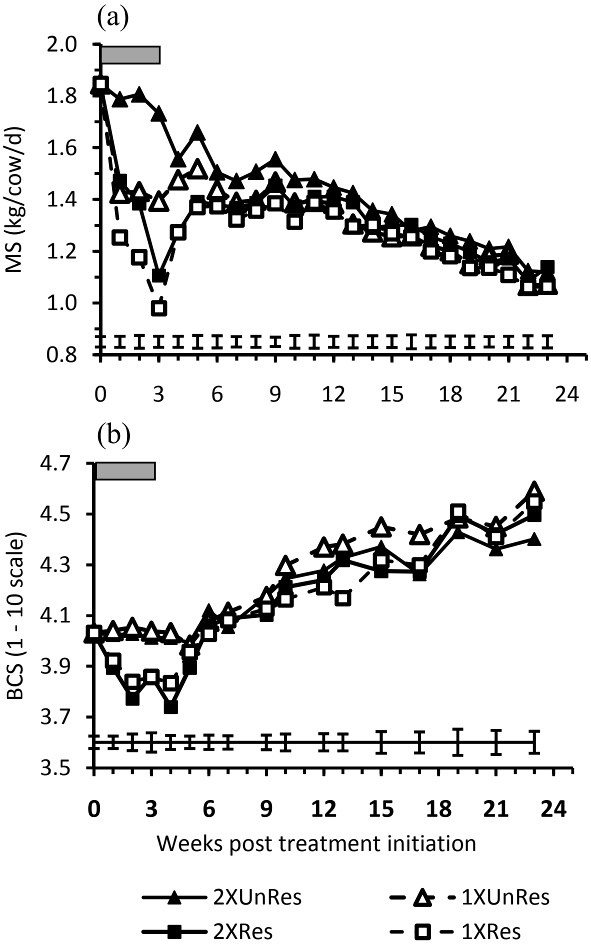
Reduced milk production with 1X milking is due to decreased udder capacity or mammary epithelial cell number (Carruthers *et al.,* 1993) and down-regulation of milk synthesis genes (Boutinaud *et al*., 2008; Littlejohn *et al*., 2010; Grala *et al*., 2011). This combination of decreased cell number and activity may be responsible for the negative milk production carry-over effect when 1X cows are returned to 2X milking. In the present study, from Weeks 4-12 (1-8 weeks after returning to 2X milking), cows milked 1X produced less milk (6%), fat (4%), protein (5%) and lactose (6%) than those continually milked 2X and these differences remained during weeks 13-23 (Table 1). The temporal milksolids (MS; fat + protein) production pattern is depicted in Figure 1a; the negative carry–over effect of three weeks of reduced MF, continued until Week 20 (17 weeks after the cows had returned to 2X milking). Grala *et al*. (2011) reported that transcription of genes involved in milk synthesis remained lower for at least three to six weeks after cows milked 1X for three or six weeks were switched to 2X milking, indicating a sustained reduction in mammary cell activity. The longer-term production loss may also be a consequence of decreased udder capacity (Carruthers *et al*., 1993) due to increased net cell turnover rate. This results in increased cell death via apoptosis relative to cell proliferation, in cows milked 1X compared with 2X

TABLE 1: Milk production from cows (34 ± 6 days in milk) consuming an unrestricted (UnRes: 14.3 ± 3.6 kg DM/cow/d) or restricted (Res: 8.3 ± 2.9 kg DM/cow/d) pasture intake and milked once (1X) or twice (2X) daily for three weeks. Following the treatment period (Weeks 1-3), all cows were offered a generous pasture allowance (Residuals >1,600 kg DM/ha) and milked 2X for 20 weeks. Mean values and standard error of the difference (SED) are presented. Bolding of P values indicates significance (P <0.05). MF = Milking frequency; FL = Feeding level.

Variable	Week[1]	Treatment				SED	P value		
		2XUnRes	1XUnRes	2XRes	1XRes		MF[2]	FL[3]	MFxFL
Milk (kg/d)	1-3	22.65	17.81	16.30	14.09	0.41	**<0.001**	**<0.001**	**<0.001**
	4-12	19.15	17.75	17.56	16.69	0.46	**<0.001**	**<0.001**	0.42
	13-23	15.12	14.22	14.68	13.83	0.52	**0.02**	0.26	0.95
Fat (%)	1-3	4.37	4.44	4.98	4.87	0.15	0.77	**<0.001**	0.38
	4-12	4.33	4.36	4.30	4.45	0.10	0.20	0.70	0.40
	13-23	4.60	4.61	4.62	4.81	0.13	0.31	0.25	0.34
Protein (%)	1-3	3.55	3.54	3.18	3.28	0.03	**0.01**	**<0.001**	**0.02**
	4-12	3.70	3.72	3.64	3.72	0.04	**0.03**	0.19	0.26
	13-23	3.86	3.85	3.82	3.90	0.05	0.37	0.96	0.26
Lactose (%)	1-3	4.92	4.71	4.92	4.73	0.03	**<0.001**	0.53	0.62
	4-12	4.83	4.84	4.79	4.78	0.02	0.86	**<0.001**	0.90
	13-23	4.79	4.80	4.79	4.81	0.03	0.42	0.88	0.79
Fat (kg/d)	1-3	0.98	0.79	0.80	0.68	0.03	**<0.001**	**<0.001**	0.15
	4-12	0.82	0.77	0.75	0.73	0.02	**0.05**	**<0.001**	0.29
	13-23	0.68	0.65	0.67	0.65	0.02	0.10	0.81	0.62
Protein (kg/d)	1-3	0.80	0.63	0.52	0.46	0.01	**<0.001**	**<0.001**	**<0.001**
	4-12	0.70	0.66	0.64	0.62	0.02	**0.01**	**<0.001**	0.25
	13-23	0.58	0.54	0.56	0.53	0.02	**0.03**	0.23	0.77
Lactose (kg/d)	1-3	1.11	0.84	0.80	0.67	0.02	**<0.001**	**<0.001**	**<0.001**
	4-12	0.92	0.86	0.84	0.80	0.02	**<0.001**	**<0.001**	0.52
	13-23	0.72	0.68	0.70	0.66	0.03	**0.03**	0.28	0.98

[1]Week relative to treatment initiation; Weeks 1−3 = Treatment period and Weeks 4−12 and Weeks 12-23 = Post-treatment periods.

FIGURE 1: (a): Milksolids (MS: fat + protein; kg/cow/d) and (b) Body condition score (BCS; 1–10 scale, 1 = Emaciated; 10 = Obese) from cows (34 ± 6 days in milk) consuming an unrestricted (UnRes; 14.3 ± 3.6 kg DM/cow/d) or restricted (Res; 8.3 ± 2.9 kg DM/cow/d) pasture intake and milked once (1X) or twice (2X) daily for three weeks. Following treatment, all cows were offered a generous pasture allowance (residuals >1,600 kg DM/ha) and milked 2X for 20 weeks. The solid horizontal bar highlights the three week treatment period. Mean values and error bars showing the standard error of the difference are presented.



(Bernier-Dodier *et al.*, 2010; Grala *et al.*, 2011). In contrast, although there was a negative carry-over effect of feed restriction in Weeks 4-12, there was no effect of FL on the yield of milk or milk components (Table 1) during Weeks 13-23 (10-20 weeks post-treatment) and Figure 1a illustrates that by Week 11, (8 weeks post-treatment) there was no effect of FL on MS production. Vetharaniam *et al.* (2003) used a mathematical model based on previous data (Davis *et al.,* 1999; Carruthers *et al.,* 1993) to determine that a lower dietary energy intake promotes active secretory cells to enter a quiescent non-secretory state, from which point they can either senesce via apoptosis or be reactivated. This reactivation, may explain the earlier recovery in milk yield in Res cows once energy intakes were normalised.

Figure 1b presents the effect of MF and FL on BCS. During the treatment period, Res cows lost 0.2 more BCS units than UnRes cows, and during Weeks 4-12, (1-8 weeks after returning to generous pasture allowance) Res cows remained 0.1 BCS unit less than UnRes cows. However, by Week 13-23 (10-20 weeks post-treatment) there was no effect of pasture restriction on BCS. There was no effect of MF on the immediate or carry-over BCS loss caused by the pasture restriction, nor on BCS throughout the experiment. These results indicate that grazing cows subjected to a severe, but relatively short, three week feed restriction in early lactation will still mobilise body tissue, even when energy demand for milk production is reduced by milking 1X. However, BCS is a relatively crude measure of energy balance. Previous research has reported improvements in energy balance, either calculated (energy output less intake) and/or indicated by plasma metabolite and hormone data such as non-esterified fatty acids, β-hydroxybutyrate, glucose and insulin, when MF was reduced during a feed restriction (Auldist & Prosser, 1998; Remond *et al.*, 2002). In the present experiment, as DMI is estimated on a group basis from pre and post-grazing paddock herbage mass these data can not be used to calculate energy balances for individual cows. Additionally, plasma samples have not yet been analysed for metabolites and hormones involved in energy metabolism. When analysed these samples may indicate subtle changes in energy balance at a physiological level that are not detectable by coarse BCS measurements. Remond *et al.* (2002) also reported improved reproductive performance with reduced MF during a feed restriction; however, in the present experiment there was no effect of MF or FL on post-partum anoestrus interval. The average post-partum anoestrus interval was 34, 34, 33 and 35 ± 9 days for 2XUnRes, 1XUnRes, 2XRes and 1XRes, respectively; and these occured prior to the start of the treatment period (~35 DIM). Data on other reproduction parameters such as submission, conception and pregnancy rates, are not available as cows were not mated during the course of the experiment.

In summary, reducing MF and imposing a feed restriction in early lactation, results in both immediate and carry-over milk production losses. While reducing MF may be a viable strategy to manage labour shortages in early lactation or to decrease energy output during a feed restriction, results indicate that the effects of reduced MF or FL on MS production continue after cows are switched back to normal intakes and 2X milking. Additionally, milking 1X did not alleviate the immediate BCS loss caused by a short-term feed restriction and the MF–induced milk production losses appeared to be separate and additive to any losses caused by pasture restriction. These results need to be considered when determining management strategies in early lactation.

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REFERENCES

Auldist, M.J.; Prosser, C.G. 1998: Differential effects of short-term once-daily milking on milk yield, milk composition and concentrations of selected blood metabolites in cows with low or high pasture intake. *Proceedings of the New Zealand Society of Animal Production* **58**: 41-43.

Bernier-Dodier, P.; Delbecchi, L.; Wagner, G.F.; Talbot, B.G.; Lacasse, P. 2010: Effect of milking frequency on lactation persistency and mammary gland remodeling in mid-lactation cows. *Journal of Dairy Science* **93**: 555-564.

Boutinaud, M.; Ben Chedly, M.H.; Delamaire, E.; Guinard-Flament, J. 2008: Milking and feed restriction regulate transcripts of mammary epithelial cells purified from milk. *Journal of Dairy Science* **91**: 988-998.

Broster, W.H. 1974. Response of the dairy cow to level of feeding. *Biennial Review of the National Institute of Research in Dairying*: 14-34.

Bryant, A.M.; Trigg, T.E. 1979: Immediate and longer term response of dairy cows to level of nutrition in early lactation. *Proceedings of New Zealand Society of Animal Production* **39**: 139-147.

Burke, C.R.; Williams, Y.J.; Hoffmann, L.; Kay, J.K.; Phyn, C.V.C.; Meier, S. 2010: Effects of an acute feed restriction at the on-set of the seasonal breeding period on reproductive performance and milk production in pasture-grazed dairy cows. *Journal of Dairy Science* **93**:1116-1125.

Carruthers, V.R.; Davis, S.R.; Bryant, A.M.; Henderson, H.V.; Morris, C.A.; Copeman, J.A. 1993: Response of Jersey and Friesian cows to once a day milking and prediction of response based on udder characteristics and milk composition. *Journal of Dairy Research* **60**:1-11.

Clark, D.A.; Phyn, C.V.C.; Tong, M.J.; Collis, S.J.; Dalley, D.E. 2006: A systems comparison of once-versus twice-daily milking of pastured dairy cows. *Journal of Dairy Science* **89**: 1854-1862.

Davis, S.R.; Farr, V.C; Stelwagen. K. 1999: Regulation of yield loss and milk composition during once-daily milking: a review. *Livestock Production Science* **59**:77-94.

Grala, T.M.; Phyn, C.V.C.; Kay, J.K.; Rius, A.G.; Littlejohn, M.D.; Snell, R.G.; Roche, J.R. 2011: Temporary alterations to milking frequency, immediately post-calving, modifies expression of milk synthesis and apoptosis genes in the mammary gland. *Proceedings of the New Zealand Society of Animal Production* **71**: 3-8.

Guinard-Flament, J.; Delamaire, E.; Lamberton, P.; Peyraud, J.L. 2007: Adaptations of mammary uptake and nutrient use to once-daily milking and feed restriction in dairy cows. *Journal of Dairy Science* **90**: 5062-5072.

Guinard-Flament, J.; Delamaire, E.; Lemosquet, S; Boutinard, M; David, Y. 2006: Changes in mammary uptake and metabolic fate of glucose with once-daily milking and feed restriction in dairy cows. *Reproduction, Nutrition and Development* **46**: 589-598.

Lacy-Hulbert, S.J.; Woolford, M.W.; Nicholas, G.D.; Prosser, C.G.; Stelwagen, K. 1999: Effect of milking frequency and pasture intake on milk yield and composition of late lactation cows. *Journal of Dairy Science* **82**: 1232-1239.

Littlejohn, M.D.; Walker, C.G.; Ward, H.E.; Lehnert, K.B.; Snell, R.G.; Verkerk, G.A.; Spelman, R.J.; Clark, D.A.; Davis, S.R. 2010: Effects of reduced frequency of milk removal on gene expression in the bovine mammary gland. *Physiological Genomics* **41:** 21-32.

O'Brien, B.; Gleeson, D.; Mee, J.F. 2005: Effect of milking frequency and feeding level on milk production, somatic cell count and reproductive performance of dairy cows over the first 20 weeks of lactation. *Proceedings of Irish Grassland Agricultural Research Forum*. Tullamore, Co. Offaly, Ireland. p. 28.

Payne, R.W.; Murray, D.A.; Harding, S.A.; Baird, D.B.; Soutar, D.M. 2009: GenStat for Windows, 12th Edition. Introduction. VSN International Ltd., Hemel Hempstead, Hertfordshire, UK.

Phyn, C.V.C.; Kay, J.K.; Rius, A.G.; Davis, S.R.; Stelwagen, K.; Hillerton, J.E.; Roche, J.R. 2010: Review: Impact of short-term alterations to milking frequency in early lactation. *In:* Meeting the challenges for pasture-based dairying. Proceedings of the 4th Australasian Dairy Science Symposium. Edwards, G.R.; Bryant, R.H. eds. Lincoln University, Christchurch, New Zealand. p. 156-164.

Remond, B.; Aubailly, S.; Chilliard, Y.; Dupont, D.; Pomies, D.; Petit, M. 2002: Combined effects of once-daily milking and feeding level in the first three weeks of lactation on milk production and enzyme activities, and nutritional status in Holstein cows. *Animal Research* **51**: 101-117.

Rhodes, F.M.; Clark, B.A.; Macmillan, K.L.; McDougall, S. 1998: Use of once-daily milking or treatment with progesterone and oestradiol benzoate in anoestrus cows. *Proceedings of the New Zealand Society of Animal Production* **58**: 44-46.

Roche, J.R. 2007: Milk production responses to pre- and post-calving dry matter intake in grazing dairy cows. *Livestock Science* **110**: 12-24.

Roche, J.R.; Dillon, P.G.; Stockdale, C.R.; Baumgard, L.H.; VanBaale, M.J. 2004: Relationships among international body condition scoring systems. *Journal of Dairy Science* **87**: 3076-3079.

Stelwagen, K. 2001: Effect of milking frequency on mammary functioning and shape of the lactation curve. *Journal of Dairy Science* **84** (Electronic supplement):E204-E211.

Vetharaniam, I.; Davis, S.R.; Upsdell, M.; Kolver, E.S.; Pleasants, A.B. 2003: Modelling the effect of energy status on mammary gland growth and lactation. *Journal of Dairy Science* **86**: 3148-3156.

Wall, E.H.; McFadden, T.B. 2007: The milk yield response to frequent milking in early lactation dairy cows is locally regulated. *Journal of Dairy Science* **90**: 716-720.