

Effect of increased milking frequency during early lactation on health and performance of lactating dairy cattle

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TAKE HOME MESSAGES:

1. Cows milked 3X daily produced more milk (3.7 lbs/d) than cows milked 6X
2. Cows milked 6X daily for 21 days and administered rbST at 63 days in milk produced more milk (10.1 lbs/d) throughout lactation than cows not given rbST
3. Heifers milked 3X daily produced more milk (12.6 lbs/d) than those milked 4X & 2X
4. Cows milked 3X daily produced more milk (9.5 lbs/d) than those milked 4X & 2X
5. Fat percentages were higher for cows milked 4X & 2X (4.05 vs. 3.64%) vs. 3X
6. Our data clearly indicate that IMF (6X vs. 3X or 4X and 2X vs. 3X) immediately after calving does not enhance performance, reproduction, or herd health parameters

INTRODUCTION

Increasing milking frequency (IMF) has enhanced milk yield by 7.7 and 10.8 lbs/day (d) when cows were milked twice daily (2X) compared to milking 3 times (3X), or 4 times (4X), respectively, and there was a tendency for milk fat and protein percentages to be reduced (Erdman and Varner, 1995). Although IMF has been demonstrated to increase milk synthesis during an established lactation (Erdman and Varner, 1995) its effects on milk yield when implemented for a short period (< 21 d) during early postpartum (PP) lactation are less clear. Bar-Peled et al. (1995), and Sanders et al. (2000) observed a 16.1 and 13.2 lbs/d increase in cows milked 6 times daily (6X) for the first 42 days in milk (DIM) compared to cows milked 3X. Hale et al. (2003) demonstrated that IMF (2X vs. 4X) in early lactation (1 to 21 DIM) enhanced milk yield (> 15.4 lbs/d) during 4X milking and the increase persisted (post-IMF) for up to 252 DIM. In addition, a recent field study reported IMF (6X vs. 3X) in early lactation (1 to 21 DIM) increased milk yield > 17.6 lbs/d and the enhanced production remained > 11 lbs/d after IMF ended according to the first 10 monthly DHIA tests (Dahl et al., 2004). However, few studies have looked at the length of time IMF is required to have an immediate and subsequent effect on milk yield. Minimal research has compared cows milked 4X compared to 3X (Hillerton et al., 1990) and the combination of 4X followed by 2X has not been compared to 3X milking previously. From an economic perspective it is important that research is focused on comparing the 4X and 2X combination compared to 3X, because more cows can be milked within a facility with the combination of 4X and 2X milking compared to 3X alone.

Between November 2003 and March 2005 we conducted two IMF experiments (6X vs. 3X and 4X and 2X vs. 3X) to evaluate IMF effects on health, reproduction, and performance parameters of lactating cows. The 6X compared to 3X study objectives were to 1) determine if cows milked 6X during early lactation would produce more milk during the IMF routine; 2) evaluate the time necessary to milk cows 6X during early PP to have a positive impact on peak milk yield and lactation persistency, and 3) to determine if IMF in early lactation and subsequent rbST administration during established lactation are additive. The 4X and 2X compared to 3X study objectives were to 1) determine the effects of 4X milking immediately PP followed by 2X

and its impact on milk yield compared to 3X milking; 2) determine the timing at which IMF treatment must be conducted PP to elicit carryover effects on lactation milk yield.

COWS AND TREATMENTS

6X vs. 3X

Three hundred multiparous Holstein cows were randomly assigned to 1 of 5 treatment (trt) groups at freshening. All cattle freshened between November 4, 2003 and February 10, 2004. Cows milked 6X were housed in a separate pen from cows milked 3X (during the IMF routine; \leq 21 DIM) and both pens were located near (287 ± 119 ft) the milking parlor (Table 1). Cows milked 6X for 7, 14, and 21 d were moved to the pen housing the 3X cows immediately after their 6X regimens had ended (d 8, 15, or 22), and all cows were co-mingled in a single pen at 29 DIM through the remainder of the 305-d trial. The 5 trt groups (60 cows/trt) were: 1) cows milked 3X/d through 305 DIM, 2) cows milked 6X/d for 7 DIM and then 3X/d through 305 DIM, 3) cows milked 6X/d for 14 DIM and then 3X/d through 305 DIM, 4) cows milked 6X/d for 21 DIM and then 3X/d through 305 DIM, and 5) cows milked 6X/d to 21 DIM and then 3X/d through 305 DIM. Cows assigned to treatments 1, 2, 3, and 4 were administered rbST (Posilac, Monsanto, Saint Louis, MO) at 14-d intervals beginning at 63 ± 3 DIM and remained on rbST throughout the 305-d study. Treatment 5 did not receive rbST. The 3X group was milked at 8-hour (h) intervals and groups 2 to 5 (6X) were milked at 4-h intervals during the IMF regimen.

4X and 2X vs. 3X

Two hundred multiparous and 100 primiparous Holstein cows were randomly assigned to 1 of 5 trts at freshening. All cattle freshened between December 16, 2004 and January 12, 2005. Cows milked 2X were housed in a separate pen from cows milked 4X (during the IMF routine; \leq 40 DIM) and all pens of cows were located near (376 ± 149 ft) the milking parlor (Table 2). Cows milked 4X for 10, 20, 30 and 40 d were moved to the pen housing 2X cows immediately after their 4X regimen ended (d 11, 21, 31 or 41), and all 2X cows were co-mingled in a single pen throughout the remainder of the 119 d trial. The 5 trt groups (60 cows/trt) were: 1) cows milked 3X/d through 119 DIM, 2) cows milked 4X/d for 10 DIM and then 2X/d through 119 DIM, 3) cows milked 4X/d for 20 DIM and then 2X/d through 119 DIM, 4) cows milked 4X/d for 30 DIM and then 2X/d through 119 DIM, and 5) cows milked 4X/d to 40 DIM and then 2X/d through 119 DIM. The 3X group was milked at 8h intervals, the 4X group was milked at 6h intervals during the IMF regimen and the 2X group was milked at 12h intervals, respectively.

Measurements

In both studies cows calved at a commercial dairy near Buckeye, AZ and were fed a TMR formulated to meet or exceed NRC, (2001) requirements daily at 0400, 1200, and 2000 and were housed in dry lot corrals (permitting 530 sq. ft/cow) with shades (29.0 ft wide by 13.0 ft high; permitting 480 sq. ft/cow). Use of animals on both trials was approved by the University of Arizona Institute of Animal Care and Use Committee. Daily milk weights were measured electronically by Boumatic Computer software (Madison, WI) for each animal milking throughout the 305 and 119 day trials. Monthly milk composition analysis and somatic cell count (SCC) was conducted at Arizona Dairy Herd Improvement Association (DHIA, Tempe, AZ). Energy corrected milk (ECM) was calculated from milk, protein and fat volumes ($0.7 \times \text{milk lbs} + 28.5 \times \text{fat lbs} + 15.9 \times \text{protein lbs}$). Pen DMI was monitored and recorded using PROFEED2000 (DairyWorks, Tempe, AZ) feed management software. Cows were scored for

body condition at freshening and at 28 ± 4 d intervals. Body condition scores (BCS) were based on a 5-point scale (Wildman et al., 1982). Blood samples were collected from the coccygeal vein from a subset of 15 cows milked 6X or 3X (trial 1), or 27 cows milked 3X or 4X for 40 d (trial 2) at parturition and on d 7, 14, 21, 28 and 35 ± 1 PP and the harvested plasma was immediately frozen at -20°C until analyzed for non-esterified fatty acids (NEFA) concentration.

Statistical Analyses

All performance data from the 6X vs. 3X trial were analyzed for the first 9 wk PP and then from wk 10 to 44 to evaluate the effects of IMF during early lactation and the potential carry over effects after peak milk. Treatments 1, 2, 3, and 4 were analyzed to evaluate the impact of IMF and trts 4 and 5 were analyzed separately to determine the impact of 6X milking for 21 d PP with and without rbST administration beginning at 63 DIM. Performance data for the 4X and 2X vs. 3X trial were analyzed through 119 DIM to evaluate the effects of IMF during early lactation and the potential carry over effects after peak milk. Treatments from both trials were analyzed to evaluate the impact of IMF using PROC MIXED of SAS (1999, Cary, NC) and previous 305 d mature equivalent milk yields were included as a covariate in each analysis of multiparous cows.

RESULTS

6X vs. 3X

Cows milked 3X daily tended ($P = 0.08$) to produce more milk (95.2 vs. 91.5 and 90.2 ± 2.43 lbs/d; Figure 1) and more ($P < 0.01$) ECM (99.2 vs. 93.9 and 91.7 ± 1.5 lbs/d; Table 3) during the first 9 wk of lactation compared to cows milked 6X for 7 or 21 DIM, respectively. Milk yields did not differ among milking frequency trts (84.4 ± 1.5 lbs/d) after wk 9 (Table 3 and Figure 1). Yields of ECM were higher ($P < 0.05$) after wk 9 for cows milked 3X daily compared to 6X for 21 DIM (86.2 vs. 84.2 ± 0.9 lbs/d). However, 3X cows had similar ECM yields as those milked 6X for 7 or 14 DIM, (85.5 lbs/d; Table 3). Percentages of milk fat (3.80 ± 0.12) and protein (2.90 ± 0.06) did not differ between trts during the first 9 wk after calving (Table 3). However, cows milked 3X daily had increased ($P = 0.01$) yields of fat compared to cows milked 6X for 7 or 21 DIM (3.7 vs. 3.4 ± 0.1 lbs/d; Table 3). In addition, cows milked 3X or 6X for 7 or 14 DIM produced more ($P < 0.01$) protein compared to cows milked 6X for 21 DIM (2.8 vs. 2.6 ± 0.07 lbs/d). Somatic cell count (257×10^3 cells/ml) and BCS (3.60) did not differ between treatments during the first 9 wk after calving (Table 3). However, SCC were higher ($P = 0.02$) between wk 10 and 44 for cows milked 3X and 6X for 21 DIM compared to cows that were milked 6X for 14 DIM (595 and 526 vs. $294 \times 10^3 \pm 111$ cells/ml; Table 3). Plasma NEFA concentrations ($477 \mu\text{eq/L}$) did not differ between the 3X and 6X 21 d trts (Table 3).

Early lactation milk yield (92.4 lbs/d) did not differ statistically between cows milked 6X for 21 DIM that eventually received rbST starting at 63 DIM compared to those not assigned rbST administration (Table 4 and Figure 2). Cows treated with rbST and milked 6X for 21 DIM produced more ($P < 0.01$) milk (85.5 vs. 75.4 ± 1.9 lbs/d; Figure 2) and ECM (86.0 vs. 76.1 ± 0.9 lbs/d) compared to cows milked 6X for 21 DIM and not provided rbST between wk 10 and 44 of lactation (Table 4). Milk fat (3.71) nor protein (2.86) percentages nor yields of fat (3.44 lbs/d) differed during the first 9 wk after calving between the 2 treatment groups milked 6X for 21 DIM (Table 4). Average SCC (226×10^3 cells/ml) and BCS (3.58) were also similar between the 2 treatments during the first 9 wk after calving. Milk fat and protein percent (3.62 and 2.91) were not affected by rbST administration, however yields of milk fat (3.02 vs. 2.78 ± 0.07 lbs/d)

and protein (2.47 vs. 2.21 ± 0.07 lbs/d) were higher ($P < 0.01$) for cows administered rbST and milked 6X for 21 DIM compared to cows not receiving rbST (Table 4). Overall milk SCC (528 vs. $252 \times 10^3 \pm 102$ cells/ml) were increased ($P < 0.01$) and BCS decreased (3.53 vs. 3.56 ± 0.01 ; $P < 0.05$) for cows administered rbST after being milked 6X for 21 DIM compared to cows milked 6X for 21 DIM and did not receive rbST at 63 DIM.

The percentage of cows pregnant (37%) within 65 d of the voluntary waiting period (80 DIM), average DIM at pregnancy (126) and average service per conception (2.28) did not differ between treatments ($X^2 = 0.96$). The number of cows that were sent to the hospital pen during the 305 d trial for mastitis (97), digestive disorders (14), respiratory issues (9), lameness (22), and/or retained placenta (16), were not affected by milking frequency or somatotropin trts ($X^2 = 0.49$; Table 5).

4X and 2X vs. 3X

Primiparous cows milked 3X daily ($P < 0.01$) produced more milk (81.4 vs. 68.8 ± 1.5 lbs/d) and tended ($P = 0.08$) to produce more ECM (86.0 vs. 77.6 ± 4.2 lbs/d) during the 17 wk trial compared to cows milked 4X for 10, 20, 30, or 40 DIM, respectively (Figure 3; Table 6). Cows milked 4X for 10 or 20 DIM were lower (65.7 ± 1.5 lbs/d; $P < 0.01$) from cows milked 4X for 30 or 40 (68.8 ± 1.5 lbs/d) DIM, however calculated yields of ECM were not different among trts ($P = 0.08$; Table 6). Percentages of milk fat ($P = 0.05$) were higher for cows milked 4X for 10, 20, 30, or 40 DIM compared to cows milked 3X (3.72 vs. 4.04 ± 0.11). There was no difference between yields of fat (3.02 lbs/d ± 0.11), SCC (194 SCC/ml ± 63), and NEFA (557 μ eq/L ± 40) concentration, respectively. Percentage of protein ($P = 0.02$) was higher for cows milked 4X for 10 or 20 DIM (2.86 ± 0.06 ; $P = 0.02$) however there was no difference between cows milked 3X or 4X for 30 or 40 DIM.

Multiparous cows milked 3X daily ($P < 0.01$) produced more milk (98.6 vs. 89.1 ± 2.43 lbs/d; Table 6; Figure 4) and more ($P = 0.04$) ECM (103.8 vs. 99.0 lbs/d ± 3.7 lbs/d) during the 17 wk trial compared to cows milked 4X for 10, 20, 30, or 40 DIM, respectively. Cows milked 4X for 10 DIM followed by 2X thereafter had similar ECM (103.8 lbs/d ± 3.7 lbs/d) as cows milked 3X. Fat percentages ($P = 0.01$) were higher for cows milked 4X for 10, 20, 30, or 40 (4.05 vs. 3.64 ± 0.12) DIM than cows milked 3X. Yields of protein ($P = 0.01$) were higher for cows milked 3X than for cows milked 4X for 10, 20, 30, or 40 DIM (1.28 vs. 1.19 ± 0.07), however, they were similar between cows milked 4X for 30 or 40 DIM compared to cows milked 3X or 4X for 10 or 20 DIM. There was no difference between yields of fat (3.84 ± 0.11), protein percentages (2.80 ± 0.04), SCC (481 SCC/ml ± 132), BCS (3.11 ± 0.01).

The percentage of cows pregnant (52.8 %) within 65 d of the voluntary waiting period (80 DIM), average DIM at pregnancy (127) and average service per conception (1.74) did not differ between treatments ($X^2 = 0.81$). The number of cows that were sent to the hospital pen during the 305 d trial for mastitis (85), digestive disorders (0), respiratory issues (3), downer (5), lameness (5), and retained placentas (22); were not different between trts ($X^2 = 0.17$; Table 7).

DISCUSSION

During established lactation, IMF consistently enhances milk synthesis, and recent data indicate that IMF during early lactation has an immediate impact on milk yield and this response is maintained even after the IMF regimen has ceased. However, previous early lactation IMF studies have not directly evaluated the time (i.e. how many DIM) IMF needs to be implemented

PP in order to obtain a beneficial effect on immediate milk yield, peak milk levels and lactation persistency. We evaluated the effect of milking cows 6X for as few as 7, 14, or 21 d PP and our data indicate IMF did not enhance milk yield during the IMF (≤ 21 d) regimen (78.9 lbs/d), nor during the first 9 wk of lactation (93.3 lbs/d) or after wk 9 (wk 10 to 44; 84.4 lbs/d). This is in contrast to findings by others (Bar-Peled et al., 1995; Sanders et al., 2000), who reported 16.1 lbs/d (21%) and a 13.3 lbs/d increase (16%) in production when cows were milked 6X for 42 DIM compared to 3X.

When we evaluated the effect of milking cows 4X for 10, 20, 30, or 40 d PP compared to 3X and our data indicated that IMF did not enhance milk yield during the IMF regimen, or over the first 17 wks of lactation. However, when cows milked 4X were changed to 2X, milk yield reduced ($8.1 \text{ lbs/d} \pm 5.9$) in primiparous and multiparous ($6.5 \text{ lbs/d} \pm 2.5$) cows, indicating that cows milked 4X were at an advantage every day they were on 4X vs. 2X. Our findings are similar to, Hale et al. (2003) who milked cows 2X vs. 4X for 21 DIM, and reported that cows milked 4X immediately PP produced up to 19.4 lbs/d more milk than those milked 2X during early lactation and this response in lactation persisted until 252 DIM.

In the 6X vs. 3X study, monthly milk fat (3.8) and protein (2.9) percentages were not affected by IMF during the first 9 wk PP or from wk 10 to 44, which agrees with Bar-Peled et al. (1995) and Sanders et al. (2000). In our study and others (Bar-Peled et al., 1995; Sanders et al., 2000), milk samples were analyzed on a monthly basis (therefore the range in DIM at sampling was ± 28 d) and there were no differences observed in milk composition, whereas Hale et al. (2003) sampled weekly and detected differences. In our 4X and 2X vs. 3X study, monthly milk fat percentages increased during the 4X and 2X regimen compared to cows milked 3X in both primiparous (0.32 ± 0.05) and multiparous (0.41 ± 0.01) cows, since cows milked 4X did not have higher yields (although higher percentages) of milk fat this most likely resulted from fat being more concentrated within milk.

In the 6X vs. 3X study, milk SCC were not affected by IMF during the first 9 wk PP and this agrees with other IMF trials (Bar-Peled et al., 1995; Sanders et al., 2000; Hale et al., 2003). In contrast to early lactation, during wk 10 to 44 of lactation, cows previously milked 6X for 7 or 14 DIM had reduced milk SCC compared to 3X or 6X for 21 DIM (Table 3); however the biological significance (if any) is difficult to interpret. Primiparous cows milked 4X and 2X vs. 3X had lower ($P = 0.07$) SCC (93 vs. 219×1000 cells/ml ± 73) compared to those milked 4X followed by 2X. There was no difference among SCC for multiparous cows milked 4X and 2X vs. 3X; respectively.

Reasons why our IMF milk production data do not agree with others (Bar-Peled et al., 1995; Sanders et al., 2000; Hale et al., 2003; Dahl et al., 2004) are not clear, but facility logistics may have contributed to the discrepancies. Unlike previous reports, where cows were housed in a tie-stall environment or individual pens, (or where the impact of facility is not reported) cows used in our study were not individually penned. Instead, they were housed in a dry lot facility located 287 ± 119 ft (6X vs. 3X) or 376 ± 149 ft (4X, 3X, and 2X) from the milking parlor (Tables 1 and 2). Thus, cows in the 6X and 4X milking regimen walked further each day and spent on average 6.5 h/d outside of their pen and consequently were physically away from feed and water longer than cows milked 2 or 3 times daily. Incidentally, although increased walking may have impacted estimated maintenance requirements the calculated increase was small (1.12 vs. 0.80 Mcal/d; 2001 NRC), and compared to typical large commercial dairies, the total walking distance was nominal even for cows being milked 6X daily. Although cows milked 6X were

away from feed longer, average pen DMI during the 6X IMF regimen did not differ (42.5 lbs/d). In contrast, Bar-Peled et al. (1995) reported an increase in DMI (37.0 vs. 42.8 lbs/d, from 0 to 42 DIM; and 44.3 vs. 49.4 lbs/d from 49 to 70 DIM) for cows milked 3X daily compared to 6X. Incidentally, DMI (for 6X and 3X treatments) immediately postpartum (0 to 21 DIM) in our study was similar to that reported for the cows milked 6X in Bar-Peled et al. (1995) in later lactation (i.e. ~ 40 DIM). In contrast to Bar-Peled et al. (1995), our cows consumed similar amounts of feed while experiencing differing IMF regimens and cows milked 3X (on the current study) produced more milk than cows milked 6X on Bar-Peled et al. (1995) study, possibly suggesting that 3X milking under conditions utilized in this experiment maximized the potential for changes in milking frequency to enhance production during early lactation.

As previously stated, the exact reason why our milking frequency data doesn't agree with others is currently not known. An inadequate nutritional status is not likely as cows across trts averaged over 28,660 lbs of 3.5% fat corrected milk throughout lactation and overall nutritional status was certainly sufficient during established lactation as exogenous rbST increased (12%) milk yield consistently throughout lactation (Figure 2). Furthermore, environmental factors such as heat stress were not an issue as the majority of the trials were conducted during the winter months and ambient temperatures (57 ± 15 °F) and humidity ($50 \pm 25\%$) were low, respectively. A possible explanation may be that cows in all treatments had an average milk production of 78.9 lbs/d during the first 21 DIM and this is difficult to compare because other studies (Bar-Peled et al., 1995 and Sanders et al., 2000) administering a 6X milking regimen milked cows 6X through 42 DIM. As a consequence, high producing cows, especially high producers that have a steep slope of milk yield increase may not be as responsive to increased milking frequency immediately after calving (≤ 21 DIM). Although, Hale et al. (2003) demonstrated an increase in as little as 21 d after calving, their cows were milked 4X compared to 2X, not 6X compared to 3X.

Exogenous bovine somatotropin consistently increases milk yield by 10 to 15% /cow (Bauman and Vernon, 1993). In our study, 2 groups of cows were milked 6X for 21 DIM, 1 group received rbST beginning at 63 DIM the other did not. We did not see a milk response to IMF, however cows milked 6X and treated with rbST (wk 10 to 44) produced more milk (12%) and more ECM (12%) than cows that did not receive rbST. It has been demonstrated that somatotropin does not alter the normal relationship between milk yield and mastitis (White et al., 1994). In the current study, mastitis was the number one reason for cows visiting the hospital pen in all treatments; however, there was no difference in clinical mastitis incidence between cows given rbST and those not given rbST. As previously reported (White et al., 1994) with respect to mastitis, the performance and health of cows administered rbST appears to be similar to that of cows not administered rbST but yielding similar amounts of milk.

Reproductive variables (cows pregnant within 65 d of the voluntary waiting period, average DIM at pregnancy; and average service per conception) and the number of cows that were sent to the hospital pen (for mastitis, digestive disorders, respiratory issues, lameness, and/or retained placenta) were not affected by IMF (6X, 4X, 3X, or 2X) nor with use of somatotropin during our trials. This suggests that IMF with or without rbST had no impact on animal health and reproductive performance. Therefore, the decision to implement a 6X milking regimen for 7, 14, or 21 d or a 4X milking regimen 10, 20, 30, or 40 d PP in an attempt to immediately increase milk synthesis or enhance lactation persistency compared to 3X does not appear to be warranted under the management conditions of the current study.

CONCLUSIONS

Our data clearly indicate that IMF (6X vs. 3X or 4X and 2X vs. 3X) immediately PP did not affect performance, reproduction, herd health, or culling variables. Cows receiving rbST (at 63 DIM) produced 10.1 lbs/d more milk compared to cows milked 6X for 21 DIM and not administered rbST. Although there is scientific evidence to support 4X vs. 2X milking for 21 DIM followed by 2X milking thereafter and/or 6X milking for 42 DIM followed by 3X, our data indicate that 6X milking for either 7, 14, or 21 or 4X for 10, 20, 30 or 40 DIM compared to 3X milking is not effective. More research needs to be conducted to evaluate milking intervals (hours between milkings) during different IMF regimens and the number of d that 4X and/or 6X milking might be conducted after calving to elicit the potential increases in milk and subsequent carryover effects.

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Table 1. Impact of facility on cows milked 3 times (3X) or 6 times (6X) daily during early lactation

Item	Milking Frequency	
	3X	6X
Number of cows in pen		
Average	39	44
Range	20 to 70	1 to 70
Walking distance (feet)		
1-way	168	406
Per d	1,008	4,872
Time out of pen (minutes)		
Average	195	390
Range	180 to 225	270 to 450
Time milking (minutes)		
Range	60 to -75	45 to 75

Table 2. Impact of facility on cows milked 2X, 3X, or 4X daily

Item	Milking Frequency		
	2X	3X	4X
Number of cows in pen			
Average	240	240	150
Range	220-250	220-250	1-168
Walking distance (feet)			
One way	227	227	525
Per d	908	1,362	4,200
Time out of pen (minutes)			
Average	185	278	230
Range	180-190	270-285	220-240
Time milking (minutes)			
Range	90-95	90-95	55-60

Table 3. Performance of cows milked 3 times (3X, control) or 6 times (6X) daily for 7, 14 or 21 DIM followed by milking 3X daily thereafter

Item	6X milking (DIM)				SEM	P
	0 (control)	7	14	21		
First 9 wk postpartum						
Milk, lbs/d	95.2	91.5	95.7	90.2	2.4	0.08
ECM, lbs/d	99.2 ^a	93.9 ^b	97.9 ^a	91.7 ^b	1.5	< 0.01
Pen DMI ¹ lbs/d	42.6	42.7	42.7	42.7	NA	NA
Fat, %	3.90	3.77	3.77	3.74	0.12	0.50
Fat, lbs/d	3.70 ^a	3.44 ^{bc}	3.62 ^{ac}	3.37 ^b	0.11	0.01
Protein, %	2.90	2.95	2.89	2.85	0.06	0.40
Protein, lbs/d	2.78	2.71	2.78	2.56	0.07	< 0.01
SCC/ml	347	255	136	288	119	0.32
BCS	3.60	3.63	3.58	3.57	0.03	0.29
NEFA, μ eq/L	489	-	-	464	73	0.74
Wk 9 to wk 44						
Milk, lbs/d	85.3	83.6	84.9	84.0	1.5	0.60
ECM, lbs/d	86.2 ^{ac}	84.4 ^{ab}	86.2 ^c	84.2 ^b	0.9	0.04
Fat, %	3.63	3.64	3.69	3.60	0.07	0.70
Fat, lbs/d	3.11	3.04	3.13	3.02	0.07	0.27
Protein, %	2.93	2.95	2.95	2.93	0.04	0.82
Protein, lbs/d	2.51	2.60	2.49	2.47	0.02	0.26
SCC/ml	595 ^a	362 ^{bc}	294 ^b	526 ^{ac}	111	0.02
BCS	3.54 ^a	3.57 ^b	3.54 ^a	3.53 ^a	0.01	< 0.01

^{a,b,c} Means with unlike superscripts within row differ $P < 0.05$.

¹ Total feed fed to the 3X and 6X pen minus refusals divided by the number of cows in the pen daily.

Table 4. Production variables of cows milked 6 times (6X) daily for 21 DIM followed by 3 times (3X) daily thereafter

Item ¹	21 DIM of 6X milking		SEM	<i>P</i>
	No rbST	rbST		
First 9 wk postpartum				
Milk, lbs/d	94.6	90.2	2.06	0.12
ECM, lbs/d	95.5	91.7	1.59	0.03
Fat, %	3.68	3.74	0.13	0.68
Fat, lbs/d	3.48	3.37	0.11	0.38
Protein, %	2.86	2.85	0.06	0.80
Protein, lbs/d	2.69	2.56	0.04	0.02
SCC/ml	164	288	125	0.34
BCS	3.59	3.57	0.04	0.79
Wk 9 to wk 44				
Milk, lbs/d	75.4	85.5	2.0	< 0.01
ECM, lbs/d	76.6	86.0	0.9	< 0.01
Fat, %	3.64	3.60	0.07	0.63
Fat, lbs/d	2.73	3.09	0.07	< 0.01
Protein, %	2.90	2.92	0.04	0.53
Protein, lbs/d	2.21	2.47	0.07	< 0.01
SCC/ml	252	528	102	< 0.01
BCS	3.56	3.53	0.01	0.03

¹Both groups were milked 6X for 21 DIM and then 3X thereafter. Starting on d 63, recombinant bovine somatotropin (rbST) was administered at 14-d intervals to cows in the group scheduled to receive rbST.

Table 5. Reproductive, herd health and culling measures for cows milked 3 times (3X, control) or 6 times (6X) daily for 7, 14 or 21 DIM followed by 3X milking thereafter

Item	6X milking (DIM)				
	0 (control)	7	14	21	21 no rbST
Reproduction					
% Pregnant by VWP+65 d ¹	37	43	40	31	32
DIM @ pregnant	125	124	127	127	127
Times bred	2.33	2.03	2.24	2.47	2.31
%, and total cows that visited hospital²					
Mastitis	50 (16)	65 (22)	58 (19)	67 (22)	60 (18)
Digestive	9 (3)	9 (3)	6 (2)	6 (2)	13 (4)
Respiratory	9 (3)	6 (2)	9 (3)	3 (1)	0
Lame	19 (6)	15 (5)	6 (2)	18 (6)	10 (3)
Retained placenta	13 (4)	6 (2)	12 (4)	6 (2)	13(4)
Other ³	0	0	9 (3)	0	3 (1)
%, and total that left the herd					
Diarrhea	25 (1)	0	0	25 (1)	50 (2)
Downer	14 (1)	30 (2)	30 (2)	14 (1)	14 (1)
<i>E. Coli</i> Mastitis	0	33 (2)	0	17 (1)	51 (3)
Lame	13 (2)	44 (7)	13 (2)	25 (4)	6 (1)
Pneumonia	50 (2)	25 (1)	25 (1)	0	0
Reproduction	25 (1)	25 (1)	0	0	50 (2)

¹Percentages of cows pregnant within 65 d of the voluntary waiting period did not differ across groups ($X^2 = 0.96$), ² Percentages of cows that visited the hospital did not differ across groups ($X^2 = 0.49$), ³ 2 cows on 6X milking for 14 DIM were downer cows and 1 cow had a skin condition; the 1 cow on 6X milking for 21 DIM, no rbST was also a downer cow.

Table 6. Performance of Primiparous and Multiparous cows milked 3X (control) or 4X daily for 10, 20, 30, or 40 DIM followed by 2X daily thereafter

Item	Milking Frequency					SEM	<i>P</i>
	Control (3X)	4X-10 d	4X- 20 d	4X-30 d	4X- 40d		
Primiparous							
Milk, lbs/d	81.4 ^a	64.8 ^b	66.8 ^b	71.0 ^c	72.5 ^{cd}	1.5	< 0.01
ECM, lbs/d	86.0	76.3	76.7	81.1	76.7	4.2	0.08
Fat, %	3.72 ^a	4.15 ^b	3.94 ^{ab}	4.02 ^b	4.05 ^b	0.15	0.05
Fat, lbs/d	3.15	2.93	2.88	3.09	2.93	0.11	0.39
Protein, %	2.76 ^a	2.91 ^b	2.82 ^{ab}	2.76 ^{ac}	2.76 ^{ac}	0.06	0.02
Protein, lbs/d	2.36 ^a	2.07 ^b	2.09 ^b	2.16 ^{ab}	2.03 ^b	0.11	0.03
SCC x 1,000/ml	93	329	160	160	226	63	0.07
BCS	3.09 ^a	3.12 ^a	3.07 ^a	3.05 ^{ab}	3.02 ^{bc}	0.02	0.02
NEFA, μ eq/L	535				579	40	0.44
Multiparous							
Milk, lbs/d	98.5 ^a	91.5 ^b	89.9 ^{bc}	86.7 ^c	88.2 ^{bc}	2.4	<0.01
ECM, lbs/d	103.8 ^a	103.8 ^a	101.2 ^{ab}	95.2 ^b	96.3 ^b	3.7	0.04
Fat, %	3.64 ^a	4.03 ^b	3.99 ^b	4.13 ^c	4.06 ^{bc}	0.12	0.01
Fat, lbs/d	3.79	4.15	3.86	3.68	3.70	0.11	0.42
Protein, %	2.77	2.82	2.79	2.82	2.78	0.04	0.26
Protein, lbs/d	2.82 ^a	2.78 ^a	2.69 ^a	2.51 ^b	2.54 ^b	0.09	0.01
SCC x 1000/ml	303	561	566	389	587	132	0.40
BCS	3.13	3.13	3.11	3.07	3.12	0.01	0.09
NEFA, μ eq/L	523				428	60	0.08

Means with unlike superscripts within row differ $P < 0.05$.

Table 7. Reproductive, herd health and culling parameters of 3X (control) or 4X daily for 10, 20, 30 or 40 DIM followed by 2X daily thereafter.

Item	Milking Frequency				
	Control (3X)	4X-10d	4X-20d	4X-30d	4X-40d
Reproduction					
% Pregnant by VWP+65 d ¹	40	60	62	54	48
DIM @ pregnant	127	122	128	129	127
Times bred	1.65	1.81	1.79	1.70	1.74
%, and total cows that visited hospital²					
Mastitis	88 (15)	67 (20)	79 (23)	55 (11)	59 (16)
Digestive	0	0	0	0	0
Respiratory	0	0	0	10 (2)	4 (1)
Downer	0	3 (1)	3 (1)	0	11 (3)
Lame	6 (1)	0	7 (2)	5 (1)	4 (1)
Retained placenta	6 (1)	30 (9)	10 (3)	30 (6)	22 (6)
%, and total that left the herd					
Digestive	10 (1)	20 (2)	30 (3)	20 (2)	20 (2)
Downer	0	0	100 (1)	0	0
<i>E. Coli</i> Mastitis	27 (3)	18 (2)	27 (3)	18 (2)	9 (1)
Lame	0	25 (1)	0	25 (1)	50 (2)
Pneumonia	67 (2)	0	33 (1)	0	0
Reproduction	0	20 (1)	20 (1)	60 (3)	0

¹Percent pregnant within 65 d of the voluntary waiting period did not differ across groups ($X^2 = 81$),

²Percentages of cows that visited the hospital did not differ across groups ($X^2 = 0.17$).

Figure 1. Milk yields during the first 9 wk of lactation and then from wk 10 to 44 postpartum for cows milked 3 (3X) or 6 times daily (6X) for 7, 14, or 21 DIM (broken vertical line indicates end of wk 9). \circ = 3X milking, \triangle = 6 X milking for 7 d postpartum, \diamond = 6X milking for 14 d postpartum, X = 6X milking for 21 d postpartum. All cows were milked 3X daily except for experimental periods in the first 3 wk .

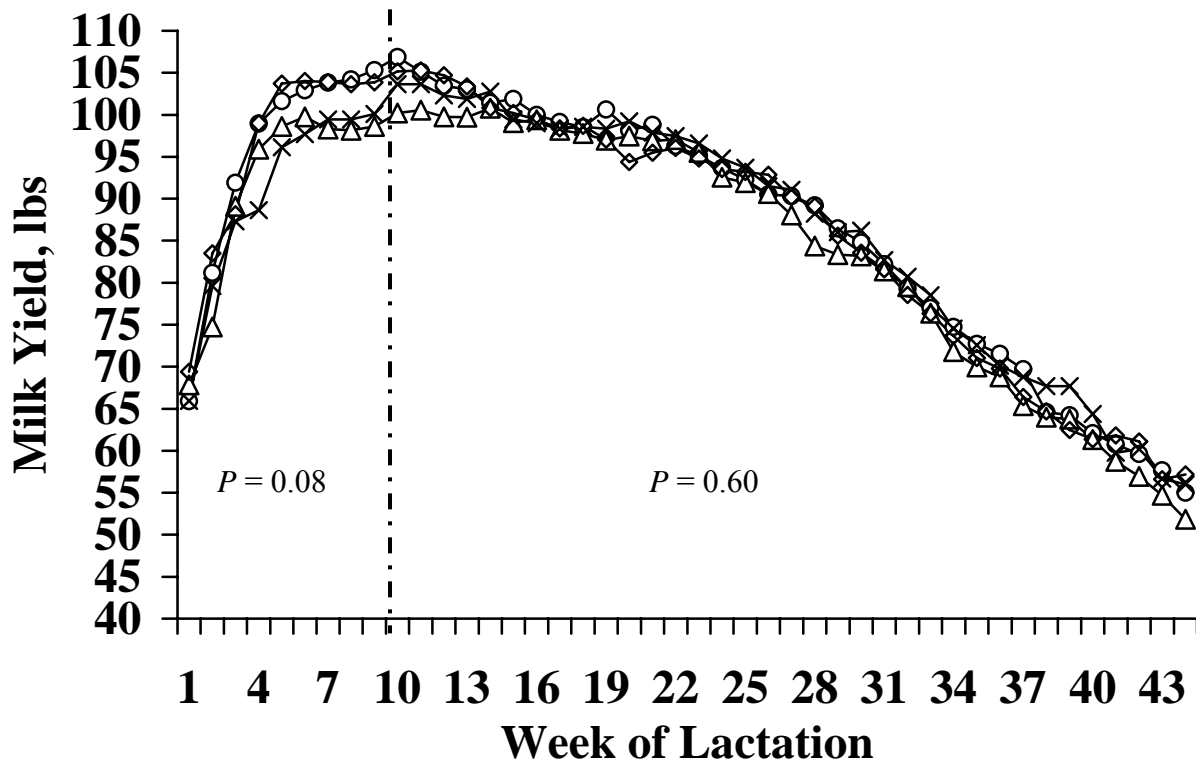


Figure 2. Milk yield of cows milked 6 times (6X) daily for 21 DIM and subsequently receiving or not receiving rbST beginning at 63 DIM. Milk yields include the first 9 wk of lactation indicated by broken vertical line and then wk 10 to 44 postpartum after initiation of injections of rbST. □ = 6X milking for 21 DIM going to receive somatotropin (rbST) beginning at wk 9, ■ = 6X milking for 21 DIM not going to receive somatotropin (rbST) throughout the 44-wk trial.

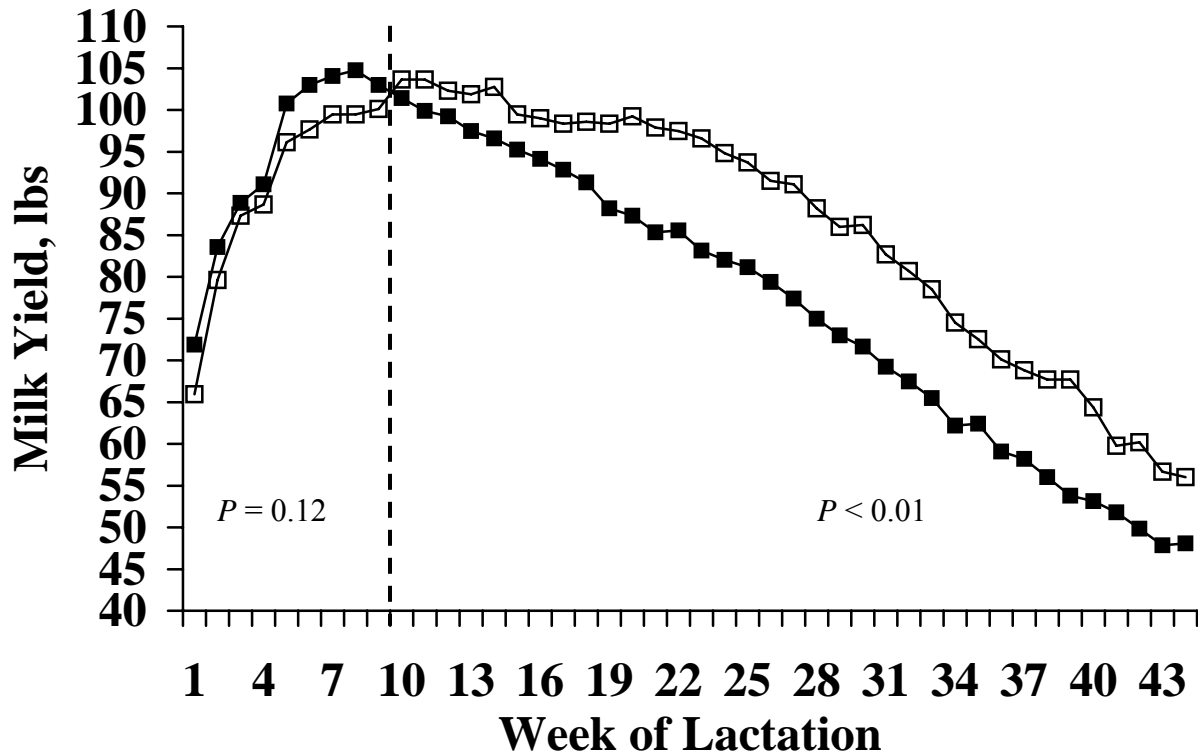


Figure 3. Milk yields of primiparous cows milked three times (control) or 4X times daily for 10, 20, 30, or 40 DIM followed by two times daily thereafter.

○ = 3 times per day milking, △ = 4 times per day milking for 10 days postpartum, ◇ = 4 times per day milking for 20 days postpartum, x = 4 times per day milking for 30 days postpartum, ■ = 4 times per day milking for 40 days postpartum.

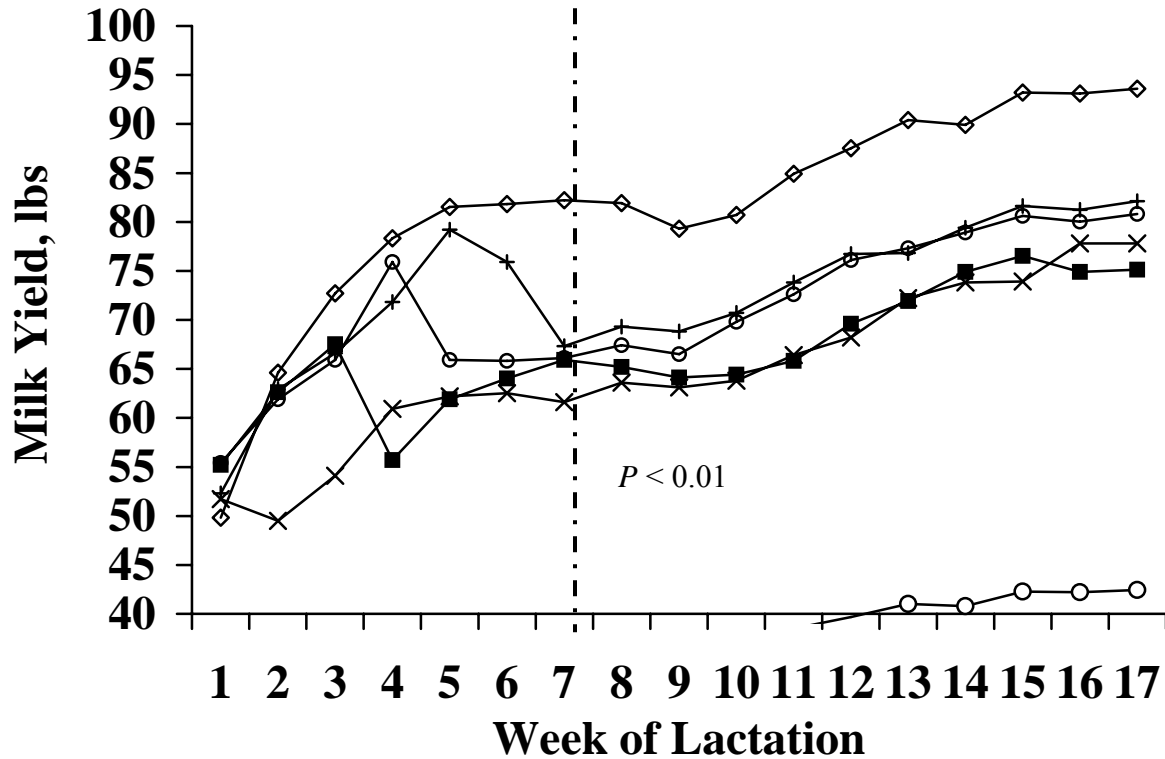


Figure 4. Milk yields of multiparous cows milked three times (control) or 4X times daily for 10, 20, 30, or 40 DIM followed by two times daily thereafter.

○ = 3 times per day milking, △ = 4 times per day milking for 10 days postpartum, ◇ = 4 times per day milking for 20 days postpartum, x = 4 times per day milking for 30 days postpartum, ■ = 4 times per day milking for 40 days postpartum.

