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Abstract

The objective of this study was to determine how milking frequency, estradiol cypionate (ECP) postpartum therapy given at 1 week after calving, and biweekly bovine somatotropin (bST) administration alter lactational and reproductive outcomes in dairy cattle. Holstein cows (n=144) were randomly assigned to eight treatments (18 cows per treatment): 1) twice daily milking frequency (2x), 10-mg injection of ECP at 1 week after calving (ECP), and bST (given biweekly according to label beginning in the ninth week of lactation); 2) 2x milked, oil (cottonseed oil vehicle for ECP), bST; 3) 2x milked, ECP, and no bST; 4) 2x milked, oil, and no bST; 5) four-times daily milking frequency (4x; first 30 days in milk then 2x thereafter), ECP, and bST; 6) 4x milked, oil, and bST; 7) 4x milked, ECP, and no bST; and 8) 4x milked, oil, and no bST. Milk yields were recorded at each milking during the first 90 days of lactation. Milk samples were collected weekly at each milking and composited to determine milk components (percentages of fat, protein, lactose, solids-not-fat [SNF], milk urea nitrogen [MUN], and somatic cell count [SCC]). Energy-corrected milk yields were calculated for the first 90 days and for whole lactation yields (305-2x- ME standardized lactation records). Ovulation before first AI was synchronized beginning between 59 and 72 DIM using 100 :g of GnRH given 7 days before 25 mg of PGF2", followed in 24 hr by 1 mg of ECP. Cows were inseminated after detected estrus or at 48 hr after ECP. Pregnancy rates were assessed by transrectal ultrasonography 28-30 days after AI. Postpartum ECP therapy increased milk production for first-lactation 2x cows, but decreased milk yields of the multiparous 4x cows until bST restored those yields. Pregnancy rates were greater for the 4x cows given the postpartum ECP therapy injection, despite fewer cows cycling before AI. In conclusion, postpartum ECP therapy increased pregnancy rates in 4x cows, but had a detrimental effect on milk yields of 4x milked cows unless bST was administered.; Dairy Day, 2002, Kansas State University, Manhattan, KS, 2002;

Keywords

Diary Day, 2002; Kansas Agricultural Experiment Station contribution; no. 03-121-S; Report of progress (Kansas State University. Agricultural Experiment Station and Cooperative Extension Service); 898; Dairy; Estradiol cypionate; bST; Milking frequency; Fertility

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MILKING FREQUENCY, ESTRADIOL CYPIONATE, AND bST ALTERS MILK YIELD AND REPRODUCTIVE OUTCOMES IN DAIRY COWS

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Summary

The objective of this study was to determine how milking frequency, estradiol cypionate (ECP) postpartum therapy given at 1 week after calving, and biweekly bovine somatotropin (bST) administration alter lactational and reproductive outcomes in dairy cattle. Holstein cows (n=144) were randomly assigned to eight treatments (18 cows per treatment): 1) twice daily milking frequency (2x), 10-mg injection of ECP at 1 week after calving (ECP), and bST (given biweekly according to label beginning in the ninth week of lactation); 2) 2x milked, oil (cottonseed oil vehicle for ECP), bST; 3) 2x milked, ECP, and no bST; 4) 2x milked, oil, and no bST; 5) four-times daily milking frequency (4x; first 30 days in milk then 2x thereafter), ECP, and bST; 6) 4x milked, oil, and bST; 7) 4x milked, ECP, and no bST; and 8) 4x milked, oil, and no bST. Milk yields were recorded at each milking during the first 90 days of lactation. Milk samples were collected weekly at each milking and composited to determine milk components (percentages of fat, protein, lactose, solids-not-fat [SNF], milk urea nitrogen [MUN], and somatic cell count [SCC]). Energy-corrected milk yields were calculated for the first 90 days and for whole lactation yields (305-2x-ME standardized lactation records). Ovulation before first AI was synchronized beginning between 59 and 72 DIM using 100 μg of GnRH given 7 days before 25 mg of $PGF_{2\alpha}$, followed in 24 hr by 1 mg of ECP. Cows were inseminated after detected estrus or at 48 hr after ECP. Pregnancy rates were assessed by transrectal ultrasonography 28-30 days after AI. Postpartum ECP therapy increased milk production for first-lactation 2x cows, but decreased milk yields of the multiparous 4x cows until bST restored those yields. Pregnancy rates were greater for the 4x cows given the postpartum ECP therapy injection, despite fewer cows cycling before AI. In conclusion, postpartum ECP therapy increased pregnancy rates in 4x cows, but had a detrimental effect on milk yields of 4x milked cows unless bST was administered.

(Key Words: Estradiol Cypionate, bST, Milking Frequency, Fertility.)

Introduction

Conception in lactating dairy cows began to decline in the mid 1970s and accelerated in the late 1980s during which time average milk yield for DHIA herds in the U.S. has increased steadily from nearly 15,000 to nearly 21,000 lb per cow. Some studies reported that the increase in milk yield is genetically based and has precipitated the decline in conception rates. Because milk yield per cow can be increased by daily milking frequency and the administration of bovine somatotropin (bST), many dairy producers have opted to milk cows three times daily and use bST to increase gross income and thereby reduce fixed costs that are associated with empty milking parlors.

Another common practice on many dairies is the use of a prophylactic postpartum therapy of estradiol cypionate (ECP) to assist transition cows that may have had calving difficulties and retained placentae. Injections of ECP facilitate uterine contractions by increasing endometrial oxytocin receptors that respond to endogenous secretion or injections of oxytocin. Legal label indications for ECP include: 1) correcting anestrus (absence of heat period) in the absence of follicular cysts in some cases; 2) treating cattle having a persistent corpus luteum due to certain causes; 3) expulsion of purulent material from the uterus in pyometra of cows; and 4) stimulate uterine expulsion of retained placentas and mummified fetuses.

In light of these changes in management, little is known about the direct or indirect effects of increased milking frequency and ECP on various reproductive traits, such as resumption and occurrence of estrous cycles (estrus and ovulation), conception rates, and calving intervals. Further, some studies report positive effects of bST on fertility while others show negative effects.

Our objective was to determine the effect of increased milking frequency (2x to 4x/day) during the first 30 days in milk, treatment of bST, and treatment of postpartum ECP therapy on lactation and reproductive outcomes.

Procedures

Holstein cows (n=144) were assigned randomly to a 2×2×2 factorial experiment consisting of eight treatments (18 cows per treatment). The main effects (Figure 1) were milking frequency, postpartum injection of 10 mg of estradiol cypionate (ECP; Pharmacia Animal Health, Kalamazoo, MI) at 8 ± 2.6 days in milk, and bST (administered according to label indications beginning in the 9th week of lactation). Cows with previous lactations were paired based on their previous 305-2x-ME record, heifers were paired based on their predicted transmitting ability (PTA) for milk, and pairs were assigned randomly to either of the milking frequency treatments. The eight treatments were: 1) twice daily milking frequency (2x), 10-mg injection of ECP at 1 week after calving (ECP), and bST (given according to label beginning in the 9th week of lactation); 2) 2x milked, oil (cottonseed oil vehicle for ECP), bST; 3) 2x milked, ECP, and no bST; 4) 2x milked, oil, and no bST; 5) four-times daily milking frequency (4x; first 30 days in milk then 2x thereafter), ECP, and bST; 6) 4x milked, oil, and bST; 7) 4x milked, ECP, and no bST; and 8) 4x milked, oil, and no bST.

Milk yields were recorded at each milking during the first 90 days of lactation. Milk samples were collected weekly at each milking and composited to determine milk components ([percentages of fat, protein, and lactose], solids-not-fat, milk urea nitrogen, and somatic cell count). Energy-corrected milk yields were calculated for the first 90 days and for whole lactation yields (305-2x-ME standardized lactation records). Body weights and body condition scores (1 = thin and 5 = fat) were assessed weekly in all cows during the first 10 weeks after calving.

Blood samples were collected twice weekly from 82 cows beginning after calving until just before first inseminations were scheduled to occur between 69 and 82 days in milk. Blood serum was harvested and analyzed for concentrations of progesterone. Concentration of progesterone for each cow was graphically plotted and occurrence of ovulation was estimated from patterns of serum progesterone that occur during a normal estrous cycle. Based on these patterns, days from calving to first and second ovulation were calculated.

Ovulation before first AI was synchronized (Heatsynch) beginning between 59 and 72 DIM using 100 µg of GnRH given 7 days before 25 mg of $PGF_{2\alpha}$, followed in 24 hr by 1 mg of ECP. Cows were inseminated after detected estrus, or in the absence of estrus, at 48 hr after ECP (timed artificial insemination [TAI]). Blood was collected prior to each hormonal injection for later analysis of serum concentrations of progesterone. From these concentrations, the percentage of cycling and anestrous cows were determined before ovulation synchronization and AI. Those cows that had resumed estrous cycles had elevated concentrations of progesterone in any of the three samples (cycling), whereas those that remained anestrus had low concentrations of progesterone in all three samples. Pregnancy rates were assessed by transrectal ultrasonography 28 to 30 d after AI.

Based on visual observation of estrus during the period that included ovulation synchronization, the following were calculated: days to first postpartum estrus, the percentage in estrus after the ECP treatment injection, percentage of cows in estrus at least once before ovulation synchronization, percentage in estrus prior to TAI, and hours from breeding protocol injection of ECP to estrus.

In general, data were analyzed as a factorial experiment using analysis of variance (General Linear Models procedure of SAS). When interactions of main effects were detected, eight treatments replaced main effects. For those data collected prior to first administration of bi-weekly injections of bST (60 days in milk or the first 8 weeks of milk data), only two main effects (milking frequency and postpartum ECP therapy treatment) were included in models. When significant F ratios were determined by analyses of variance, mean separations were made using the Tukey-Kramer adjustment for multiple mean comparisons.

Results and Discussion

Reproductive Traits

Among the ovulatory traits assessed, days to first and second postpartum ovulation were not different among treatments (Table 1) in the reduced number of cows (n=82) in which blood was collected twice weekly during the first 60 days postpartum. In contrast, in all 143 cows (blood samples missing from one cow), 20% fewer ($P<0.01$) cows in the 4x + ECP treatment were cycling before initiating the Heatsynch breeding protocol.

Among estrual traits assessed by visual observation, both milk frequency and ECP treatment influenced behavior. Milking 4x daily compared to 2x daily reduced ($P<0.01$) days to first estrus and the percentage of cows in estrus after ECP treatment. All of which implies that estrus behavior was reduced by increased milking frequency. Treatment with ECP compared to oil at the end of the first week postpartum, decreased ($P<0.01$) days to first behavioral estrus and increased ($P<0.01$) the percentage of cows in estrus after ECP. These effects of ECP reflect its estrogenicity and ability to induce sexual behavior. The percentages of cows in estrus at least once before TAI or after $PGF_{2\alpha}$, or the timing of the estrus after $PGF_{2\alpha}$ were not affected by treatments.

Pregnancy rates averaged 41% (Figure 2). Of the four treatment groups, the 4x + ECP cows had the greatest ($P<0.05$) pregnancy rate of 53%. A three-way interaction ($P<0.05$) of milking frequency, ECP therapy, and occurrence of estrus after $PGF_{2\alpha}$ was detected. The difference was observed in the oil-treated groups (2x + oil and 4x + oil), where pregnancy rates of the 4x milked cows after TAI group were the lowest and those of the 2x milked cows after detected estrus were the lowest. It seems that the ECP therapy injection produced more consistent pregnancy rates regardless of milking frequency or whether inseminations were made after detected estrus, or by TAI, in the absence of estrus.

Body Weight and Condition

Average body weights and body condition scores during the first week after calving are summarized in Table 2 along with the average percentage change in both traits during the first 10 weeks of lactation. Neither milking frequency nor ECP treatment had any effect on body weight change in first-lactation cows. However, a tendency (*P*=0.07) was detected for an interaction between milking frequency and ECP therapy treatment for change in body condition score. Cows milked 4x daily and treated with oil lost the most body condition compared to the least condition loss for cows milked 2x daily and treated with oil.

Losses in body weight were greater (*P*<0.05) for multiple-lactation cows that were milked 4x daily than those milked 2x daily. A tendency (*P*=0.06) for a treatment × week interaction also was detected. The interaction was caused by a more rapid increase in body weight gain in the cows in the 2x + oil treatment. By week 5 of lactation, these cows had exceeded their first week's body weight by the fifth week (exceeded 100%), whereas that only occurred in two of the three remaining treatments by the tenth week. Treatment had no effect on changes in body condition scores in multiple-lactation cows.

Lactational Traits

First-lactation cows. During the first 8 weeks of lactation before initiation of bST treatment, first-lactation cows treated with ECP produced more (*P*<0.05) ECM than cows treated with oil. However, a treatment × week interaction (*P*<0.001) also was detected (Panel A; Figure 3). Increased milking frequency did not consistently improve yields because only 4x + ECP cows produced more ECM than cows in all other treatment during the first 2 weeks; more than 4x + oil and 2x + oil, treated cows during week 3; and more than 4x + oil cows during week 4. The interaction was partly caused by the convergence of yields to similar values after day 30 after which time all cows were milked 2x daily.

Yields of first-lactation cows after initiation of bST treatment were unaffected by treatments only among the cows previously milked 4x daily. The cows in the 4x + oil + no bST and 4x + ECP + bST produced (*P*<0.001) more ECM during 60 to 90 days in milk than the 4x + ECP + no bST cows. These results are consistent treatment effects on yields during the first 60 days in milk and reinforce the negative effect of ECP on milk secretion of 4x cows. Further, it demonstrates that bST treatment restored yields of cows previously treated with ECP equal to oil-treated, 4x + no bST levels.

Standardized energy-corrected 305-2x-ME yields of first-lactation cows were affected by an interaction (*P*<0.05) of milking frequency × ECP therapy (Table 2). Among 2x milked cows, ECP stimulated greater standardized ECM yields, whereas among 4x milked cows, ECP reduced whole lactation yields.

Multiple-lactation cows. The main effects of milking frequency (*P*=0.05) and ECP (*P*<0.01) were both significant, but a strong interaction (*P*<0.001) of treatments and a treatment × week interaction (*P*<0.001) was detected (Panel B; Figure 3). Yield of ECM for 4x + oil cows was clearly elevated during the first 6 weeks of lactation, with treatment of ECP inhibiting the ability of 4x cows to produce more milk than 2x cows during the first 30 days in milk. During the 4x milking period, the 4x + oil cows differed from the other treatments during weeks 2 and 4; with all treatment groups converging during weeks 7 and 8, except for the 4x + ECP cows in which ECM was clearly lowest of all treatments.

During the 60- to 90-day period after bST treatments began, ECM was affected by the main effects of ECP and bST, but no interaction of treatment × week was detected (Table 2). For older cows, ECP treatment reduced (P<0.01) ECM yields uniformly except in the 2x + ECP + no bST cows, where yield seemed to be greater than that for 2x + oil + no bST cows (Table 2). Treatment with bST increased (P<0.001) ECM yields in all but the 2x + ECP + bST cows compared to the 2x + ECP + no bST cows (Table 2).

For older cows, whole lactation yields were only affected by bST (Table 2). Treatment with bST consistently increased (P<0.01) yields in all treatments, even to restore reduced yields of 4x + ECP cows to levels equal to that of 4x cows treated with oil.

Conclusions

Milking first-lactation cows 4x daily increased yields during the first 30 days of lactation, but only in the absence of ECP. Postpartum ECP therapy stimulated yields in first-lactation cows, but decreased yields in older cows. Treatment with ECP continued to suppress yields in older cows after initiating bST treatments in the 9th week lactation. The long-term carry-over effect of the early lactation injection of ECP affected the entire lactation yield. Whole lactation yields followed similar trends with ECP inhibiting yields in 4x milked cows but enhancing them in 2x milked cows. Administration of bST restored yields of 4x milked + ECP cows to the levels equal to or greater than contemporaries treated with oil or bST. Based on our results, when ECP is used as a postpartum therapy, bST must be used to prevent reduction in whole lactation yields. Increased milking frequency during the first 30 days in milk provided no milk production benefit to cows based on the measures examined in our study.

Reproductive outcomes were generally positive with ECP therapy increasing pregnancy rates at first service and making pregnancy rates consistent after detected estrus or TAI. These positives occurred despite the fact that ECP decreased the number of cows cycling prior to first insemination. Treatment with ECP seemed to have no effect on the occurrence of postpartum ovulation in 82 of the cows blood sampled twice weekly, but negatively affected characteristics related to estrus activity. Increasing milking frequency and use of ECP did not reduce reproductive efficiency.

Table 1. Reproductive Characteristics of Lactating Dairy Cows after Treatments

Item	Treatment[1]			
	2x + oil	2x + ECP	4x + oil	4x + ECP
	---------- Mean ± SE or % (no.) ----------			
Ovulatory traits[1]				
Days to first ovulation	26 ± 3 (20)	25 ± 3 (21)	24 ± 3 (23)	26 ± 3 (18)
Days to second ovulation	44 ± 3 (14)	44 ± 3 (16)	48 ± 3 (20)	45 ± 3 (14)
Percentage cycling before TAI[a]	100 (33)	97 (36)	97 (36)	78 (36)
Estrual traits[2]				
Days to first behavioral estrus[b]	30 ± 4 (17)	17 ± 3 (23)	47 ± 4 (20)	34 ± 4 (21)
Percentage in estrus after postpartum ECP[c]	11 (35)	42 (36)	0 (36)	14 (37)
Percentage in estrus at least once before TAI	49 (35)	64 (36)	56 (36)	57 (37)
Percentage in estrus after $PGF_{2\alpha}$	65 (34)	64 (36)	53 (36)	47 (38)
Hours to estrus after $PGF_{2\alpha}$	27 ± 5 (22)	30 ± 6 (17)	23 ± 6 (15)	28 ± 6 (16)

[1]See experimental protocol in Figure 1.

[2]Based on serum progesterone.

[3]Based on visual observation.

[a]Fewer (P<0.01) 4x + ECP cycling cows.

[b]4x milked (40.2 ± 2.6 days; n=41) differed (P<0.01) from 2x milked (23.7 ± 2.6 days; n=40). ECP (25.4 ± 2.5 days; n=44) differed (P<0.01) from oil (38.5 ± 2.7 days; n=37). Lactation 1 (36.5 ± 2.4 days; n=47) differed (P<0.01) from lactation 2+ (26.6 ± 2.8 days; n=34).

[c]4x milked (7%; n=73) differed (P<0.01) from 2x milked (27%; n=71). ECP (27%; n=73) differed (P<0.01) from oil (6%; n=71).

Table 2. Average Body Weights and Body Condition Scores

	Treatment[1]			
Item	2x + oil	2x + ECP	4x + oil	4x + ECP
	---------------- Mean ± SE -------------------			
Body weight, lb				
First lactation	1182 ± 106[3]	1195 ± 92	1116 ± 152	1206 ± 129
	98 ± 1.4[4]	96 ± 1.3	96 ± 1.2	97 ± 1.1
Multiple lactation[a]	1487 ± 182	1459 ± 211	1570 ± 292	1409 ± 179
	101 ± 1.7	98 ± 2.0	96 ± 1.7	96 ± 1.6
Body condition score[2]				
First lactation[b]	2.7 ± 0.3[3]	2.8 ± 0.4	2.5 ± 0.4	2.7 ± 0.3
	91 ± 2.3[4]	89 ± 2.1	84 ± 2.1	89 ± 1.9
Multiple lactation	2.8 ± 0.4	2.5 ± 0.4	2.8 ± 0.7	2.4 ± 0.5
	89 ± 3.0	86 ± 3.7	86 ± 3.0	82 ± 2.8

[1]See experimental protocol in Figure 1.

[2]Thin=1 and fat=5.

[3]Mean body weight or body condition score during the first week after calving.

[4]Average percentage change in body weight or body condition score during the first 10 weeks after calving.

[a]2x milked multiple-lactation cows lost less ($P<0.05$) body weight than 4x multiple-lactation cows.

[b]A tendency ($P=0.08$) for an interaction between milking frequency and postpartum ECP therapy injection for body condition scores.

Table 3. Energy-Corrected Milk (ECM) Yields (lb) of Lactating Dairy Cows

	ECM (60-90 days in milk)		ECM 305-2x-ME	
Treatments[1]	Lactation 1	Lactation 2+[a]	Lactation 1[b]	Lactation 2+[c]
	----- Mean (no.) -----		----- Mean (no.) -----	
2x + ECP + bST	75 (11)	90 (9)	31,633 (11)	31,315 (9)
2x + Oil + bST	76 (11)	99 (8)	29,034 (11)	32,328 (8)
2x + ECP + no BST	75 (9)	99 (7)	31,069 (9)	28,958 (7)
2x + Oil + no BST	75 (9)	93 (6)	29,239 (9)	27,749 (6)
4x + ECP + bST	78 (11)	98 (9)	32,680 (11)	32,729 (9)
4x + Oil + bST	73 (12)	104 (9)	32,279 (12)	30,625 (9)
4x + ECP + no BST	68 (10)	86 (7)	28,326 (10)	25,411 (7)
4x + Oil + no BST	78 (9)	90 (6)	32,652 (9)	28,698 (6)

[1]See experimental protocol in Figure 1.

[a]Oil vs. ECP (P<0.01). No bST vs. bST (P<0.0001).

[b]Milking frequency × ECP interaction (P<0.05): 2x milked + oil = 29,137 ± 945 lb (n=20); 2x milked + ECP = 31,351 ± 945 lb (n=20); 4x milked + oil = 32,465 ± 927 lb (n=21); and 4x milked + ECP = 30,505 ± 919 kg (n=21).

[c]bST effect (P<0.01): No bST = 27,703 ± 1,077 lb (n=26) vs. bST 31,749 ± 927 lb (n=35).

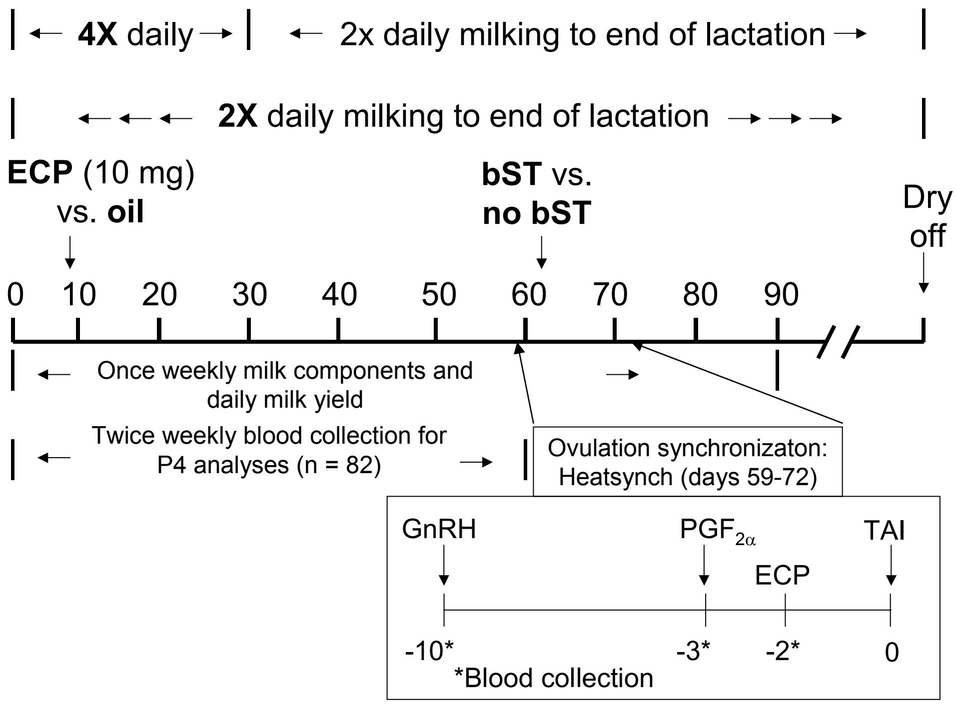

Figure 1. Experimental Protocol.

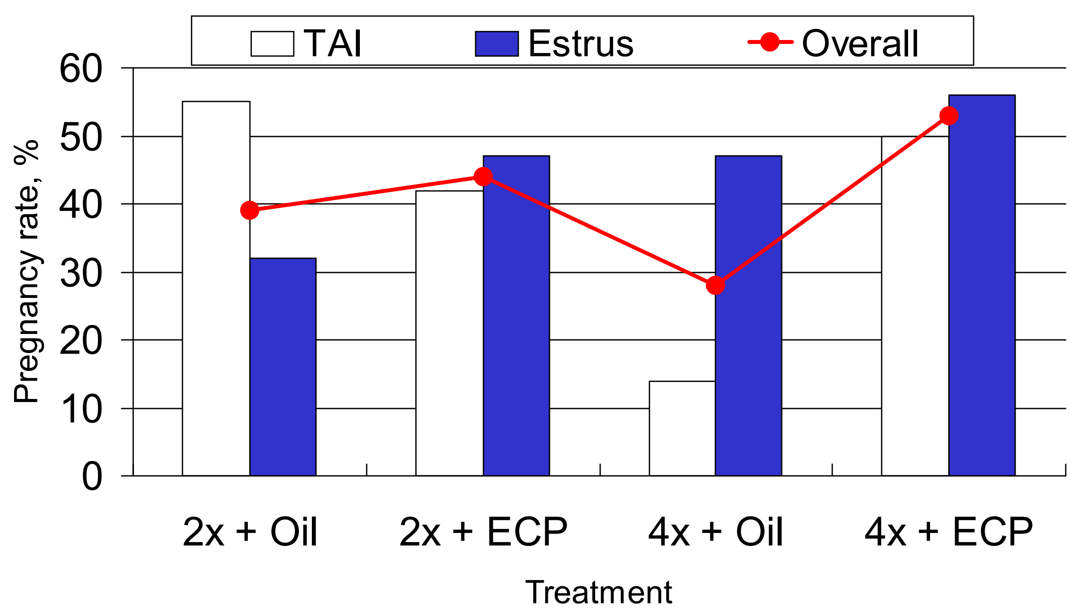

Figure 2. Pregnancy Rate of Dairy Cows 28 to 30 Days After Timed Artificial Insemination (TAI). The overall pregnancy rate differed (P<0.05) among treatments with cows milked 4x daily + ECP having the greatest pregnancy rates. In addition, a three way interaction (P<0.05) was detected for milking frequency, postpartum ECP therapy, and occurrence of estrus after $PGF_{2\alpha}$.

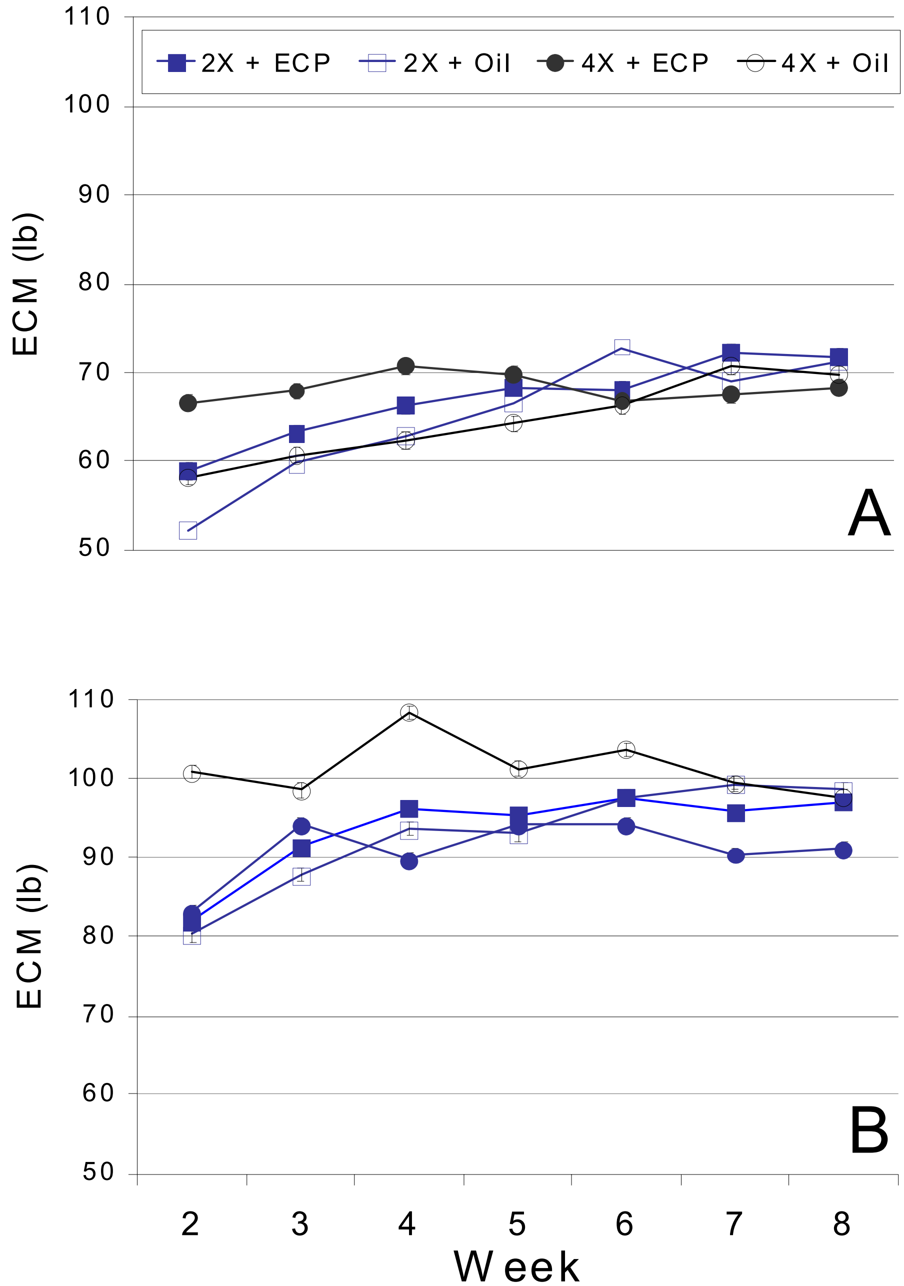

Figure 3. Energy Corrected Milk Yield of First-Lactation (Panel A) and Multiple-Lactation Cows (Panel B) During the First 8 Weeks of Lactation.