Effect of Weaning System on Milk Composition and Distribution of Milk Fat within the Udder of East Friesian Dairy Ewes

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ABSTRACT

We investigated whether the inhibition of milk ejection during and/or between machine milkings is responsible for the low milk fat observed in commercial milk obtained from dairy ewes managed with a mixed system (MIX) of partial daily suckling (10 h) and once daily machine milking (after 14 h of udder filling). East Friesian crossbred dairy ewes were randomly allocated postpartum to the MIX system (n = 9) or to exclusive twice-daily machine milking (DY1, n = 8). Following wk 4, MIX ewes were permanently weaned from their lambs and milked twice daily. All ewes were injected with saline, oxytocin, or an oxytocin-receptor antagonist prior to three morning milkings during wk 2, 4, and 6 of lactation to study cisternal and alveolar milk distribution. Overall milk yield (cisternal + alveolar) for MIX ewes was 42% greater than for DY1 ewes during wk 2 and 4, which demonstrates the beneficial effect of lamb suckling on milk production of dairy ewes. However, during normal machine milking, only the cisternal fraction was obtained from MIX ewes, confirming that milk ejection did not occur for as long as these ewes remained in partial daily contact with their lambs. Although the volume of milk stored within the cistern, and its concentration of milk protein was similar for the two weaning systems, milk of MIX ewes was significantly inferior in cisternal milk fat concentration and yield compared to DY1 ewes. This provides evidence that not only is there inhibition of milk ejection during machine milking of MIX ewes, there is additional inhibition of transfer of milk fat, but not milk protein, from the alveoli to the cistern during the evening when MIX ewes are separated from their lambs. Following weaning of MIX ewes, the majority of lactation traits studied were similar compared to DY1 ewes.

(**Key words:** East Friesian, milk ejection, milk fat, weaning system)

Abbreviation key: AT = Atosiban, an oxytocin receptor antagonist, C18:1 = octadecenoic acid, DY1 = d-1 weaning system, MIX = mixed weaning system, OT = oxytocin, SAL = saline.

INTRODUCTION

For dairy sheep producers, a mixed system (MIX) of suckling only during the day and once daily machine milking during the morning for the first 30 d of lactation is common (Folman et al., 1966; Papachristoforou, 1990; Gargouri et al., 1993). This MIX system has been shown to be economically superior, in terms of lamb and milk production, to both the traditional system of lamb suckling and no machine milking during the first 30 d of lactation, and to a system (DY1) where lambs are weaned at 24 h postpartum and the ewe is machine milked twice daily (McKusick et al., 2001b). The main disadvantage of the MIX system, however, is that the commercial milk obtained during the first 30 d of lactation (during the period of partial lamb contact) is low in fat content (Gargouri et al., 1993; Fuertes et al., 1998; McKusick et al., 2001b), which could potentially disadvantage this milk for cheese making (Requena et al., 1999).

Reasons for low milk fat in MIX ewes could involve one or all of the following three physiologic mechanisms: 1) milk ejection during machine milking does not occur, 2) milk fat synthesis is inhibited, and 3) milk fat transfer from the alveoli to the cistern between milkings does not occur. Marnett and Negrão (2000) have addressed the first possibility, and have demonstrated that plasma oxytocin concentrations do not increase above baseline levels during machine milking of MIX ewes; which causes failure of milk ejection during milking, but not during suckling, for as long as these ewes remain in partial daily contact with their lambs. Therefore, only the cisternal milk fraction is assumed to be available during machine milking of MIX ewes, because removal of the alveolar milk fraction would require active myoelectric

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thelial contraction (Bruckmaier et al., 1994). It has been estimated that up to 75% of the total fat yield within the udder is associated with the alveolar milk fraction, and thus is only obtainable when milk ejection occurs during machine milking (Labussière, 1969). However, this estimation was made without any pharmacological blockade of milk ejection, and relied solely on the researcher's subjective ability to identify ewes that did not present a milk ejection reflex during a milk flow emission recording (unimodal milk flow emission). Furthermore, in ewes with large cisterns and high levels of milk production such as the East Friesian (Bruckmaier et al., 1997; McKusick et al., 1999), bimodal milk flow emission may not be visible, therefore making it difficult to study milk fraction differences (Marnet et al., 1998).

The second possibility is supported by some research suggesting that fat synthesis in the mammary gland might be inhibited by certain medium or short chain fatty acids (Levy, 1964; Williamson et al., 1995). If milk ejection and distribution is altered in ewes managed with the mixed system of suckling and machine milking during early lactation, it is possible that milk fat synthesis could be inhibited or modified in MIX ewes due to milk fat stasis in the alveoli (McKusick et al., 2001a). Finally, we hypothesize that reduced milk fat transfer from the alveoli to the cistern, and thus milk stasis, might be due to changes in milk fluidity and fatty acid distribution within the udder.

There exist no reports in the literature on how milk is stored within the udder for animals managed with different weaning systems. With the availability of a new technique that involves an oxytocin receptor antagonist (Atosiban), a more accurate determination of cisternal and alveolar distribution of milk can be determined in dairy ruminants (Knight et al., 1994; Wellnitz et al., 1999). The objectives of the present experiment were to study milk ejection, milk storage, and milk yield within the udder of dairy ewes managed with the MIX or DY1 systems. More specifically, differences between cisternal and alveolar milk yield, milk fat and protein concentrations, milk fatty acid composition, and somatic cell count (SCC) were estimated.

MATERIALS AND METHODS

Seventeen fourth parity East Friesian crossbred dairy ewes (50 to 75% East Friesian and 25 to 50% Dorset, Polypay, and/or Rambouillet) in the first 6 wk of lactation were studied during the spring of 2001. Ewes had been selected from the University of Wisconsin-Madison's main dairy ewe flock at the Spooner Agricultural Research Station and synchronized for lambing. Ewes were kept in an indoor laboratory facility on the University of Wisconsin-Madison campus; each weaning system group

was housed separately in two rooms and fed a 16% crude protein grain concentrate and alfalfa haylage in excess of NRC requirements for lactating ewes. The milking machine (Coburn Co., Inc., Whitewater, WI, and Interpuls Inc., Albinea, Italy) was set to provide 165 pulsations per minute in a 50:50 ratio with a vacuum level of 37 kPa.

Ewes were randomly allocated to one of two weaning systems by the order that they lambed: 1) beginning 24 h postpartum, ewes were separated from their lambs for 14 h during the evening (1630 to 0630), ewes were machine milked once daily the following morning (0630), and their lambs were allowed to suckle for 10 h during the day (MIX, $n = 9$); or 2) ewes were weaned from their lambs at 24 h postpartum, ewes were machine milked twice daily (0630 and 1630), and their lambs were raised artificially (DY1, $n = 8$). Permanent weaning of MIX ewes occurred at the end of wk 4, and MIX ewes were subsequently milked twice daily.

On Monday, Wednesday, and Friday, during wk 2, 4, and 6 of lactation, three injection treatments were randomly administered to ewes in both weaning system groups immediately prior to the morning milking in a 3×3 Latin square design: 1) an intravenous injection of sterile physiological saline (SAL, 1 ml/ewe), 2) an intravenous injection of an oxytocin receptor antagonist (Atosiban, AT, 1 mg/ewe, Ferring Research Institute, Inc., San Diego, CA), or 3) an intramuscular injection of oxytocin (OT, 2 IU/ewe, Vedco, Inc., St. Joseph, MO). In the evening (1630) prior to a treatment morning, 2 IU of OT were administered intravenously to each ewe, followed by machine milking to empty the udders of ewes in both treatment groups as completely as possible. As a result of oxytocin receptor antagonism with the AT treatment, milk ejection during machine milking would presumably not occur, and only the cisternal milk fraction would be obtained. A dose of 1 mg/ewe of AT had been tested in our flock, and is a reliable antagonist for approximately 15 min. The time between AT injection and milking of an individual ewe never exceeded 15 min. Conversely, as a result of OT treatment, milk ejection would presumably occur and thus both cisternal and alveolar fractions would be obtained together during machine milking. Saline injections were administered as a control. Individual ewe milk yield during each treatment was recorded.

After milk yield measurements and milk samples were taken for the AT treatment only, an additional injection of 2 IU of OT was administered to reestablish milk ejection and allow the alveolar milk fraction to be measured and sampled. Milk samples were analyzed for percentage of milk fat and protein and Fossomatic SCC by a State of Wisconsin certified laboratory. SCC was transformed to logarithms of base ten. Milk fat and protein yield was calculated by multiplying milk yield by milk

fat or protein percentage. Total milk yield and milk fat and protein yield were calculated by adding cisternal and alveolar milk together. Total percentages of milk fat and protein were calculated by dividing total milk fat or protein yield by total milk yield, and total SCC was calculated by a weighted average of the cisternal and alveolar SCC. Alveolar and cisternal milk fraction data obtained during wk 2 and 4 were averaged for some of the analyses for comparison with data obtained during wk 6, the time when both MIX and DY1 ewes were exclusively machine milked.

At 12 h postpartum, and on Tuesday of wk 1, 3, and 6, milk samples were collected with an aseptic technique from each udder half of every ewe for routine aerobic bacteriological culture.

During wk 4, a 50 ml sample of both cisternal and alveolar milk from each ewe was pooled within weaning system and milk fraction. Samples of raw milk were cooled and the fat fraction separated by centrifugation for 30 min at $6000 \times g$ and 4°C . The fat fraction was collected from the vessel and extracted with diethyl ether. The extract was dried under a stream of nitrogen, and the oil residue frozen at -80°C . Gas chromatography was performed on the extracted milk fat samples in triplicate according to Alonso et al. (1999) to determine the proportions of several fatty acids.

ANOVA were conducted with the general linear models procedure of SAS (1999) for a 3×3 Latin square experimental design. Milk yield data presented in Figure 1 were analyzed with a model that included the following independent variables and interactions: weaning system (DY1 or MIX), ewe within weaning system, treatment (AT, OT, or SAL), day (Monday, Wednesday, or Friday), week (2, 4, or 6), weaning system $\times$ treatment, weaning system $\times$ treatment $\times$ week, and residual error. Significance of the weaning system $\times$ treatment $\times$ week interaction was tested with residual error. Analyses of fractional milk (alveolar vs. cisternal) data in Table 1 were conducted with a model that included the following independent variables and interactions: fraction (alveolar or cisternal), weaning system, fraction $\times$ weaning system, ewe within weaning system, period (wk 2 and 4 averaged together, or wk 6), fraction $\times$ period, weaning system $\times$ period, fraction $\times$ weaning system $\times$ period, and residual error. Significance of the fraction $\times$ weaning system $\times$ period interaction was tested with residual error. The model used to analyze total milk data in Table 1 included the following independent variables and interactions: weaning system, ewe within weaning system, period, weaning system $\times$ period, and residual error. Significance of the weaning system $\times$ period interaction was tested with residual error. The model used to analyze milk fatty acid data in Table 2 included the following independent variables and interactions: weaning sys-

tem, fraction, weaning system $\times$ fraction, day, weaning system $\times$ day, fraction $\times$ day, weaning system $\times$ fraction $\times$ day, and residual error. Significance of the weaning system $\times$ fraction interaction was tested with residual error. Within all models, because the interaction of interest was found to be significant, no other parts of the models were tested. Statistical significance between least squares means was tested with Fisher's LSD test.

RESULTS

The weaning system $\times$ injection treatment $\times$ week interaction was a significant source of variation for milk yield, and data are presented in Figure 1. During wk 2 and 4, milk yield was highest for MIX ewes treated with OT, intermediate for DY1 ewes treated with SAL or OT, and lowest for MIX ewes treated with SAL or AT, and for DY1 ewes treated with AT. Treatment of MIX ewes with OT increased milk yield by 60% compared to MIX ewes treated with SAL during wk 2 and 4, however, for DY1 ewes, milk yield was similar between OT and SAL treatments. Milk yield was similar between MIX and DY1 ewes treated with AT during all experimental weeks, which did not differ during wk 2 and 4 from MIX ewes treated with SAL. Following weaning of MIX ewes, milk yield did not differ between MIX and DY1 ewes during SAL or OT treatment, whereas treatment with AT decreased milk yield by 45% regardless of weaning system treatment at wk 6.

The weaning system $\times$ time-period interaction for cisternal, alveolar, and total milk fractions were significant sources of variation with respect to milk yield, milk composition, and SCC; Table 1 presents the least squares means. Total milk yield for MIX ewes was 42% higher compared with DY1 ewes during wk 2 and 4, however, following weaning of MIX ewes, milk yield was not different between weaning system groups at wk 6. Total milk yield for MIX and DY1 ewes decreased by 40 and 23%, respectively, from wk 2 and 4 to wk 6. During wk 2 and 4, the amount of milk stored within the cistern for DY1 and MIX ewes was similar. Cisternal milk represented more of the total milk volume (55%) in DY1 ewes, whereas relatively less milk was stored in the cisternal fraction (43%) of MIX ewes. During wk 6, milk storage in the alveolar and cisternal compartments was similar within and between weaning system groups.

During wk 2 and 4, average total percentage of milk protein was similar between weaning systems, yet milk protein percentage was higher in cisternal milk compared to alveolar milk. During wk 6, percentage of milk protein did not differ between weaning system or milk fraction. Total milk protein yield was higher for MIX ewes during wk 2 and 4 compared to DY1 ewes but similar between weaning systems during wk 6. Alveolar

Table 1. Least squares means and SEM for morning milk yield and composition for the weaning system treatment by time-period combinations.

Factor	Fraction ¹	Period			
		Wk 2 and 4		Wk 6	
		DY1 ²	MIX ³	DY1	MIX
Milk yield, kg	Alveolar	1.03 ^d	1.86 ^a	0.87 ^d	1.05 ^{cd}
	Cisternal	1.28 ^{bc}	1.42 ^b	0.90 ^d	0.90 ^d
	SEM	0.12			
	Total	2.31 ^b	3.28 ^a	1.77 ^c	1.95 ^c
	SEM	0.08			
Milk protein, %	Alveolar	4.24 ^c	4.30 ^c	4.22 ^c	4.31 ^c
	Cisternal	4.59 ^{ab}	4.65 ^a	4.39 ^{cd}	4.49 ^{bc}
	SEM	0.06			
	Total	4.45 ^a	4.44 ^a	4.25 ^b	4.38 ^{ab}
	SEM	0.04			
Milk protein, g	Alveolar	44.0 ^c	81.7 ^a	34.8 ^c	44.5 ^c
	Cisternal	58.2 ^b	64.0 ^b	38.2 ^c	39.6 ^c
	SEM	5.5			
	Total	102.1 ^b	143.9 ^a	73.0 ^c	84.1 ^c
	SEM	3.8			
Milk fat, %	Alveolar	8.25 ^a	6.87 ^b	6.96 ^b	6.97 ^b
	Cisternal	3.48 ^c	2.09 ^d	3.64 ^c	3.76 ^c
	SEM	0.25			
	Total	5.48 ^a	4.77 ^b	5.23 ^a	5.50 ^a
	SEM	0.18			
Milk fat, g	Alveolar	81.6 ^b	128.3 ^a	58.5 ^{cd}	73.6 ^{bc}
	Cisternal	42.8 ^{de}	29.9 ^c	32.2 ^e	33.2 ^c
	SEM	6.7			
	Total	124.2 ^b	154.6 ^a	88.3 ^d	106.9 ^c
	SEM	4.6			
SCC, log units	Alveolar	5.06 ^a	4.86 ^{bc}	4.85 ^{bc}	5.06 ^a
	Cisternal	4.71 ^{cd}	4.61 ^d	4.69 ^{cd}	4.90 ^{ab}
	SEM	0.08			
	Total	4.88 ^{ab}	4.72 ^c	4.74 ^{bc}	4.98 ^a
	SEM	0.05			

^{a,b,c,d,e}Means within rows or milk fractions for an individual factor with different superscripts differ ($P < 0.05$).

¹Milk fraction. Immediately prior to milking, ewes were injected with an oxytocin receptor antagonist for the removal of cisternal milk, and then injected with oxytocin for the removal of alveolar milk. Total milk is the sum (or average) of alveolar and cisternal milk fractions.

²DY1 ewes ($n = 8$) were weaned from their lambs within 24 h postpartum and machine milked twice daily (0630 and 1630).

³MIX ewes ($n = 9$) suckled their lambs during the day, were separated from their lambs in the evening (1630), and machine milked once daily in the morning (0630). MIX ewes were permanently weaned at the end of wk 4.

milk from MIX ewes contained the most milk protein, cisternal milk from MIX or DY1 ewes was intermediate, and alveolar milk from DY1 ewes contained the least amount of milk protein during wk 2 and 4. During wk 6, milk protein yield did not differ between weaning system or milk fraction.

Average total percentage of milk fat within the udder of MIX ewes was lower than for DY1 ewes during wk 2 and 4, but similar between weaning systems during wk 6. The cisternal milk fraction of MIX ewes contained a lower concentration of milk fat than the cisternal milk fraction of DY1 ewes. Regardless of weaning system during wk 2 and 4, the alveolar milk fraction had a higher percentage of milk fat compared to the cisternal fraction, although MIX ewes had less alveolar milk fat percentage

than DY1 ewes. During wk 6, the cisternal milk fraction continued to be less concentrated with milk fat than the alveolar fraction, but there were no significant differences between weaning system for total milk fat percentage. Although total milk fat yield during wk 2 and 4 was higher for MIX ewes compared to DY1 ewes, only 19% of the total fat yield was present in the cisternal fraction of MIX ewes compared to 35% in DY1 ewes. During wk 6, MIX ewes had higher total milk fat yield compared to DY1 ewes, and milk fat yield continued to be lower in the cisternal milk fraction compared to the alveolar fraction regardless of weaning system.

During wk 2 and 4, average SCC for MIX ewes compared with DY1 ewes was lower, yet higher during wk 6. Regardless of weaning system during wk 2 and 4,

Table 2. Least squares means and SEM for individual fatty acid concentrations expressed as a percentage of total lipids by the weaning system $\times$ milk fraction combinations.

Fatty acid	Fraction ¹	Weaning system			Fatty acid	Fraction	Weaning system		
		DY1 ²	MIX ³	SEM			DY1	MIX	SEM
C4:0	Alveolar	4.14	4.13	0.01	C16:0	Alveolar	25.72	25.35	0.15
	Cisternal	4.12	4.13			Cisternal	25.37	25.51	
C6:0	Alveolar	2.43	2.44	0.02	C16:1	Alveolar	3.29	3.30	0.02
	Cisternal	2.42	2.41			Cisternal	3.28	3.27	
C8:0	Alveolar	2.38	2.36	0.01	C18:0	Alveolar	8.22	8.24	0.01
	Cisternal	2.38	2.35			Cisternal	8.24	8.25	
C10:0	Alveolar	9.54	9.53	0.02	C18:1	Alveolar	19.06 ^{ab}	19.24 ^a	0.30
	Cisternal	9.56	9.57			Cisternal	18.99 ^{ab}	18.46 ^b	
C12:0	Alveolar	5.52	5.53	0.01	C18:2	Alveolar	2.75	2.74	0.02
	Cisternal	5.49	5.51			Cisternal	2.77	2.77	
C14:0	Alveolar	10.22	10.21	0.01	C18:3	Alveolar	1.06	1.06	0.01
	Cisternal	10.18	10.20			Cisternal	1.05	1.05	
C14:1	Alveolar	4.72	4.72	0.02	C20:0	Alveolar	0.05	0.04	0.01
	Cisternal	4.74	4.72			Cisternal	0.04	0.04	

^{a,b}Means within an individual fatty acid with different superscripts differ ($P < 0.10$).

¹Milk fraction. Immediately prior to milking, ewes were injected with an oxytocin receptor antagonist for the removal of cisternal milk, and then injected with oxytocin for the removal of alveolar milk.

²DY1 ewes ($n = 8$) were weaned from their lambs within 24 h post-partum and machine milked twice daily (0630 and 1630).

³MIX ewes ($n = 9$) suckled their lambs during the day, were separated from their lambs in the evening (1630), and machine milked once daily in the morning (0630). MIX ewes were permanently weaned at the end of wk 4.

SCC was lower in the cisternal milk fraction compared to the alveolar fraction. With respect to milk fraction during wk 6, SCC was similar within weaning system.

Incidence of intramammary infection, based on bacteriology, was low for both groups and nonsignificant between groups (data not shown). One udder half of one DY1 ewe tested positive for *Corynebacterium bovis* during

wk 1 and 6, and one udder half of one MIX ewe tested positive for *C. bovis* during wk 6. The other udder halves for these two ewes and all other ewes' udder halves tested negative for routine mastitis pathogens at all testings.

Individual fatty acid concentration expressed as a percentage of total lipid content is summarized in Table 2. For the majority of fatty acids, there were no significant differences due to weaning system or milk fraction. For MIX ewes, concentration of octadecenoic acid (C18:1) tended to be higher in the alveolar, compared to the cisternal milk fraction.

DISCUSSION

Milk Ejection

The fact that milk yield of MIX ewes treated with the oxytocin receptor antagonist (AT) was not different from MIX saline treated controls (SAL) or DY1 ewes treated with AT, and also that exogenous oxytocin did increase milk yield of MIX ewes, definitively demonstrates a failure of milk ejection at machine milking of dairy ewes managed with the MIX weaning system during the first 4 wk of lactation (Figure 1). This explains why only 40 to 60% of the total milk yield is available during machine milking of MIX ewes (McKusick et al., 2001b) and cows (Bar-Peled et al., 1995). Conversely, ewes managed with the DY1 weaning system have normal milk ejection during machine milking because DY1 ewes treated with

Figure 1. Morning milk yield for ewes managed with a mixed system of suckling during the day, separation from their lambs at night, and machine milking once daily in the morning (MIX, $n = 9$) and for ewes managed with no suckling and twice daily machine milking (DY1, $n = 8$). Permanent weaning of MIX ewes took place at the end of wk 4. Injection treatments were randomly administered to both weaning system groups in a 3×3 Latin square design prior to milking: oxytocin (OT), atosiban (an oxytocin receptor antagonist, AT), and saline (SAL). Error bars represent the pooled standard error of the least squares means obtained from the weaning system $\times$ injection treatment $\times$ wk interaction.

oxytocin (OT) had similar milk yield as DY1 ewes treated with SAL. Milk ejection during machine milking was reestablished for MIX ewes following weaning, as evidenced by similar milk yield between MIX and DY1 ewes treated with either SAL or OT at wk 6. The present experiment thus compliments and confirms previous observations of reduced commercial milk yield in MIX managed ewes compared to DY1 managed ewes for as long as MIX ewes remain in partial daily contact with their lambs (Marnet and Negrão, 2000; McKusick et al., 2001b). Moreover, this reduction in milk yield is clearly due to inhibition of milk ejection, because alveolar milk is not obtained during machine milking of MIX ewes.

Milk Synthesis and Storage

The present experiment shows clear differences between weaning systems in milk yield and milk composition and their distribution among the cisternal and the alveolar compartments of the mammary gland. The high level of milk production of MIX ewes prior to weaning was probably at the limit of udder storage capacity. It is likely that the increased milk secretion during wk 2 and 4 observed for MIX ewes is because of delayed weaning of the lambs. This phenomenon has also been observed in partially suckled high producing dairy cows (Bar-Peled et al., 1995). The fact that cisternal milk volume was similar between MIX and DY1 ewes, yet alveolar milk volume was much greater for MIX ewes, indicates that alveolar secretory capacity of these ewes was superior. The mammary gland may synthesize more milk as a result of increased udder stimulation and evacuation during partial daily suckling (Labussière et al., 1974; Walsh, 1974; McKusick et al., 2001b). The beneficial effect could be explained by a reduction in the putative feedback inhibitor of milk lactose and protein synthesis (Wilde et al., 1987), or better hormonal maintenance of lactation due to the favorable effects of prolactin and cortisol (Marnet and Negrão, 2000), or oxytocin (Balous et al., 1993) on milk synthesis. This beneficial effect is removed at weaning, and thus we observed a marked drop in milk production (approximately 40%) for MIX ewes between the preweaning (wk 2 and 4) and postweaning (wk 6) period. This agrees with the earlier observations in dairy ewes of Labussière and Pétrequin (1969).

Total percentage of milk protein did not differ between weaning system in either period, and is consistent with other observations in mammals indicating that the mammary gland is capable of producing milk with similar protein concentration regardless of differences in environment or management (Cowie and Tindal, 1971). Additionally, because milk protein is within the aqueous fraction of milk, the casein micelle had no difficulty in

being transferred from the alveoli to the cistern between milkings.

Compared to the cow, the average size of the milk fat globule in sheep is large (Muir et al., 1993). Large fat globules require active expulsion from the alveoli, usually in conjunction with myoepithelial contraction during milk ejection (Linzell, 1955), for their transfer to the cistern and subsequent removal from the udder. Thus, cisternal milk is normally lower in milk fat concentration than alveolar milk (Labussière, 1969). Because milk ejection was not present during machine milking of MIX ewes and only the cisternal milk fraction was obtainable, a large amount of milk fat was left behind in the udder, resulting in a lower percentage of milk fat compared to DY1 ewes. This alveolar milk of MIX ewes is however available to the lambs during suckling due to normal milk ejection (Marnet and Negrão, 2000), and thus will favor lamb growth (Papachristoforou, 1990; McKusick et al., 2001b) which is one of the important attributes of the "dual-purpose" MIX system.

We have observed a marked reduction in the total amount and percentage of milk fat within the cisternal fraction of MIX ewes, which suggests to us that there was a reduction in the amount of fat transferred to the cistern during the time when MIX ewes were separated from their lambs in the evening. Although the mechanism is presently unknown, one could imagine that the stress associated with separation of MIX ewes from their lambs every evening might have played a role in inhibiting fat transfer. However, we observed that by wk 2, lambs were usually found waiting to be separated in the creep pen, away from the ewes at the 1630 separation time, and there were no behavioral indicators of stress from the ewes (e.g. no increased vocalization or unwillingness to leave the pens and enter the milking parlor). Additionally, at least one report in dairy ewes demonstrated that cortisol concentration for MIX ewes during machine milking was similar to that during suckling (Marnet and Negrão, 2000). Finally, OT release during the period between milkings for ewes managed with the MIX system may have been suppressed due to maternal behavior as suggested by Marnet and Negrão (2000). Olfactory cues perceived by the ewe serve to strengthen the milk ejection reflex during suckling (Marnet et al., 1999). In the present experiment, these cues would have been absent during the evening, and pulsatile release of OT may have been suppressed. Moreover, the effects of possibly higher circulating catecholamines on central and peripheral mechanisms for inhibiting milk ejection (Lefcourt et al., 1997; McKusick and Marnet, 2001) remain to be demonstrated in the context of milk fat transfer and merits further investigation.

We also hypothesized that another mechanism concerning the fluidity of milk due to alteration in individual

milk fatty acid composition might have played a role in prohibiting milk fat transfer from the alveoli to the cistern between milkings. The short chain and unsaturated fatty acid content in milk is important in determining the fluidity of milk fat secretion (Parodi, 1982). However, there were not large differences in the percentage of fatty acids between the cisternal and alveolar milk fractions for ewes managed with either the DY1 or MIX system.

In contrast, the low milk fat observed in milk of MIX ewes may be explained by reduced fat synthesis. Levy (1964) was one of the first to report that certain free fatty acids in milk could inhibit fatty acid synthesis at weaning in rats. Later, Williamson et al. (1995) proposed that medium chain fatty acids played a regulatory role in mammary lipid metabolism of rats, independent of the putative feedback inhibitor of protein synthesis previously described by Wilde et al. (1987). Davis and Brown (1970) hypothesized that one of the reasons for milk fat depression in dairy cows (diet induced low fat milk syndrome) was an increase in trans C18:1 fatty acid content. This has been confirmed by Griinari et al. (1998) who showed that decreased milk fat yield was due to specific increases in trans-10 C18:1 content in milk. The results of the present experiment provide some evidence for an inhibitory role of C18:1 in the alveolar milk fraction. Although total milk fat yield during wk 2 and 4 was higher for MIX ewes compared to DY1 ewes, alveolar and total milk fat concentrations were significantly reduced, implying that the large amount of fat retained within the alveoli of MIX ewes may have participated to some extent in the reduction of milk fat synthesis and merits further investigation.

Considerable research has been aimed at modifying milk fat in dairy cows via nutritional manipulation (see review by Bauman and Griinari, 2001), primarily in an attempt to reduce milk fat content and to identify milk components that improve human health. Some authors feel that milk contains too much fat and that there is now a strong demand for low-fat milk (Boland et al., 2001). Use of the MIX system could provide a nonnutritional technique for obtaining low fat milk during early lactation in dairy ewes and possibly in other dairy species.

Somatic cell count and incidence of intramammary infection for both treatment groups were low during all weeks of the experiment. Regardless of milk fraction, SCC was lower for MIX ewes during the period of partial suckling and once daily milking (wk 2 and 4) compared to DY1 ewes and agrees with other reports in dairy ewes (McKusick et al., 2001b) and dairy cows (Krohn, 1999), possibly due to more frequent udder evacuation. This explanation is supported by the fact that we observed an increase in SCC for MIX ewes during wk 6 of the

experiment when lambs had been permanently weaned and udder evacuation frequency was decreased to twice daily. Collectively, it appears that SCC can increase in dairy ewes without intramammary infection, when udder storage capacity is not sufficient for a given level of milk production or when the time between udder evacuations (suckling or milking) is too infrequent. Our findings of increased SCC in the alveolar, compared to the cisternal milk fraction, regardless of weaning system, are consistent with observations of increased proteolysis in milk with high SCC due to increases in tight junction permeability (Pirisi et al., 1996; Stelwagen et al., 1997) and might provide another reason for lower milk protein percentage in the alveolar fraction.

CONCLUSIONS

The results of the present experiment demonstrated a clear failure of milk ejection during machine milking of East Friesian dairy ewes that are managed by the MIX system in early lactation which resulted in recuperation of only 40 to 60% of the total milk yield for as long as MIX ewes remained in partial contact with their lambs. For ewes managed with the DY1 system, milk ejection was normally present during machine milking. It appears that the transfer of milk fat, but not milk protein, from the alveoli to the cistern was impaired and resulted in poor milk fat concentration of the cisternal milk fraction. Reduced milk fat transfer from the alveoli to the cistern was more severe in ewes managed with the MIX system. Milk ejection is therefore obligatory in dairy ewes for the recuperation of milk that is rich in total solids. Producers who utilize the MIX system during early lactation should expect commercial milk to be low in fat content, which may affect cheese processing characteristics.

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