



# Changes in Udder Compartments (Alveolar and Cisternal) Depending on Lactation Stage and Parity in Crossbred Cows

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## Abstract

This study investigates morphological and compositional variations in the mammary gland of crossbred dairy cows, with specific attention to the udder cisterns and alveoli across different stages of lactation and parities. The primary objectives were: (1) to assess the dimensional changes in the udder cisterns and alveoli in relation to lactation stage and parity, and (2) to analyze variations in milk composition within the cisternal and alveolar compartments. Twenty-eight crossbred dairy cows, comprising both primiparous and multiparous individuals, were evaluated across three distinct lactation phases: early (1–3 months), mid (4–6 months), and late (7 months to drying off). The investigation spanned an 11-month period from January to November 2022. Milk samples from the udder and teat cisterns were analyzed using thermogravimetric methodology.

Findings revealed that cisternal milk constituted approximately 31.35–31.95% of total udder milk in both primiparous and multiparous cows. In contrast, the alveolar compartments in multiparous and primiparous cows contributed up to 70% and 72% of total milk fat, respectively ( $P < 0.05$ ). Notably, no statistically significant differences ( $P > 0.05$ ) were observed in average fat, protein, lactose, and solids-not-fat (SNF) percentages between the cisternal and alveolar compartments in primiparous cows. Cisternal milk volume increased during early and mid-lactation but declined in late lactation, whereas alveolar milk volume demonstrated an inverse trend. A significant decrease of 18% ( $P < 0.05$ ) in alveolar milk volume was observed from early to late lactation, with no further reduction noted from mid to late stages. Cisternal milk volume remained stable from early to mid-lactation but declined by 16% ( $P < 0.05$ ) in the late phase.

In conclusion, the crossbred dairy cows exhibited favorable udder morphology, characterized by medium-sized cisterns and teats, suggesting suitability for mechanical milking. Multiparous cows showed superior performance in both cisternal and alveolar milk yield compared to primiparous counterparts. These findings underscore the productive potential of crossbred cows, although enhancements in nutritional management are recommended to optimize milk composition. Further investigations with larger sample sizes are warranted to substantiate these preliminary observations.

**Keywords:** Cisternal; Alveolar; Stage lactation; Milk fraction; Crossbreed cow

## 1. Introduction

Milk storage within the bovine udder is commonly conceptualized through a two-compartment model, comprising the cisternal and alveolar regions [1,2]. The cisternal compartment encompasses the large ducts, gland cisterns, and teat cisterns, while the alveolar compartment refers to milk secreted and retained within the small ducts and alveoli. Species-specific differences in the proportional distribution of milk between these compartments have been well documented. In dairy cows, less than 30% of the total milk volume is typically stored in the cisternal region [3], whereas in dairy goats, this figure ranges between 57% and 88% [4]. Among sheep, the proportion exceeds 50% in dairy breeds [5,6] but falls

below 30% in meat breeds [7], following conventional milking intervals of 8 to 16 hours.

The compositional characteristics of milk are significantly influenced by its site of storage within the udder. Numerous studies have indicated that the relative distribution of cisternal and alveolar milk fractions fluctuates depending on the milking interval [3,8]. The hormone oxytocin (OTH) plays a pivotal role in facilitating milk ejection, primarily by inducing the contraction of myoepithelial cells encasing the alveoli, thereby promoting the transfer of alveolar milk into the cistern [9,10]. An understanding of how lactation stage and parity affect the volume of the cisternal compartment is essential, particularly in light of the milk yield

losses that may occur during the interval between milk ejection and complete evacuation. Prolonging milking intervals has been proposed as a strategy to enhance the efficiency of milk removal and to stimulate additional milk synthesis.

Accordingly, the present study aims to: (1) examine morphological changes in the size of udder cisterns and alveoli across different lactation stages and parities, and (2) evaluate the compositional differences in milk collected from the alveolar and cisternal compartments in crossbred dairy cows.

## 2. Materials and Methods

### 2.1 Animals and Management

This study was conducted at the University of Khartoum Educational Farm using twenty-eight crossbred dairy cows maintained under uniform nutritional and managerial conditions. The experimental cohort included cows at varying parities: twelve primiparous cows (six in their first lactation and six in their second), and sixteen multiparous cows (eight in their sixth lactation and eight in their seventh). All animals were categorized into three lactation stages: early (1–3 months postpartum,  $n = 28$ ), mid (4–6 months,  $n = 28$ ), and late (7–9 months,  $n = 28$ ). The observational period spanned 11 months, from January to November 2022.

### 2.2 Experimental Procedures

A completely randomized design (CRD) was employed to assess the effects of parity and lactation stage. Measurements of cisternal and alveolar milk volumes were performed at 12:00 PM, approximately 12 hours after the morning milking session, to ensure clear differentiation between milk compartments [11,12]. To inhibit premature milk ejection and thereby avoid overestimation of cisternal milk volume, cows were intravenously administered Atosiban at a dosage of 10 µg/kg body weight, an oxytocin receptor antagonist, in line with the protocol described by [14].

Cisternal milk was subsequently extracted from each quarter using a sterile teat cannula (10 cm in length), and its volume was recorded. Ten minutes following the cisternal milk removal, each cow received an intravenous injection of oxytocin (10 IU) to negate the effects of Atosiban and stimulate the ejection of alveolar milk. Alveolar milk was then manually collected and measured. For each udder quarter, milk fractions were weighed individually, and a 50 mL representative sample was collected and preserved for laboratory analysis.

### 2.3 Milk Composition Analysis

Milk samples were transferred to the Dairy Laboratory of the Department of Dairy Production, College of Animal Production (Shambat), where they were analyzed using a Milko Scan ultrasonic analyzer. Prior to testing, the 50 mL milk samples were equilibrated to room temperature and homogenized to prevent stratification of milk fat. The analyzer provided digital readings for key compositional parameters, including fat, protein, lactose, solids-not-fat (SNF), and density.

### 2.4 Statistical analysis

Data were subjected to analysis using the General Linear Model (GLM) framework as specified below:

$$Y_{ijk} = \mu + a_i + b_j + \varepsilon_{ijk}$$

where  $\mu$  represents the general mean,  $a_i$  is the effect of parity (primiparous or multiparous),  $b_j$  is the influence of stage lactation (early, mid, and late), and  $\varepsilon_{ijk}$  is the random error term. Differences between treatment means were evaluated using Duncan's multiple range test, with statistical significance determined at  $P < 0.05$  [15].

## 3. Results and Discussion

The findings of this study revealed no statistically significant effect of parity ( $P > 0.05$ ) on cisternal size, cisternal milk percentage, alveolar size, or alveolar milk percentage in crossbred dairy cows (Table 1). On average, cisternal milk constituted approximately 31.35–31.67% of total udder milk, regardless of parity.

These results align with earlier studies reporting minimal influence of parity on cisternal milk yield and its proportional contribution to total udder milk. For instance, [15] observed that cisternal milk fractions tend to be lower in primiparous cows, while multiparous cows typically exhibit increased cisternal volumes and yield. Similarly, [16] documented that

primiparous cows demonstrated reduced cisternal milk output and proportion, corroborating the present study's findings.

The physiology underlying these observations may be attributed to the structural development of the udder over successive lactations. Cows with relatively smaller cisternal capacity experience faster filling, which increases intramammary pressure. This, in turn, can exert a suppressive effect on alveolar milk secretion, as reported by [17]. Consequently, cows with smaller cisterns often exhibit a more pronounced milk yield response to increased milking frequency (e.g., thrice-daily milking) than those with larger cisternal reservoirs [18], likely due to the alleviation of inhibitory pressure on the alveolar tissue.

**Table 1:** Effect of parity on cisternal and alveolar milk volumes and composition in crossbred dairy cows.

Traits	Cistern		Alveolar	
	Multiparous	Primiparous	Multiparous	Primiparous
<b>Milk production, (L/AM)</b>				
Milk volume-12h, L	1.74±0.05 <sup>a</sup>	1.42±0.05 <sup>b</sup>	3.75±0.09 <sup>a</sup>	3.08±0.10 <sup>a</sup>
Milk volume-12h, %*	31.64 <sup>b</sup>	31.67 <sup>b</sup>	68.36 <sup>a</sup>	68.38 <sup>a</sup>
<b>Milk compositions (%)</b>				
<b>Fat</b>	2.89±0.27 <sup>b</sup>	2.54±0.23 <sup>b</sup>	7.04±0.50 <sup>a</sup>	7.27±0.43 <sup>a</sup>
<b>Protein</b>	3.69±0.11	3.56±0.09	3.22±0.06	3.64±0.05
<b>Lactose</b>	5.03±0.08	4.97±0.07	4.48±0.09	4.54±0.08
<b>SNF</b>	9.39±0.16	9.24±0.14	8.67±0.20	8.49±0.18
<b>Density</b>	33.54±0.56 <sup>a</sup>	33.36±0.48 <sup>a</sup>	28.68±0.88 <sup>b</sup>	27.66±0.76 <sup>b</sup>

<sup>a,b</sup> Means in the same row within each period carrying different superscripts differ ( $P < 0.05$ ). \*Percentage values transformed to arcsine before statistical analysis. SNF: non-fat souled.

The results of this study demonstrated that parity did not exert a statistically significant effect ( $P > 0.05$ ) on the relative distribution of cisternal and alveolar milk volumes in crossbred dairy cows (Table 1). On average, cisternal milk comprised approximately 31.64–31.67% of total udder milk, while alveolar milk accounted for 68.36–68.38%, irrespective of parity. However, multiparous cows produced significantly greater volumes of both cisternal and alveolar milk compared to primiparous cows, indicating enhanced secretory capacity and udder development in later lactations.

A positive correlation was observed between cisternal milk volume and total milk yield ( $P < 0.05$ ), suggesting that cisternal size may serve as a predictive marker for overall milk production potential in crossbred cows. This aligns with earlier reports which indicate that cows with larger cisternal capacity produce more milk, milk faster, and adapt better to extended milking intervals [3, 23].

Regarding milk composition, fat content was significantly higher in alveolar milk than in cisternal milk across both parity groups ( $P < 0.05$ ), with alveolar compartments accounting for up to 70% of total milk fat in multiparous cows and 72% in primiparous cows. These findings are consistent with prior research in dairy cows [19] and ewes [5], and can be explained by the physicochemical properties of milk fat. Specifically, the alveolar compartment tends to retain a larger proportion of milk fat due to the higher viscosity and larger size of fat globules [21]. A similar pattern has been reported in dromedary camels [20], as well as in Najdi and Naeimi sheep, where alveolar milk fat concentrations were 41% and 37% higher, respectively, than in the cisternal fraction [22].

Cisternal milk also exhibited significantly higher density values compared to alveolar milk in both primiparous and multiparous cows, likely reflecting differences in component concentration and distribution. This difference has practical implications for milk processing and storage stability.

In contrast, protein, lactose, and solids-not-fat (SNF) contents did not significantly differ ( $P > 0.05$ ) between the cisternal and alveolar fractions. This uniformity in composition supports the notion that these components are more homogeneously distributed within the colloidal phase of milk and less influenced by anatomical compartmentalization [20, 26]. Similar patterns have been observed in both cows and sheep, where protein levels

remained stable following a 12-hour milking interval, and protein yield changes were predominantly associated with overall milk volume [5, 21, 24, 25].

Collectively, these findings emphasize that while parity influences the absolute volumes of milk produced, it does not significantly alter the proportional or compositional characteristics between udder compartments, apart from fat content. Moreover, the data reinforce the relevance of cisternal volume as an indicator of milk yield efficiency and its potential utility in selective breeding for improved dairy traits

Lactation stage had a statistically significant effect on both the absolute volumes and the relative proportions of cisternal and alveolar milk in crossbred cows (Table 2). Total milk yield over a 12-hour interval declined progressively from early ( $5.54 \pm 0.08$  L) to mid ( $5.10 \pm 0.07$  L) and late lactation ( $4.61 \pm 0.04$  L). Alveolar milk volume decreased by 18% between early and late lactation ( $P < 0.05$ ), while cisternal milk volume remained relatively stable between early and mid-lactation but declined by 16% from mid to late lactation ( $P < 0.05$ ). These shifts corroborate prior findings that show a marked decline in milk volume stored in the cistern as lactation advances [19, 27].

**Table 2:** Effect of lactation stage on the cisternal and alveolar milk volumes in crossbred cows.

Traits	Milk volume-12h, L			Milk volume-12h, %*	
	Cisternal	Alveolar	Total	Cisternal	Alveolar
Early 1-3 months	1.76±0.05 <sup>a</sup>	3.78±0.07 <sup>a</sup>	5.54±0.08 <sup>a</sup>	31.89 <sup>b</sup>	68.09 <sup>b</sup>
Mid 4-6 months	1.74±0.04 <sup>a</sup>	3.36±0.05 <sup>b</sup>	5.10±0.07 <sup>b</sup>	34.24 <sup>a</sup>	65.76 <sup>c</sup>
Late 7- end of drying	1.31±0.02 <sup>b</sup>	3.30±0.04 <sup>b</sup>	4.61±0.04 <sup>c</sup>	28.79 <sup>c</sup>	71.20 <sup>a</sup>

\* Percentage values transformed to arcsine before statistical analysis.

<sup>a,b</sup> Means in the same Column within each period carrying different superscripts differ ( $p < 0.05$ ).

Notably, cisternal milk as a percentage of total udder milk increased from 31.89% in early lactation to 34.24% in mid-lactation, before decreasing sharply to 28.79% in late lactation. This temporal pattern aligns with observations in dairy cows and sheep, where cisternal milk proportions generally range from 20% to 35% during a 12-hour milking interval [4, 15, 19]. It contrasts with the findings of [3], who reported a lower cisternal proportion of 17% under similar conditions. Conversely, [1] reported an incremental increase in cisternal proportion over the course of lactation, attributing this to a sharper decline in alveolar milk.

The reduction in both cisternal and alveolar milk volumes during late lactation can be attributed to physiological changes, particularly the apoptosis of secretory epithelial cells, which reduces the capacity for milk synthesis [16, 28]. Similar reductions in cisternal milk storage have been documented in dairy sheep using ultrasonography, affirming this pattern across species [6].

These findings emphasize the dynamic interplay between lactation stage and udder compartmental storage. The temporary increase in cisternal proportion during mid-lactation may reflect adaptive responses in milk storage mechanisms, possibly to compensate for declining alveolar activity. As lactation progresses, the udder undergoes structural and functional regression, leading to an overall reduction in secretory capacity and milk yield. Understanding these shifts is essential for optimizing milking schedules and assessing mammary gland health across lactation.

#### 4. Conclusion

This study found that cisternal milk constituted roughly 31–32% of total udder milk in crossbred cows, with no significant difference due to parity. However, cisternal volume was positively associated with total milk yield ( $P < 0.05$ ), indicating its potential as a marker of productivity. Fat content was predominantly concentrated in the alveolar compartment, accounting for up to 70% in multiparous and 72% in primiparous cows. This confirms the alveolar region's primary role in fat storage. As lactation progressed, both alveolar and cisternal milk volumes declined significantly. Cisternal milk proportion peaked during mid-lactation but dropped in late lactation, whereas the alveolar percentage increased. Alveolar milk volume decreased by 18% from early to late lactation. Cisternal volume remained stable between early and mid-lactation but

decreased by 16% between mid- and late-lactation stages. Overall, these results highlight the interplay between lactation stage, udder morphology, and milk composition, suggesting that management and breeding strategies should take these dynamics into account to optimize yield.

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#### Author Contributions

Conceptualization, methodology, validation, formal analysis, and investigation were carried out by Al-Hayani A.A. The original draft was prepared by Al-Hayani A.A., with review and editing contributions from Al-Hayani A.A., Abu Nihaila A.M., and Tarig A.A. Visualization was led by Al-Hayani A.A., under the supervision of Abu Nihaila A.

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#### Conflict of Interest Declaration

The authors declare that there are no conflicts of interest regarding the publication of this paper.

#### Ethical standards

Not applicable.

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