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Review

Milk Properties and Morphological Characteristics of the Donkey Mammary Gland for Development of an Adopted Milking Machine—A Review

Shehadeh Kaskous [1,*] and Michael W. Pfaffl [2]

1 Department of Research and Development, Siliconform, Schelmengriesstrasse 1, 86842 Turkheim, Germany
2 Department of Animal Physiology and Immunology, TUM School of Life Sciences, Technical University of Munich, Weihenstephan, Weihenstephaner Berg 3, 85354 Freising, Germany; michael.pfaffl@tum.de
* Correspondence: skaskous@siliconform.com

Abstract: Donkey milk (DM) has been known in the world for 5000 years for its benefits for human nutrition and health. Nowadays, DM has become more and more attractive as a commercial product. DM contains several physiologically functional components, including high-quality whey proteins, vitamins, important minerals, unsaturated fatty acid and bioactive components. Therefore, it is not only consumed as food but also as a remedy. The average daily milk yield of a female donkey over the entire lactation season was 1.57 ± 1.12 kg/day and fluctuated between 0.20 and 6.00 kg/day. Average milk concentrations (±SD) of fat, protein, lactose, total solids and ash in DM were 0.63 ± 0.41%, 1.71 ± 0.24%, 6.34 ± 0.37%, 9.11 ± 0.95% and 0.39 ± 0.04%, respectively. Interestingly, DM is similar in composition to mare's milk, and both are similar to mother's milk. The anatomical and morphological properties of the mammary gland of the female donkey are special and can be compared with those of mare udders. However, the cistern cavity of the mammary gland of female donkeys is characterized by the presence of multiple pockets that open directly into the teat, instead of a single cistern cavity. Therefore, the mammary gland capacity in donkey mare is low and milking technique and routine are of most importance. So far there is no special milking machine for female donkeys and mares. The milking machines used nowadays were initially designed for smaller sheep and goat udders. The company Siliconform, Germany, has set itself the task of developing an optimized milking machine for donkey mares, which is adapted to the anatomical and morphological properties of the donkey mammary gland. Furthermore, it should achieve a physiologically ideal milking process meeting high animal welfare standards for increased milk production with high quality standards.

Keywords: donkey milk; milk yield; milk composition; morphology; milking machine

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1. Introduction

Donkey milk (DM) is produced by the female animals of the house donkey (*Equus asinus*). Compared to cows, donkey mares give low amounts of milk [1,2], which initially serves as food for the donkey offspring. The small but valuable milk of the female donkey is particularly noticeable in the prices of the products. However, donkey milk production has recently become more and more an issue [3–5].

Donkey mares have been known for thousands of years for their valuable milk [6,7]. Thus, DM has been known since the time of ancient Egypt [2,6]. Hippocrates (460–370 BC) was the first to write about the medicinal properties of DM [8]. Furthermore, Cleopatra, the former queen of Egypt, bathed in the milk of female donkeys to preserve the beauty and youth of her skin [9]. In ancient Rome, DM was also used for cosmetic purposes [10]. In the 19th century, the French Dr. Parrot breast-fed orphaned babies directly on female donkeys, as DM is most similar to human breast milk [11]. DM was then sold well into the late 20th century to feed and heal orphaned infants and cure weak and sick children, as well as the elderly [7]. Much research has been carried out on DM in recent years, particularly with regard to its properties and chemical composition [12–18]. However, there is a lack of information on the morphological properties of donkey udder anatomy and the use of suitable milking machines to increase milk yield and to meet donkey milk requirements. Therefore, the aim of this study was to provide information about milk properties and morphological characteristics of the donkey mammary gland for the development of an adapted milking machine.

2. Milk Yield of Female Donkey

The average daily milk yield over the entire lactation season was 1.57 ± 1.12 kg/day and fluctuated between 0.20 and 6.00 kg/day (Table 1). However, the daily milk yield in donkeys is influenced by many factors, such as breed, management of dams and foals, feeding, stage of lactation, lactation number, foaling season, milking procedure, number of milkings per day, milkability degree of donkey mares and production country [1,17,19–22]. During lactation season, the milk yield of the Italian Ragusana breed steadily decreased from 1.98 to 1.28 kg/day [21], whereas Salimei et al. [20] reported that the milk yield per milking did not vary significantly during lactation season. Muhatai et al. [23] reported that during lactation season the daily milk yield decreased from 3.30 kg/day at the start of the lactation to 2.20 kg/day by the end of the lactation season. Based on the observations of Doreau et al. [19] it is assumed that the diet plays a relevant role in the milk yield and quality of female donkeys, while Salimei et al. [24] found no significant differences in milk yield and ingredients in two feed rations. It is noteworthy that the daily milk yield of donkey mares, the length of lactation season and the milk characteristics varied depending on the foaling season [21]. However, donkeys foaling in winter and in summer produced more milk than those foaling in spring and autumn [21]. Moreover, studies have shown that the average amount of milk yield for morning milking was statistically lower than that for afternoon milking (549.2 mL vs. 949.3 mL) [20]. In addition, it was found that parity affects the daily milk yield and parity 3 donkeys produced 22% more milk than parity 4 donkeys (3.30 vs. 2.70 kg/day) [23].

Table 1. Average daily milk yield of the donkey mare during the lactation season.

Location	Breed or Type	Daily Milk Yield (kg/day)	Range of Milk Yield (kg/day)	Reference
Africa	Local type	1.50	*	[6]
China	Jiangyue	3.00 (4 milkings/day)	2.20–3.30	[23]
China	Jiangyue	1.28 ± 0.17	1.16–1.54	[1]
Croatia	Littoral Dinaric	0.17 (1 milking/day)	*	[25]
Croatia	Istrian	0.75 ± 0.30 (1 milking/day)	*	[26]
	Littoral Dinaric	0.32 ± 0.15 (1 milking/day)	*	[26]
India	Local type	1.00	*	[7]
Italy	Local breeds	1.36 ± 0.51	*	[27]
Italy	Ragusana	1.64 ± 0.79 (2 milkings/day)	0.38–3.30	[17]
Italy	Ragusana	1.70 ± 0.10 (2 milkings/day)	1.28–1.98	[21]
Italy	Ragusana	1.71 ± 0.66	1.25–2.35	[14]
Italy	Amiata	0.70 ± 0.10	0.52–0.76	[28]
Italy	Martina Franca	1.50 ± 0.08	1.27–1.47	[24]

Table 1. *Cont.*

Location	Breed or Type	Daily Milk Yield (kg/day)	Range of Milk Yield (kg/day)	Reference
Italy	Martina Franca and Ragusana	1.48 ± 0.06	1.30–1.80	[20]
Italy	Ragusana	4.87 ± 0.12 (8 milkings/day)	*	[29]
Italy	Different races	2.68 ± 1.96	0.72–6.00	[22]
Jordan	Local type	1.00	*	[30]
Mean ± SD		1.57 ± 1.12		

SD: standard deviation; * not specified.

D'Alessandro and Martemucci [31] reported that the daily milking management influences milk yield and udder health condition. Thus, the observed differences in milk yield can be attributed to the number of milkings per day and milking interval. However, milking three times a day with a 3 h interval showed a higher milk yield per milking compared to three times a day with a 2 h interval ($p < 0.01$). In addition, the milk yield per milking increased with an increase from one to three milkings per day, but the values for this variable fluctuated strongly from 40 to 1.640 mL/milking (Table 7).

Furthermore, no differences in milk yield were found between the right and left udder halves of the female donkey, while the effects of lactation day and the individual were significant ($p < 0.01$) [31].

3. Lactation Curve in the Female Donkey

Following the daily milkings shows a typical development of milk production from birth to the end of lactation. The course of lactation can be represented graphically as a curve for a donkey mare [31] (Figure 1).

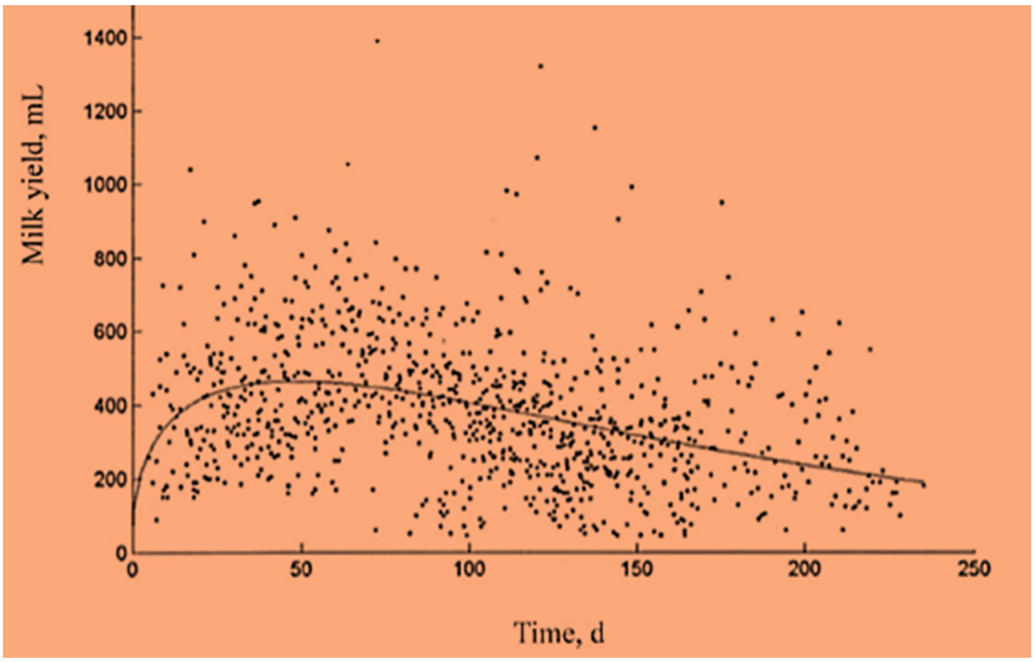

Figure 1. Lactation curve from milk yield data (morning milk) in donkey mares (n = 12) with regression [31], by application of Wood's model (Wood, 1967): $Y_t = a \times t^b \times e^{-ct}$ ($r^2 = 0.11$), where Y is milk yield on day t of lactation, a is the constant level of initial yield, b is the rate of increase to peak, e is the mathematical constant, c is the rate of decline after peak and peak in days (t) is given by b/c.

According to the results shown in Figure 1, the lactation peak was reached after 48 days. However, most female donkeys (42%) showed their maximum milk yield between 64 and 73 days of lactation. Likewise, milk yield decreased progressively and significantly ($p < 0.01$) until the end of lactation, as shown in the figure (Figure 1). According to Malacarne et al. [17], the daily milk yield in donkey mare was around 2 kg/day for up to 160 days and then gradually decreased to 0.13 kg/day at the end of lactation (250–279 days). The effect of the mechanical milking management was also examined, and it was found that the milking management had no significant influence on the total milk yield in the entire lactation [32].

4. Physiochemical Properties of Donkey Milk

DM composition has been studied in different parts of the world [1,17,18,33–35]. However, data from the literature indicated ranges of variation in DM composition. The mean values of the DM composition are given in Table 2.

Table 2. Physicochemical composition parameters of donkey milk.

Fat (%)	Protein (%)	Casein (%)	Lactose (%)	Dry Matter (%)	Ash (%)	pH	Reference
0.16	1.40	*	*	8.74	*	7.03–7.06	[34]
0.28 ± 0.05	1.59 ± 0.02	0.72	6.73 ± 0.06	8.91 ± 0.03	0.32 ± 0.02	7.01 ± 0.01	[36]
1.16	1.57	0.82	6.33	9.53	0.40	7.18	[1]
0.44 ± 0.10	1.9 ± 0.02	0.88	6.40 ± 0.10	*	*	*	[21]
0.33	1.55	*	6.28	8.80	*	*	[25]
0.49 ± 0.02	2.03 ± 0.15	*	5.70 ± 0.14	10.71 ± 0.64	0.42 ± 0.02	7.16 ± 0.02	[37]
1.20	2.14	0.88	5.90	7.50	*	*	[38]
0.16	1.34	0.56	6.07	8.19	0.36	*	[17]
1.21	1.74	*	6.23	9.60	0.43	*	[39]
0.30–1.80	1.50–1.80	0.78	5.80–7.00	8.80–11.70	*	7.00–7.20	[7]
0.38	1.72	0.87	6.88	8.84	0.39	*	[20]
0.67 ± 0.55	1.88	0.79	6.84 ± 0.15	*	0.41	7.20	[35]
0.63 ± 0.41	1.71 ± 0.24	0.79 ± 0.11	6.34 ± 0.37	9.11 ± 0.95	0.39 ± 0.04	7.12 ± 0.08	Mean ± SD

SD: standard deviation; * not specified.

DM is characterized by little fat, protein and casein and high levels of lactose and whey proteins and is rich in lysozyme [1,20,36,40]. DM composition was found to be less than other species such as bovine milk [20]. However, variations observed in DM composition could be attributed to several factors such as breeds or type, stage of lactation, age of animals, lactation number, season and country [41]. Breed and seasonal variation were found to be the most effective factors in DM composition (Table 3). It is noteworthy that three hours was described as an ideal milking interval for obtaining the best organoleptic characteristics of DM, while an interval of 5 to 8 h between milking times resulted in a decrease in milk fat and lactose content [42].

Table 3. Effect of the production season on the main qualitative variables [21].

Season	Daily Milk Yield * (kg/day)	Fat (%)	Protein (%)	Lactose (%)	SCC ** (log 10 n × 10^3/mL)
Spring	1.85 ^{a***}	0.39 ^{ab}	1.93 ^{b}	6.65 ^{a}	5.35 ^{a}
Summer	1.44 ^{b}	0.23 ^{b}	1.81 ^{ab}	6.05 ^{c}	24.05 ^{b}
Autumn	1.41 ^{b}	0.38 ^{ab}	1.78 ^{a}	6.34 ^{bc}	9.210 ^{a}
Winter	1.47 ^{b}	0.50 ^{a}	1.92 ^{b}	6.50 ^{ab}	24.60 ^{b}

* Daily milk yield (kg/day): the daily milk yield was achieved by mechanical milking twice a day with an inter-milking interval of 5 h. ** SCC: somatic cell count. *** Means in the same column and same parameter with different superscripts a, b, c are significantly different at $p < 0.05$.

4.1. Fat Content

The mean content of fat in DM during the lactation season was 0.63 ± 0.41% (Table 2), and fat content ranged between 0.16 and 1.21% [1,17,20,37,43], with an increasing, non-linear, trend from partum to the end of lactation [1,20]. However, the fat content of DM depends on many factors such as stage of lactation, lactation season, feed, country, breed or type, milking methods [34,44]. Malacarne et al. [17] reported a very low fat content (0.16 ± 0.06 g/100 g milk) in Italian Ragusana DM. Ivankovic et al. [25] also observed that the fat content of DM was low (0.33%). Giosue et al. [21] indicated that the season had an influence on the milk fat content in DM, as the female donkeys foaled in spring had 0.28% of fat in DM, while the milk of those foaled in autumn had 0.52% of fat.

4.2. Protein Content

The mean value of protein content in DM was 1.71 ± 0.24% (Table 2), and protein content ranged between 1.34 and 2.20% [20,21,25,33]. This protein concentration in DM was similar to that of breast milk and much lower than that of cow, goat or sheep milk [1]. Compared to cow milk, DM is characterized by low casein (42–52% of total protein) and high whey protein content (48–58% of total protein) [1,17,36,38]. However, the casein to whey protein ratio of 52:48 was between the lower value of human milk and the higher value of cow milk [1]. A specific protein profile with the following DM whey proteins was also found in DM: 4.48% lactoferrin, 6.18% serum albumin, 29.85% β-lactoglobulin, 21.03% lysozyme and 22.56% α-lactalbumin [20]. It has also been found that the protein content of DM decreases significantly during lactation [20,34]. This may be due to the differential expression of milk protein synthesis genes during the lactation period [1,34]. On the other hand, the protein content in the milk produced by female donkeys foaling in winter and summer was greater than the ones foaling in autumn and in spring [21]. Furthermore, Guo et al. [1] reported that DM had higher levels of amino acid such as serine (6.2%), glutamic acid (22.8%), arginine (4.6%) and valine (6.5%) and a lower level of cystine (0.4%).

4.3. Lactose Content

The mean value of lactose content in DM was 6.34 ± 0.37% (Table 2) which is higher than that of cow milk and very similar that of to breast milk [1,33]. However, the average values of lactose were 7.00%, 4.90% and 6.00% in DM, cow milk and human breast milk, respectively [16,45]. The lactose content of DM showed a progressive lowering from the beginning of lactation (6.30 g/100 g) to the end of lactation (5.70 g/100 g) [17]. On the other hand, the concentration of lactose in DM was nearly constant throughout the lactation season [1], since the lactose content of the milk plays an important role in the osmotic pressure of the mammary gland, which is equal to that of blood. However, due to differences in the season and donkey feeding conditions, slight changes in the lactose content of DM were found in different studies [20,21,46,47].

4.4. Mineral Content

DM contains an average of 0.39 ± 0.04% minerals (Table 2), which differ depending on the breed and stage of lactation. Malacarne et al. [17] showed similar results, and the total ash content was 0.36% and it represented 4.4% of the dry matter. Thus, most of the minerals in DM vary significantly during lactation [17,48]. However, Ca, P, Mg, Na, Fe and Zn contents were affected by the days in milk, whereas no significant variations were reported for K, Cl and Cu [17]. It is noteworthy that the concentrations of minerals in DM are similar to those in breast milk [48]. However, the concentrations of K, Na, Fe, Zn and Mg in DM appear to be similar to those in breast milk [2]. Generally, DM is a rich source of various minerals such as Ca, P, Na, K, Mg, Fe, Zn and Cu [2,17,20].

4.5. Vitamin Content

In DM, the content of B-complex vitamins such as B1, B2, B3, B6 and B9 is higher than that in breast milk [2,49]. However, the B1 and B6 concentrations in DM were very high compared to those in breast milk, and the values were 0.66 and 5.38 µM/L vs. 0.12 and 0.48 µM/L, respectively [50,51] (Table 4). Likewise, B2 (0.17 µM/L), B3 (18.75 µM/L) and B9 (0.83 µM/L) concentrations in DM were also higher than the values reported in breast milk [50,52,53]. On the other hand, vitamin B12 was not detected in DM [54]. The lack of vitamin B12 in equid milk compared to ruminant milk could be explained by the different digestive systems among these two species, and vitamin B12 is synthesized by the microorganisms of the digestive tract. In addition, the vitamin C content in DM is very similar to that in breast milk but higher than that in cow's milk (Table 4) [2,49,55,56].

Table 4. Vitamin content in donkey milk [2,49,56,57].

Water-Soluble Vitamins		**Fat-Soluble Vitamins**	
Vitamin	**Concentration µM/L**	**Vitamin**	**Concentration µg/100 mL**
B1 (thiamine)	0.66	Vitamin A	58.00 µg/100 mL
B2 (riboflavin)	0.17	Vitamin D	2.23 µg/100 mL
B3 (niacin)	18.75	Vitamin E	5.20 µg/100 mL
B6 (pyridoxine)	5.38	Vitamin K	*
B9 (folic acid)	0.83		
B12 (cyanocobalamin)	*		
Vitamin C	57.00 **		

* Not detected, ** mg/L.

The vitamin A content in DM (58 µg/100 mL) is slightly lower compared to that in breast milk (60 µg/100 mL) [57], while the vitamin D content in DM is higher than the values found in the milk of many other mammals and in breast milk [45]. Compared to breast milk, DM has a very low vitamin E content, and the values were 237 and 5.2 µg/100 mL, respectively [57] (Table 4). It is noteworthy that vitamin K has never been detected in DM.

4.6. Total Solids

The mean content of dry matter in DM was 9.11 ± 0.95% (Table 2), and dry matter content ranged between 8 and 11%. Malacarne et al. [17] observed that the dry matter of DM showed a progressive lowering from the beginning of lactation (8.60 g/100 g) to the end of lactation (7.7 g/100 g), whereas Salimei et al. [20] reported that day of lactation and other investigated factors of variability, breed, year of lactation and milking times, did not influence the dry matter content of the DM.

4.7. The pH Values

The average pH value of DM was 7.12 ± 0.08 (Table 2), which is slightly higher than cow milk (6.6) [1,34]. However, the high pH value in DM compared to cow milk is due to the lower casein and phosphate content in DM than in cow's milk [20]. Further studies have reported that the pH value was not influenced by breed or stage of lactation [1,36,40].

4.8. Somatic Cell Count (SCC) in Donkey Milk

According to the current state of knowledge, international reference values for normal physiological SCC in DM have not been determined yet, whereas these values have long been known for cow's milk [58]. Pilla et al. [59], Sarno et al. [60] and Tavsanli et al. [61] observed that the estimated SCC values were always below 50×10^3 cells/mL in DM. On the other hand, it was observed by Beghelli et al. [9] that the mean values of the SCC in DM were below 100×10^3 cells/mL milk. Ivankovic et al. [25] found that the average SCC in DM was 12.5×10^3 cells/mL during the lactation season (150 days). Malissiova et al. [62] reported that SCC was between 5×10^3 and 13×10^3 cells/mL in 90 raw donkey milk samples. Based on the observations of Fantuz et al. [63], it was found that a low SCC (7.3×10^3 cells/mL) and a low total bacterial count (3.4×10^3 CFU/mL) were detected in bulk milk from female donkeys. Furthermore, investigations in some farms in Italy clearly showed that the SCC in DM was $22.9 \times 10^3 \pm 34.8 \times 10^3$ cells/mL and the bacterial count was $134.2 \times 10^3 \pm 229.4 \times 10^3$ CFU/mL [27].

It was also detected that the stage of lactation did not significantly influence the SCC in DM. However, the highest SCCs were registered at the beginning and at the end of lactation [9]. Tavsanli et al. [61] have shown that SCC is linked to udder health and hygiene. That is why SCC is a very important criterion for safe raw milk production. Pilla et al. [59] suggest that DM could be safe food if the mammary gland is healthy and the animals are milked in proper hygienic conditions. In addition, an investigation of DM identified four major somatic cell types: lymphocytes, macrophages, polymorphonuclear neutrophils and epithelial cells (Table 5) [9].

Table 5. Mean values of SCC and differential fraction of SCC during three stages of lactation [9].

Stage of Lactation	SCC × 1000/mL Milk	Lymphocytes %	Macrophages %	PMN %	Epithelial Cells %
1	43.400	10.0	21.5	18.0	50.6
2	14.095	17.0	19.4	46.6	17.7
3	25.875	12.0	12.5	54.0	22.5

5. Morphological Properties of the Mammary Gland in Female Donkey

The mammary gland in female donkeys, as in other dairy animals, is an exocrine gland and is morphologically greatly altered. The donkey's lactating mammary gland comprises one pair of mammas, each with a teat. However, each mamma is usually drained by two independent mammary units, although three may rarely occur. Thus, each teat has two orifices through which the main ducts discharge [3]. This means that the donkey's mammary gland possesses at least four (2 × 2) independent milk units. The synthesized milk accumulates in the udder of female donkeys between the events of milk extraction by suckling or milking and is stored within two compartments: the cistern and the alveoli (Figure 2) [64].

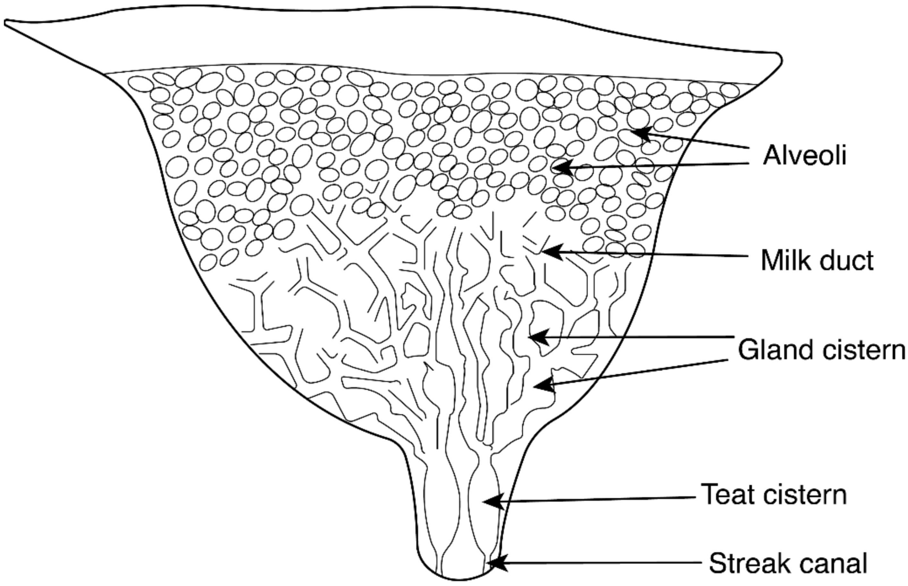

Figure 2. Schematic representation of a half udder of a donkey mare (own drawing Kaskous, S., 2022). Between the events of milk removal by suckling or milking, milk accumulates in the udder and is stored within two compartments: the cistern (including teat and gland cistern and in large and medium milk ducts) and the alveoli (alveoli and small milk ducts). The udder cistern of donkey mare is very small and consists of several pockets that open directly into the teat, rather than a single cistern cavity.

It is noteworthy that the cistern cavity of the mammary gland of donkeys is characterized by the presence of multiple pockets that open directly into the teat, instead of a single cistern cavity (Figure 2) [3]. Moreover, compared to ruminants, equids have a different structure and a low storage capability for their udders [6], and mares [65] and donkeys [6] are more adapted to frequent milk removal (2 or 3 h after separation from the foal). Accordingly, the donkey mare has to be milked several times a day due to the relatively small amount of milk per milking [66,67]. Thus, the udder cistern of a donkey mare has a very small volume; therefore, only a small cisternal fraction of milk is available, and the alveolar fraction of milk is large (70–85%) [67].

Since the milk storage capacity of this species is low, milk production depends on it being removed from the mammary gland by milking or suckling [68,69]. On the contrary, it was observed that an increase in milk yield by extending the milking interval from 3 to 8 h is due to the expandability of the cistern, which leads to a compensatory effect with regard to the decrease in milk synthesis [31]. Long intervals between milking events may cause a rise in intra-udder pressure, inducing early cessation of glandular activity, due to the udder size and its low storage capacity [29]. It was found that 70–85% of secreted milk is stored in the alveoli [70] and can be drawn only if milk ejection occurs [3]. The milk ejection reflex can be triggered if the female donkey always has visual contact with her foal [6,71]. However, milking donkeys by humans is easier to manage for the safety of humans and animals and for optimal milk production when foals are not physically present [72]. Furthermore, studies have shown that the udder shape of the female donkey was classified as 89% “bowl” and 11% “globular”, while the teat shape was classified as 78% “conical” and 22% “cylindrical” [3]. That means the predominant shapes are bowl for the udder and conical for the teat. Further measurements were also carried out on the donkey’s udder (Table 6).

Table 6. Measurement of udder and teat properties before and after machine milking in female donkeys [3].

Parameters	Before Milking	After Milking	Significance Level
	Udder (cm)		
Length	16.7 ± 1.7	13.8 ± 1.2	**
Width	11.8 ± 1.8	9.4 ± 1.4	**
Depth	11.2 ± 1.6	9.7 ± 1.6	**
	Teats (cm)		
Length	4.6 ± 1.0	3.9 ± 1.0	ns
Diameter at the base	3.3 ± 0.7	2.4 ± 0.8	*
Diameter at the apex	2.3 ± 0.5	1.9 ± 0.5	*
Teat distance at the base	5.7 ± 0.8	4.2 ± 0.8	**
Teat distance at the apex	6.6 ± 1.0	5.3 ± 1.0	**

ns: not significant, * $p < 0.05$, ** $p < 0.01$.

In fact, the morphological features of the udder and teat of lactating donkeys are considered to be fundamental characteristics for milk production and milking efficiency [73].

6. The Use of the Milking Machine for Donkey Mares

The history of mechanical milk extraction of donkey mares is not very old, as many owners have milked their donkeys by hand and found that the use of milking machines would be traumatic for the donkey mares [6]. On the contrary, donkey mares in Italy were milked by machine for years, although the foals were always there during milking. However, such studies have reported that after a training period, daily milking was performed with a modified mechanical sheep milking machine in the presence of the foal [20,24,31]. However, the presence of the foal during milking does not affect the milk yield of donkeys that are adapted to the milking process [74]. The milking machine used was set with the following operating parameters: vacuum level 42 kPa, pulse ratio 50% and pulse rate 120 cycles/min. Furthermore, D'Alessandro et al. [3] reported that the pulsation rate of 120 cycles/min resulted in a higher daily milk yield in comparison with the pulsation rates of 90 and 150 cycles/min, reduced the residual milk fraction and thus improved milking efficiency. In addition, due to the rapid milk ejection of 40 to 90 s per milking process, the use of a pulsation rate (120 cycles/min) is recommended in order to avoid both liner backflow and milk stress [75]. An interesting aspect is reported in a field study [76] in which the authors measured milk flow curves and milk production in lactating donkey mares to assess the milkability after using the milking machine set at a vacuum level of 42 kPa, 60 cycles/min and pulsation ratio 60:40. The observed flow curves were characterized by a short plateau phase (0.08 ± 0.13 min), a long increase phase (0.47 ± 0.27 min) and a long decrease phase (0.54 ± 0.37 min), while the total milking time was 1.75 ± 0.49 min.

Moreover, it is also noted that many Italian farmers used milking machines modified for goats [27]. Salimei [10] reported that while manual milking can be just as efficient as machine milking in terms of the amount of milk extracted per milking, machine milking donkeys produce lower fluctuations, and the risk of contamination is also less compared to manual milking [77].

The latest information showed that some farmers are using simple milking machines for donkey mares (Figure 3) [30]. However, we have no further information about the effectiveness of these milking machines and whether problems with udder health can occur if they are used for a long time.

Figure 3. Donkey mare during machine milking at a Jordanian donkey farm [30].

Table 7. Effects of daily milking number and interval between milkings on milk yield per milking in donkey mares [31].

Group	Milking		Milk Yield per Milking (mL)	
	Number/day	Interval (h)	Mean	Range
1	1	3	346.0 ± 7.2 [a]	40–1220
2	3	3	512.2 ± 7.6 [b]	50–1500
3	3	2	325.5 ± 10.9 [a]	40–1640
4	6	2	282.3 ± 7.7 [c]	40–1330

Mean values with different letters are significant ($p < 0.01$).

Under these conditions, the company Siliconform, Germany, set itself the task of developing a milking machine for donkey mares, as the company has been in milking technology for 50 years and has a great amount of experience with all lactating animals such as cows, sheep, goats and camels [78–83]. The company's aim is to develop a milking machine for female donkeys that is suitable for all teat shapes and anatomical measurements, needs a low vacuum to open the teat effectively and has the right kind of pulsation for a stable vacuum on the teat area during milking. Furthermore, the milking machine should have the right type of teat cup liner to quickly and completely extract the milk from the udder, trigger the milk ejection reflex without the presence of the foal during milking and provide the right pulsation rate and ratio to achieve an ideal milking process. The goal is to keep the animal welfare at a high level while meeting the milk quality standards.

7. Discussion

Donkey and horse milks are known for their nutritional and nutraceutical properties when compared to milk traditionally used for human consumption [22,84–87]. The realistic prospect of using this food in a balanced diet worldwide, especially for infants and the elderly, requires in-depth knowledge of the physicochemical properties of the milk produced and the possibility of increasing the milk yield per animal through machine milking. However, donkey and horse milks are produced and consumed in almost all European countries, as well as in Africa and Latin America [10,15,33,38,88,89]. It is noteworthy that horse mare's milk, like DM, is rich in whey proteins [20,40,85,88], polyunsaturated fatty acids such as linoleic acid C18:2 and linolenic acid C18:3 [20,40,90–92] and vitamin C [93,94]. Since horse and donkey milks contain a low level of casein and a high level of whey proteins, they are easier to digest for humans [95]. Furthermore, horse and donkey milks have a sweet taste because they contain a high quantity of lactose.

Horse and donkey milks, compared to milk from other dairy animals such cow, buffalo, goat and sheep, are most likely to resemble human breast milk [2,7,95,96].

In addition, various studies have reported that the average lysozyme concentration of DM was approximately 1.07 g/L, similar to that of horse milk (0.80–1.10 g/L) and human milk (0.30–1.10 g/L) and higher than that of cow, goat or sheep milk, in which negligible amounts of lysozyme are found [1,40,97–102]. The high lysozyme content may be the cause of the low bacterial count of donkey and horse milks and also makes these milks useful for preventing intestinal infections in infants [37,40]. Due to the significantly lower casein content, many people with a cow's milk protein allergy can tolerate donkey and horse milks, as they find that donkey and horse milks do not trigger an allergy reaction [88,103,104].

Furthermore, the mechanization of milking female donkeys and mares is necessary to improve milk productivity and quality [105,106]. Since the milk yield is closely related to the morphological characteristics of the mare's and donkey's udders, these must be taken into account in machine milking [107].

Our literature review clearly shows that donkey and horse keepers have major problems with milking machines because they do not use milking machines that are adapted to the morphological and physiological characteristics of the udders of female donkeys and horses. Salimei and Park [87] reported that the milking machine used for mares was a sheep milking machine with some modifications as for female donkeys. Therefore, the company Siliconform, Germany, has set itself the task of developing a milking machine for horses and donkeys.

8. Conclusions

- The average daily milk yield of a donkey mare is low and depends on many factors, particularly the breed, the number of milkings per day and the milkability of the donkey mare.
- Donkey milk is characterized by little fat, protein and casein and high levels of lactose and whey proteins and is rich in lysozyme.
- The cistern cavity of the mammary gland of donkeys is characterized by the presence of multiple pockets that open directly into the teat, instead of a single cistern cavity.
- The udder shape of the female donkey was classified as 89% "bowl" and 11% "globular", while the teat shape was classified as 78% "conical" and 22% "cylindrical".
- There is no donkey milking machine on the market. So far, adjusted but suboptimal sheep or goat milking machines have been used.
- The company Siliconform, Germany, set itself the task of developing a milking machine for donkeys. These specific milking machines take into account all the morphological, anatomical, physiological and economic aspects to allow fast and complete milk extraction and maintain good udder health.

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