



Nutritional Quality of Mare's Colostrum and Milk: Potential Impact on Blood Glucose Values in Fresh and Cold-stored Status in Mice

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10.18805/IJAR.BF-2036

ABSTRACT

Background: Mare's milk has been indicated to improve blood glucose value, liver and kidney functions.

Methods: Thirty male albino mice were distributed over three groups; control, mare's colostrum (fresh and cold-stored) and mare's milk (fresh and cold-stored). Chemical composition and immunoglobulin content of mare's milk and colostrum were determined through milkoscan and Brix refractometer, respectively. Body weights (BW), rectal temperature, pulse rate and peripheral oxygen saturation (SPO2), blood glucose values were recorded.

Result: The results showed that mare's colostrum had higher ($P < 0.0001$) SNF, protein and lactose but comparable fat contents compared to mare's milk. Consumption of *ad libitum* fresh mare's colostrum resulted in significant decrease in blood glucose values compared to fresh mare's milk. On the other hand, consumption of cold-stored mare colostrum and milk resulted in significant increase in blood glucose values compared to fresh state of colostrum, milk and control one. Milk group had higher body temperature compared to colostrum and control groups. Pulse rate and SPO2 values did not differ among groups. It could be concluded that fresh state of mare's colostrum gave hypoglycemic effect whereas cold-stored of mare's colostrum and milk resulted in hyperglycemic effects.

Key words: Cold-Stored, Colostrum, Fresh, Glucose, Mare, Milk.

INTRODUCTION

Cow's milk represents the vast majority of global milk production (Sheng and Fang, 2009; Claeys *et al.*, 2014), with mare's milk accounting for less than 0.1% (Faye and Konuspayeva, 2012). Despite this, mare or equine's milk has gained popularity in Europe and central Asia, expanding to several countries. Accurate worldwide production and consumption data are scarce, as much of it is likely consumed locally. Estimates range from 1-1.3 million liters in Europe to 8 million liters in Mongolia, highlighting the difficulty in quantifying its global usage (Minjigdorj and Austbø, 2009; Uniacke-Lowe and Fox, 2012).

Mare's colostrum is a highly specialized secretion providing essential immunoglobulins, concentrated nutrients and bioactive factors that support the immediate survival and development of newborn foals (Reiter and Reed 2023; Akköse 2025). The composition of colostrum changes rapidly after foaling, transitioning to mature milk within the first 24-48 hours (Derisoud *et al.*, 2023). The concentration of immunoglobulins and other bioactive factors decreases significantly during this period, while lactose content increases. The quality of colostrum, particularly its immunoglobulin concentration, is crucial for the foal's health. Factors such as the mare's age, health, nutrition and vaccination status can influence colostrum quality (Soufleri *et al.*, 2021; de Sobral *et al.*, 2023). It's important for foals to ingest an adequate amount of high-quality colostrum within the first few hours of birth

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How to cite this article: Mohammed, A.A., Shawky, M., Alyousef, Y., Almuyidi, A., Almarri, R., Alkhalifah, H., Alhamd, A. and Mohammed, M.A. (2025). Nutritional Quality of Mare's Colostrum and Milk: Potential Impact on Blood Glucose Values in Fresh and Cold-stored Status in Mice. *Indian Journal of Animal Research*. 1-6. doi: 10.18805/IJAR.BF-2036.

Submitted: 28-06-2025 **Accepted:** 25-08-2025 **Online:** 08-09-2025

to ensure proper transfer of passive immunity (Lopez-Rodriguez *et al.*, 2020; de Sobral *et al.*, 2023).

Furthermore, mare's milk has garnered attention recently due to its rich nutrient profile and potential health benefits. Notably, its chemical composition closely resembles human milk, making it a valuable alternative for infant feeding

and a dietary option for individuals with specific health needs (Bakshi *et al.*, 2023). Chemically, mare's milk consists of ~89.7% water, 2.3% protein and notable concentrations of ascorbic acid, phosphorus, potassium, magnesium and calcium, often surpassing levels found in other livestock milk (Musaev *et al.*, 2021; Faccia *et al.*, 2020). The growing market reflects this interest, with mare's milk appearing in various food and cosmetic products (Jastrzębska *et al.*, 2017). Mares continue to be milked extensively in central Asia, including Mongolia, Kazakhstan, Kyrgyzstan, Russia and China. This practice is essential to the food economies of these areas (Blanco-Doval *et al.*, 2024). Mare's colostrum and milk are still not well-known products, although it is part of the daily diet in Asian countries. In addition, it is worth noting that while mare's milk is valued for its potential benefits, it is not widely available and is generally more expensive than cow's milk. The question arose, whether mare's colostrum and milk are able to promote health status, in fresh and cold-stored states. Therefore, the aims of the current study were to investigate the nutritional quality of mare's colostrum and milk and their effects on blood glucose values and physiological parameters upon *ad libitum* consumption in fresh and cold-stored states in mice.

MATERIALS AND METHODS

The experimental procedures were approved by the ethical committee of King Faisal University [KFU-REC-2025-APR-EA3261]. The fresh and cold-stored mare's colostrum and milk were obtained from a farm located in the Al-Ahsaa region of KSA. Experiments were conducted in the experimental animal laboratories within the Agriculture and Food Sciences College at King Faisal University in Saudi Arabia.

Colostrum and milk samples' collection, cold-storage and analyses

Five purebred mares were chosen for colostrum and milk collection. The mares were 6-10 years old. The mares gave birth naturally without cases of dystocia. The collections took place between the months of March to April. Colostrum samples were obtained within six hours of parturition by manual milking. The udder was cleaned prior to milking and the samples were placed in sterilized plastic bottles. The samples then underwent chemical analysis to determine their composition (fat, protein, lactose, total solids and minerals) in addition to Brix percentage *via* refractometer (High brix Refractometer ORZ 117) (Akköse 2025). Milk samples were collected 10 days after birth. To ensure an adequate milk volume, foals were separated from their mares for two hours before each collection. Prior to milking, the mares' udders were cleaned with a 70% alcohol compress and the milker's hands were washed with neutral soap and water, dried and sanitized with 70% alcohol. The first three milk streams were discarded and the remaining milk was then collected into an autoclaved glass container by fully emptying the udder. The colostrum and milk samples were kept cold-stored for four days in refrigerator at 4°C.

Mice and experimental groups

Thirty albino male mice (28.90±0.45) were kept in cages (40.0 × 24.0 × 18.0 cm) for control and the colostrum and milk groups (Fig 1).

The colostrum and milk groups were offered *ad libitum* mare's fresh colostrum and milk at day0 followed by cold-stored colostrum and milk at, day 1, day 2, day 3 and day 4 whereas the control group was offered *ad libitum* water.

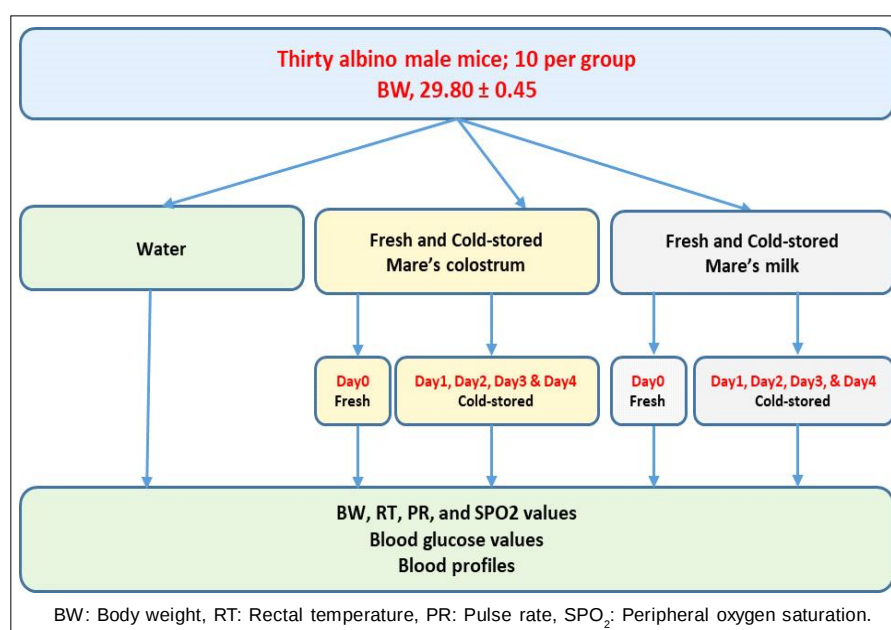


Fig 1: Effects of *ad libitum* consumption of fresh and cold-stored mare's colostrum and milk on blood glucose values, physiological parameters, hematological and biochemistry profiles in mice.

The mice were fed pellet diet composed of 18.0 protein, 3.20 fiber, 2.90 fat, 1.0 mixture of minerals and vitamins and energy of 3300 kcal/kg. The control group was given *ad libitum* water whereas mare colostrum and milk groups were provided *ad libitum* mare colostrum and milk diluted with water (1:1 volume) through bottles with automated nipple. The mice consumed daily ~8.0 ml of diluted milk and colostrum, respectively. This diluted and *ad libitum* method was chosen to allow the animals to regulate their intake according to their voluntary needs and reduces stress. The mare colostrum and milk were given either fresh or cold-stored. Animals had free access to diet. Mice were kept controlled under 12h light and 12 h dark cycles starting at 8:0a.m. The temperature (°C) and relative humidity (%) values during the study were controlled to 24.00±1.5°C and 40.0±6.0%, respectively.

Monitoring body weight, rectal temperature, heart rate, peripheral oxygen saturation and blood glucose values

Body weights (g) of control, mare's colostrum and milk groups were recorded using digital balance (Sartorius balance, Azulmart-KSA). Rectal temperature, heart rate, peripheral oxygen saturation and blood glucose values were recorded at day0, day1, day2, day3 and day4. The mice of control, mare's colostrum and milk groups were sedated using 26.6 mg/kg BW xylazine for immediate recording of rectal temperature, pulse rate and SPO2. Rectal temperatures were monitored using clinical thermometer (Citizen). Peripheral oxygen saturation and pulse rates were monitored using pulse oximeter apparatus (CMS60D-VET). The blood glucose values were recorded using blood glucose meter (ICare, Taiwan) without sedation. The tail vein of mice was punctured and 10 µl tiny blood sample put on ICare glucose strips for recording blood glucose values (Mohammed *et al.*, 2024, 2025).

Statistical analysis

Chemical composition of mare's colostrum and milk were statistically analyzed using T-test procedure. In addition, body weight, rectal temperature, SPO2 and pulse rate and blood glucose values of control group, colostrum and milk groups were statistically analyzed using ANOVA procedure (SAS 2008) according to the following model:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where,

μ = Mean.

T_i = Effects of fresh and cold-stored mare's colostrum and milk consumption.

e_{ij} = Standard error.

Duncan's multiple range test (Duncan 1955) was used to compare between means of control, mare's colostrum and milk groups.

RESULTS AND DISCUSSION

Chemical composition of mare colostrum and milk

Chemical composition of mare's colostrum and milk is shown in (Table 1). The results showed that mare colostrum had higher ($P<0.0001$) SNF (11.94 vs. 10.24), protein (4.56 vs. 3.80) and lactose (6.66 vs. 5.56) but similar fat contents (2.45 vs. 2.46) compared to mare's milk, respectively. Moreover, the Brix value (%) of immunoglobulin content in mare's colostrum is 18.0%.

Body weight, rectal temperature, SPO2 and pulse rate

Changes of rectal temperature, pulse rate and SPO2 among control, mare's colostrum and milk groups, fresh and cold-stored, are presented in Table 2. The rectal temperature (°C), pulse rates and SPO2 values did not differ among control, mare's colostrum and milk groups, fresh and cold-stored groups.

Monitoring blood glucose levels

Blood glucose values (mg/dl) per day of mice consuming fresh and cold-stored mare's colostrum and milk are presented in (Fig 2). There were no significant differences in blood glucose levels between any of the groups at the start of the experiment (day 0). The control group's glucose levels remained relatively stable over the four days of experiment. All groups had similar starting glucose levels around 133-135 mg/dl (day 0). At day 1, the fresh mare's colostrum group showed the lowest blood glucose value ($P<0.0001$) followed control and by mare's milk groups, respectively. At day 2, day 3 and day 4, mare's colostrum and milk cold-stored were given to mice. The blood glucose values were the highest ($P<0.0001$) in cold-stored mare's milk group followed by cold-stored colostrum compared to control group at day 3 and day 4.

Table 1: Chemical composition of mare's colostrum and milk.

Parameters, %	Colostrum	Milk	SEM	P value
Fat	2.45	2.46	0.4	<0.95
SNF	11.94 ^a	10.24 ^b	1.38	<0.0001
Protein	4.56 ^a	3.80 ^b	1.23	<0.0001
Lactose	6.66 ^a	5.56 ^b	0.45	<0.0001
Density	1.0394	1.0327	0.01	0.100
Freezing point	-0.419 ^b	-0.525 ^a	0.01	<0.0001
Mineral	0.160 ^b	0.67 ^a	0.01	<0.0001
%Brix Immunoglobulin	18.0	-	-	-

^{a,b}Values between groups in the same row with different superscripts significantly differ at $P<0.05$. SEM: Standard error of means.

The results of the current experiments are shown in (Fig 2) and (Table 1-2) indicating the chemical composition of mare's colostrum and milk and their effects in fresh or cold-stored states on physiological parameters and blood glucose profiles. The obtained results showed potential effects of fresh mare's colostrum in decreasing blood glucose values compared to mare's milk and control groups (Fig 2). Mare's milk is still not widely recognized globally despite its established use in Asian diets. The growing interest of mare's milk in Western Europe and the USA is primarily due to its suitability for individuals with allergies (Dankow *et al.*, 2012; Pieszka *et al.*, 2016; Blanco-Doval *et al.*, 2024).

Chemical composition of mare's colostrum and milk

Chemical composition of mare's colostrum and milk is shown in (Table 1). The results showed that mare colostrum had higher ($P<0.0001$) SNF (11.94 vs. 10.24%), protein (4.56 vs. 3.80%) and lactose (6.66 vs. 5.56%) but similar fat contents (2.45 vs. 2.46) compared to mare's milk, respectively. Moreover, the Brix value (%) of immunoglobulin content in mare's colostrum is 18.0%. This result is consistent with other studies (Csapó *et al.*, 1995; Santos *et al.*, 2005; Barreto *et al.*, 2020; Blanco-Doval *et al.*, 2024). Mare's colostrum, the initial secretion following parturition, differs significantly from mature milk (Barreto *et al.*, 2020; Polidori *et al.*, 2022). These compositional

variations underscore colostrum's vital role in providing essential immune protection and concentrated nutrients to the newborn foal, facilitating its transition to extra-uterine life. Colostrum has a dry matter much higher than milk (11.94 vs. 10.24%), especially the high protein content which composed of 80.0% immunoglobulins (Csapó *et al.*, 1995). Colostrum has about 20.0% higher fat content than milk fat, which produced in the initial third of lactation stage (Pecka *et al.*, 2012). In addition, bioactive peptide precursors, such as α -lactoalbumin and β -lactoglobulins are present in colostrum in considerable quantities (Fessas *et al.*, 2001).

Rectal temperature, pulse rate and peripheral oxygen saturation

Changes of rectal temperature, pulse rate and SPO2 among control, mare's colostrum and milk groups, fresh and cold-stored, are presented in Table (2). The rectal temperature ($^{\circ}\text{C}$), pulse rates and SPO2 values were not differed among control, mare's colostrum and milk groups, fresh and cold-stored groups. This could be attributed to the body's robust thermoregulatory system actively works to maintain a stable core temperature, overriding minor fluctuations that might be caused by food or drink intake. In addition, due to the short period of experiment, colostrum and milk substances don't directly provide significant amounts of substances that would immediately alter respiratory function or oxygen-

Table 2: Changes in rectal temperature ($^{\circ}\text{C}$), pulse rate and peripheral oxygen saturation (%) of mice receiving *ad libitum* fresh and cold-stored mare's colostrum and milk.

Parameters	Control	Colostrum	Milk	SEM	<i>P</i> value
Initial body weight, g	28.80	28.70	29.30	0.66	0.95
Rectal temperature, $^{\circ}\text{C}$	35.70	35.75	35.79	0.06	0.70
Pulse rate/min.	167.2	161.3	164.5	6.30	0.93
Peripheral oxygen saturation, %	86.30	87.80	89.50	2.90	0.09

^{a,b}Values between groups in the same row with different superscripts significantly differ at $P<0.05$. SEM standard error of means.

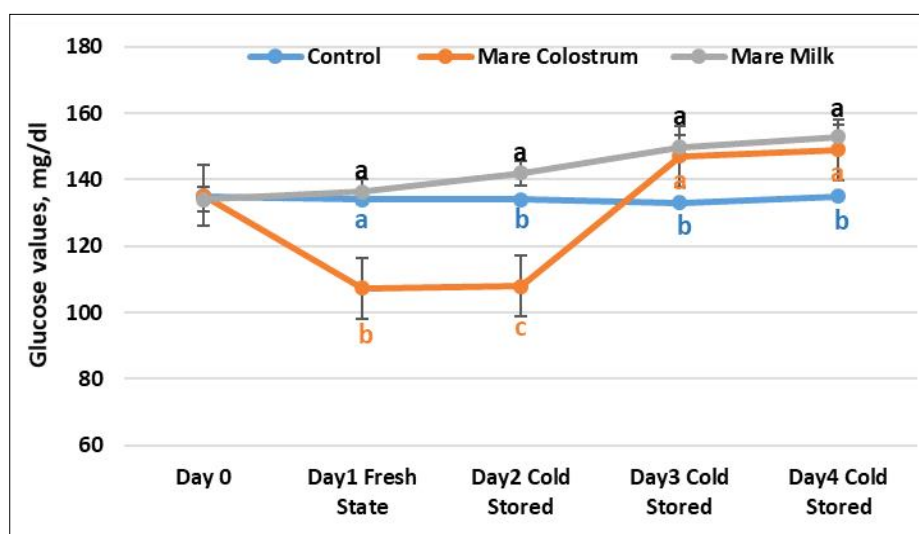


Fig 2: Effects of mare's colostrum and milk on blood glucose values in fresh and cold-stored status.

carrying capacity of the blood in healthy mice. Hence, consumption of mare's colostrum and milk is unlikely to trigger a rapid and significant change in pulse rate in healthy individuals.

Blood glucose levels

Blood glucose values per day of mice consuming fresh and cold-stored mare's colostrum and milk are presented in (Fig 2). There were no significant differences in blood glucose levels between any of the groups at the start of the experiment (day0). The control group's glucose levels remained relatively stable over the four days of experiment. All groups had similar starting glucose levels around 133-135 mg/dl (day 0). At day 1, the fresh mare's colostrum group showed the lowest blood glucose value ($P<0.0001$) followed by mare's milk and control groups, respectively. At day 2, day 3 and day 4, mare's colostrum and milk cold-stored were given to mice. The blood glucose values were the highest ($P<0.0001$) in cold-stored mare's milk group followed by cold-stored colostrum compared to control group at day 3 and day 4. These effects could be attributed to the ingredients in mare's colostrum and milk (Table 1).

Both mare's colostrum and milk contain various biological compounds, some of which can potentially affect on blood glucose levels and body functions (Kazimierska *et al.*, 2021; Musaev *et al.*, 2021; Chen *et al.*, 2024). Mare's milk use in people with diabetes helps reduce the dose of insulin and improves the glycemic index (Jastrzębska *et al.*, 2017). Cold storage is a common method for preserving food and biological samples, which can have a significant effect on mare's colostrum and milk (Cuttance *et al.*, 2024). Mare's milk is sensitive to heat above 40°C, so it requires quick cooling and should be used in liquid form within 6-9 hours of milking (Dankow *et al.*, 2006; Teichert *et al.*, 2020; Hachana *et al.*, 2022). In addition, short-term refrigeration (up to 48 hours) generally preserves the concentration of most immunological factors (Vincent *et al.*, 2018; Straat *et al.*, 2022). However, some studies have shown a decrease in certain cytokines even after relatively short periods of refrigeration (Pawłowska *et al.*, 2022).

CONCLUSION

The current study indicates that fresh mare's colostrum and milk has strong hypoglycemic effect whereas cold-stored mare's colostrum and milk has hyperglycemic effect upon *ad libitum* consumption.

ACKNOWLEDGEMENT

The authors want to thank and acknowledge Deanship of Scientific Research, King Faisal University, Saudi Arabia for funding and support (KFU252410).

Funding

The study was funded by Funding of Scientific Research Deanship of King Faisal University (KFU252410).

Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the official stance of their affiliated institution.

Informed consent

Ethical Approval of Scientific Research Deanship Committee of King Faisal University (ETHICS3145).

Conflict of interest

The authors have no conflicts of interest to disclose.

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