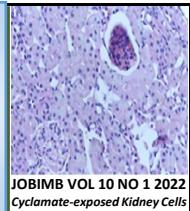


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Cyclamate-exposed Kidney Cells

Evaluation of Nutrients Composition, Minerals, Vitamins and Bioactive Components of Camel and Cow Milk Sold in Katsina Metropolis

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ABSTRACT

Milk is considered as a nearly complete food since it is a good source of protein and major minerals. The consumption of camel and cow milk is becoming more popular, and this is by the perception of the nutritional and therapeutic benefits of the milks. Therefore, the aim of this research is to evaluate the nutrients composition, minerals, vitamins and bio-active components of camel and cow milk. Two samples of camel and cow milk were collected from Katsina central abattoir and transported to Al-Qalam University Katsina Biological Sciences Laboratory and were stored in an icebox. The determination of minerals composition, proximate analysis, Vitamins content and bio-active components were carried out in triplicates using standard analytical procedures. The minerals composition obtained from camel and cow milk were Impressive and Appreciable. The proximate analysis gave a high protein content of 13.00 ± 1.120 in camel milk and 9.00 ± 1.326 in cow milk, Ash content of 14.00 ± 0.06 in camel milk and 18.333 ± 0.577 in cow milk and fat content of $(18.700 \pm 1.664$ in camel and 20.366 ± 0.635 in cow milk and low moisture content of 64.066 ± 3.421 in camel milk and 49.800 ± 2.771 in cow milk respectively. Vitamins content determined shows high content of vitamin c ($1.0366 \pm 0.0115\%$) in camel milk and lower 0.960 ± 0.121 in cow milk. the results obtained from bio-active components shows flavonoids, terpenoids and tannins absent, while Alkaloids, steroids and glycosides present. Therefore, camel and cow milk revealed the therapeutic and nutritional properties which are widely exploited for human health.

INTRODUCTION

Milk from goats, sheep, and cows has been consumed by humans since prehistoric times. These days, when someone says "milk," they almost always mean cow's milk. When different kinds of animal milk are sold commercially, they are named specifically, such sheep milk and goat milk. Milk, the natural production of the breast, is extracted from the mammary glands after one or two actions, with nothing added or taken away, and is then consumed as is or processed further [2]. Milk is practically a complete food since it contains all the essential nutrients, including protein, fat, and important minerals [2]. For the most vulnerable members of society, including infants, school-aged children, and the elderly,

milk and milk products are indispensable staples in the daily diet [2]. Multiple studies have analyzed the distribution and occurrence of the necessary components in various animal milks, and they all agree that milk is one of the most important nutritious food sources for infants and babies, second only to breast milk. In fact, recent studies have linked the consumption of dairy products to positive health outcomes [2].

Vitamins, minerals, enzymes, dissolved gases, and other lipids are just a few of the hundreds of substances that contribute to its biological and technical properties. Lipase, phosphatase, and catalase are the three enzymes. Casein (S1-Casein, S2-Casein, -Casein, and -casein) is the primary protein found in

milk. Whey proteins consist of alpha-lactalbumin and beta-lactoglobulin [3]. Milk contains a variety of nutrients including proteins, lactose, calcium, magnesium, potassium, vitamins A, B1, B2, C, and D, and other substances and minerals in a fat emulsion. The percentage of solids in milk is 13% [2], with fat at 4%, protein at 3.5%, and lactose at 5%.

Clinical trials have shown that camel milk can reduce the risk of developing cancer, allergies, and diabetes. Due to its low concentration and lack of lactoferrin, immunoglobulin, lysozyme, and vitamin C, camel milk has anti-allergic and anti-cancer properties that can help prevent allergy and cancer [4]. It has been proven that infants and young children can benefit from a diet of camel milk. These days, camel milk is pasteurized and packaged in a number of countries, including Saudi Arabia, the United Arab Emirates, Kazakhstan, Mauritania, and Zaire [4].

Camel milk contains a variety of immunological protein components that can kill or neutralize harmful bacteria and viruses. There are several types of immunoglobulins found in camel milk, including IgM, IgG, IgA, and even IgD [5]. Milk and its derivatives are pivotal to a balanced diet, as they are the finest natural food for proper growth and development, strong muscles, strong teeth and bones, clear vision, and robust health [6]. Despite the fact that mammals make milk to sustain their young, people continue to consume milk throughout their lives in many parts of the world, with global milk production reaching 730 million tons annually [7].

Cow milk is a comprehensive diet that satisfies practically all of the requirements of the human body. Specifically, it contains proteins, lactose, lipids, phosphate, calcium, and vitamins (B2, A and mainly D). It has a lot of calcium and the amino acid lysine, which is frequently absent from plant proteins. The minerals are dominated by calcium and phosphorus, which facilitate the body's absorption of the mineral [3]. There was no scientific basis for the long-standing tradition of utilizing camel milk for medical purposes in the former Soviet Union, the Middle East, and several countries of Africa and Asia [8].

However, based on the knowledge currently available, camel milk may be used as a source of bio-active compounds with medicinal characteristics in addition to nutrition [8]. More scientific studies are required to pinpoint the chemical composition with greater accuracy globally [8]. Although camel milk has a long history and has drawn increasing interest recently, there is little information available about its composition and quality in Nigeria. Therefore, it is essential to assess the quality of camel milk in a large district to make sure that this lovely animal keeps its special position in the hearts of future generations [8].

The perception of the nutritional and therapeutic benefits of the milks has led to an increase in the intake of camel and cow milk [9]. Consequently, this study's focus is on the quantitative relevance of the milks' effects on human health. This study aims to compare the nutritional value, minerals, vitamins, and bio-active elements of camel and cow milk.

MATERIALS AND METHOD

Sample collection

From the Katsina central Abattoir, located next to the Gwagware filling station in Katsina town, Katsina State, fresh camel, and cow milk were collected by hand milking into sterile rubber bottles and transported in an icebox to the Al-qalam University

Katsina Biological Science laboratory, where they were stored in a refrigerator at 4 °C.

Determination of Nutrients Composition of camel and cow milk

Determination of Moisture Content

After about an hour in the drying oven at 1000°C, the crucible was ready to be used. After thirty min in a desiccator, the crucible was weighed. This procedure was repeated until a consistent weight was achieved as (W1). After the sample was properly combined, two milliliters of the liquid were placed in a crucible that had already been weighed (W2). For another 24 h, the sample was heated to 1050 degrees Celsius in the oven. The desiccators received the crucible with the dried sample. A desiccant sample was placed in a crucible, allowed to cool for 30 min, and then weighed (W3) [8]. The following formula was then used to get the relative humidity:

$$\text{Moisture (g/100g)} = \frac{W2 - W3}{W2 - W1} \times 100$$

Where: W1 = weight of empty crucible (g)

W2 = weight of crucible + sample before drying (g)

W2 - W1 = weight of sample

W3 = weight of crucible + sample after drying (g)

W2 - W3 = loss of weight (g)

Determination of Ash Content

Marked crucible was heated in a furnace at 550 °C for 1 hour. The furnace temperature was lowered to 180 °C and the crucible transferred into desiccator, cooled for 30 min and weigh. The sample procedure was repeated to obtain constant weight (W1). 2 g of the sample was measured in and duplicated into the pre-weight crucible (W2). The sample was incinerated in a furnace at 550 °C until the residue is uniformly white. The crucible was transferred into desiccators for 30 min and the crucible was weighed as (W3) [8]. Ash content was calculated by the following formula:

$$\text{Ash per 100g} = \frac{W3 - W1}{W2 - W1} \times 100$$

Where W1 = weight of crucible

W2 = weight of crucible + sample

W3 = weight of crucible + ash

Determination of Fat Content

Each sample was weighed out to be exactly 100 mL, then transferred to an evaporating dish and placed in a water bath to evaporate the water. The 2.0 g of evaporated sample was placed on a sheet of filter paper and weighed. The filter paper was then folded and placed within the Soxhlet extraction apparatus's porous thimble. A spherical bottle flask holding 500 mL was filled with 250 mL of petroleum ether. In total, it took 3.5 h to set up and connect all the equipment. After 15 min, the filter paper with the sample was withdrawn from the thimble and placed in a small beaker before being placed in a preheated 70 °C oven. The weight of the extraction flask after oil had been poured into it revealed the amount of crude fat extracted [8]. The following formula was used to derive the percentage of fat:

$$\text{Fat \%} = \frac{W2 - W1}{W3} \times 100$$

Where, W1 = weight of the fat

W2 = weight of sample

W3 = weight of sample after dried

Determination of Fiber

After accurately weighing 2.0 g of each sample, it was added to 100 mL of distilled water and 20 mL of 20% sulfuric acid in a 250 mL conical flask and cooked on low heat for 30 min. Even after cooling and filtering, the leftover residue was scraped back into the flask. To this, we added 100 mL of distilled water and heated it on low for 30 min. Once again, it was cooled and filtered, the residue cleaned with hot water and then rinsed once with ethanol before being filled with petroleum ether. They scraped it into a dry, sterile crucible after drying it off. The crucible's contents were baked at 105 °C for 12 h to speed up the drying process. It was desiccated for further analysis and weighed afterward (W1). Then, after being roasted at 550 °Celsius for 90 min, it was moved to a muffle furnace. Finally, after being dried in a desiccator, it was reweighed as (W2) [8].

Fiber% = Weight of ether extract /Weight of sample x 100

Where: Weight of ether extract = WF+WS-WR

WF= Weight of filter paper (g)

WS= Weight of sample (g)

WR= Weight of residual sample (g)

Determination of Crude Protein

The method [8] for calculating crude protein involved the following three steps:

Digestion: Each sample was weighed at 1 gram, and then added to a digestion flask containing a 5-gram Tablet of Kjedhal catalyst and 15 milliliters of strong sulfuric acid (H₂SO₄). The mixture was heated on a hot plate till the digest color turned to clear solution. The cooled digest was transferred to a 100 mL volumetric flask and brought to the correct level with distilled water [8].

Distillation: The Kjeldahl distillation flask was filled with 20 mL of 45 percent Sodium hydroxide (NaOH) and 10 mL of sample. The flask and condenser were linked to the transport tube. After 15-20 minutes of distillation using a mixed indicator containing a few drops of each color, the result was 10 mL of 2% boric acid [8].

Titration: The distillate was titrated using 0.12N HCl to a green color which signified the endpoint [8].

Protein was calculated as:

%Protein = (b - a) × (0.1 × 14.00) / W × 100 × 6.25/1000

Where:

1000 = the conversion of m gN/100 g to gN /100 g sample

6.25 = the protein-nitrogen conversion factor for milk and its by-products.

Determination of Carbohydrate

The samples were analyzed, and then the carbohydrate content was determined quantitatively. Total accessible carbohydrate in the sample was calculated by subtracting the results of protein, fat, ash, and moisture analysis from 100 [8].

I.e. 100 - %protein + %fat + %ash + %moisture = %carbohydrate

Determination of Minerals Content

Minerals were determined first through digestion of sample with nitric acid and perchloric acid at a ratio of 1:2. The mixture were then digested at the temperature of 150°C on a hot plate.

Clear solution was acquired then the digestion flask was removed from hot plate and allowed to cool. The content of each sample was transferred into sample bottles. The sample were analyzed for their mineral content of interest using Atomic Absorption Spectrophotometer (AAS). [10].

Determination of Vitamins Content

Extraction and Determination of β-Carotene

For 30 minutes, while keeping the temperature between 70 and 800 degrees Celsius and stirring the flask occasionally, 10 milliliters of the milk sample were placed in a conical flask holding 100 milliliters of 95 percent ethanol. We drained the supernatant, let it cool, and then measured its volume with a measuring cylinder to use as the starting point for our data. The mixture's ethanol concentration was increased to 85% by adding 15mL of distilled water, and it was chilled in a container of ice water for about 5 minutes. As a final step, the cooled ethanol was poured over the mixture in the separating funnel, which had previously included 25mL of petroleum ether (pet-ether). We first achieved a homogeneous mixture by gently whirling the funnel; then we allowed it to settle until two different layers developed.

The liquid was collected in a 250 mL conical flask and the sediment was transferred to a beaker. Three times with 10 mL of pet-ether in the funnel, the bottom layer was scooped up and re-extracted. A total of 10 mL of 80% ethanol was added to the pet-ether that had been collected in a separating funnel from a 250 mL conical flask. Sample vials were then filled with the final extract and metered out [11]. The extracts were analyzed by a spectrophotometer, and their absorbance at 450 nm was determined. The spectrophotometer was zeroed using a pet-ether (blank)-filled cuvette. The extract samples were placed in cuvettes, and the readings were collected once the numbers on the display window stabilized. Three independent measurements were taken for each sample, with the average used [11]. Bear-Lamberts Law, which stipulates that absorbance (A) is proportional to concentration (C) of the pigment, was used to determine the -carotene concentration, as shown in the equation:

$A \propto L$ (if concentration (C) is constant).

A=ECL; C=A/EL

Where:

C= concentration of carotene

A= absorbance

E=extinction coefficient

L= thickness of cuvettes (path length) =1cm

E of β-carotene = $1.25 \times 10^4 \mu\text{g/L}$

Determination of Ascorbic acid

Each sample were accurately taken as 10 mL, and then transferred into a test tube, 10 mL of standard ascorbic acid solutions was put into the test tube, and then 1 mL of potassium permanganate (KMnO₄) was added. Each test tube's contents were thoroughly combined. After 5 minutes, the solution was thrown away. Standard solutions were measured for absorbance at 530 nm [12]. The spectrophotometer's zero point was determined using a cuvette holding a blank (standard) Ascorbic acid solution. The extract samples were placed in cuvettes, and the readings were collected once the numbers on the display window stabilized. For each sample, the procedure was done three times, and the average values were recorded [11].

Determination of Bio-active Component

Milk sample was tested for the presence of bio-active compounds by using the following standard methods:

Test for Flavonoids

Exactly 2 mL of Milk sample was mixed with 4 drops of concentrated sulfuric acid (H_2SO_4) and orange color indicates the presence of flavonoids [13].

Test for Alkaloids

Exactly 2 mL of milk sample was mixed with 2mL HCl and heated gently. Wagner's reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids [13].

Test for Terpenoids

Exactly 2 mL of milk sample was dissolved in 2mL of chloroform and evaporated to dryness. To this, 2 mL of concentrated H_2SO_4 was added and heated for about 2 min. A grayish color indicated the presence of terpenoids [13].

Test for Steroids

Exactly 2 mL of milk sample was mixed with 2mL of chloroform and concentrated sulfuric acid was added sidewise. A red color produced in the lower chloroform layer indicated the presence of steroids [13].

Test for Tannins

Two milliliters of the milk sample were combined with two milliliters of a $FeCl_3$ solution. The presence of tannins was identified by a blue-green or black hue [13].

Test for Glycosides

Chloroform and acetic acid were added to a milk sample that measured exactly 2 milliliters in volume. The ice bath was used to chill the mixture. Acidic sulfuric acid was added after being carefully concentrated. The presence of the steroid nucleus, represented by the glycoside part of the glycoside, resulted in a shift in color from violet to blue to green [13].

Statistical Analysis

The results were analyzed in triplicates where it was subjected to Analysis of Variance (ANOVA).

RESULTS AND DISCUSSION

The findings from the analysis of the minerals present in camel and cow milk shows the presence of copper, manganese, zinc, magnesium, and iron. The analysis of each sample was carried out in triplicate. **Table 1**'s findings showed that there was copper in both cow milk (0.0285 ± 0.0005 mg/g) and camel milk (0.029 ± 0.002 mg/g), which is statistically similar at $p < 0.05$. Compared to the average, which is 0.44 mg/L, this is lower [14].

Table 1. The mineral composition of camel and cow milk.

Mineral Elements	Camel Milk (Mg/g)	Cow Milk (Mg/g)
Copper (Cu)	0.029 ± 0.002^a	0.0285 ± 0.0005^a
Manganese (Mn)	0.022 ± 0.0005^a	0.013 ± 0.0004^a
Zinc (Zn)	0.106 ± 0.003^a	0.142 ± 0.0002^b
Magnesium (Mg)	0.365 ± 0.127^a	0.326 ± 0.1260^a
Iron (Fe)	0.450 ± 0.0017^a	0.367 ± 0.0002^b

Keys: Values are means of 3 triplicate determinations \pm S.D, means with different superscript along the same horizontal array differ significantly ($P < 0.05$).

Copper is an essential nutrient for the body to form red blood cells, it helps to maintain healthy bones, blood vessels, nerves and immune function [10]. A diet rich in copper may help prevent osteoporosis and cardiovascular disease [10]. Manganese was also determined to be (0.022 ± 0.0005 mg/g) in camel milk and (0.013 ± 0.0004 mg/g) in cow milk which are statistical similar at $p < 0.05$. Consumption of manganese is believed to support the immune system, it regulates the blood sugar levels, the production of energy and cell reproduction [18]. The concentration of zinc was determined to be (0.106 ± 0.003 mg/g) in camel milk and (0.142 ± 0.0002 mg/g) in cow milk which are statistical different at $p < 0.05$ and less than average value of which (2.00 mg/L) [14].

Due to the cow's dietary habits, increased zinc levels were found in the animal. The cow tends to eat food that is higher in zinc content than the camel's diet. According to some sources, zinc is a necessary component that promotes vitamin activity and the production of red and white blood cells [10]. Concentration of magnesium was also observed to be (0.365 ± 0.127 mg/g) in camel milk and (0.326 ± 0.1260 mg/g) in cow milk which are statistical different at $p < 0.05$. The average concentration is (11.82 mg/L) [5]. Magnesium plays a significant role in the human body and its deficiency led to severe diarrhea and persistent migraines [10]. Iron concentration of the milks was observed to be (0.450 ± 0.0017 mg/g) in camel milk and (0.367 ± 0.0002 mg/g) in cow milk which are statistical different at $p < 0.05$ significant level. Camel milk was observed to be more present than the cow milk but less than average concentration of (1.00 mg/L) [5]. According to some sources, iron is crucial for the diets of pregnant women, nursing mothers, newborns, and the elderly in order to prevent anemia and other connected disorders [10].

Table 2. Proximate analysis of camel and cow milk.

Parameters	Camel Milk	Cow Milk
Moisture content (%)	64.066 ± 3.421^a	49.800 ± 2.771^b
Crude Fibre (%)	1.300 ± 0.1000^a	9.666 ± 0.577^b
Crude Fat (%)	18.700 ± 1.664^a	20.366 ± 0.635^b
Crude Protein (%)	13.000 ± 1.123^a	9.000 ± 1.326^b
Ash content (%)	14.00 ± 0.06^a	18.333 ± 0.577^b
Carbohydrate (%)	8.700 ± 1.779^a	7.200 ± 1.628^b

Keys: Values are means of 3 triplicate determinations \pm S.D, means with different superscript along the same horizontal array differ significantly ($P < 0.05$).

The findings of the proximate analysis of camel and cow milk are presented in **Table 2**. The results revealed that the crude protein content of camel milk ($13.00 \pm 1.120\%$) and cow milk ($9.00 \pm 1.326\%$) which are statistical different at $p < 0.05$. However, the protein content of camel milk was considerably higher than that of cow milk. This may be because the animals' diets differed and because of environmental changes; the camel may have had more protein-rich forage than the cow. This outcome is consistent with other research' findings that camel milk's protein concentration was 3.27 percent and cow milk's protein content was 2.04 percent [8]. The reduced protein level of the samples above must be due to the dry season, when there is a lack of food in the natural environment and poor economic conditions among Nigerians, making it difficult for people to provide food for the camels [8]. Protein is an essential dietary component for humans since it is required for the replacement of tissues and the supply of enough energy, however, protein deficiency cause growth retardation [10]. Moisture content was observed to be ($64.066 \pm 3.421\%$) in camel milk and ($49.800 \pm 2.771\%$) in cow milk.

The moisture content of camel milk was highly present compared to that of the cow milk. The standard range for moisture is (85-95%) [8]. Low moisture content is influenced by the animals' diet and the quality of the water they drink [8]. Moisture content indicates the susceptibility towards spoilage [15]. Ash content was observed to be (14.00 \pm 0.06%) from camel milk and (18.333 \pm 0.577%) from cow milk. It was observed higher than the reported work of (0.5%) [8] and (0.75%) was observed from camel milk and (0.71%) from cow milk [15]. The excessive camel grazing on bushes or plants cultivated in the desert region may be the cause of the high ash content that was detected [24].

Ash content is an indication of inorganic elements level such as zinc, magnesium, copper in a milk [10]. The result of fat content revealed that (18.700 \pm 1.664%) from camel milk and (20.360 \pm 0.635%) from cow milk. The findings indicate that camel milk has a lower crude fat level than cow milk. The reported fat content in this study was higher than that in other studies, where the observed fat contents were (4.2%) for camel milk and (4.14%) for cow milk [15]. The high fat level seen in this study may be due to a natural phenomenon where the animal's diet and water content have an impact on its fat content [15]. The fiber content observed was (1.300 \pm 0.1000%) from camel milk and (9.666 \pm 0.577%) from cow milk. Cow milk has been found to contain higher fiber than camel milk. Fiber makes stools softer, which prevents constipation and shields humans from colon cancer [10].

The vitamins content such as vitamin A (beta-carotene) and vitamin C (ascorbic acid) of camel and cow milk were determined as shown from **Table 3**. Vitamin C (ascorbic acid) content observed from camel milk as (1.0366 \pm 0.0115mg/L) and cow milk as (0.960 \pm 0.121 mg/L). The amount of vitamin C (ascorbic acid) in camel milk is higher than that in cow milk. Between camel and cow milk, there is an average difference of 25.00 mg/L of vitamin C [4].

Table 3. Vitamins content in camel and cow milk.

Vitamins	Camel Milk	Cow Milk
Beta-carotene (ug/g)	0.150 \pm 0.0435 ^a	0.120 \pm 0.0346 ^a
Ascorbic acid (mg/mL)	1.0366 \pm 0.0115 ^a	0.960 \pm 0.121 ^b

Keys: Values are means of 3 triplicate determinations \pm S.D, means with different superscript along the same horizontal array differ significantly ($P < 0.05$).

Ascorbic acid, a form of vitamin C, helps keep joints, lungs, gums, and teeth healthy. It also helps to cleanse blood [10]. Vitamin C is a powerful antioxidant property [4]. Vitamin A (Beta-carotene) content observed from camel milk (0.150 \pm 0.0435 ug/L) and from cow milk (0.120 \pm 0.03ug/L). Camel Milk shows the highest content of vitamin A than Cow milk. The average content of vitamin A from camel milk is (60.90 \pm 25.00 ug/L) and cow milk is (20.10 \pm 10.00 ug/L) [4]. Vitamin A promotes growth, resistance to diseases and, delays ageing. It also promotes the health of the eyes, skin, nails and hair [10]. The Bio-active component of camel and cow milk was summarized in **Table 4**.

Table 4. Bioactive component in camel and cow milk.

Parameters	Camel Milk	Cow Milk
Flavonoids	-	-
Alkaloids	+++	++
Terpenoids	-	-
Steroids	+++	++
Tannins	-	-
Glycosides	++	+

Keys: +++= Highly Present, ++= Moderately Present, += Minimal Present and -= Not Present

The findings showed that certain bioactive compounds have been identified. The type of plant food consumed by the animals, as seen, may be the cause of the presence of these bioactive compounds. Alkaloids were discovered in both milks, although camel milk had a high observed presence (++) compared to cow milk's (++), which had a moderate presence. Workers in the field have noted that alkaloids have analgesic and antibacterial effects. Both camel and cow milk were found to be devoid of flavonoids. Terpenoids were also lacking in camel (-) and cow (-) milk. Steroids were also detected, with a high concentration (++) in camel milk and a lower concentration (+). Anabolic steroids, which are related to substances like sex hormones, have been demonstrated to have antibacterial effects [10]. No tannins were detected in either the camel (-) or the cow (-) milk. In contrast to cow's milk, which had a negligible number of glycosides (+), camel's milk had a significant amount (++) . Numerous studies [10] have shown that glycosides successfully reduce hypertension. So, the counterfactual is confirmed.

CONCLUSION

In the Holy Qur'an and the hadith of the Prophet Muhammad (PBUH), both camel and cow are described as miracles and are recommended as treatments for diseases. This study has shown the minerals, vitamins, proximate and bio-active composition of camel and cow milk which revealed the rich source of the macro and micronutrients, potential medicinal or therapeutic properties and usefulness of camel and cow milk. Therefore, in light of the above evidence, it is crystal clear that camel and cow milk has valuable nutritional and therapeutic properties which can be widely exploited for human health.

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