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DISSERTATION

Presented by

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In partial fulfillment of the requirements for the

Master's Degree in agronomy sciences

In Animal production and dairy processing

Title

Physicochemical, biochemical characteristics and Antioxidant Properties of Camel Milk and Urine mixed and separated

Defended on 01/07/2024 before the jury composed of:

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بِسۡمِ ٱللَّهِ ٱلرَّحۡمَٰنِ ٱلرَّحِيمِ

﴿أَفَلَا يَنظُرُونَ إِلَى الْإِبِلِ كَيْفَ خُلِقَتْ﴾

Then do they not look at the camels - how they are created?

[الغاشية: 17]

Acknowledgments

First, I would like to thank Allah, my Almighty God, for helping me to accomplish this work.

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Dedication

To the people of GAZA

Our brothers and sisters in Philistine the steadfast warriors,

Whose painful events have accompanied every step of this work,

To the fighters who defend on our third holiest place and our first Qibla "EL QUDES"

Amira wafaa

ملخص

يشتهر حليب الإبل بخصائصه الغذائية والعلاجية، بينما يتم استخدام بول الإبل أيضًا لتأثيراته العلاجية. ومع ذلك، هناك القليل من الدراسات العلمية المتعمقة حول التأثيرات المشتركة لهذين المنتجين البيولوجيين.

تهدف هذه البحث إلى استكشاف أولاً الخصائص الفيزيائية والكيميائية لحليب الإبل، وبول الإبل، ومزيجهما؛ وثانيًا الخصائص المضادة للأكسدة لحليب الإبل وفقًا لمرحلة الرضاعة، وبول الإبل، وكذلك تأثيرات مزجهما والبسترة

تظهر النتائج أن إضافة البول إلى الحليب يزيد من الحموضة ويغير قليلاً من درجة الحموضة. يتغير التركيب الكيميائي للحليب وفقًا لمراحل الرضاعة، حيث تزداد نسبة الدهون والملح وتقل المواد الصلبة غير الدهنية واللاكتوز والبروتين. يزداد محتوى البروتين في الخلائط بينما ينخفض فيتامين C. تتفاوت تركيزات الفلافونويدات والبوليفينول بين الخلائط. تكون النشاطات المضادة للأكسدة هي الأعلى في نهاية فترة الرضاعة، مع أن لبول الإبل أعلى نسبة تثبيط. تقلل البسترة من النشاط المضاد للأكسدة في الخلائط. تؤكد النتائج على الخصائص العلاجية الكبيرة لبول وحليب الإبل، خاصة في صورتهما الخام، نظرًا لثرائهما بالجزيئات العضوية وغير العضوية. تؤثر البسترة سلبًا على هذه الخصائص، مما يشير إلى أن الخلائط الخام قد توفر فوائد صحية أكبر.

الكلمات المفتاحية

حليب الإبل , مرحلة الإرضاع ,بول الإبل ، نشاط مضادات الأكسدة، التركيب الفيزيوكيميائي

ABSTRACT

Camel milk is renowned for its nutritional and therapeutic properties, while camel urine is also used for its curative effects. However, there are few in-depth scientific studies on the combined effects of these two biological products.

This research aims to explore, firstly, the physicochemical properties of camel milk, urine, and their mixture; and secondly, the antioxidant properties of camel milk according to the lactation stage and of urine, as well as the effects of their mixture and pasteurization

The results show that adding urine to milk increases the acidity and slightly changes the pH. The chemical composition of milk changes according to the stages of lactation, with an increase in fat and salt content and a decrease in non-fat solids, lactose and protein. The protein content increases in the blends, while vitamin C decreases. Concentrations of flavonoids and polyphenols vary between the blends. Antioxidant activity is highest at the end of lactation, with camel urine having the highest inhibition percentage. Pasteurisation reduces the antioxidant activity of the mixtures. The results highlight the significant therapeutic properties of camel urine and milk, especially in raw form, due to their richness in organic and inorganic molecules. Pasteurisation negatively affects these properties, suggesting that raw mixtures may provide greater health benefits.

Keywords: camel milk, Stage of lactation, Physicochemical Composition, camel urine, activity antioxidant

RESUME

Le lait de chamelle est réputé pour ses propriétés nutritionnelles et thérapeutiques, tandis que l'urine de chamelle est utilisée aussi pour ces effets curatifs. Cependant, il existe peu d'études scientifiques approfondies sur les effets combinés de ces deux produits biologiques.

Cette recherche vise à explorer, premièrement, les propriétés physicochimiques du lait de chamelle, de l'urine et de leur mélange ; deuxièmement, les propriétés antioxydantes du lait de chamelle selon le stade de lactation et de l'urine, ainsi que les effets de leur mélange et de la pasteurisation.

Les résultats montrent que l'ajout d'urine au lait augmente l'acidité et modifie légèrement le pH. La composition chimique du lait change selon les stades de lactation, avec une augmentation de la teneur en matières grasses et en sels, et une diminution des solides non gras, du lactose et des protéines. La teneur en protéines augmente dans les mélanges, tandis que la vitamine C diminue. Les concentrations des flavonoïdes et des polyphénols varient entre les mélanges. L'activité antioxydant est la plus élevée en fin de lactation, avec l'urine de chamelle ayant le pourcentage d'inhibition le plus élevé. La pasteurisation réduit l'activité antioxydant des mélanges. Les résultats soulignent les propriétés thérapeutiques significatives de l'urine et du lait de chamelle, en particulier sous forme crue, en raison de leur richesse en molécules organiques et inorganiques. La pasteurisation affecte négativement ces propriétés, suggérant que les mélanges crus peuvent offrir de plus grands bénéfices pour la santé.

Mots-clés : Lait de chamelle, urine de chamelle, stade de lactation, Composition physicochimique, activité antioxydant

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List of Abbreviations

UHT: ultra-high temperature treatment

CM: Camel milk

RM: Raw milk

PM: Pasteurized milk

RMU/ RM+U: raw milk urine

PMU/ PM+U : pasteurized milk urine

FPM: Freezing point of milk

WAP: Whey Acidic Protein

PGRP: Peptidoglycan Recognition Protein)

HTST: High-temperature short-time

LTLT: Low-Temperature long time

HCAbs: Heavy Chain Antibodies

VHHs: Variable Heavy Chain Antibodies

PGRP: Peptidoglycan Recognition Protein

NAGase: N-acetyl-glucosaminidase

IgG1: immunoglobulin, Gamma

HCAbs: Heavy-chain antibodies

LPO: Lactoperoxidase

ACE; Angiotensin-converting enzyme

HepG2: Hepatic Cell line #2

MCF7: Michigan Cancer Foundation

mRNA: messenger Ribonucleic acid

CAT: Catalase

SOD: Superoxide Dismutase

GPx: Glutathione Peroxidase

Prevent CCL4-induced oxidative stress and increase antioxidant activity in liver

CCL4: Carbon tetrachloride

CU: Camel urine

PMF: Protein molecular fraction

U251: Brain cancer (glioma) cell line

HCT116: Colon/colorectal cancer cell line

HEPG2: Liver cancer (hepatocellular carcinoma) cell line

AA: arachidonic acid

ADP: adenosine diphosphate

WRS: Water-immersion restraint stress

Le PFA-100 : Platelet Function Analyzer-100

Literature Review

General introduction

Camels are utilized as working animals that are well-suited for the arid desert environment and play a crucial role in the transportation of both passengers and goods; hence, they are integral in sustaining the pastoral way of life by meeting essential livelihood needs. The domestication of camels has been a longstanding practice, offering valuable food resources such as milk and meat, in addition to supplying fiber and felt derived from their hair for the production of textiles. (Tharwat et *al.,* 2023)

The dromedary camel, commonly known as Camelus dromedarius, accounts for approximately 94% of the global camel population, making it the most renowned among the three camel species, with the Bactrian camel, scientifically referred to as Camelus bactrianus, representing around 6% of the total population. According to the Food and Agriculture Organization (FAO) in 2013

Camel milk and urine are among the traditional therapeutic products used since ancient times in different communities for the treatment of many diseases. It is a scientific miracle of the Sunna that the Prophet Mohammed (peace be upon him) mentioned camel urine as a remedy in ancient times, as reported in Sahih Bukhari and also Sahih Muslim. Anas reported: Some people from the tribe of `Ukl came to the Prophet (pbuh) and embraced Islam. The climate of Medina did not suit them, so the Prophet (pbuh) ordered them to go to the (herd of milch) camels of charity and to drink, their milk and urine (as a medicine). They did so, and after they had recovered from their ailment (became healthy)

Camel milk is one of the most valuable food resources in the nomadic society and has high nutritive and therapeutic values, due to its essential elements like minerals, vitamins, fatty acids, carbohydrates and protective proteins such as, lactoferrin, lacto peroxidase, lysozyme, peptidoglycan recognition protein and immunoglobulins. Due to its components, camel's milk is important for the treatment of diseases like, dropsy, jaundice, spleen ailment, tuberculosis, asthma, anemia, autoimmune diseases (autism), constipation, crohn's diseases, and liver cirrhosis also serve as beauty products.(Zerihum, 2023)

Camel urine is considered the prototype of urotherapy and has more beneficial effects than other animal urines. It consists of various chemical constituents that contribute to its anticancer, antiplatelet, gastroprotective, and hepatoprotective effects (Salam et *al.,* 2021).

Although camel urine may seem unappealing, it has been widely consumed for a long time in the Arabian Peninsula, as well as in the United States, the United Kingdom, and other

European countries. Its use is considered an available option for managing various health disorders such as fevers, colds, and tumors. People generally take it directly or occasionally mix a few drops with camel milk (Abdel Gader and Alhaider, 2016).

Drinking camel urine alone or mixed with milk is a widely recognized practice in traditional medicine. Based on this principle, the aim of this study is to investigate and compare the physico-chemical properties, chemical compositions, and antioxidant capacities of camel urine and camel milk, both separately and combined.

The present work is structured around those axes:

- ✓ A literature review;
- ✓ Physico-chemical characterization of camel milk and urine, as well as their mixture
- ✓ Determination of the chemical composition of camel milk and the quantification of flavonoids, polyphenols, vitamin C, and proteins in camel urine and their mixture.
- ✓ Evaluation of the antioxidant activities of camel milk and urine, and their mixture

Chapter I: Overview on Camels

I. 1.Generality

The camels are the most dominant and widely distributed animal in tropical and subtropical continents of Africa and Asia. They make an important contribution to human survival and utilization of these lands, where climatic conditions are at their most severe, characterized, in particular, by very high temperatures and very low and irregular rainfall. (Chehma et *al.*, 2023; Fayera and Zerihun, 2023)

The dromedary stands out as the singular species of livestock with the ability to utilize and improve upon the extensive Saharan territories. Numerous research endeavors have underscored the anatomical and physiological adjustments of this creature to arid conditions, facilitating the preservation of its energy reserves. Possessing distinctive climate adaptability, it exhibits the capacity to endure extended periods without access to water, rendering it of significant worth in regions where water availability is limited. (Kandil et *al.,* 2023; Chehma et *al.,* 2023)

The significant camelids, namely the dromedary and Bactrian camel, represent a longstanding lineage of domesticated creatures that have served humanity for millennia. These animals play a crucial role as providers of milk, meat, hides, wool, and their dung, which serves as a valuable fertilizer and fuel source. Moreover, they fulfill diverse functions such as transportation, draft power, and riding, serving as both an investment opportunity and a long-term asset that enhances the prestige of their owners. The trade in live camels constitutes a sizable market, while camel-derived products, including milk, meat, and even urine, exhibit therapeutic properties for various human ailments. Specifically, camel meat is renowned for its ability to bolster disease resistance, fortify muscles and bones, hydrate the skin, and alleviate internal discomfort. Furthermore, the fat obtained from the camel's distinctive hump is utilized for its potent anti-inflammatory properties, effectively mitigating pain and swelling. (Fayera and Zerihun, 2023; Faye and Ratto, 2022)

I. 2.Origins and Taxonomy

Camelids were likely one of the final significant species to be domesticated by humans. The estimated period of domestication is approximately 4000 years prior or slightly earlier. The presumed region of domestication is the Southern Arabian Peninsula, specifically the regions of Yemen and Oman. (Fayera and Zerihun, 2023)

According to Khomeiri (2015) many people believed the myth that camels originated from a rabbit because a camel has grooved upper lip like of rabbits. The camelids are classified under the order Artiodactyla, specifically the sub-order Tylopoda (Figure 01). Within this sub-order, they are grouped alongside Suiformes (resembling pigs) and are considered ruminating animals that are closely related to, yet distinct from, the suborder Ruminantia. Discrepancies in factors such as foot structure, digestive system, and the lack of horns serve as evidence supporting this distinction. The schism between Tylopoda and Ruminantia can be traced back to ancient times due to the significant differences observed between camels and ruminant species. Various scholars have pointed towards the origins of camels being linked to the Protylopus, a small animal resembling a rabbit in size, possessing four-toed feet and low-crowned teeth. This creature inhabited the North American continent during the Eocene period approximately 45-50 million years ago.

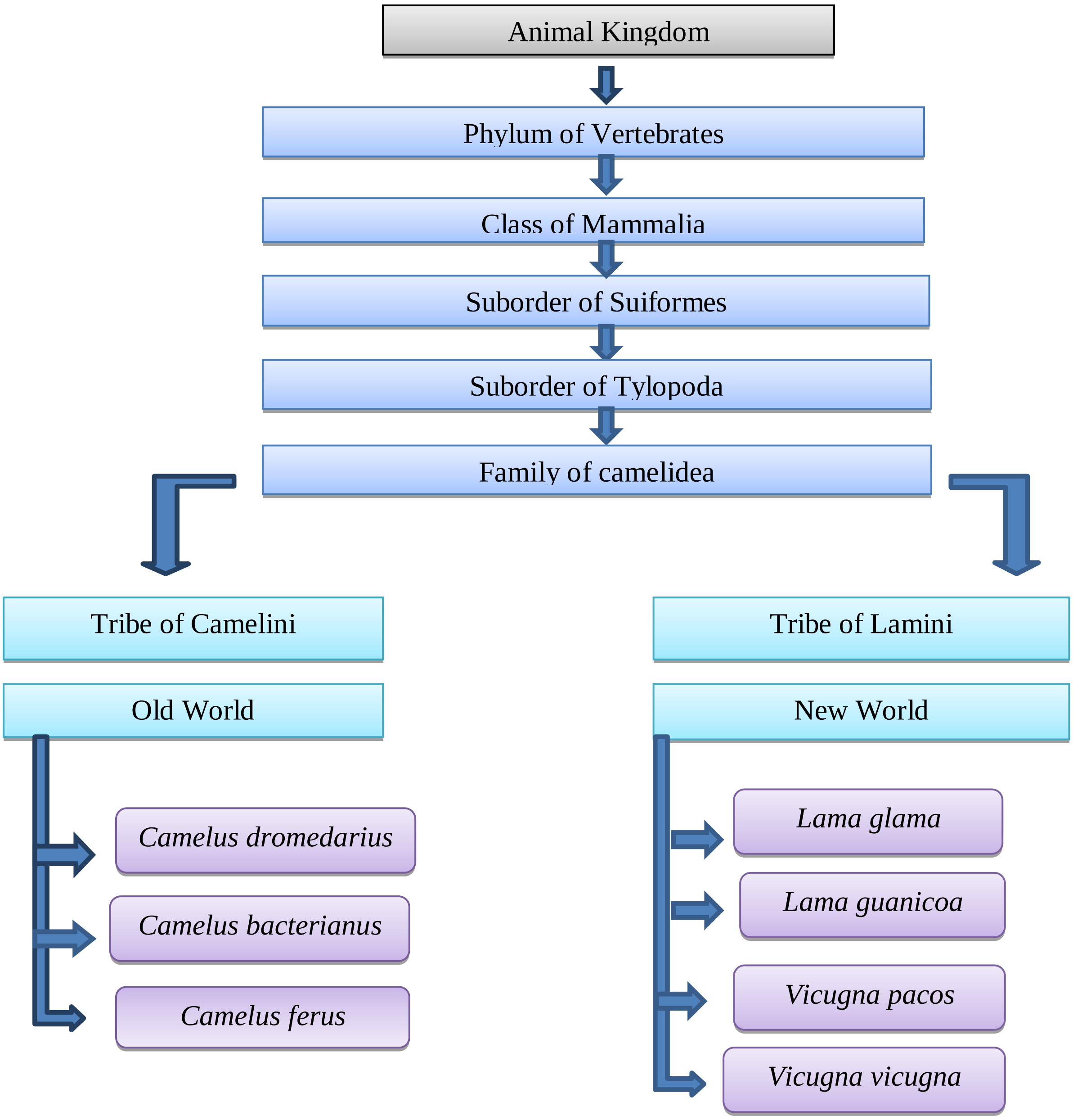


Figure 1 : Classification of camelids (Khalafalla and Hussein, 2021)

The old-world camelids include the dromedary camel (*Camelus dromedarius*), the domesticated Bactrian camel (*Camelus bactrianus*) and the endangered wild Bactrian camel. (MacKay et al., 2022)

- Dromedary or one-humped camel (*Camelusdromedarius*) :

Arabian camels are vital domestic animal. He is the most important livestock animal in the semi-arid areas of Northern and Eastern Africa as well as in the Arabian Peninsula and Iran. The one-humped camel was domesticated about 3000 B.C.E. In south Arabia mainly for its meat and milk (Figure 02). (Fayera and Zerihun, 2023; Khomeiri, 2015)

The name of the dromedary derived from the Greek, “dromeus” which means runner or droma-running. (Khomeiri, 2015)

- Bactrian camel Bactrian or two-humped camel(*Camelus bactrianus*):

Exist in the cold deserts and dry steppes of Asia. The name of Bactrian camel comed from the area of Bactriana in Asia that was the old name of Iran (Bakhtar or Bactar). He was domesticated on the border of Iran and Turkmenistan and spread to an area bordered by the Crimea, southern Siberia, Mongolia and China. These animals are stockier than the dromedary and covered by a thicker wool (Figure 03). Also in the desert Gobi there is still a population of wild Bactrian camels classified as Camelus ferus (Figure 04), the wild Arabian camel became extinct. (Khomeiri, 2015; Martini, 2017)

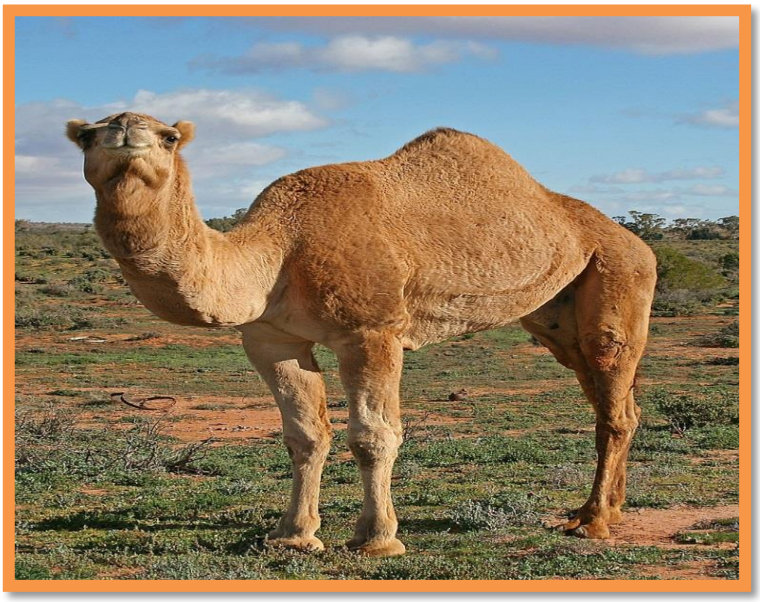


Figure 2: Camelus Dromedarius (Ouologuem and Moussa, 2020)

Figure 3: Bactrian Domestic Camel (Ouologuem and Moussa, 2020)

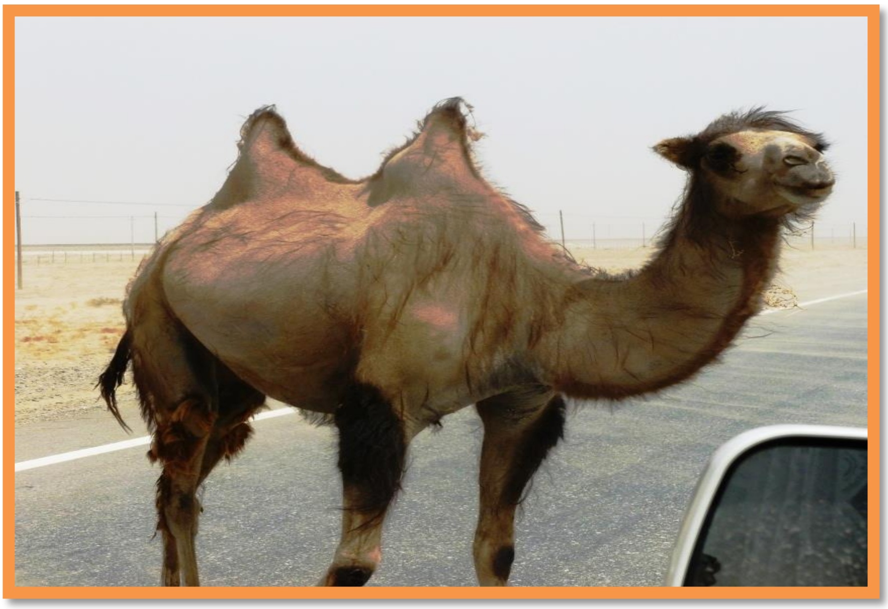


Figure 4: Wild Bactrian Camel (Ouologuem and Moussa, 2020)

I. 3.Camel population

A- Worldwide

Worldwide according to FAO there are 41,772,353 camels, with highest numbers found in Africa (figure 5), further it was recorded that 35,100,908 camels found in Africa while 6,665,043 in Asia in 2022.

The first country which has the highest number of camels is Somalia 7, 5 million, the second is Sudan which has 4,9 million, and the third is Kenya 4,6 million.

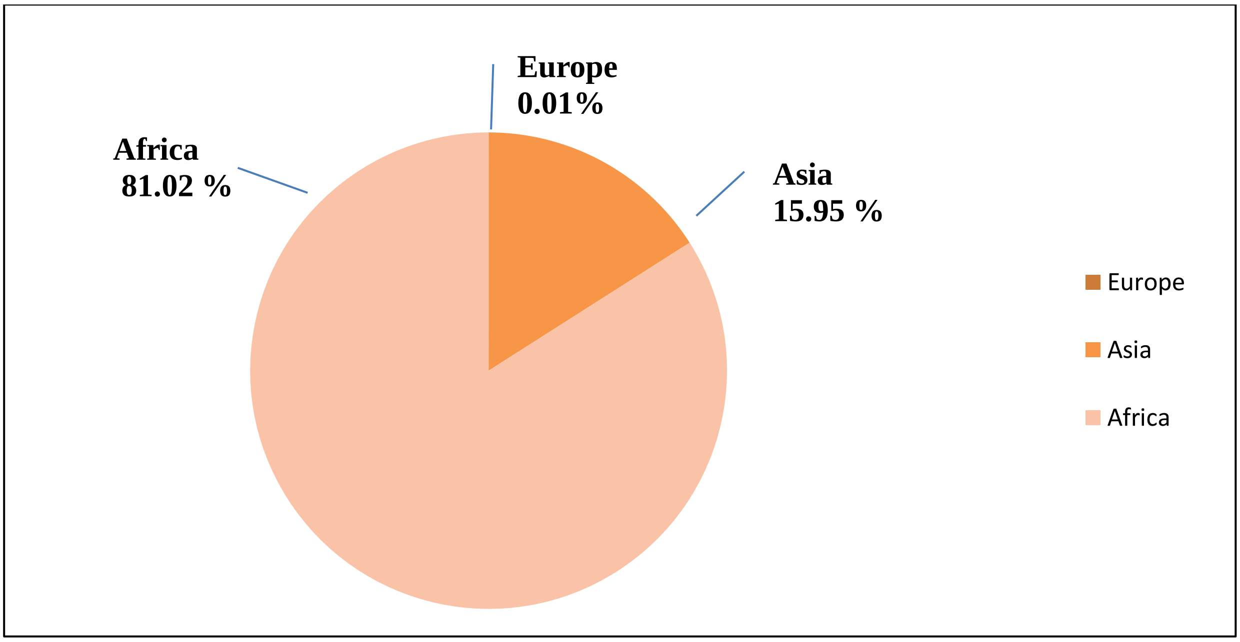


Figure 5: Camels population by region, Sum 2022, Source: FAOSTAT (March 20, 2024)

B- In Alegria

The camel population in Algeria is estimated at 459616 heads in 2022. It represent 1.10% of the world population, 1.30% of the African population, and 7.72% of the Maghreb population, with a mean annual growth corresponding to 7, 41% % recorded between 1961 and 2022. (FAO, 2022)

Therefore, the camel herd in Algeria has experienced an increase of approximately 35.14% between 2012 and 2022.

The dromedary is present in 17 Wilayas (8 Saharan and 9 Steppic). There are three main distribution areas. With 56% of the national livestock, the central Sahara is the major distribution area of the dromedary in Algeria. The two other areas are the Northern Sahara (37%) and the steppe (7%). The main camel breeds in Algeria are the Chaambi, Ouled Sidi

Cheikh, Ait Khebbach, The steppe camel, Saharaoui, Targui, Ajjer, Reguibi and Ftouh. (Moula, 2023)

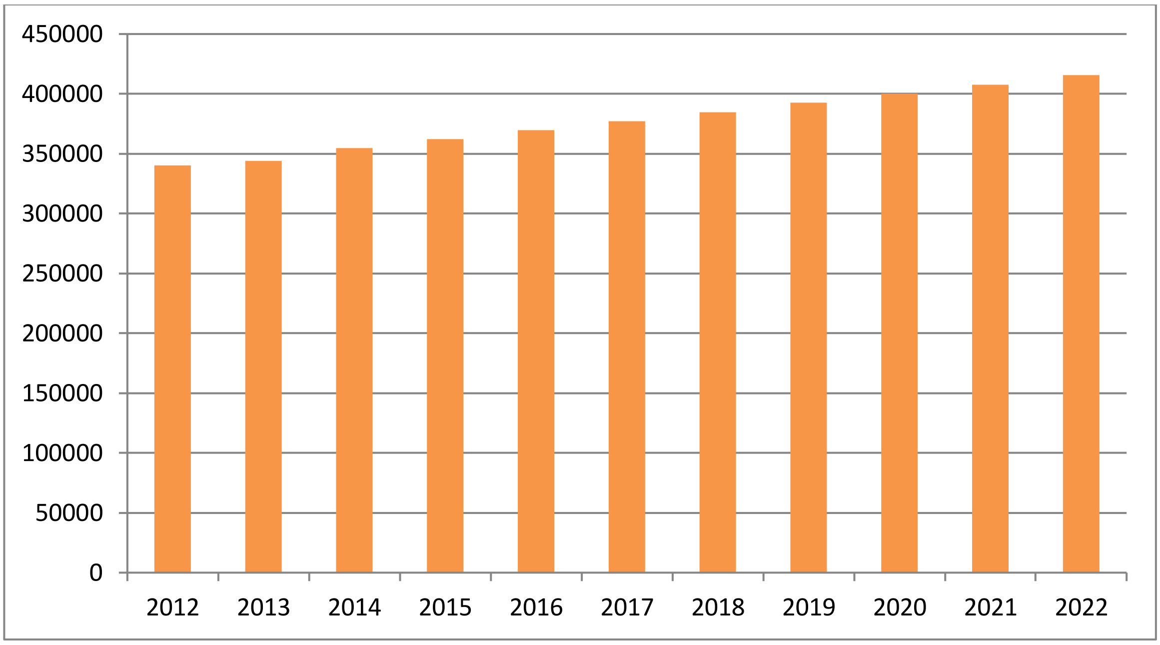


Figure 6: Evolution of camel population in Algeria (FAOSTAT, 2022)

I. 4.Camel breeds

There are, officially, 46 national entities in the world declaring camel stock. Among them, 20 countries are in Africa, 125 in Asia and one in Europe (Ukraine2). Regarding the geographical distribution of the two involved species, only dromedaries are found in African countries and in near and Middle Eastern and Southern Asian countries and only Bactrians inhabit Central Asia. The two species, however, are cohabiting in a few countries only, mainly in Kazakhstan (Figure 07). (Faye, 2020)

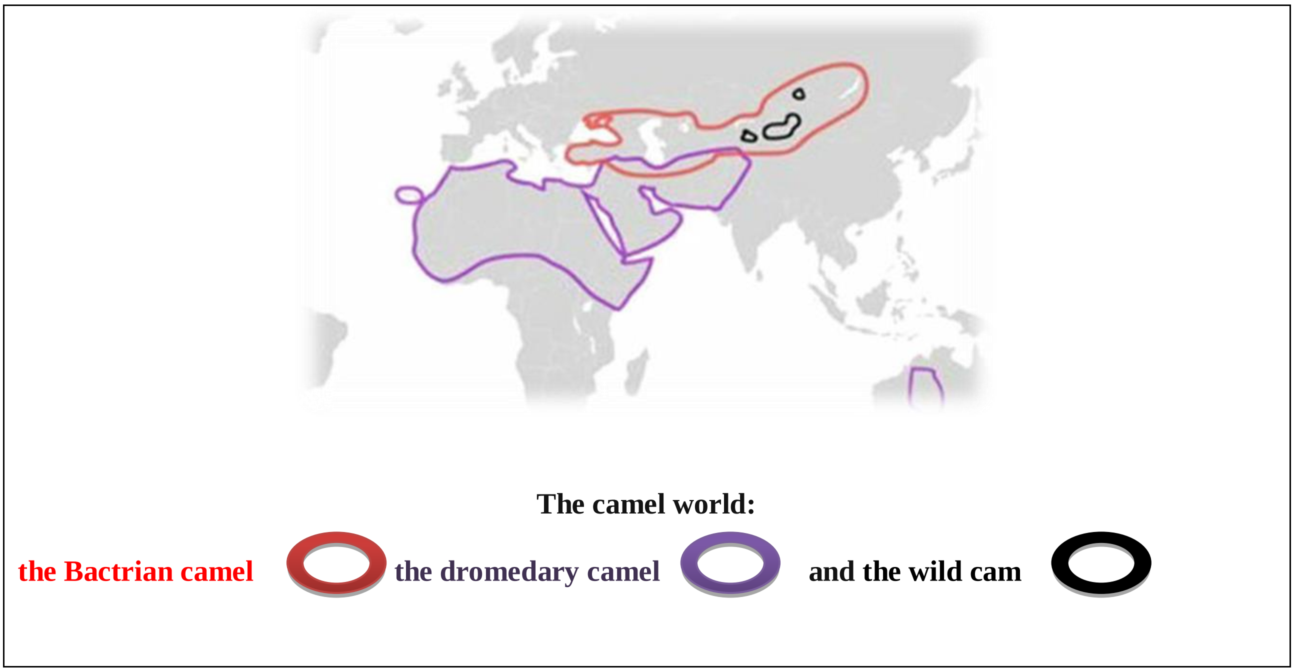


Figure 7: Distribution Maps of Camels Species in the world (Faye, 2020)

A- In Algeria

The breeds encountered in Algeria are found in the three North African countries; these are saddle, pack, and draft breeds (Bekki, 2023).

The breeds in Algeria:

1- Chaâmbi:

A heavy animal commonly used for transportation. It is only exceptionally used for riding and has a medium size. Its coat ranges from bay to ash with very dense short tufts of hair, particularly at the top of the hump and in the region of the withers and parotids. In the Southeast region, it is a meat breed. (Bekki, 2023)

2- Ouled Sidi Cheikh

It is a riding or pack animal, suitable for both stone and sand. It is quite large .It is found in the high plateaus of the Grand Erg Occidental. (Bekki, 2023)

3- Sahraoui

This breed is also a good milk producer and gains weight quickly. This population is found in all Saharan regions, as well as in Morocco, Algeria, Mauritania, and Mali. Its distribution territory extends from the Great Western Erg to the center of the Sahara. It originates from the crossbreeding of the Chaambi and Ouled Sidi Cheikh breeds. This is a fairly early-maturing breed, as females can be bred as early as three years old. They are considered as good milk Producer. (Babelhadi et *al.,* 2016)

4- Berberi

A slender animal, with well-muscled hindquarters, mainly found in Saharan and Tellian regions. It is very close to the Chaâmba and Ouled Sidi Cheikh. (Bekki, 2023)

5- Ait Khebbach

It's a powerful pack animal, it is short-legged and of medium size. It's found in the Southwest area. (Bekki, 2023)

6- The Steppe Camel

It's a common dromedary, small and stocky. It's used for close-range nomadism. It is found at the southern limits of the steppe. (Bekki, 2023)

7- Targui or Tuareg North breed

A dromedary of the Tuareg of the North. It's an excellent méhari but above all a renowned riding animal. Founded in the central Sahara, the Hoggar and the extreme south of Algeria (Tamanrasset). It's often encountered a little further north, because it is also widely used as a breeder. Indeed, it is the quintessential racing dromedary; it is very tall on fine and dry limbs, with a very short and fine grey coat. (Babelhadi et *al.,* 2016)

8- Ajjer

It is the dromedary of Tassili, resembling the Targui, differing only in size as it is shorter and has longer hair than the Targui. It is a riding dromedary, but is more often used as a pack animal. It is found in the Tassili region and in the South of the wilayas of Tebessa, El-Oued, and Biskra (Bekki, 2023)

9- Reguibi

It is a medium-sized dromedary with an ash-colored coat ranging from light to dark shades, used interchangeably for transportation or riding. It is found in the Southwest of Algeria, in the region of Bechar, Tindouf, and up to the Adrar region. (Bekki, 2023)

10- Aftoyth Camel

Draft and pack animal in the region of Tindouf and Bechar. The term Aftoyth is a genetic term that encompasses several types of dromedaries from the Western Sahara region and is characterized by a wide variety of coat colors ranging from light yellow to dark blackish. (Bekki, 2023)

I.5.Reproduction

Camels should begin coupling shortly after reaching sexual maturity or during subsequent breeding cycles during the first and second days of the oestrus if observable, as this period coincides with the maximum proportions of anatomical structures favoring fertilization. In case of undetectable estrus, it is advisable to introduce the male into the female group to facilitate fertilization. A male breeder can serve several times a day, with a ratio of one male for 25 to 30 females in a herd (Faye et *al.*, 2022)

Table 1: Some reproduction parameters in large camelids (interval and average). (Faye et *al.*, 2022)

Parameters	Interval	Average
Age of puberty	30-48 month	36 month
Age of the first calving	42-72 month	54 month
Gestation length	370-390 day	380 day
Interval between two calvings	15-36 month	24 month
Number of births per lifetime	3-8	6
Annual fertility rate	30-45%	35%

I. 6.Gestation

The reproductive system of the camel is characterized by a bifid but asymmetrical uterus. The developmental disparity is observed between the left and right uterine horns, with the first of them showing slightly higher progression. Despite the equal functionality of the right and left ovaries, the vast majority of pregnancies, 99%, occur in the left cornea of the uterus. (Bello and Bodinga, 2020).

The duration of the camel's gestation is long and usually lasts about 13 months on average. (Faye et *al.*, 2022)

I. 7.Breastfeeding

Milk production begins at the time of breastfeeding, but the amount of milk produced fluctuates throughout the period of lactation. This result depends on a myriad of factors: genetic, diet, interval between breeds, quality of stimulation, weather conditions, stress and know-how of the traitor or parameters of the machine to be drawn, the presence of diseases, especially mammitis, and of course the physiological stage of the female, i.e. beginning, middle or end of lactation. (Faye et *al.*, 2022)

The average duration of lactation ranges from 6 to 18 months (Alhadrami and Faye, 2022)

Chapter II: General Information about camel milk

II. 1.Definition of Milk

Milk is considered a fundamental element of human nutrition and plays an important role in launching a wide range of human productive activities. (Gradinaru, 2015)

Milk is produced by secretory cells in mammalian mammary glands. The purpose of this milk is mainly to feed infants. It contains a significant amount of protein and carbohydrates. Many dairy products are made from milk. (Vilain, 2010)

The Codex (Standard 206-1999) defines milk as the normal mammary secretion of trade animals obtained from one or more treatments, without adding or subtracting anything, intended for consumption as liquid milk or for subsequent processing.(FAO/OMS, 2014)

According to the International Congress for the Suppression of Fraud in 1909, milk is the product obtained through the complete and continuous trade of a healthy, well-being, well fed and not over-trained cow. It must be collected properly and free of colostrum. The Decree of 25 March 1924 specifies that the definition of "milk", without mention of the animal species of origin, is reserved for cow's milk. (Snappe et *al.,* 2010)

Liquid milk obtained from farm animals (such as cows, sheep, goats, and buffalo). Milk is generally heat-treated by pasteurization, ultra-high temperature treatment (UHT) or sterilization. This includes dried milk, partially dried, low in fat or whole. (FAO/OMS, 2014)

Raw milk, in its natural form, is considered a delicate physiological fluid due to the presence of microorganisms, the quantity and characteristics of which are influenced by various factors: the sanitary conditions of the animals and the people involved in the collection process, the cleanliness of equipment such as milking machines, buckets and tanks, the ambient temperature, and the time delay between collection and delivery to the processing plant. The primary objective of industrial processes is to purify the milk and improve its shelf life. (Kebir, 2018)

II. 2.Camelmilk

Camel milk is a traditional food that plays a crucial role in the diet of people living in semi-arid and arid regions. It has been consumed for centuries by nomadic populations and is recognized for its medicinal value. (Arain *et al.* 2023)

CM, recognized as the "white gold of the desert", enjoys great popularity among the Kazakh and Mongolian populations of Xinjiang and Inner Mongolia. In China, this milk stands out for its exceptional quality, offering significant nutritional benefits and positive health impacts. It is notably distinct from the milk of other ruminants. (Liu *et al.,* 2023; Saibhavana *et al.* 2023)

II. 3 Organoleptic Characteristics

CM, characterized by its white and opaque appearance, has a taste profile influenced by camels' eating habits and water consumption. The salinity and presence of natural foam are among the attributes inherent in camel milk. The distinctive olfactory characteristics of camel milk have deterred a large number of people from fully appreciating its flavor. (AL-Moosawi et *al.,* 2023; Baroin, 2011)

CM exhibits qualities of purity, a white hue, and a sweet taste. The consumption of desert shrubs by camels can result in the production of salty milk. Various physiological and genetic elements play a role in influencing milk production. (Al-Abdali, 2010)

The milky-white and opaque appearance is due to the fine dispersion of lipids in the milk, while flavor variations result from the composition of the feed and the quality of the water. (Abrhaley and Leta, 2018)

II. 4. Physico-chemical characteristics

Physical properties include measurements of the overall behavior of milk and its response to energy, while physicochemical properties study the relationship between this behavior and the energetic interactions concerning the colloidal constituents of milk, in particular the particles, molecules, atoms and ions. (McCarthy, 2021)

II. 4. 1. Density

The density of milk is influenced by various factors such as temperature, processing conditions, and animal type, with higher temperatures resulting in decreased density levels. (Parmar et *al.,* 2021; Parmar et *al.,* 2020)

The density of milk has been evaluated using three distinct analytical methodologies: a portable densimeter, a conventional desktop densimeter, and calibrated 100 cm3 glass pycnometers. (Parmar et *al.,* 2021)

There is a direct correlation between protein content and milk density, as well as an inverse correlation between milk temperature and milk density. An increase in protein content results in a corresponding increase in milk density. Conversely, a decrease in milk temperature results in an increase in milk density. (Suhendra et *al.,* 2020)

Abrhaley and Leta (2018) mention that the variation in the density of camel milk ranges from 1.026 to 1.035. Additionally, a study conducted by Tameur et *al.* (2021) reported a density of 1.030 for milk, which indicates a slight increase compared to that of cow's milk

II. 4. 2. Freezing Point

The determination of the freezing point of milk is of paramount importance as a key parameter of milk quality. This analysis primarily aims to detect the presence of adulterated water in milk and to quantify the level of water content present. (Zagorska and Ciprovica, 2013)

The freezing point of milk (FPM) is an immediate indicator of compromised technological quality of raw milk, particularly regarding dilution. The FPM may exhibit variability due to various factors related to changes in milk composition and properties. In addition to seasonal influence, stage of lactation, breed, milk yield, subclinical mastitis, among others, the consequences of nutritional deficiencies and metabolic irregularities play a significant role. The FPM represents a relatively constant physical attribute, influenced by osmotically active components. (Chládek and Čejna, 2005)

The FPM of camel is higher than that of bovine milk, ranging between -0.57 and -0.61°C. (Swelum et *al.*, 2021; Konuspayeva et *al.*, 2023)

II. 4. 3. pH

The pH scale quantifies the level of hydrogen ion activity in a solution by calculating the negative logarithm, base 10, of the hydrogen ion concentration present in the solution. (Aydogdu et *al.*, 2023)

The acidity or alkalinity level of milk is indicated by its pH. This parameter quantifies the presence of hydrogen ions in milk. The pH scale ranges from 0 to 14, where neutrality is represented by 7, acidity by values less than 7, and alkalinity by values greater than 7. In the field of milk fermentation, the evaluation of pH values plays an important role in assessing the quality of fermented dairy products and their acceptance by consumers. Jotić (2019)

The acidity or alkalinity level of milk is indicated by its pH. Camel milk has a pH range of 6.2 to 6.5, which is lower than that of cow milk. The highest buffering capacity of skim milk occurs at a pH of 4.95, determined by the concentration of hydrogen ions present in the milk. The pH values are influenced by these factors. (Abrhaley and Leta, 2018)

II. 4. 4. Titratable Acidity

Titratable acidity refers to the measurement of the total amount of acid present in a substance, which can be determined by titration. In the context of camel milk, the Titratable acidity has been found to be high, with values ranging from 0.71% to 0.87%. (Al-Zoreky and Al-Otaibi, 2015)

II. 5. Chemical and Biochemical Composition of camel milk

The composition of milk varies by species, geographical location, and the needs of the offspring. Milk is enriched with components that promote rapid growth of newborns; the sugar content of milk is associated with cognitive development, and the presence of milk fats is higher in animals living in colder climates. (Grădinaru, 2015)

The proportion of water present in CM fluctuates depending on the volume of water consumed by the camel; it can reach 89% in milk with daily water consumption by camels, or up to 91% with weekly one-hour water consumption by camels. Evidently, the camel tends to excrete more water when the natural environment is scarce, in order to ensure the well-being of the offspring or humans in general. (Al-Abdalall, 2010)

The complete chemical composition of camel milk compared to various species is presented in (Table 2).

Table 2 : composition of milk of different Species (Kebir, 2018)

Species	Water %	Protein %	Fat %	Ash %	Lactose %
Camel	86-88	3.0-3.9	2.9-5.4	0.6-0.9	3.3
Cow	85-87	3.2-3.8	3.7-4.4	0.7-0.8	4.8-4.9
Buffalo	82-84	3.3-3.6	7.0-11.5	0.8-0.9	4.5-5.0
Sheep	79-82	5.6-6.7	6.9-8.6	0.9-0.1	4.3-4.8
Goat	87-88	2.9-3.7	4.0-4.5	0.8-0.9	3.6-4.2
Human	88-89	1.1-1.3	3.3-4.7	0.2-0.3	6.8-7.0

Table 3 : Differences between camel and cow's milk (Ashour and Abdel-Rahman, 2022)

Parameters	Camel Milk	Cow Milk
Water (%)	90	87
Total Solid (%)	10.00	13.00
Fat (%)	2.00	4.00
Insulin (µu/ml)	40.50	16.30
Iron	0.05	0.27
Calcium (Mg/100g)	0.05	0.27
Potassium (Mg/100g)	152	140
Zinc (Mg/100g)	0.50	0.40
Vitamine C (Mg/ml)	35	10
Niacin (Mg/ml)	4.60	0.60
Panthothenic acid (Mg/ml)	0.90	3.80
B-lacto-globulin (Mg/ml)	0	3500
Whey acide protein (Mg/ml)	157	0
Peptydoglycon recognition	107	0
Protein (Mg/ml)		
B-lacto albumin (Mg/ml)	3500	1200
Kappa casein (%)	5.00	14.00
Casein micelles (µm)	320	160
Why protein (%)	1.00	0.80
Omega-6 (%)	3.50	5.20
Omega-7 (%)	11.60	2.30

Camel milk is a valuable reservoir of essential minerals, including calcium, magnesium, potassium, sodium, iron, zinc, and copper, as well as unsaturated fatty acids, low cholesterol, and various vitamins (see table 4). Furthermore, it contains proteins whose properties can prevent the growth of antibacterial impurities such as lactoferrin. Additionally, camel milk contains a wide range of bacteria, which suggests its potential use in the field of food production. (Boudalia et *al.*, 2023)

Table 4 : Contenu des minéraux et vitamines dans le lait de chamelle (Alhadrami and Faye, 2022)

	$Mg\ L^{-1}$
Minerals	
Calcium	1060.0 – 1570.0
Inorganic phosphate	580.0 – 1040.0
Copper	13.0 – 1.8
Iron	1.3 – 2.5
Magnesium	75.0 – 160.0
Manganese	0.1 – 0.2
Sodium	360.0 – 620 .0
Zinc	4.0 – 5.0
Les vitamines	
Retinol (A)	0.10 -0.15
Thiamine (B1)	0.33 – 0.60
Riboflavin(B2)	0.42 – 0.80
pyridoxine (B6)	0.52
Cobalamin (B12)	0.002
Niacin	4.6
Folic Acide	0.004
Pantothenic Acid	0.88
Tocopherol	0.53
Ascorbic Acid	24 - 36

CM is devoid of beta-lactoglobulin, a protein known for its strong allergenicity, but it is richer in lactoferrin and immunoglobulins, with a lower concentration of caseins, and is characterized by the presence of particular peptides such as WAP or PGRP. (Alhadrami and Faye, 2022)

Camel milk contains a higher concentration of phospholipids than cow and goat milk. Furthermore, an observation has been made regarding the atypical nature of phospholipid fatty acids in camel milk, deviating from the norm observed in ruminant herbivores. The phospholipid fatty acids present in camel milk are notably rich in linoleic acid (C 18:3 n-3) and long-chain polyunsaturated fatty acids. It is important to note that the fatty acid composition of camel milk is likely to be modified depending on the dietary intake. (Alhadrami and Faye, 2022)

Camel milk is rich in various vitamins, including vitamin A, vitamin E, and vitamin C. The concentration of vitamin A in camel milk depends, in particular, on the techniques used for its production and the parity order. (Mohamed and El Zubeir, 2023)

The amounts of Na, K, Zn, Fe, Cu, Mn, niacin, and vitamin C present in CM are high, while those of thiamine, riboflavin, folacin, vitamin B12, pantothenic acid, vitamin A, lysine, and tryptophan are comparatively reduced compared to cow milk. The milk fat presents a composition of 26.7% palmitic acid, 25.5% oleic acid, 11.4% myristic acid, and 11.0% palmitoleic acid. (Nikkhah, 2014)

II. 6. Microbiological Characteristics of Camel Milk

Microbes are responsible for the fermentation of milk and various biochemical processes, while playing a vital role in milk transformation. (Pepi and Focardi, 2022)

Recent research using high-throughput 16S rRNA sequencing has examined the composition of the microbial population present in milk, revealing the presence of diverse microbial communities. The favorable temperature conditions and the abundant nutritional composition of the mammary gland create an environment conducive to the proliferation of microorganisms. (Zhu et *al.*, 2023)

In pasteurized and raw milk, the residual quantity of microorganisms from different categories has been determined: mesophilic, psychrotrophic, lactic acid, heat-resistant, and spore-forming. An observation revealed that during the pre-pasteurization of raw milk, the main part of the microflora was composed of psychrotrophic and mesophilic microorganisms, representing up to 70%, while the share attributed to the lactic microbiota reached 25%; heat-resistant bacteria and spore-forming bacteria were observed at 4% and 0.8%, respectively. (Kukhtyn et *al.*, 2023)

II. 6. 1. Lactic Acid Microflora

The Lactic Acid Flora of Camel Milk Often Exhibits Notable Diversity, Characterized by the Presence of Various Bacteria Such as Lactococci, Enterococci, Lactobacilli, Leuconostocs, Streptococci, and Weissella. These Bacterial Species Include Lactococcus lactis subsp. Lactis, Lactococcus lactis subsp. Cremoris, Lactococcus lactis subsp. Lactis biovar diacetylactis, Enterococcus faecium, Enterococcus faecalis, Enterococcus duralis, Enterococcus durans, Lactobacillus cremoris, Lactobacillus lactis, Lactobacillus plantarum, Lactobacillus acidophilus, Lactobacillus acidophilus, Lactobacillus, among others, Lactobacillus casei, Lactobacillus sakei, Lactobacillus Bavaricus, Lactobacillus brevis, Lactobacillus pentosus, and Lactobacillus rhamnosus. These strains have been frequently identified in raw or fermented camel milk in various geographical regions. (Saidi, 2020)

II. 6. 2. Contamination Microflora

It is composed of pathogenic flora and spoilage flora.

II. 6. 2. 1. Pathogenic Flora

Pathogenic bacteria transmitted through the consumption of milk can cause many serious outbreaks. (Onoharigo et *al.*, 2022)

Bovine mastitis, a major disease in the dairy industry, is caused by various pathogens, including Streptococcus dysgalactiae, Escherichia coli, Prototheca spp., and Streptococcus uberis, which are primarily of environmental origin. (Lucheis et *al.*, 2023)

II.6. 2. 2. Spoilage Flora

Microorganisms responsible for deterioration, such as Klebsiella spp., Escherichia coli, Enterococcus faecalis, and Streptococcus faecalis, can spoil both branded and unbranded milk, underscoring the importance of appropriate handling and storage techniques. (Onoharigho et *al.*, 2022)

Colonization modifies the nutritional characteristics of edible products, including their taste, composition, and nutritional content, and sometimes generates harmful secondary compounds that can lead to food spoilage. (Onoharigho et *al.*, 2022)

II. 7. Pasteurization of Camel Milk

Pasteurization is a widely used technique for sanitizing foods, implemented with the aim of eradicating harmful bacteria and reducing enzymatic processes. Additionally, it allows for extending the storage life of goods for a limited period. Traditional pasteurization methods employ continuous heat transfer processes to eliminate microorganisms present in liquid food products such as milk and fruit juices. The traditional inactivation of these organisms is achieved through a heat treatment that may modify the sensory characteristics and nutritional composition of the product. (Azizi-Lalabadi et *al.*, 2023)

Pasteurization techniques differ in terms of temperature and duration to achieve specific degrees of microbial safety and product quality. Various methods are commonly used, including Low-Temperature pasteurization (15 to 30 minutes/60 to 65°C), high-temperature short-time (HTST) pasteurization (15 to 40 seconds/70 to 75°C), and high-temperature pasteurization (1 to 2 minutes/85 to 95°C). (Chethouna et *al.*, 2022)

Figure 8: Pasteurized camel milk (Dongo, 2022)

II. 7. 1. Low-Temperature Pasteurization

Low-Temperature pasteurization, generally around 60 to 65°C for duration of 15 to 30 minutes, represents a frequently used technique to extend the shelf-life of various food products. (Inanoglu et *al.*, 2022)

According to (Chethouna et *al.*, 2022), the application of a gentle pasteurization treatment to camel milk at 63°C for 30 minutes was effective in preserving the nutritional properties of the milk. This approach also ensures the stability of the physicochemical and chemical composition of the milk.

This approach helps to preserve essential elements such as vitamin C, proteins, and fats. Furthermore, cold pasteurization is considered the most appropriate option for preserving the antioxidant qualities of camel milk, making it a promising natural reservoir of antioxidants for the food and nutraceutical industries. (Hamouda et *al.*, 2022)

II. 7. 2. High-Temperature Short-Time (HTST) Pasteurization

High-temperature short-time (HTST) pasteurization is a preferred technique for rapidly pasteurizing donor milk, as it effectively ensures microbiological safety while preserving bioactive components. (Escuder Vieco et *al.*, 2023)

HTST pasteurization involves subjecting the milk to temperatures ranging from 72 to 75°C for 15 seconds, which is a much faster procedure compared to traditional techniques such as

Low-Temperature long-time (LTLT) pasteurization. (Nanthagopal and Ramaveerapathiran, 2022)

The application of HTST treatment has the potential to improve the rheological characteristics of fermented camel milk by increasing viscosity and particle size, while preserving its shear-thinning properties. Additionally, the use of HTST pasteurization in camel milk processing could lead to improved sensory characteristics and reduced syneresis, suggesting promising advantages for the quality and stability of the final product. (Arslan amin et *al.,* 2024; Hamouda et *al.,* 2022)

II. 7. 3. High-Temperature Pasteurization

The milk is subjected to temperatures ranging from 85°C to 95°C for one to two minutes using various techniques, which may involve direct exposure to steam or indirectly through the use of heat transfer equipment such as plate heat exchangers. (Fryč et *al.,* 2023)

II. 8. Milk Production

In camel milk production, yield and milk quality are the key factors for evaluating the performance of camels, and the nutritional composition of the milk affects the milk quality. This is due to factors such as breed, their size, age, stage of lactation, level of nutrition, feeding management, and sampling techniques. (Liu et *al.,* 2023)

The lactation period of the camel varies from 9 to 18 months. The average daily production seems to be in the range of 2 to 6 liters in extensive farming. Versus 12 to 20 liters in more intensive farming (chouia and kasmi, 2022)

Individual production varies between 1,000 and 2,700 liters per lactation in Africa, but can reach 7,000 to 12,000 liters according to some sources. LHOSTE (2004)

Camel milk production varies from 3.5 to 40 L per day in intensive management. Lactation lasts between 9 and 18 months, with a peak production occurring during the first 2 to 3 months after calving. (Nikkhah, 2014)

II. 8. 1. Global milk production

Over the past decades, an increasing interest in CM, resulting from a growing market demand due to its nutritional and health-beneficial properties, has been observed. (Chamekh et *al.,* 2020)

The annual global production of CM is estimated at 4.1 million tons in 2022. With 69, 9 % of Milk production in Africa and 30, 1% in Asia (Figure 9).

Table 5: The milk production in the top 10 milk-Producing countries (FAO, 2022)

Contries	Production (ton)
Somali	10920026.55
Kenya	1018110.96
Pakistan	9740000
Mali	294248.58
Ethiopie	220446
Saudi Arabia	1371020
Niger	106597.35
United Arab Emirates	79434.44
Soudan	673509.01
Chad	30968.27

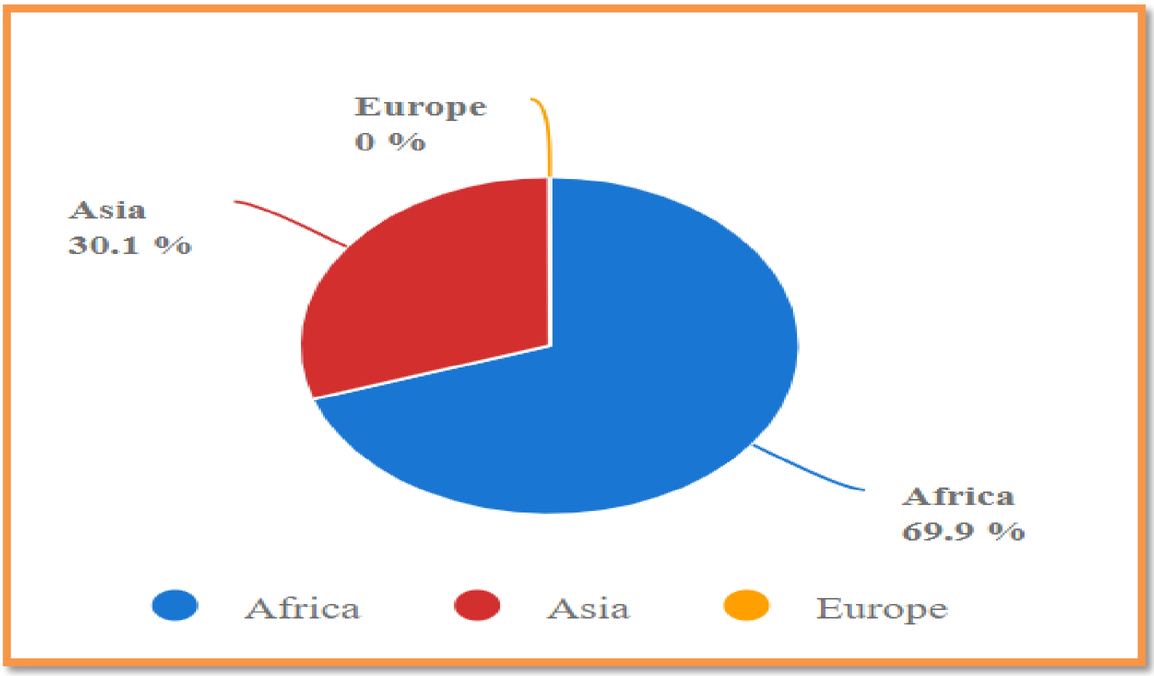


Figure 9: Production Share of Raw milk of camel, sum 2022; Source FAOSTAT (March 17, 2024)

II. 8. 2. Milk production in Algeria:

According to the statistics of FAO for 2022 there are 41,772,353 heads of camel worldwide and 459,616 heads in Algeria.

The production quantity in Algeria is expected to reach approximately 15,518.05 tons in 2022, as indicated by the Food and Agriculture Organization of the United Nations (FAO), and 88,000 tons according to estimates by specialists. This increase is significantly higher than that of the previous decade, as illustrated in the figure 10.

In the nomadic system, the average daily milk production was particularly deficient, as it was less than three liters per day; conversely, its importance was overshadowed by the transhumant system where production varied between four and five liters per day, presenting satisfactory physicochemical characteristics despite a poor bacteriological quality. (Boudalia et *al.*, 2023)

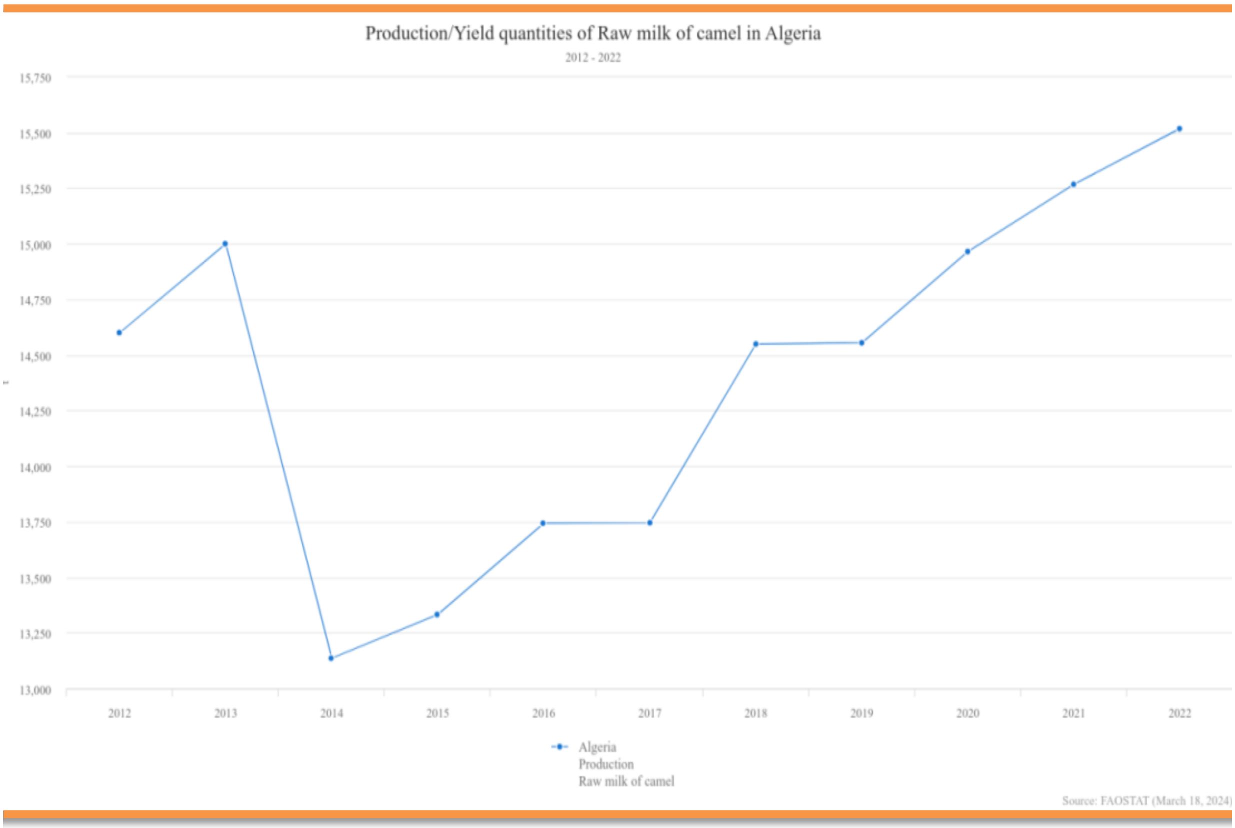


Figure 10: Production/Yield quantities of raw milk of camel in Algeria 2012-2022 (Source FAOSTAT March 18, 2024)

II. 9. Medicinal and therapeutic use of camel milk

Previous research has provided evidence highlighting the distinctive nutritional characteristics of camel milk and its derivatives, particularly a higher concentration of bioactive proteins and vitamin C compared to traditional animal milks. Furthermore, these products exhibit a range of biological functions, such as anti-diabetic, anti-infectious, anti-fatigue, anti-cancer, and anti-arthritic properties.(Li et *al.*, 2023 ; Chethouna et *al.*, 2022)

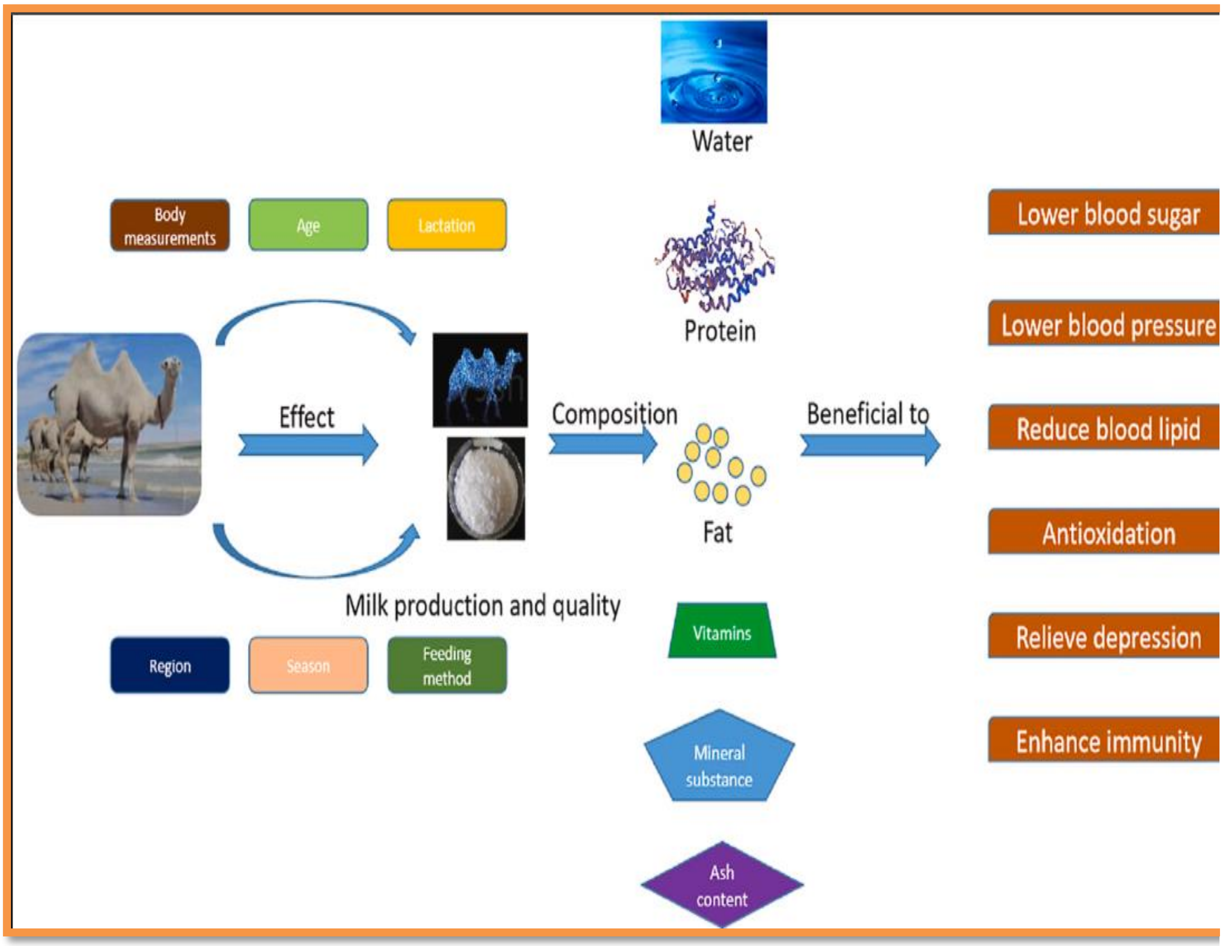


Figure 11: Body measurement and health effects of camel milk (Li et *al.*, 2023)

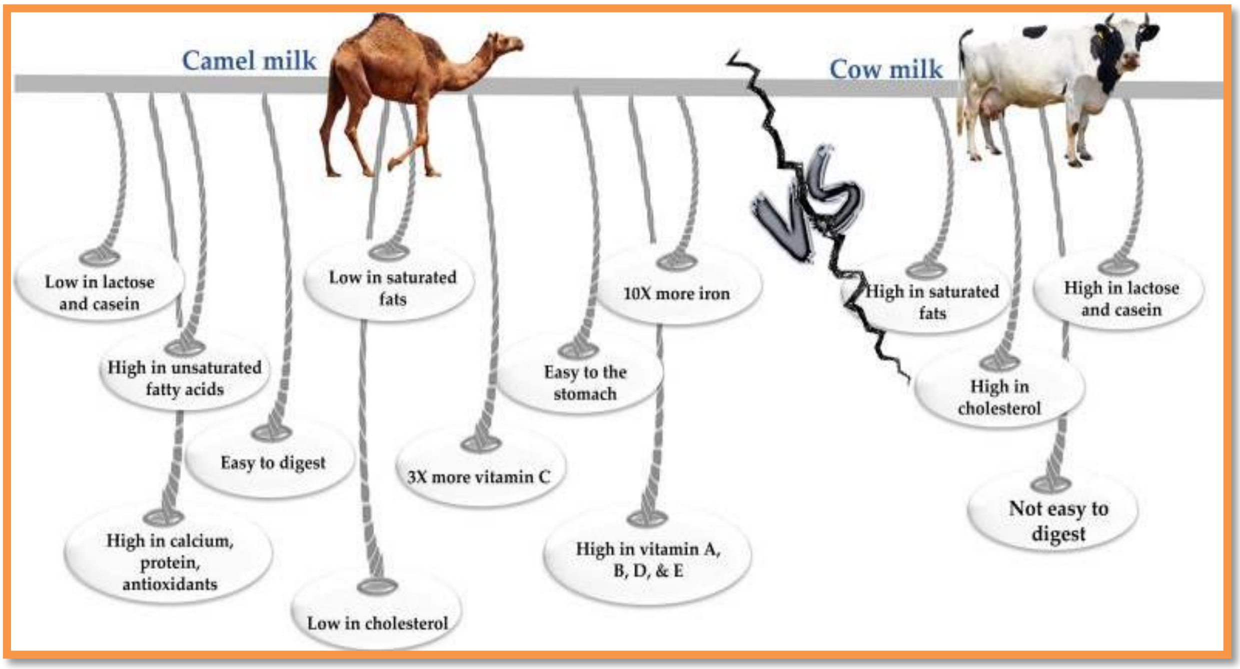


Figure 12: The Difference between camel milk and Cow milk. (Swelum, 2021)

The following table N 06 highlights the various therapeutic properties of camel milk and the specific molecules responsible for these beneficial effects. Camel milk is known for its many health benefits, especially due to its unique bioactive components.

Table 6: The using of raw camel milk as a therapeutic agent in certain diseases (Saidi, 2020)

Treatment against	Involved Molecules
Diabetes	Insulin as a molecule (Nano-bodies)
Hepatitis C virus	Amylase and Lactoferrin
Allergy	Low β-casein content and absence of β-lactoglobulin
Liver and kidney function	Alanine amino-transferase and aspartate aminotransferase
Slimming properties	Low protein and cholesterol content
Bacterial infections	Lysozyme and lactoperoxidase
Nutritional supplements	Unsaturated fatty acids
Strengthening the immune system	Peptidoglycan recognition
Lack of lactate and easy assimilation	L-lactate
Bone formation	High calcium content
Diarrhea	High sodium and potassium content

II. 9. 1. Antimicrobial en factors

Camel milk is rich in niacin (essential amino acids), valine, leucine and phenylalanine, as well as unsaturated fatty acids, antimicrobial constituents (e.g. lysozyme, lactoperoxidase and lactoferrin), prostaglandins and insulin. (Chethouna et *al.*, 2022)

Table 7: Bioactive Molecules with immunological function in camel milk (Saidi, 2020)

Molecule Name	Function
Heavy Chain Antibodies (HCAbs) or Variable Heavy Chain Antibodies (VHHs) or Nanobodies	**Capable of interacting with the least immunogenic part of the antigen Greater tissue penetration but similar specificity Rapid renal clearance in humans**
Peptidoglycan Recognition Protein (PGRP)	**Stimulates the immune response and has antimicrobial activity**
Lactoferrin	**Prevents pathogen invasion and microbial growth**
Lactoperoxidase	**Anti-tumor activity Bactericidal against Gram-negative bacteria like E. coli, Salmonella, and Pseudomonas Bacteriostatic against Gram-positive bacteria**
Lysozyme	**against Gram-positive bacteria**
N-acetyl-glucosaminidase (NAGase)	**Antimicrobial and antiviral activity**

II.9. 1. 2. Lactoferrine

Lactoferrin, a cationic glycosylated globular protein, is known for its non-toxic nature and was initially identified as the red protein present in mammalian milk. Subsequently, it has been recognized as a protein that binds iron by sequestering free Fe3+ and Fe2+ ions, thus being classified in the category of metalloproteins. This protein is present in various exocrine secretions of mammals, including colostrum, milk, uterine secretions, saliva, tears, and seminal fluid. (Steijns and Van Hooijdonk, 2000)

Lactoferrin exhibits robust antimicrobial activity against a wide range of pathogens and plays a role in targeted immune reactions. Its efficacy extends to potential therapeutic applications for various pathophysiological conditions. The essential biological roles related to lactoferrin supplementation are illustrated in a visual representation in figure N 15. (Ashraf *et al.*, 2024)

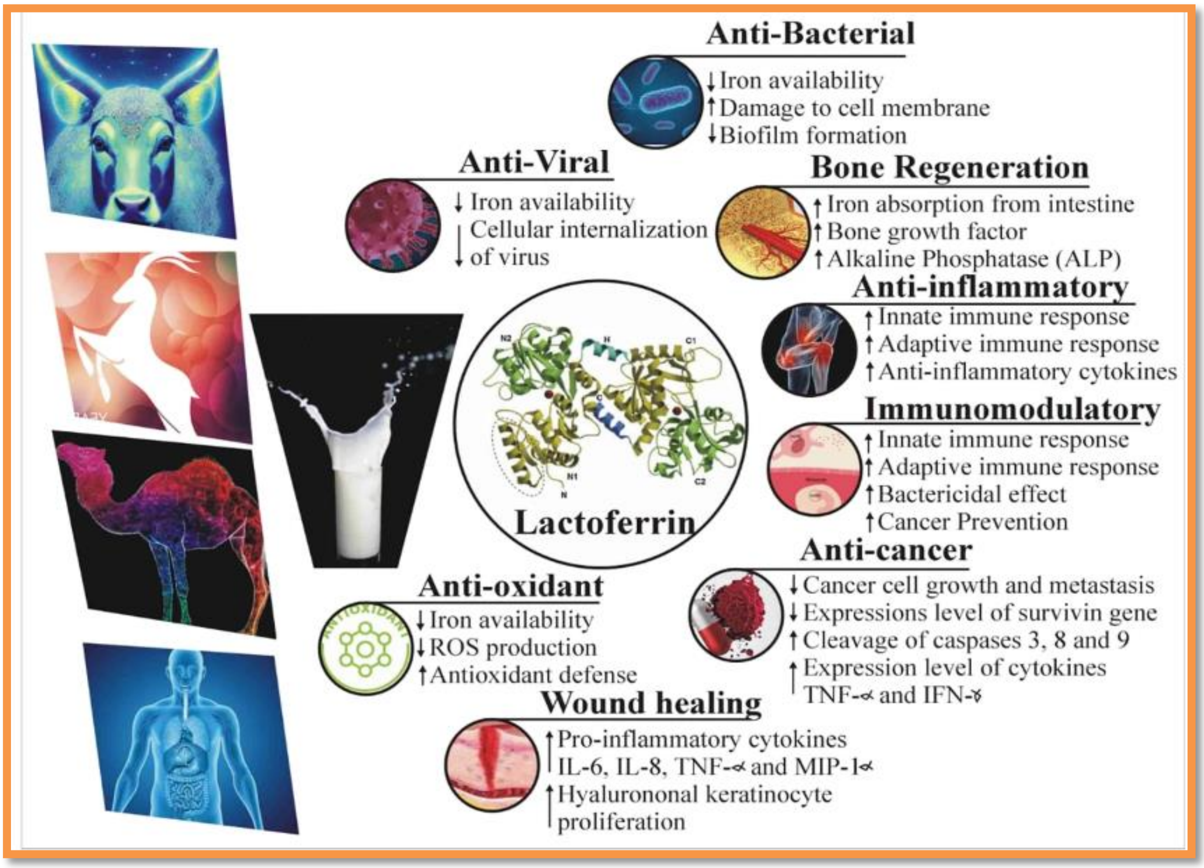


Figure 13: A schematic diagram illustrating the various therapeutic effects of Lactoferrin and the underlying mechanism of action. (Ashraf et *al.*, 2024)

Lactoferrin has demonstrated promising multidimensional and comprehensive beneficial effects, including anti-inflammatory, immunomodulatory, antioxidant, antibacterial, antiviral, neuroprotective, bone-repairing properties, and protection against various health issues such as cancer and lifestyle-related disorders like obesity, gastrointestinal problems, hypertension, diabetes, hyperlipidemia, among others.(Ashraf *et al.*, 2024; Jańczuk *et al.*, 2022)

CM contains lactoferrin, the second most abundant protein after casein, which fulfills various biological functions. More specifically, this protein is involved in the modulation of iron levels, the enhancement of immune system activity, and defense against pathogens due to its bacteriostatic and/or bactericidal properties. (Khan *et al.*, 2021)

The antimicrobial attribute of lactoferrin derived from camel milk can be attributed to its peptides, namely lactoferricin and lactoferramphin, located on the N-terminal region. Both CM lactoferrin and lactoferrin peptide hydrolysates have played a significant role in the management of typhoid fever. (Khan *et al.*, 2021)

II. 9. 1. 2. Lysozyme

Lysozyme plays a crucial role in the innate immune system, demonstrating powerful antimicrobial properties against various pathogens, including bacteria, fungi, and viruses. This protein acts as a protective agent against infections, as a natural antibiotic, and enhances the efficacy of conventional antibiotics, while also strengthening the immune response. (Regland and Criss, 2017)

In the pharmaceutical sector, lysozyme is used to alleviate various bacterial, viral, fungal, and inflammatory conditions, while demonstrating immunostimulant and antihistamine properties. (Nawaz *et al.*, 2022)

II. 9. 1. 3. Immunoglobulins

An antibody, also called an immunoglobulin, is an important Y-shaped protein that is primarily synthesized by plasma cells and used by the immune system to neutralize pathogens, including pathogenic bacteria and viruses. (Kumar *et al.*, 2022)

Immunoglobulins (Ig) are important constituents of colostrum and milk. They are essential proteins that provide crucial immunological protection against dangerous bacterial and viral diseases that pose a potential threat to the survival of the offspring. These proteins constitute the primary protective barrier. (Borad and Singh, 2022)

Camels possess two distinct categories of antibodies in their serum. In addition to the classical heterotetrameric antibodies (IgG1), camels possess exceptional antibodies characterized by a single functional chain (IgG2 and IgG3). Camel IgG2 and IgG3 lack light chains, comprising only heavy chains, and are therefore classified in the category of heavy-chain antibodies (HCAbs). (El Sheikh *et al.*, 2020)

The peak level of IgG in camel colostrum shows a notable increase, reaching 50.44 ± 3.36 mg/ml, which is significantly higher compared to the 0.26 ± 0.232 mg/ml reported in other ruminant species. Moreover, this peak typically manifests between 18 and 30 hours after birth. (Agrawal et *al.*, 2022; Sehrawat and Singh, 2004)

II. 9. 1. 4. Lactoperoxidase

Lactoperoxidase (LPO) is a glycosylated antimicrobial protein found in milk, with a molecular weight of 78 kDa. Known for its biocidal properties, it acts as both an effective antibiotic and antiviral agent. (Singh *et al.*, 2022)

LPO is the second most abundant enzyme in milk, with its primary function being to protect the mammary gland and the infant's intestines against bacterial infections. In milk, LPO exhibits remarkable thermal resilience and serves as an indicator to evaluate the efficiency of milk pasteurization; its optimal activity occurs at a pH of 6.0. Fluctuations in enzyme concentrations are subject to various factors such as the animal's reproductive cycle, the season of predominance, dietary regimes, and breed classification. The LPO system has the ability to prevent the proliferation and metabolic processes of various strains of microorganisms, demonstrating its adaptability for use at ambient temperature, thus constituting a viable substitute for refrigeration in the preservation of raw milk. Furthermore, the application of the LPO system could improve both the caliber and the safety of fermented milk derivatives. (Buys and Seifu, 2021)

LPO is a protein known for its antimicrobial properties that find applications in various industries. This protein plays an important role in the overall antimicrobial and antioxidant properties of camel milk. (Sheikh et *al.,* 2019), Furthermore, a protein analysis (proteomics) revealed the presence of the lactoperoxidase isoform 1 preproprotein in CM, highlighting its existence and potential functional importance. Despite the challenges in determining the three-dimensional configuration of LPO in camels, the use of homology modeling techniques has allowed a better understanding of its structure and interactions with substrates such as hypothiocyanate. (Sheikh et *al.,* 2019; Khan et *al.,* 2021)

II. 9. 2. The Anti-Cancer Factor

The potential anticancer properties of CM could be elucidated thanks to the existence of protective proteins such as IgG, IgA, lysozyme, lactoperoxidase, and lactoferrin. The antioxidant characteristics of camel milk protein hydrolysates have been demonstrated, indicating that they can be incorporated into functional food products. (Khan *et al.*, 2021)

Ayyash et *al.* (2018) illustrated that the use of probiotic bacteria derived from camel milk in the fermentation process enhanced the anticancer properties of camel milk. This was achieved through the inhibition of the angiotensin-converting enzyme (ACE), α-amylase, and α-glucosidase.

Camel milk plays a role in the regulation of apoptotic pathways, thus halting the growth of cancer cells. (Khan *et al.*, 2021) , The impact of camel milk on the proliferation of human cancer cells was studied by Korashy et *al.* (2012) using an in vitro model involving human hepatocellular carcinoma (HepG2) and human breast cancer (MCF7) cells. Their analysis supported the notion that camel milk prevented the proliferation of HepG2 and MCF7 cancer cells by increasing the activity of mRNA and caspase-3, as well as by stimulating death receptors within the HepG2 and MCF7 cell lines.

II. 9. 3. The antioxidant Factor

The antioxidant potential of camel milk and its ability to modulate genes involved in the inhibition or reduction of cancer cell proliferation, as well as in the suppression of genes that promote their growth, have been the subject of extensive research. (Konuspayeva *et al.,* 2007)

The table N 08 below shows the different experimental studies on the antioxidant properties of camel milk and isolated peptides from camel milk. These studies were conducted on various animal models and human cells. The results highlight the impact of camel milk on increasing antioxidant genes, preventing oxidative damage, and improving overall health.

Table 8: Antioxidant properties of camel milk (Khan et *al.*, 2021)

Agent	Properties	Experimental animal	Authors
Camel milk isolated peptides	Antioxidative	Camel	(Ganzorig *et al.*, 2020)
Camel milk isolated peptides	Antioxidative properties, increase CAT and SOD gene expression	HepG2 cells (human)	(Homayouni-Tabrizi *et al.*, 2016)
Camel milk	Antioxydative properties	Rats	(Mansour *et al.*, 2017)
Camel milk	Prevent oxidative damage, increase expression of antioxidant genes catalase, GPx and SOD	Rats	(Meena *et al.*, 2016)
Camel milk	Prevent CCL4-induced liver damage, increase antioxidant activity	Rats	(Sadek *et al.*, 2016)
Camel milk Protein hydrolysates	Antioxidative properties, increase CAT and SOD gene expression	Rats	(Kilari *et al.*, 2021)
Camel milk	Antioxidant and anti-apoptotic properties	Rats	(Almehdar et *al.*, 2019)
Camel milk	Antioxidant and decrease oxidative stress markers (malondialdehyde, myeloperoxidase and total antioxidant capacity) in lung tissue	Rats	(Zhu *et al.*, 2016)
Fermented Camel milk	Prevent CCL4-induced oxidative stress and increase antioxidant activity in liver	Rats	(Hamed *et al.*, 2018)
Fermented Camel milk	Prevent CCL4-induced oxidative stress and increase antioxidant activity in kidney	Rats	(Hamed *et al.*, 2018)
Camel milk	Antioxidant activity, prevent the damage caused by CCL4	Rats	(Hamed *et al.*, 2018)
Camel milk	Enhance the antioxidant gene expression	Rats	(Korish *et al.*, 2015)

II. 9. 4. Stimulating Factors: Vitamin C

Vitamin C, also known as ascorbic acid, is one of the water-soluble vitamins in mammals, its presence being influenced by the glucose level in the liver. There are two biologically important forms of vitamin C, the active L-ascorbic acid and the oxidized L-dehydroascorbic acid. The latter also has biological importance, although a considerable portion of vitamin C is converted to diketogulonic acid due to extensive oxidation in the rumen. Diketogulonic acid is devoid of essential functions, which makes organisms more vulnerable to vitamin C deficiency-related diseases, such as scurvy and mastitis. (Semsmia and Abed AL-Rahim, 2022)

CM presents a particularly high concentration of vitamin C compared to other animal species, exceeding cow's milk by a factor of 3 to 5. This attribute contributes significantly to its nutraceutical importance, especially for populations residing in arid regions. (Zhao et *al.*, 2015). The importance of the high concentration of this particular compound lies in its powerful antioxidant properties, which inhibit the proliferation of bacteria in the milk, thus prolonging its shelf life. Furthermore, it plays a crucial role in promoting the synthesis of collagen proteins through the hydrogenation process of the amino acids lysine and proline. (Jilo and Bai, 2016).

The concentration of vitamin C in camel milk generally ranges between 15-435 mg/l, with this variability attributed to various factors, including breed. It has been observed that the vitamin C content of milk from Bactrian camels (Camelus bactrianus) was higher than that of dromedary camels (Camelus dromedarius) and hybrid camels. Specifically, concentrations of 169 mg/l, 146 mg/l, and 133 mg/l were recorded for Bactrian camels, dromedary camels, and hybrids, respectively, as well as other variables such as seasonal fluctuations in feed quality. (Semsmia and Abed AL-Rahim, 2022)

Chapter III: General Information about Camel Urine

III. 1.Definition of Urine

Urine is a complex solution generated in the kidneys of humans and animals by the metabolism of endogenous wastes, beverages, medications, and foods. (Patel et *al.,* 2020)

Urine from different animals has been used for the treatment of mental illnesses, leprosy, leucoderma, amenorrhea, hemorrhoids, poison, tuberculosis, flatulence, loss of appetite, abdominal tumors, anemia, colic, abdominal hypertrophy, and dropsy. The animals whose urine has been used for these treatments include the donkey, camel, horse, elephant, buffalo, sheep, and goat. (Al-Abdalall, 2010; Thakur, 2004)

Urine, although a waste product of the body, nevertheless possesses many medicinal properties, and as such, is used both internally and externally as a medicine. Al-Abdalall (2010)

Urine is primarily composed of water, with small quantities of urea, uric acid, salts (phosphates, sodium oxalates, calcium, etc.), and certain hormones in varying proportions. Although it is a waste product of the body, it nevertheless possesses many medicinal properties, which have been recognized by ancient medical practitioners, and as such, it has been used both internally and externally as a medicine. In addition to human urine, the urine of animals such as goats, sheep, buffaloes, elephants, horses, camels, and donkeys was also widely used as remedies for the treatment of worms, dropsy, abdominal enlargements, flatulence, colic, anemia, abdominal tumors, loss of appetite, tuberculosis, poison, hemorrhoids, amenorrhea, leucoderma, and various mental disorders. (Thakur, 2004)

Urine is also composed of organic compounds such as creatinine, creatine, and uric acid as major components. The various components of urine are primarily influenced by the composition of the essential elements in the urine. (Ajiboye et *al.*, 2022)

III. 2. Camel Urine

Urine has been used in medical treatment, and camel (dromedary Camelus) urine is one of the animal urines that have been widely used for therapeutic purposes. (Salamt et *al.,* 2021)

Camel urine is a traditional healing liquid with powerful antioxidant properties that are beneficial for health. It exhibits significant free radical scavenging activity and reducing power, thereby contributing to disease prevention. Additionally, CU has shown cytotoxic effects against various cancer cells, protective effects against hepatotoxicity and gastric ulcers, and anti-platelet activity. (Hasni et *al.,* 2022)

III. 3. Properties and Composition of Camel Urine

Urine represents a sterile and refined substance obtained through the blood filtration process; in mammals, it is synthesized by the kidneys and scientifically termed as the plasma ultrafiltrate. Its characteristics encompass sterility, transparency, and a faint yellow hue, comprising water-soluble constituents that are readily eliminated, such as urea, creatinine, organic acids, amino acids, ammonia, and inorganic salts. (Balhara et *al.*, 2023)

The chemical components of CU were initially investigated in the early years of the previous century, revealing the presence of creatine, creatinine, urea, chlorides, hippuric acid, as well as total nitrogen and ammonia. Notably, minimal amounts of urea and ammonia were detected. The noxious smell and toxic properties of camel urine are ascribed to these minimal quantities of urea and ammonia. Its mineral salt content is reported to be ten times higher compared to that of human urine. (Tahrawat et *al.,* 2023)

According to Jamal Bayad et *al.* (2019), CU had a dark, smoked, yellow color and smelled like sweet hay. It had a specific gravity of 1.045 to 1.055.The quantity of urine produced exhibited variability based on the timing of observation.

The analysis of camel urine composition has recently been investigated through the utilization of liquid chromatography and mass spectrometry. In a study by Antakly (2012), a variety of metabolites were identified in CU, with notable components such as benzoic acid, urea, creatinine, phenylacetate, citric acid, and hippuric acid. Interestingly, the levels of these compounds were found to be comparable to those observed in elephant and rat urine; notably, camel urine exhibited a higher abundance of benzoate salt. Furthermore, gas chromatography-mass spectrometry (GC-MS) has identified phenol, salicylic acid, p-cresol, cinnamic acid, azelaic acid, and benzoic acid as the principal bioactive acids present in camel urine. (Tahrawat et *al.,* 2023)

In the case of camel urine that has been aged between 5 to 10 years, it exhibits a density ranging from 1.01 to 1.07. The urea concentration falls within the range of 18 to 36 mg/dl, while the creatinine level ranges from 0.2 to 0.5 mg/l. The pH of this aged camel urine typically varies from slightly acidic to strongly alkaline. (Tharwat et *al.*, 2023)

Table 9: Inorganic Constituents in lyophilized camel urine estimated by ICP-MS (Tharwat et *al.*, 2023)

S. No.	Element Name (Symbol)	Concentration in ppm of camel urine
1.	Lithium (Li)	0.006807
2.	Boron (B)	0.031970
3.	Sodium (Na)	647.344755
4.	Magnesium (Mg)	15.105697
5.	Aluminum (Al)	0.159909
6.	Phosphorus (P)	0.471004
7.	Chlorine (Cl)	0.0002358
8.	Potassium (K)	280.7982
9.	Calcium (Ca)	0.023656
10.	Silver (Ag)	0.000300
11.	Chromium (Cr)	1.963761
12.	Manganese (Mn)	0.017308
13.	Iron (Fe)	1.705473
14.	Cobalt (Co)	0.001637
15.	Nickel (Ni)	0.086647
16.	Copper (Cu)	0.038132
17.	Zinc (Zn)	0.05477
18.	Arsenic (As)	0.000359
19.	Selenium (Se)	0.002612
20.	Strontium (Sr)	0.103021
21.	Platinum (Pt)	0.000239
22.	Gold (Au)	0.001787
23.	Mercury (Hg)	0.000258
24.	Tin (Sn)	0.000461
25.	Antimony (Sb)	0.001004
26.	Iodine (I)	0.000256
27.	Barium (Ba)	0.013403
28.	Cadmium (Cd)	0.000086

III. 4. Therapeutic Properties of Camel Urine

Urinary therapy, also known as urotherapy, is an ancestral approach used to treat various human conditions. The use of animal urine, derived from various sources such as goats, sheep, buffaloes, horses, camels and donkeys, is traditionally used to treat conditions such as abdominal tumors, hypertrophy, tuberculosis, hemorrhoids, colic and anemia. (Al-Abdalall, 2010)

CU is considered to be the best example of urotherapy and is thought to have more beneficial properties than other animal urine. It is composed of various chemical components that contribute to its anti-cancer, anti-platelet, gastroprotective and hepatoprotective properties (Salam et *al.*, 2021). A recent study conducted by (Anwar et *al.* 2021) showed that CU has cytotoxic characteristics without inducing clastogenic effects, as demonstrated by in vitro and in vivo studies indicating the absence of clastogenicity associated with CU. This finding implies that ulcerative colitis is considered safe and poses no threat of developing genetic toxicity

While the consumption of camel urine may be perceived as disagreeable, it has been extensively utilized for an extended period in the Arabian Peninsula, as well as in the United States, the United Kingdom, and additional European nations. The utilization of camel urine is regarded as a viable approach to address a range of health issues like febrile conditions, colds, and tumors. Typically, individuals either ingest it directly or infrequently combine a small quantity with camel milk (Abdel Gader and Alhaider, 2016).

Camel urine has shown its effectiveness as an anticlastogen, making it a promising candidate for drug development. Its potential applications in the areas of anti-cancer, anti-platelet, hepatoprotective and gastroprotective therapies have been documented in various studies. (Ajiboye et *al.*, 2022)

III. 4. 1. Anticancer Properties

Cancer represents a significant challenge to global public health, resulting in considerable levels of illness and death on a worldwide scale. Annually, a substantial number of new incidences are identified with elevated rates of mortality, positioning it as a primary contributor to fatalities. (Jürgen, 2022)

The diagnosis and management of cancer pose significant challenges in the field of medical care; there is currently no medical intervention capable of selectively targeting tumor cells while preserving the integrity of healthy tissues or the functions of vital organs. The purported therapeutic properties of camel urine have been subjected to careful examination, and recent findings suggest that camel urine may effectively eliminate various cancer cells at a tolerable dose. (Tharwat, 2023)

Although considered a by-product, camel urine has been found to be effective in the treatment of various types of cancer in the human body (Alhaider et *al.*, 2011). The potential anticancer properties of camel urine may be associated with its cytotoxic effects on HeLa cells, osteosarcomas, and human leukemic cells. Reports have indicated the suppression of C tumors in onions following the application of camel urine. (Ajiboye et *al.*, 2022)

Camel urine and its PMF "Protein Molecular Fraction" fraction have inhibited the growth of malignant cells, including brain carcinoma (glioma) (U251), colon carcinoma (HCT116), hepatocellular carcinoma (HEPG2), lung cancer, and leukemia. The anticancer action of camel urine could be attributed to direct cellular cytotoxicity or via an antiangiogenic mode of action by restricting the blood supply to tumor cells, or a combined action triggered by the pH. (Tharwat, 2023)

The observations of Professor Khorshid (which have led to three patents) have been confirmed by several experiments conducted with camel urine, In camel urine and its derived fraction (PMF), detectable quantities of essential trace elements such as copper and zinc are present in their molecular composition. Various scientific works highlight the importance of zinc in crucial physiological mechanisms, including the modulation of the immune response and intracellular metabolic pathways. Furthermore, zinc is a powerful antioxidant against harmful free radicals that damage cellular integrity by interfering with the activity of reactive oxygen species. (El-Shahawy et *al.*, 2010; Ali et *al.*, 2011)

III. 4. 2. Antibacterial Activity

Camel urine possesses powerful antibacterial properties, surpassing those of bovine urine due to its high alkalinity, salt content, and low levels of uric acid. The unique composition of camel urine has been attributed to their dietary preferences and the specific plants they consume, which are rich in minerals generally avoided by felines. Studies have also highlighted the antifungal capabilities of camel urine, being more pronounced at higher concentrations than when diluted. (Ajiboye et *al.*, 2022)

Camel urine has been shown to possess efficacy as an antimicrobial agent and is purported to be devoid of adverse effects on individuals. Nevertheless, the existing evidence indicates remarkable antimicrobial properties against certain pathogenic microorganisms that affect humans, including Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, and other pathogenic microbes. (Rofaei et *al.*, 2022)

The antimicrobial efficacy of camel urine can be attributed to various factors, including high levels of salt, alkalinity, and natural bioactive compounds derived from the vegetation consumed by camels, indigenous microflora, and secreted antimicrobial substances. Unlike the urine of other bovine species, camel urine exhibits an alkaline character due to elevated levels of potassium, magnesium, and proteinaceous compounds, as well as reduced concentrations of uric acid, sodium, and creatine. (Rofaei et *al.*, 2022)

The distinct composition of camel urine, in contrast to that of bovine and caprine species, results from their dietary preferences and consumption habits. More specifically, camels prefer woody plants with high mineral content, which decrease more slowly during desiccation than grasses and other forage varieties. Uric acid, sodium, and creatine are among the major concentrated components in camel urine. (Rofaei et *al.*, 2022)

Camel urine has demonstrated inhibitory effects on the proliferation of Salmonella isolates at different concentrations (25%, 50%, 75%, and 100%), creating a significant zone of inhibition against all Salmonella isolates. The neutralization of camel urine produced results comparable to or slightly better than those of alkaline camel urine. (Rofaei et *al.*, 2022)

III. 4. 3. Antifungal Activity

The urine of camels exhibits potent antifungal properties against both C. albicans and Nonalbicans Candida. (Mostafa and Dwedar, 2016)

Camel urine can be used to treat fungal infections such as pityriasisversicolor and tinea. (Rofaei et *al.*, 2022)

The antibacterial and antifungal effects of camel urine could be attributed to the presence of a high concentration of salt and its strong alkalinity, in addition to the presence of natural bioactive components from the plants consumed by camels, as well as certain resident bacteria and excreted antimicrobial agents.

Camel urine is alkaline, in contrast to the urine of other animals. It contains high concentrations of magnesium, potassium, and albumin-like proteins, and low levels of uric acid, sodium, and creatine. (Tharwat et *al.*, 2023)

III. 4. 4. Antiplatelet Activity

Preliminary research has revealed that camel urine is composed of various chemical components that play a role in its antiplatelet properties. (Salamt et *al.*, 2021)

(Alhaidar et *al.* 2011) have shown that camel urine completely inhibited human platelet aggregation induced by arachidonic acid (AA) and adenosine diphosphate (ADP) in a dose-dependent manner. The closure time of the PFA-100 using whole human blood was prolonged in a dose-dependent manner upon the addition of camel urine. Virgin camel urine was less effective in inhibiting ADP-induced aggregation compared to urine from lactating and pregnant camels; however, all three exhibited comparable inhibitory activity. Neither human urine nor bovine urine exhibited any antiplatelet activity.

Camel urine has a potent antiplatelet activity against platelet aggregation induced by ADP (similar to clopidogrel) and AA (similar to aspirin). (Alhaidar et *al.*, 2011)

According to (Al-Ghumas, 2020), the white CU exhibits a higher level of inhibition compared to black CU. This sets the stage for additional research endeavors aimed at elucidating the characteristics of this inhibitory substance and its prospective application as an anti-platelet agent for managing thrombosis.

The inhibitory activity of CU is suggested as an important characteristic of its anti-cancer properties, as it could help prevent platelet aggregation and reduce the metastatic potential of cancer cells. Furthermore, CU has also prolonged the closure time of the PFA-100. The PFA-100 is a test that measures the duration for which platelets can block total blood flow, and it is a measure of overall platelet-related hemostasis. (Paniccia et *al.*, 2015)

The presence of benzenpropanoic acid in ulcerative colitis CU is responsible for the anti-platelet effects; however, further research is essential to validate its role as a promising therapeutic antiplatelet drug. (Ahmad et *al.*, 2017)

III. 4. 5. Gastroprotective Activity

CU exhibits a significant anti-ulcer effect in three gastroprotective models:

1) HCl/ethanol (HCl/EtOH)-induced ulcer,

2) Water-immersion restraint stress (WRS)-induced ulcer, and

3) Non-steroidal anti-inflammatory drug (indomethacin)-induced ulcer.

Ulcer formation was confirmed by the presence of lesions and inflammation in the glandular part of the stomach (HCl/EtOH model), the appearance of hemorrhagic streaks throughout the gastric mucosa (WRS model), and extensive gastric mucosal lesions with blackish-red hemorrhagic spots (indomethacin model). Treatment with 5 ml/kg of CU showed 100% protection against gastroduodenal ulcers in the HCl/EtOH and WRS models. In the indomethacin model, CU showed a 66.7% inhibition of ulcers. Furthermore, CU demonstrated a 100% healing rate with no ulcers observed in the indomethacin-induced gastric lesions (healing model), compared to cimetidine, a standard drug, which only resulted in a 60.5% healing rate. This suggests that CU possesses better healing properties than cimetidine. This potent action is attributed to the presence of antioxidants in CU. It has been shown that trace elements, such as magnesium and zinc present in CU, reduce oxidative stress and help prevent tissue damage caused by a toxic agent (Shazia et *al.,* 2012).

Therefore, CU appears to strengthen the mucosal barrier against endogenous and exogenous ulcerogenic agents and has shown a strong healing effect on indomethacin-induced gastric lesions, which could be considered a potential treatment for ulcers in the future (Salamt et *al.,* 2021).

Experimental Studies

Chapter I: Materials and Methods

I.1.Materials

I.1.1. Biological materials

I.1.1.1. Raw milk samples

16 individual milk samples were collected on May 13th from a herd of camels (Camelus dromedarius) comprising the Ouled Sid Chikh, Rguibi and Targi populations. These populations originate from the high steppe region of southwestern Algeria, specifically the El Bayadh province, particularly the El Kheiter municipality. The herd is associated with semi-intensive farming practices. The lactating camels were in a healthy physiological condition during the sampling period (36 h up to the 13th month of lactation).

I.1.1.2. Urine samples

12 individual urine samples were the subject of the investigation in the context of this particular study, all of which were obtained on May 15th. These samples came from camels belonging to the dromedary herd (Camelus dromedarius) of the Ouled Sid Chikh, Rguibi and Targi regions. The dromedaries in question are located along the routes of the commune of EL Kheiter, wilaya of El Bayadh, and the study was conducted as part of a large-scale breeding program.

I.1.2. Equipment

- Bench-top pH meter Starter 2100
- Magnetic stirrers, with and without heating
- Centrifuge
- UV-visible spectrophotometer
- Analytical balance
- Lactoscan (Milk Analyzer)

(APPENDIX 03)

I.1.3. Small equipment

Micropipettes, hemolysis tubes, beakers, Erlenmeyer flasks, volumetric flasks, burettes, graduated cylinders, gloves, 2 ml micro spectrophotometric cuvette.

I.1.4. Réactifs and solvants

Methanol, Distilled water, NaOH (0.5 M), Phenolphthalein, 2,2-Diphenyl-1-picrylhydrazyl (DPPH), Folin-Ciocalteu, Sodium carbonate, Gallic acid, Gelatin, Biuret, Ascorbic acid, TCA, Sodium nitrite ($NaNO_2$) 5%, Aluminum chloride ($AlCl_3$) 10%, Vitamin C, Sodium hydroxide ($NaOH$) 4%

I. 2. Methods

The different steps of the methodology followed in the present study are schematized in the figure below.

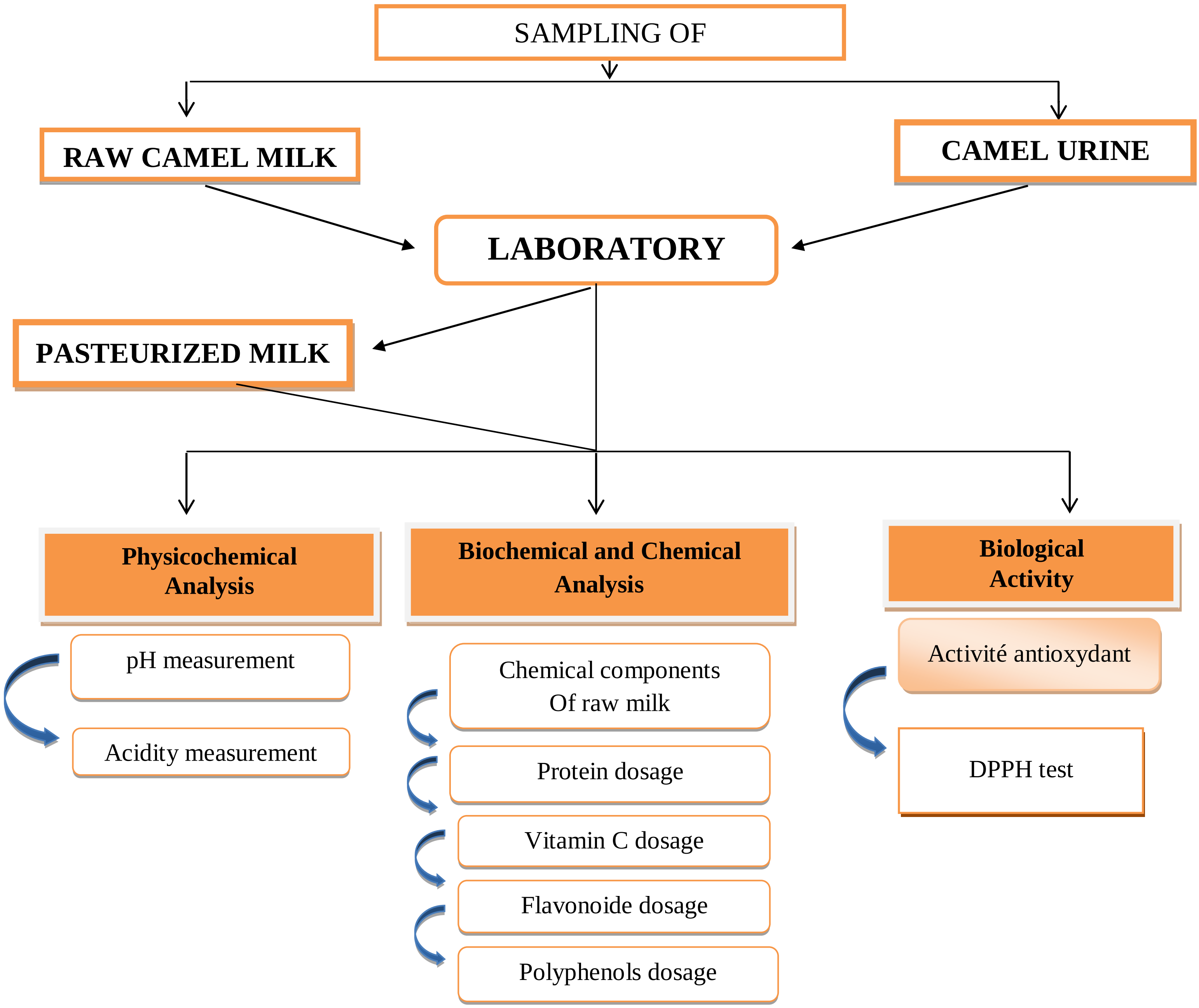


Figure 14: Experimental Protocol

I. 3.Collection of Milk

The Milk was collected in May, from healthy camels under strict hygienic conditions. This process involved wearing gloves and cleaning the udder with 70% alcohol before milking. The milk was collected in plastic flasks, which were immediately placed in a cooler containing refrigerant blocks and transported to the laboratory of the Faculty of Natural and Life Sciences at Abou Baker Belkaid University.

(APPENDIX 01)

I. 4. Collection of Urine

Urine was collected in May from healthy camels and placed in flacks, which were then transported to the laboratory of the university. (APPENDIX 02)

I. 5. Physicochemical Analysis

I. 5. 1. pH measurement

The pH (potential of Hydrogen) is a measure of the acidity or basicity of an aqueous solution. It is defined as the negative decimal logarithm of the activity of hydrogen ions (H+) in solution. In other words, pH measures the concentration of H+ ions in a solution and thus indicates its degree of acidity or basicity.

The pH is determined using a calibrated pH meter. The value is read directly on the screen of the device (Bench-top pH meter Starter 2100). (APPENDIX 02; FIGURE 02)

I. 5. 2. Determination of the Dornic acidity

The milk acidity, measured in Dornic degrees, represents the volume of 0.11 N sodium hydroxide needed to neutralize 10 ml of milk with phenolphthalene as an indicator (1°D equals 0.1 g of lactic acid per liter of Milk). Dornic acidity indicates the freshness of milk and its content of caseins, phosphates, citrate, hydrogencarbonate and lactates. (Siboukeur, 2011)

A sample of 10 ml of raw milk is placed in a beaker and then 3 drops of phenolphthalene are added. Then a titration in stirring with soda (N/9) is added (to the burette) until the turn to the rose. The pink coloration must persist for at least 10 seconds.

Acidity is given by direct reading of the volume (ml) of sodium poured and the application of the following formula: **AT=V x 10(D°).**

AT: Titratable acidity in dornic and **V** degrees: the volume of sodium in ml corresponds to the burette drop

I. 6. Biochemical and Chemical Analysis

I. 6. 1. Chemical components of raw milk

The chemical composition of the milk samples was determined using a Lactoscan. The Lactoscan is an electronic device that uses various sensors and techniques to analyze the composition of milk samples. It provides measurements for parameters such as:

- Fat
- Solids-non-fat (SNF)
- D° Dornic
- Protein
- Conductivity electric
- Freezing Point
- Sels
- Lactose.

The analysis results are displayed on the screen within 50 seconds but can also be printed on paper, as the Lactoscan has an integrated printer.

I. 6. 2. Protein dosage

The determination of protein in RCM and PCM is carried out using the Lactoscan MILK ANALYSER.

And the concentration of protein in the 50/50 mixture of PMU and RMU is determined using the biuret method.

Operating Procedure

Calibration of the instrument by using a standard protein solution at 10 g/L in Gelatin equivalent, prepare a wide range of solutions under a volume of 1 ML.

Use physiological water as a diluent.

- In a test tube, introduce: 500µl of the Sample
- After add 2ml of biuret reagent
- Then Vortex the tubes and incubate for 5 minutes.
- Read the absorbance at 540 nm against a blank.

I. 6. 3. Vitamin C dosage

Vitamin C, also known as ascorbic acid, is an enzyme cofactor for biochemical reactions catalyzed by monooxygenases, dioxygenases and mixed function oxygenases. Vitamin C Functions are: (Kükürt and Gelen, 2024)

- Serves as an antioxidative agent, safeguarding cells against oxidative harm.
- Supports the functionality of the immune system.
- Stimulates collagen synthesis, which is significant for skin, skeletal structure, and supportive tissues.
- Facilitates the absorption of iron.
- Participates in the synthesis of neurotransmitters and the operation of the nervous system.

The determination of Vitamin C is carried out using the technique of (Lowery, 1951).

Operating Procedure

- In 1ml of the sample add 0, 5 ml of 10% trichloroacetic acid (TCA) solution
- Mix it thoroughly using a vortex mixer for 1 minute.
- Allow the mixture to stand for 10 minutes at room temperature
- Centrifuge the sample at 3000 RPM for 10 minutes to separate the protein precipitate from the supernatant.
- Transfer 0.75 ml of the supernatant to a new tube. Add 0.75 ml of distilled water and 150 µl of Folin-Ciocalteu reagent (diluted 1/10) to the supernatant .Vortex the mixture and incubate it at room temperature for 15 minutes.
- Measure the optical density (OD) at 769 nm using a spectrophotometer against a blank (distilled H_2O).
- Determine the concentration of vitamin C (µg/ml) from the calibration curve.

I. 6. 4. Flavonoid dosage

Flavonoids are polyphenolic compounds responsible for the coloration of flowers, certain fruits, and are also prescribed in the treatment of disorders related to venous insufficiency, the treatment of hemorrhoidal crisis, in ophthalmology for disorders related to retinal circulation, metrorrhagia linked to the presence of an intrauterine device, and the treatment of capillary fragility at the skin and mucous membrane level. In addition to pharmacological properties: antimalarial (licocalcone A), cytotoxic, anticancer, antimicrobial, antiviral, and anti-inflammatory. (Hammoudi and Oumouna, 2021)

The determination of flavonoids is carried out using the technique of (Zhishen et *al.*, 1999).

❖ **Operating Procedure**

- A quantity of 500µl of the diluted extract is mixed with 1500µl of distilled water followed by 150µl of 5% sodium nitrite ($NaNO_2$).
- After 5 minutes, 150µl of 10% aluminum trichloride ($AlCl_3$) is added to the mixture. After 6 minutes of incubation at room temperature.
- 500µl of 4% sodium hydroxide (NaOH) is added.
- Immediately, the mixture is thoroughly agitated to homogenize the contents.
- The absorbance is determined at 510 nm against the blank using a SPECORD 200 Plus spectrophotometer. A calibration curve is prepared in parallel under the same operating conditions using catechin as a positive control (APPENDIX 04)
- The flavonoid content is expressed in milligrams (mg) of catechin equivalents per gram of dry matter (mg CE/g DM).

I. 6. 5. Polyphenols Dosage

Water-soluble polyphenolic compounds are identified for their ability to precipitate alkaloids, gelatin, and proteins; when they cause the precipitation of proteins, they are commonly referred to as antinutritional factors. These compounds exhibit various properties such as antibacterial, antifungal, wound-healing, antihypertensive, antidiarrheal, venotonic, and ophthalmic decongestant properties (Hammoudit and Oumouna, 2021).

The determination of total phenols is carried out using the Folin-Ciocalteu reagent according to the method of (Singleton and Rossi, 1965). The reagent is reduced during the oxidation of phenol, into a blue mixture of tungsten and molybdenum oxides, and the absorption is measured using a spectrophotometer at 765nm.

❖ **Operating Procedure**

- A 200µl aliquot of the diluted extract was introduced into test tubes
- Then 1000µl of the Folin-Ciocalteu reagent diluted 10 times and 800µl of sodium carbonate (Na_2CO_3) at 7.5% were added.
- The tubes were agitated and kept for 30 minutes protected from light,
- The absorbance is determined at 769 nm against the blank using a SPECORD 200 using a SPECORD 200 Plus spectrophotometer.

- A calibration curve is prepared using Gallic acid as a positive control in order to express the contents in milligrams (mg) of Gallic acid equivalents per gram of dry matter (mg GAE/g DM). (APPENDIX 03)

I. 7. Antioxidant activity

Antioxidant activity pertains to the capacity of a material, such as a chemical compound or a biological molecule, to impede or postpone the oxidation process of other molecules. Oxidation represents a fundamental chemical mechanism that can give rise to the generation of free radicals and additional reactive entities, which have the potential to induce harm to cells, proteins, and other biomolecules.

Antioxidants are substances that possess the capability to counteract or eradicate these reactive entities, thereby averting or reducing the damage caused by oxidation. The antioxidant efficacy of a material is commonly assessed through a variety of in vitro and in vivo experiments that gauge its capacity to extinguish free radicals, bind metal ions, or hinder lipid peroxidation. Substances exhibiting robust antioxidant activity are highly valued across different domains, such as food science, pharmacology, and cosmetics, owing to their promising health advantages and capacity to mitigate oxidative stress.

The antioxidant activity was evaluated using the DPPH assay method, which was first described by Blois in 1958.

I. 7. 1. DPPH (2,2-diphenyl-1-picrylhydrazy) Free Radical Trapping Test

DPPH (1,1-Diphenyl-2-picrylhydrazyl) is a stable free radical with a violet color that absorbs at 517 nm. In the presence of anti-radical compounds, the DPPH radical is reduced and changes color to yellow. The absorbance measured at 517 nm is used to calculate the percentage of DPPH radical inhibition, which is proportional to the anti-radical power of the sample (Mahmoudi, 2012).

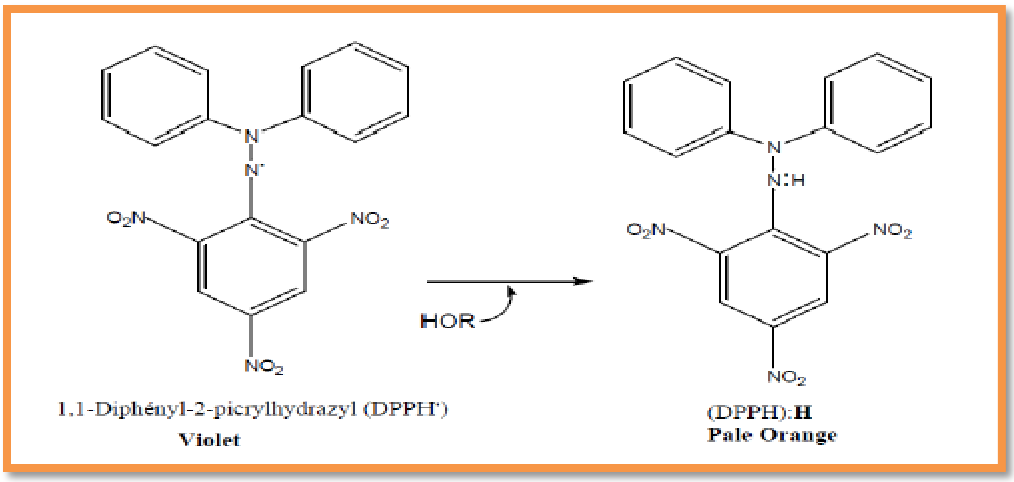


Figure 15: Reduction of DPPH Radical (Mahmoudi, 2012)

Operating Procedure

A volume of 50 µl of different concentrations (0.0625, 0.12, 0.25, 0.5, 1, 2, 4 mg/ml) of the extract is added to 1950 µl of freshly prepared methanolic DPPH solution (0.025 g/l). After incubation in the dark for 30 minutes at room temperature, the absorbance is read at 517 nm using a spectrophotometer. For the negative control, 50 µl of methanol is mixed with 1950 µl of the methanolic DPPH solution at the same concentration used. Ascorbic acid is used as a positive control.

The percentages of DPPH radical inhibition (%) are calculated using the following formula:

$$\textbf{Percentage of inhibition (I \%)} = [(AC-AE)/AC] \times 100$$

Where:

AC: represents the absorbance of the negative control

AE: represents the absorbance of the sample

I. 8. Extraction of total phenolic compound in milk and his mixture with urine:

This method is applied to measure the concentration of flavonoids, polyphenols, and the antioxidant power of milk and its mixture with urine.

Milk includes various antioxidant compounds. Exogenous compounds mainly include vitamins A, E, and C, whereas endogenous antioxidants include enzymes, glutathione peroxidase, catalase, superoxide dismutase, etc., and substances derived from proteins such as lactoferrin and coenzyme Q10. Compounds with recognized antioxidant activity include phenols, whose molecular structure includes at least one phenol group: an aromatic ring attached to at least one hydroxyl functional group. (Vázquez, 2015)

Phenols are secondary metabolites exclusive to plants whose function is mostly related to pigmentation and protection against pathogens and predators. There are over 10,000 different phenolic compounds, ranging from the simplest to the most complex, and their analysis and characteristics indicate their great diversity in nature.

The extraction of total phenolic compounds is determined by the method of (Vázquez, 2015) with modifications.

The procedures are as follows:

- Measurement and addition of 8 ml of milk in a 25 ml volumetric flask
- Addition of 10 ml of methanol-water solution (50%);
- Vortexing for 1 minute
- Stand for 6 hours
- Centrifugation (7800 x g, 15 minutes)
- Transfer of 4 ml of the supernatant to a new tube
- Obtaining the aqueous extract
- Complete the procedures of :
    1. Flavonoid concentration
    2. Polyphenol concentration
    3. Antioxidants Activity

I. 9. The pasteurization of the milk

The pasteurization of the milk was carried out following an adapted method from Chethouna et al. (2022) with modification. (Low-Temperature pasteurization)

Initially, 20 ml of milk were measured and placed in a beaker. Then placed on a magnetic stirrer with heating. A thermometer was used to monitor the temperature of the milk. Until we reached the temperature of 62°C.

The time required to reach this temperature varied between 15 and 30 minutes, depending on the specific conditions of each trial.

Chapter II: Results and discussions

II.1. Field Survey

The work carried out is located in the wilaya of El Bayadh (Figure 16) (El Kheiter commune), in the south-western part of Algeria, in the high steppe plains.

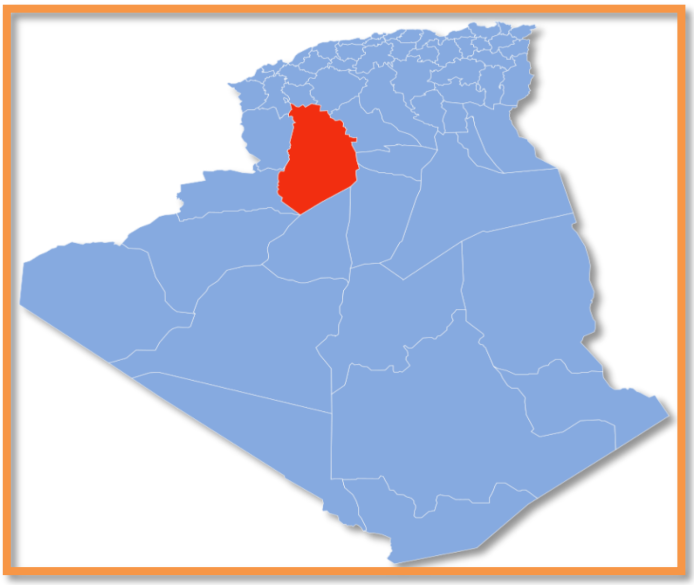


Figure 16: The Geographic map of the studies region

Our study covered an area of 1,023 km² (El Kheiter community) (Figure 17)

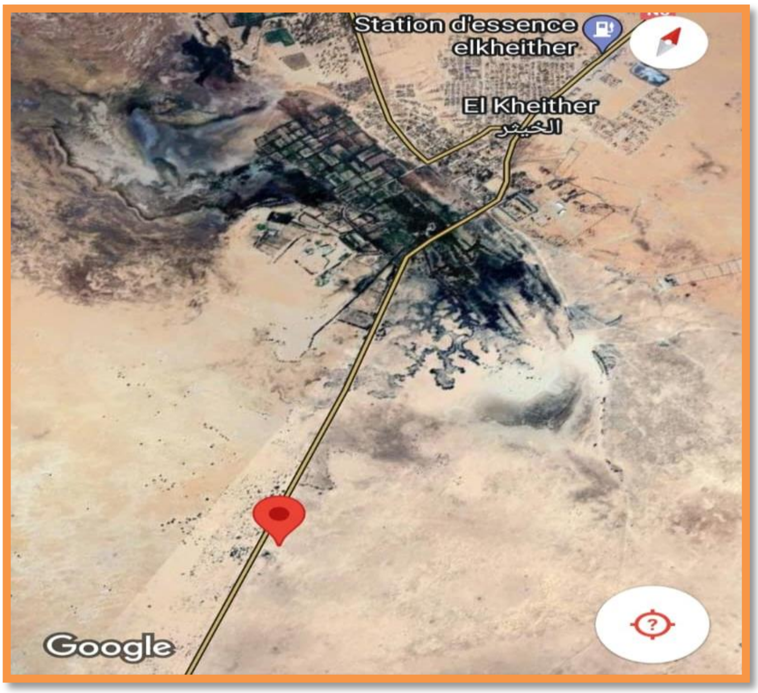


Figure 17: Geographic location of the region studied (Google Maps, 2020)

II. 2.Results of physicochemical analyze

The physico-chemical analyses of raw camel milk, pasteurized camel milk, urine, a mixture of raw camel milk and urine, and a mixture of pasteurized camel milk and urine are given in the table below.

Table 10: The result of physicochemical analyze

	Physicochemical Parameters	
	pH à 20°C	**Dornic Acidity**
Raw Milk	6,31 ±0,110	11,00±1,370
Pasteurized Milk	6,42 ±0,353	11,33± 1,154
Urine	6,56 ±0,832	_
Mixture of RM+U	6,49 ±0,575	19,83± 5,373
Mixture of PM+U	6,45 ±0,352	13 ± 1,732

II. 2. 1. pH

The average pH values obtained from the analysis are as follows: raw camel milk 6.31; pasteurized milk 6.42; urine 6.56; raw milk mixed with urine 6.49; and pasteurized milk mixed with urine 6.45.

These results indicate slight variations in pH between different samples. Raw camel milk had the lowest pH at 6.31, while urine had the highest pH at 6.56. Both raw and pasteurized milk showed an increase in pH when mixed with urine, suggesting a buffering effect of urine on milk acidity. However, when comparing the pH of raw milk (6.31) with other types of milk, camel milk is slightly more acidic than human milk (7) and cow milk (6.6).

Furthermore, the pH values of the analyzed samples of raw and pasteurized camel milk (6.31 and 6.42) are close to those reported by some authors such as (Chethouna, 2011), who reported a pH of 6.37, and (Sboui et *al.*, 2009) in Tunisia (pH 6.41).

These two milk pH values are lower than those obtained by (Khoidri and Chemla, 2020), (6.66); Siboukeur (2007), (6.6) and Khaskheli (2005) in Pakistan (6.77).

According to Debouz et *al.* (2014), the influencing factors of pH levels, such as the higher vitamin C content in camel milk, make camel milk slightly more acidic. The low pH of camel milk can be attributed to the high concentration of volatile fatty acids (Chethouna, 2011) and the type of feeding, i.e. the farming system.

The mean pH of the obtained urine (6.56) is lower than those reported by Khoidri and Chemla (2020), 6.7; Hasni and Habita (2015), 6.69; Rahem et *al.* (2016), 6.67; and Mezouari et *al.* (2020), 8.36.

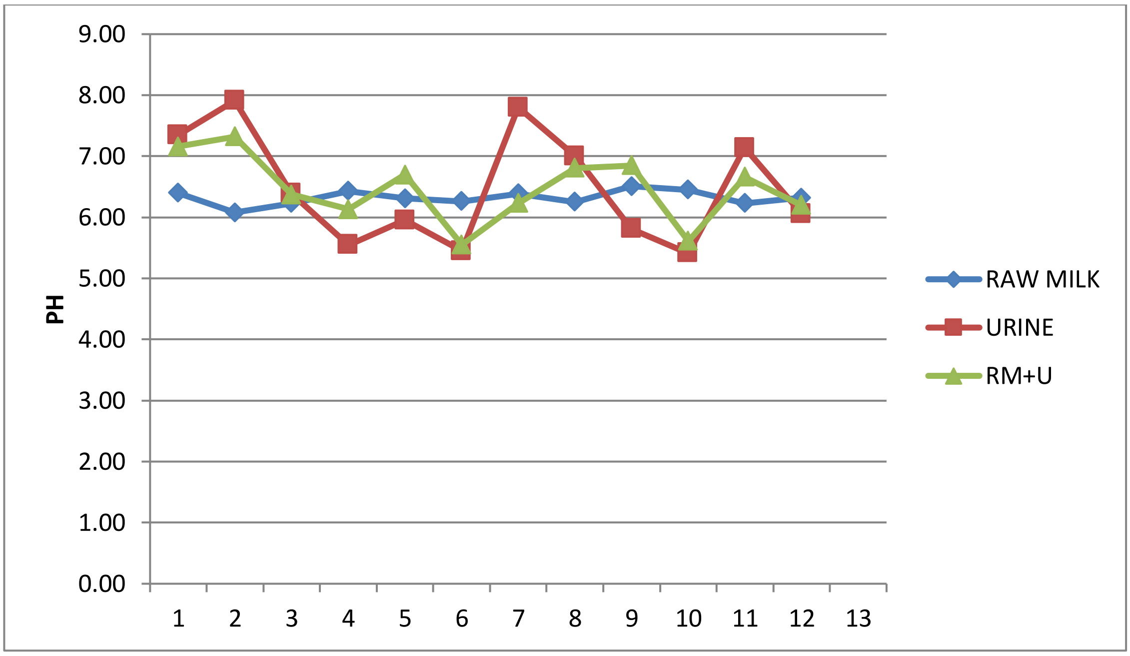


Figure 18: pH Comparison of Raw Camel Milk, Camel Urine, and Their Mixture (RM+U). (Original Figure)

- RM remains slightly acidic, consistent with previous findings that report pH values around 6.3
- Camel urine's pH fluctuates between slightly acidic to neutral, reflecting a broader range of metabolic excretions.
- The mixture's pH values typically fall within a neutral to slightly acidic range, demonstrating an averaging effect of the two components.

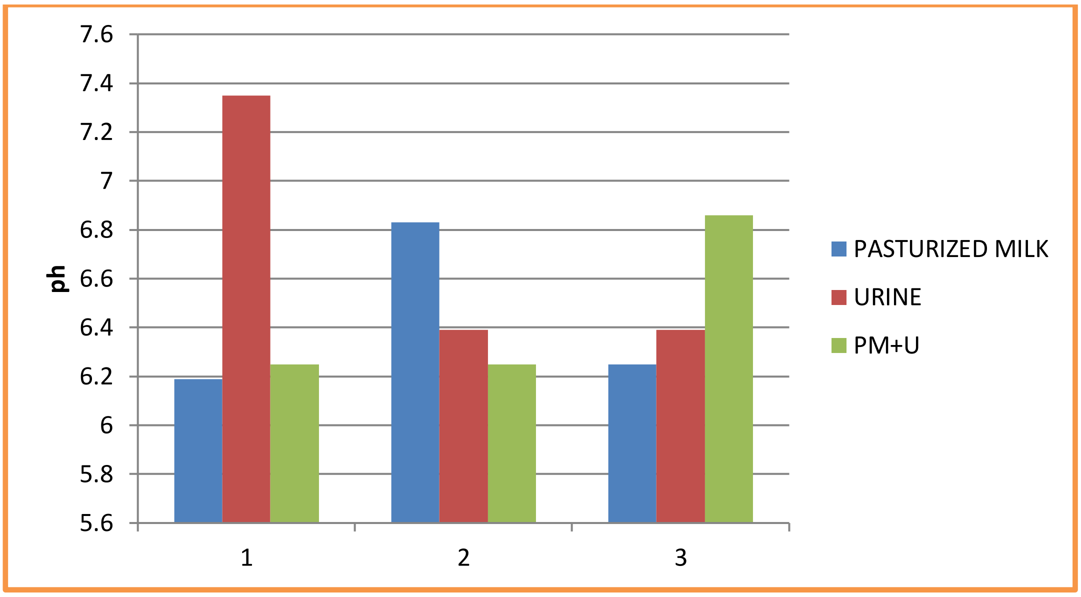


Figure 19: pH Comparison of Pasteurized Camel Milk, Camel Urine, and Their Mixture (PM+U). (Original Figure)

II. 2. 2. Dornic acidity

Before freezing the samples, the Dornic acidity of raw camel milk was measured using the Lactoscane, yielding an average of 11 D° ± 1.37. This value indicates that the milk has not undergone significant fermentation and is therefore fresh.

This acidity level is lower compared to that of bovine milk, which typically ranges around 15°D (Sawaya et al., 1984), and other reported values such as 18.7°D by Adjaine and Amiri (2013), 18.2°D by Siboukeur (2007), 17.2°D by Sboui et *al.* (2009), and 17°D by Debouz and Guerguer (2014).

The Titratable acidity of milk is contingent upon the concentration of acid molecules within the substance and exhibits an inverse relationship with its pH. Fluctuations in acidity commonly stem from differences in animal diet, environmental factors, and duration of lactation. Consequently, Titratable acidity serves as an indirect indicator of the abundance of caseins, phosphates, citrates, and bicarbonates in milk. (Mathieu, 1998)

We also measured the Dornic acid of the samples after thawing and pasteurisation to compare them with the milk-urine mixture. The results are shown in the table below.

Table 11: the Results of titrable Acidity

Samples	Titratable Acidity
Raw Milk	14,2 D°± 2,1778
Pasteurized Milk	11,3 D°± 1,1547
RM+U	19,8 D°± 5,3738
PM+U	13 D°± 1,73205

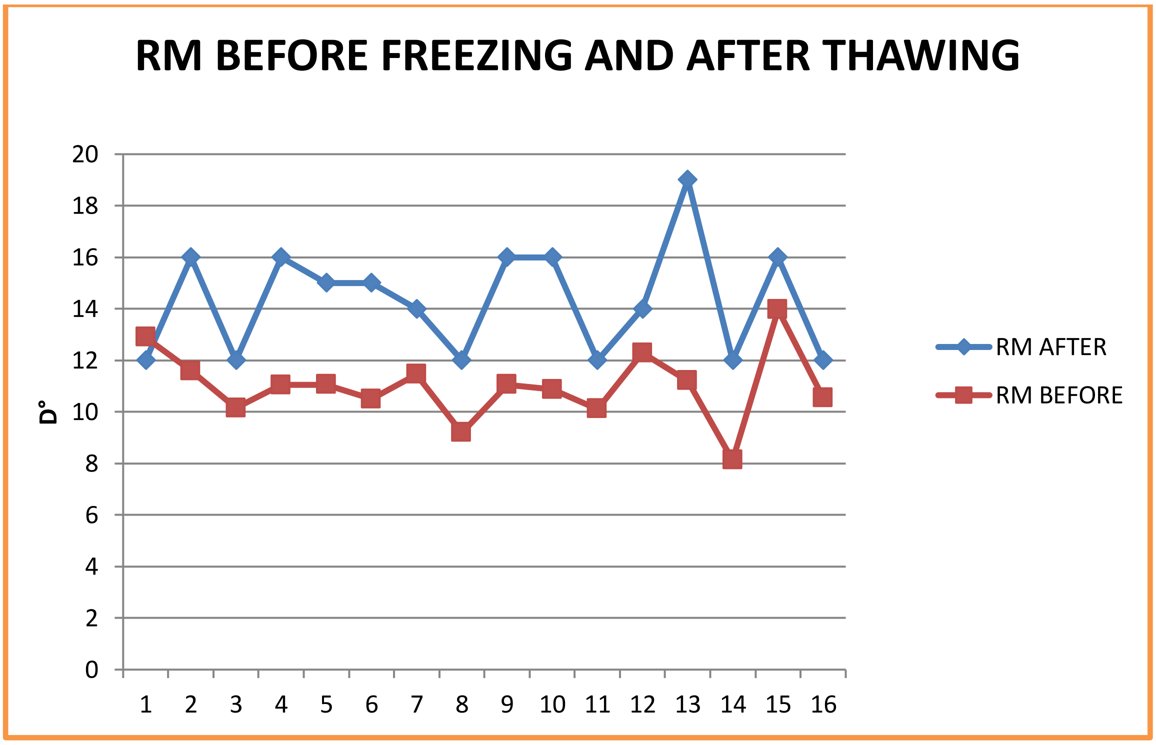


Figure 20: Variation in Milk Acidity Before freezing and after thawing (Original Figure)

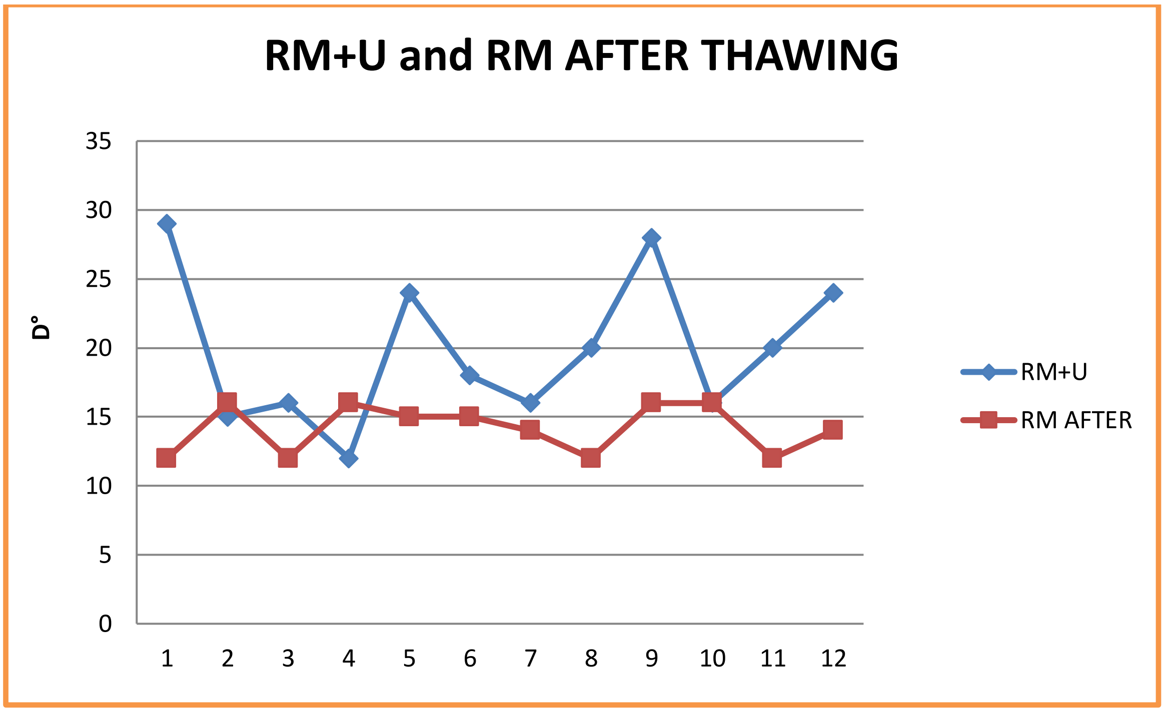


Figure 21: Variation in Milk Acidity after thawing and his mixture with urine (Original Figure)

The mixture of raw milk with urine increases the Dornic acidity, which indicates that urine influences and increases microbial activity.

Raw milk after thawing leads to a slight increase and greater variability in acidity values compared to raw milk before thawing.

The average Dornic acidity of pasteurized milk and the milk-urine mixture is shown in the following figure

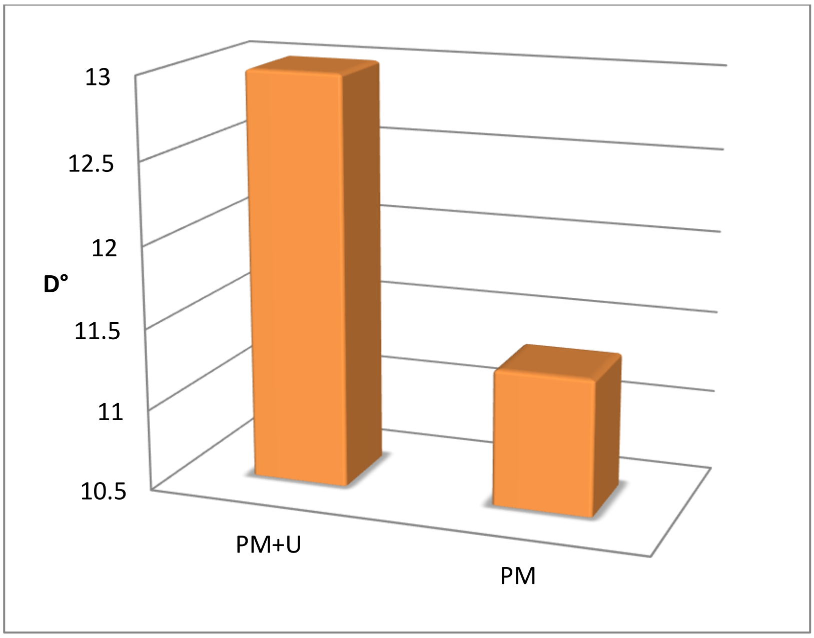


Figure 22: Comparison of Average Dornic Acidity between Pasteurized milk and his mixture with urine (Original Figure)

II. 3. Results of Biochemical and Chemical Analysis

II. 3. 1. Chemical components of raw milk

The table below represents the results of the analysis of the chemical composition of milk in relation to the stages of lactation (beginning of lactation, mid-lactation, and end of lactation).

Table 12: The results of the analysis of the chemical composition of milk

	Beginning Of Lactation(Colostrum)	Mid-Lactation	End Of Lactation
Fat (%)	**0,05**	**2,52 ± 1,3171**	**2,86 ± 1,7154**
Solids-Non-Fat (%)	**10,99**	**8,95±0,3853**	**7,64±0,7850**
Lactose (%)	**6,05**	**4,92±0,1997**	**4,07 ± 0,3166**
Protein (%)	**4,05**	**3,27±0,1468**	**2,71 ±0,2194**
Conductivity Electric (mS/cm)	**6,48**	**6,62 ±1,61**	**8,13 ± 1,38**
Freezing Point °C	**-0,692**	**-0,568 ±0,01991**	**-0,463 ±0,0356**
Sels (%)	**0.89**	**0,72 ±0,0312**	**0,6 ±0,046**

II. 3. 1. 1. Fat content

The fat content is very low at the beginning of lactation (colostrum) with 0.05% and gradually increases during lactation, reaching 2.52 ± 1.3171% in mid-lactation and 2.86 ± 1.7154% at the end of lactation. (Figure 23)

The results of fat content in g/l (25.2 g/l; 28.6 g/l) are close to those obtained by Siboukeur (2007), which are around 28 g/l. (Mint Miloud et *al.*, 2011) in Mauritania 29.02 g/l; (EL-hatmi et *al.*, 2006) in Morocco 26.50 g/l; (Kouniba et *al.*, 2005) in Tunisia 30 g/l; (Alloui-Lombarkis et *al.*, 2007) , (37.44 g/l) and very low compared to the result of (Khoidri and Chemla, 2020) (97.72- 69.59 g/l). compared with cow's milk and human milk (35 g/L - 45 g/L) and sheep's milk (60 g/L - 70 g/L), the results of fat content are low than all the types of milk, especially the colostrum.

Lipids are the most variable components of milk both quantitatively and qualitatively; they depend on the breed and the milking rank, which influences the fat content (Debouz et *al.*, 2014).

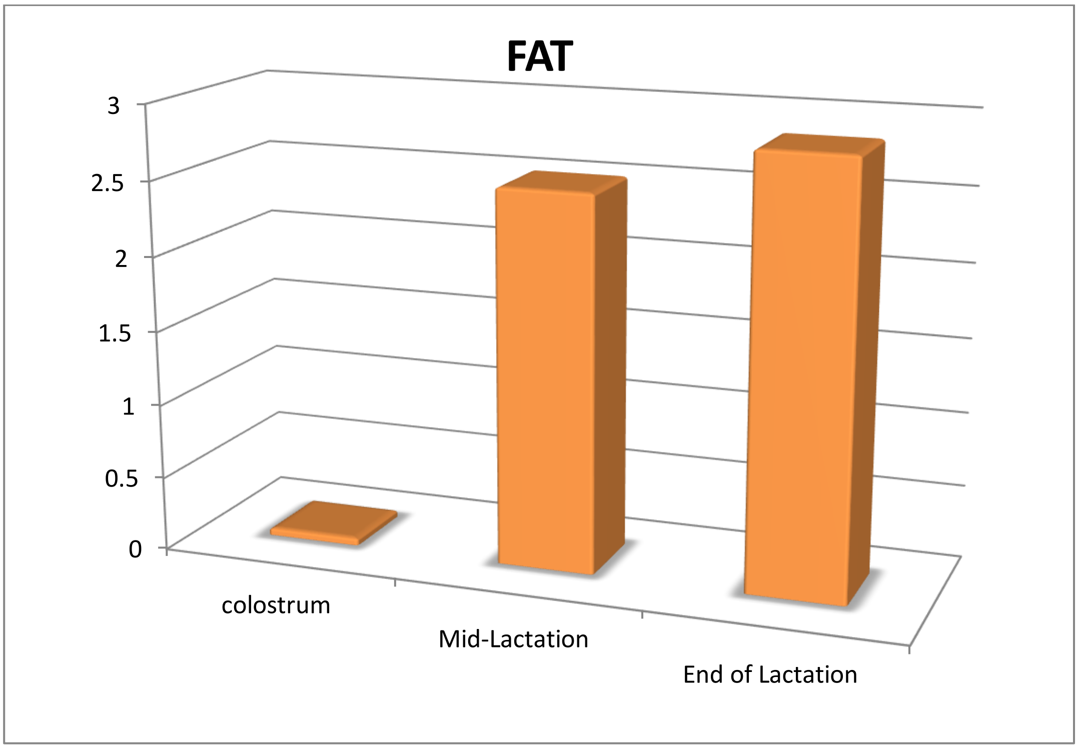


Figure 23: The changes in Fat content in relation to the lactation stage (Original Figure)

II. 3. 1. 2. Solids-non-fat (SNF) content

The solids-non-fat content is high in the beginning of lactation and decreases over time, with a concentration of 10.99% in colostrum, 8.95% in mid-lactation, and 7.64% at the end of lactation. (Figure 24)

The solids-non-fat content in colostrum is close to that of sheep's milk (10-11%), while in mid-lactation, it is similar to that of cow's milk (8.5-9%). At the end of lactation, the solids-non-fat content is the lowest compared to all the types of milk mentioned.

Those results are more high than the result of (Ali et *al*., 2019) (0,35-0,95%)

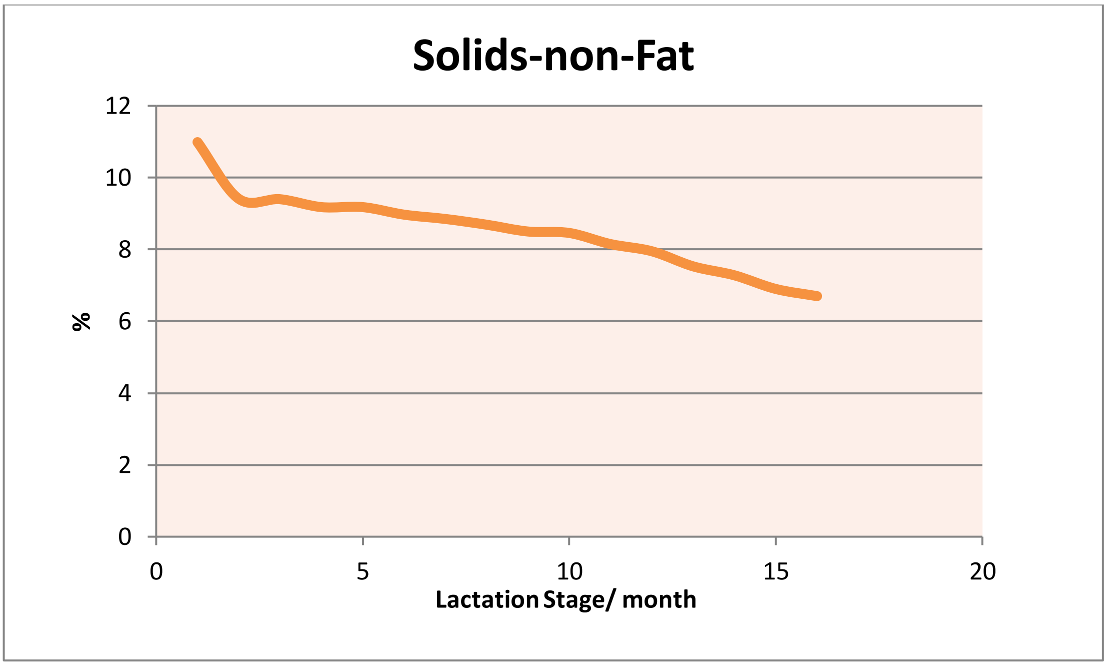


Figure 24: The changes in Solids-non-Fat Content in relation to lactation stage (Original Figure)

II. 3. 1. 3. Lactose Content

The lactose content in colostrum is higher compared to mid-lactation and the end of lactation (6.05 versus 4.92 ± 0.1997 and 4.07 ± 0.3166, respectively). The percentage decreases as the lactation period progresses (Figure 25).

Translating these values to g/l (60.5; 49.2 and 40.7 g/l), the results indicate that the lactose content in milk at different stages of lactation is consistently higher compared to cow's milk, sheep's milk, and human milk. The highest lactose concentration is observed in colostrum (60.5 g/l), decreasing through mid-lactation (49.2 g/l), and reaching its lowest at the end of lactation (40.7 g/l)

The lactose concentration in colostrum is 60.5 g/l, which is lower than that in human milk (68-70 g/l) but higher than in cow's milk (40-50 g/l) and sheep's milk (45-48 g/l). Conversely, the lactose content in mid-lactation (49.2 g/l) closely resembles that of the latter two types of milk mentioned.

The results in mid-lactation and the end of lactation are lower compared to those mentioned by (Bendellali and Mahammed) (2017) (56.68 – 52.4 g/l).

The value in mid-lactation is similar to the result of (Mint Miloud et *al.*, 2011) in Mauritania with 49 g/l , and in the end of lactation the value is similar to (Kouniba et *al.*, 2005) in Tunisia 40,50 g/l , the both are high than the results of (Alloui-Lombarkis et *al.*, 2007) with 34,20g/l and (EL-hatmi et *al.*, 2006) in Tunisia (31g/l).

The lactose content in camel milk seems to depend not only on the breed but also on the stage of lactation and the state of hydration. It is low during the first hours following calving and increases by 36% from the initial content 24 hours later. (Yagil and Etzion, 1980)

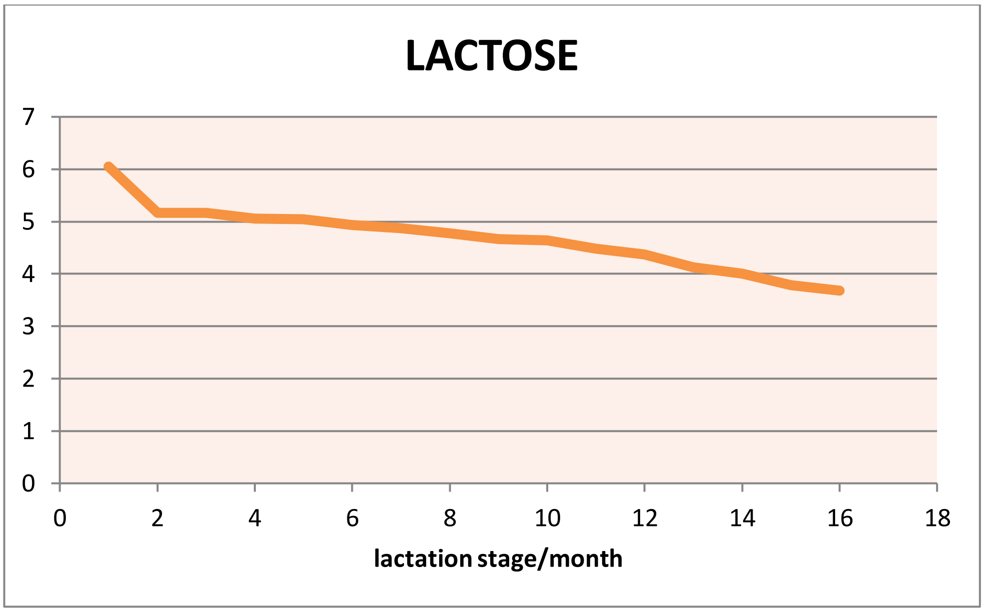


Figure 25: The changes in Lactose content in relation to lactation stage (Original Figure)

II. 3. 1. 4. Protein dosage in milk

With a concentration of 40.5 g/l in colostrum, 32.7 g/l in mid-lactation, and 27.1 g/l at the end of lactation, the percentage of proteins decreases as the lactation period progresses (Figure 26).

The protein concentration in the end of lactation falls within the range cited by Mohamed et *al.* (1989), (46 g/l and 21.5 g/l). These results align with those obtained by (Ghalem, 2016) 28.25 g/l; Chethouna (2011) 28.25 g/l; (Kuoidri and Chemla, 2020) 28.81 g/l; (Elamine and Wilcox, 1992) 28.0 g/l; and (Mehaila et *al.*, 1995) for the Majaheem and Hamra breeds (29.1 g/l and 25.2 g/l respectively). In another study, (Debouz and Guerguer, 2014) cited a range from 28.1 g/l to 10 g/l.

Conversely, the protein content in mid-lactation is close to those cited by Siboukeur (2007) 35.68 g/l; (Sboui et *al.*, 2009) 34.15 g/l; (Attia et *al.*, 2001) 30.72 g/l; Kamal et *al.* (2007) 33 g/l; Shamsia (2009) 34.6 g/l and (Sboui et *al.*, 2016) 34,15.

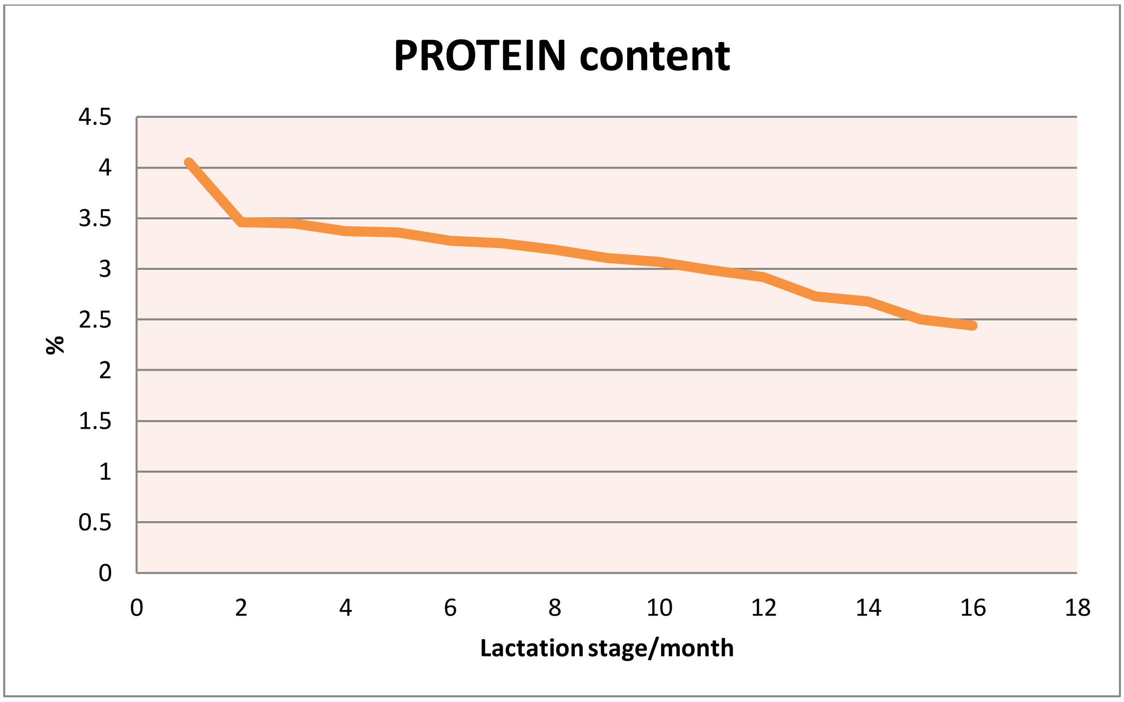


Figure 26: the changes in Protein content in relation to lactation stage (Original Figure)

II. 3. 1. 5. Conductivity electric

The electrical conductivity values of camel milk at the end of lactation is higher compared to colostrum and mid-lactation, with levels of 6.48 mS/cm for colostrum, 6.62 mS/cm ± 1.61 for mid-lactation, and 8.13 mS/cm ± 1.38 for the end of lactation. These three values are higher compared to cow's milk and human milk (4.0 - 5.0 mS/cm), and they are quite similar to sheep's milk (6.0 - 7.0 mS/cm).

In comparison to sheep's milk and the colostrum and mid-lactation stages (6.0 - 7.0 mS/cm vs. 6.48 mS/cm; 6.62 mS/cm), the values are quite similar.

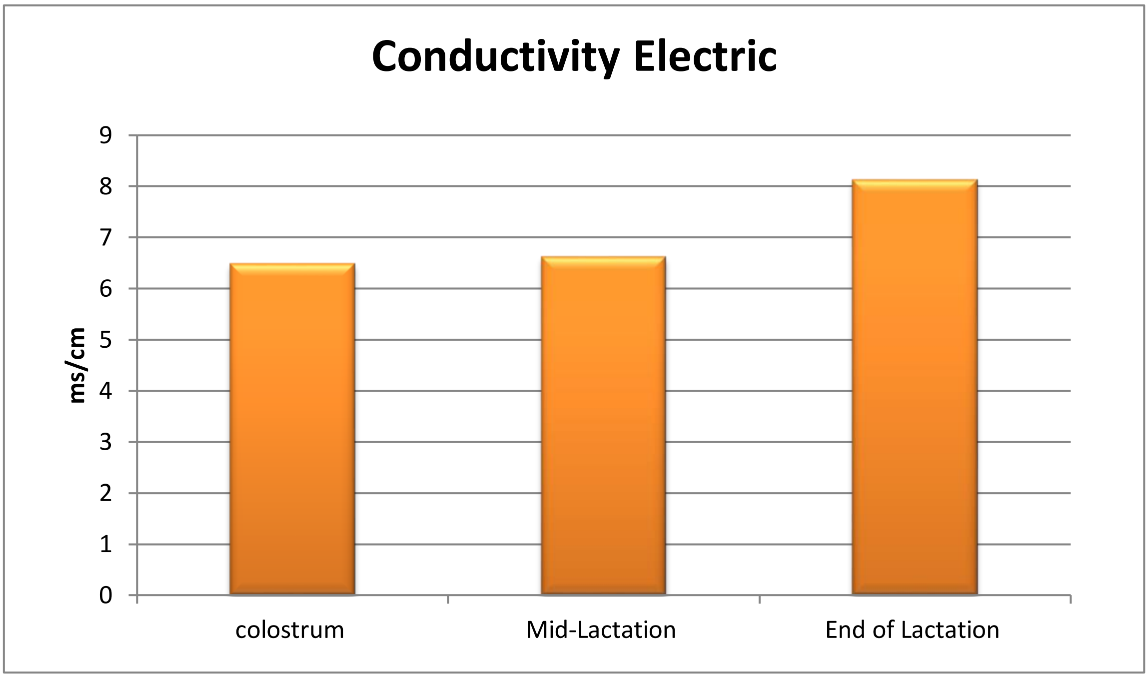


Figure 27: Comparison between Conductivity Electric of colostrum, mid lactation, end lactation (Original Figure)

The electrical conductivity values recorded for camel milk are higher than those reported by (Chethouna, 2011) (4.5 mS/cm), (Bensadek and Hadef, 2019) (6,12 mS/cm) and fall within the range of results obtained by (Bendellali and Mahammed, 2017) (7.11 ± 1.017 - 6.09 ± 0.000 mS/cm).

Camel milk contains a large quantity of ions and mineral salts (sodium, potassium, chloride, etc.), which explain its high electrical conductivity and thus favor electrical conduction. The low fat content also increases conductivity. (Muhammad et *al.*, 2023)

II. 3. 1. 6. Freezing Point

The freezing point of colostrum (-0.692°C) is lower compared to the two later lactation stages, mid-lactation (-0.568°C) and end of lactation (-0.463°C) (figure 29).

These results indicate a higher concentration of dissolved solutes in comparison to end lactation stages, as the freezing point increases, reflecting a decrease in solute concentration.

Comparing with other types of milk, cow's milk generally has a freezing point of -0.53 to -0.55°C, sheep's milk from -0.56 to -0.58°C, and human milk from -0.52 to -0.54°C. Colostrum has a lower freezing point than all these types. Mid-lactation milk has a freezing point comparable to cows and sheep's milk, while end of lactation milk shows a higher freezing point than cows, sheep's, and human milk, suggesting a lower solute concentration at the end of lactation.

The freezing point in the end of lactation is close to the result of (Muhammad et *al.*, 2023) with -0,485.

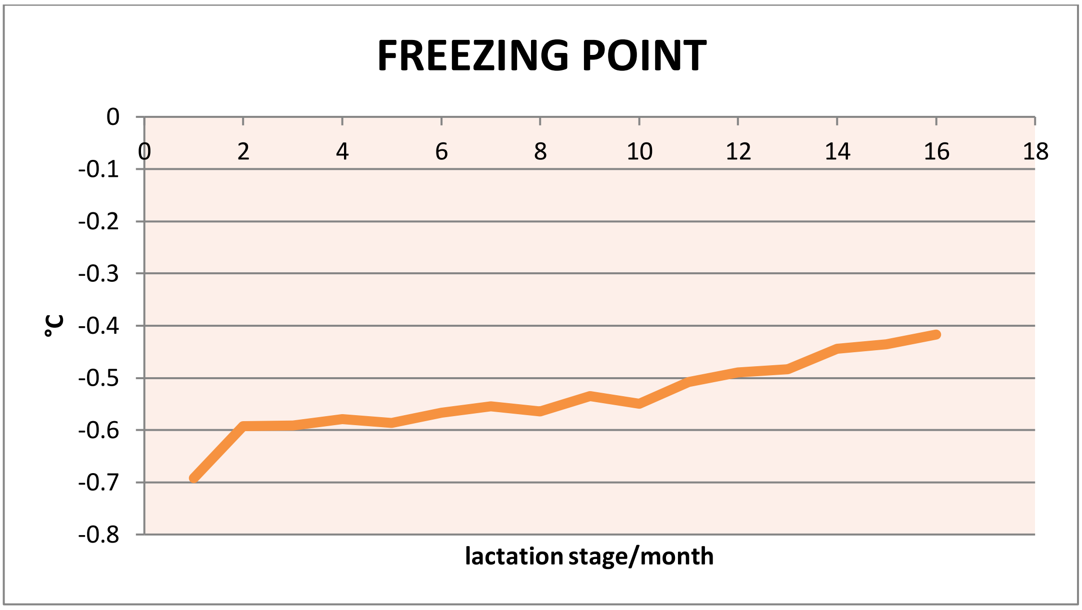


Figure 28 : The changes in Freezing point values in relation to lactation stage (Original Figure)

II. 3. 1. 7. Sels

The salt content in colostrum is 0.89%, which is higher than the salt content during mid-lactation (0.72 ± 0.0312%) and end of lactation (0.6 ± 0.046%).

Comparing those values with other types of milk, cow's milk typically has a salt content of 0.7 - 0.75%, sheep's milk ranges from 0.9 - 1.0%, and human milk is from 0.2 - 0.3%.

The salt content in colostrum is the highest at 0.89%, similar to that of sheep's milk (0.9 - 1.0%) and higher than that of cow's milk (0.7 - 0.75%) and human milk (0.2 - 0.3%). This content decreases to 0.72% during mid-lactation, which is comparable to cow's milk and higher than human milk. The salt content further decreases in late lactation to 0.6%, which remains higher than human milk but lower than both cow's and sheep's milk.

The highest salt content in colostrum indicates a high concentration of mineral ions at the beginning of lactation. This content gradually decreases over the course of lactation. The salt levels in these three types of milk are higher than those in human milk, reflecting the specific nutritional needs of calves and lambs compared to human infants.

Salts in milk primarily include mineral ions such as sodium, potassium, calcium, magnesium, phosphate, chloride, and bicarbonate. These salts play a crucial role in various biological and nutritional functions. (Bouland et *al.*, 2022)

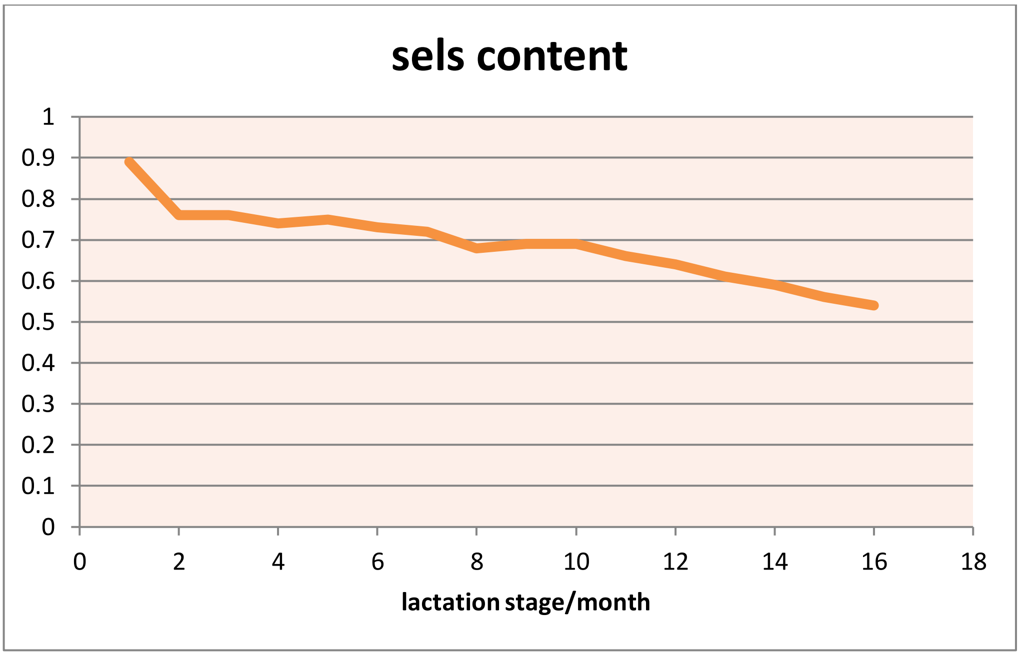


Figure 29: The Changes in Sels content in relation with lactation stage (Original Figure)

II. 3. 2. Protein dosage in the mixture of milk and urine:

The dosage of Protein was determined using the biuret method, Gelatin used as the standard.

The equation of calibration curve: y=0,2175 x -0,177 ; the correlation coefficient: : R^2 = 0.9951

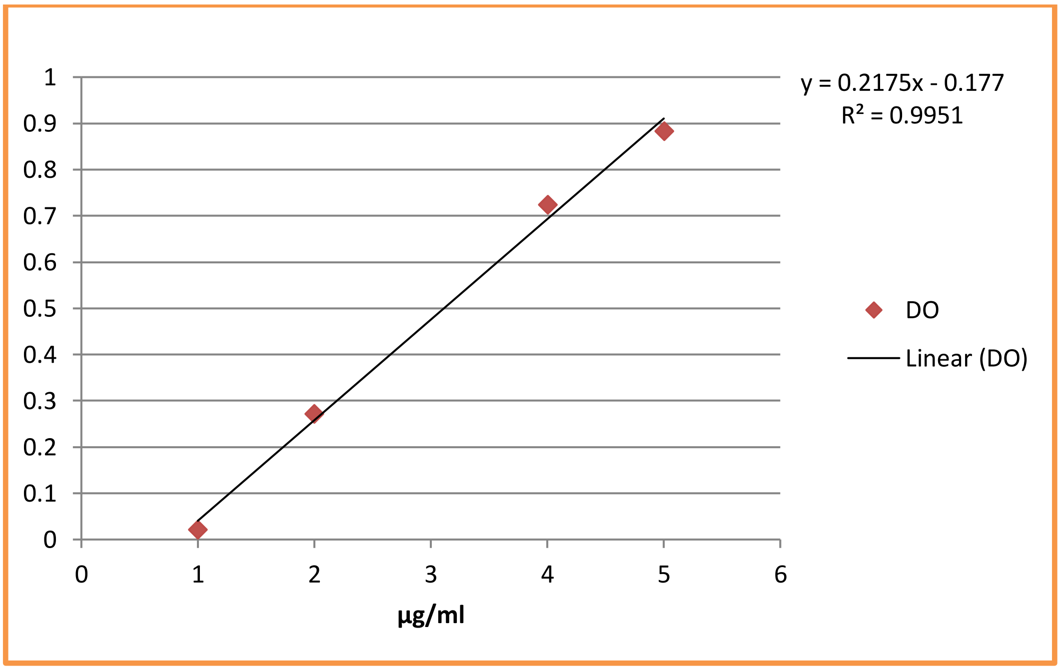


Figure 30: Calibration Curve of Gelatin (Original Figure)

The protein content in the mixture of RM U and PM U have been determined and classified in the following table:

Table 13: The resultsof the protein content in the mixture of milk urine

Samples	The wavelength (nm)	Absorbance in (1/6; 1,8) dilution (Average)	Concentration (g/l) (Average)
Mixture RM+U	540	1,166 ± 0,31822	53,59 g/l ±13,71397
Mixture PM+U	540	1,216 ± 0,56305	52,82g/l ± 20,95096

Figure 31: Comparison between protein content in RM+U, PM+U, RM (Original Figure)

The addition of urine to both pasteurized and raw milk results in a significant increase in protein content, reaching approximately 55 g/L for both mixtures. In contrast, the protein content of raw milk alone is lower at approximately 35 g/L. This suggests that the presence of urine substantially elevates the measurable protein levels in the mixture, and that he contains another type of proteins detected by this method.

A study of the urinary proteome of the dromedary camel identified 147 protein descriptors from 735 peptides. (Alhaider et *al.*, 2012)

In another study, Salwa et *al.* (2004) confirmed that camel urine contains higher levels of total proteins compared to other types of urine (bovine, goat, and human).

The mixture of RMU, PMU raises the concentration of proteins, and this does not have a negative effect.

II. 3. 3. Vitamin C Dosage:

The concentration of vitamin c is determined by using vitamin C calibration curve:

y= 0,043ln(X) +0,2123

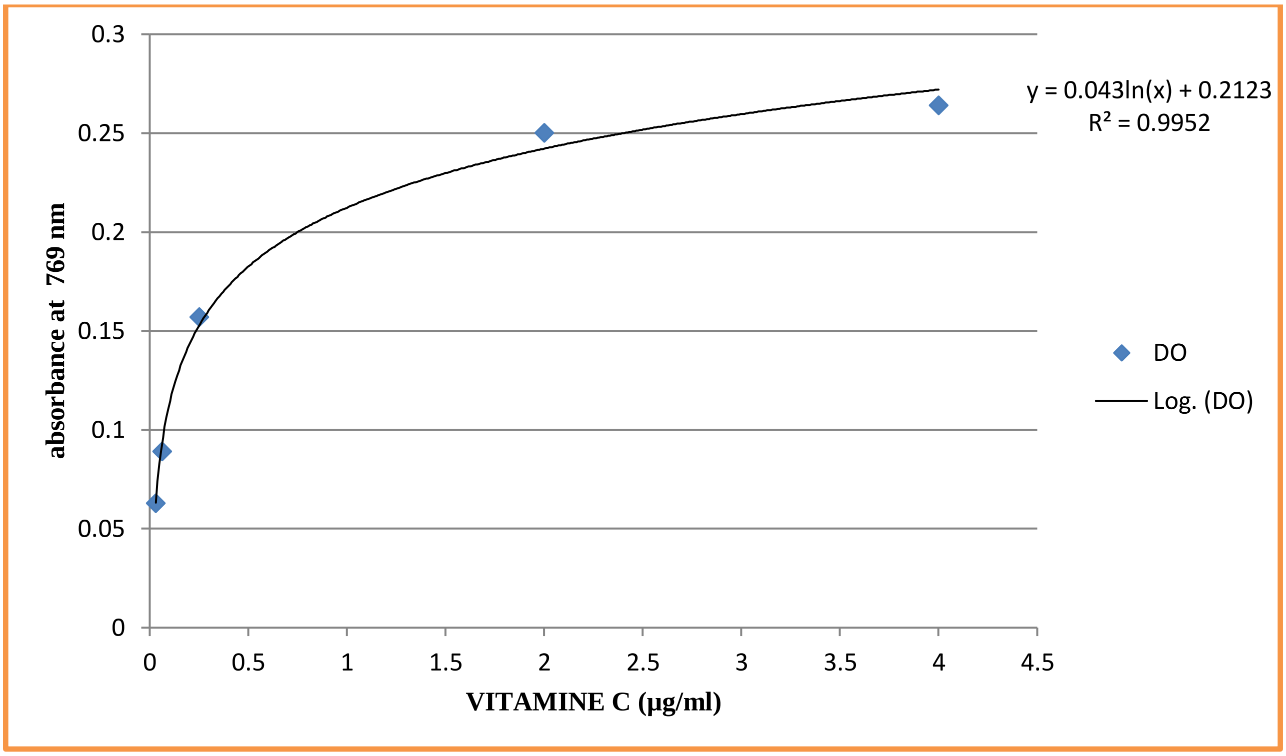


Figure 32: Calibration Curve of Vitamin C (Original Figure)

Table 14: The results of vitamin C concentration

Samples	Absorbance	Concentration (µg/ml) (Average)
RM	0,392	20,81 ± 4,19
Mixture RM+U	0,658	14,41 ± 5,14
URINE	0,265	3,42 ± 3,47

The vitamin C concentration in camel milk and its mixture with urine was conducted after a specific period following the thawing of the milk samples.

This finale negatively impacted the results and reduced the measured vitamin C content in raw milk. The findings indicate that the vitamin C levels are slightly higher in the camel milk-urine mixture compared to camel urine alone.

Comparing the milk concentration with other authors results, (20 mg/l) is low than the results of (Bendellali and Mahammedi, 2018) 25,92 mg/l and (Siboukeur ,2007) (41.40 mg/l).

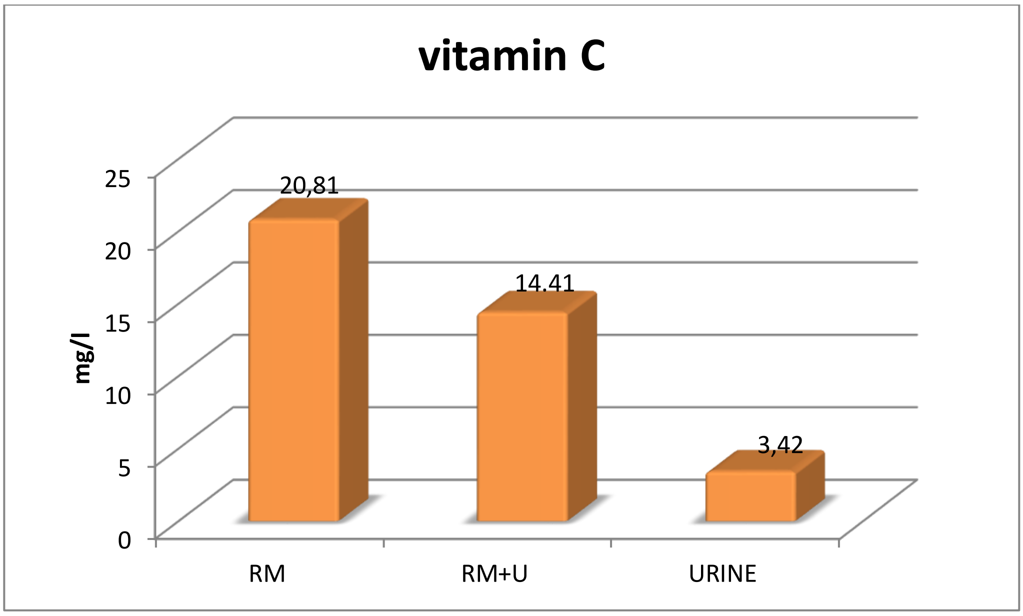


Figure 33: Comparison between Vitamin C concentration in RM, Urine and their mixture (Original Figure)

The obtained results fall within the average range of concentrations of vitamin C present in milk and urine.

II. 3. 4. Flavonoid Dosage

The concentration of flavonoids was carried out according to the aluminum chloride (AlCl3) method, with Quercetin used as the standard.

A calibration curve with the equation: $y=1.588x +0.0494$ and the correlation coefficient: $R^2 = 0.9981$

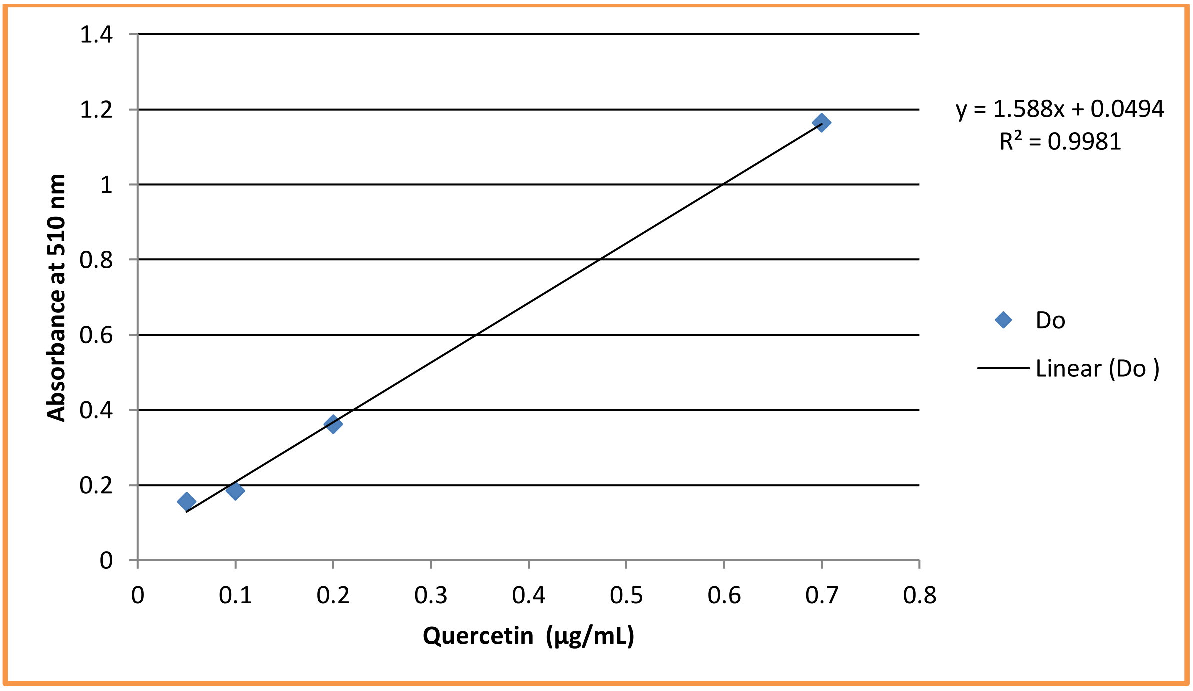


Figure 34: Calibration Curve of Quercetin (Original Figure)

The concentrations of flavonoids present in camel urine have been determined and classified in the following table (15, 16, 17)

Part 01:

Table 15: The results of the concentration of Flavonoids

Samples	Absorbance (Average)	Concentration (µg/ml) (Average)
Urine	1,113	0,6117±0,2032

The table shows that the average of Flavonoide concentration in urine is 0,6117 µg/ml.
0,6117µg/ml = 0,2032 mg/l

The result is low than the Results of (Hammoudi, 2021) 67,43 µg/ml . And also than the results of (Mercha et *al.*, 2019) (7.75 ± 0.15 ; 4.87 ± 0.07 mg QE/g of DW)

Part 02:

Table 16: the Results of the concentration of Flavonoide in RM, RM+U

Samples	Absorbance (Average)	Concentration (µg/ml) (Average)
Mixture RM+U	1,068	0,6414±0,2890
RM	1,0344	0,5295 ±0,1677

The concentration of flavonoid in RM is 0,5295µg/ml and in the mixture of RM+U is 0,64.

Part 03:

Table 17: the Results of the concentration of Flavonoide in PM, PM+U

Samples	Absorbance (Average)	Concentration (µg/ml) (Average)
Mixture PM+U	1,32	0,8043±0,2874
PM	1,66	1,0142 ±0,3381

The results shows that the concentration of Flavonoide in PM+U (1,0142 µg/ml) is significantly lower than PM (1,0142 µg/ml).

THE COMPARISON OF THE RESULTS:

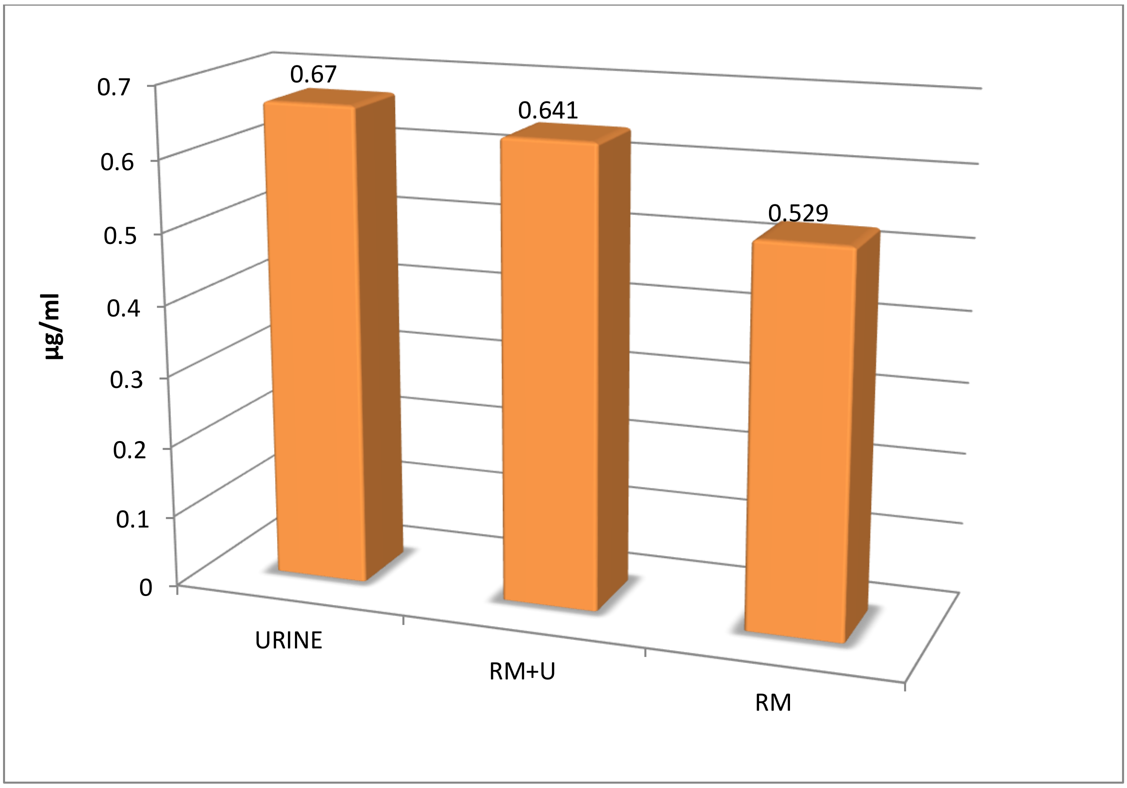


Figure 35: Comparison between Flavonoide concentration in urine and his mixture with RM and PM (Original Figure)

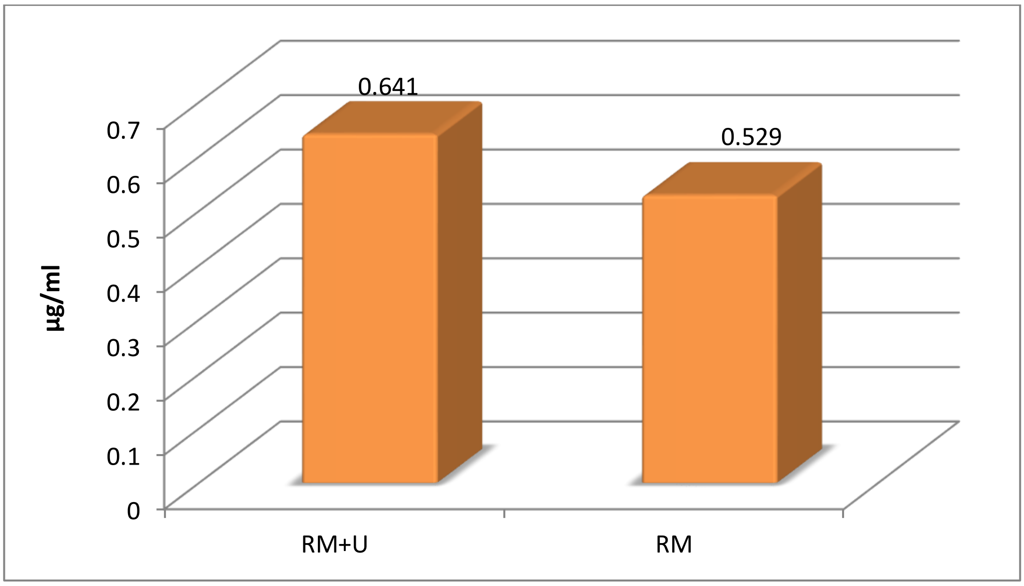


Figure 36: Comparison between Flavonoide concentration in RM and his mixture urine (Original Figure)

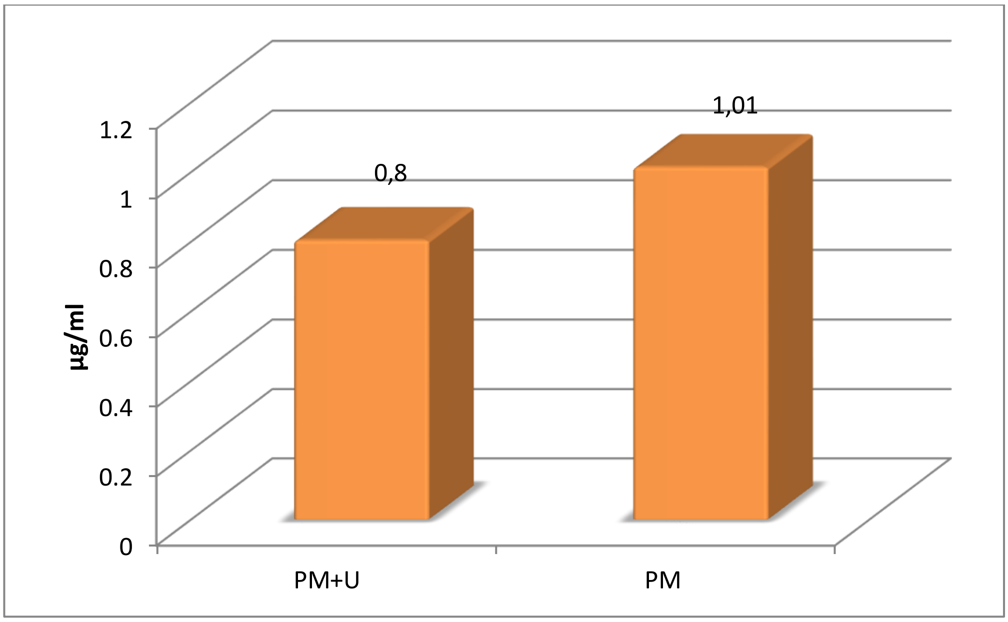


Figure 37: Comparison between Flavonoide concentration in PM and his mixture urine (Original Figure)

The obtained results of the mixture RMU and PMU fall within the average range of concentrations of Flavonoide present in milk and urine

II. 3. 5. Polyphenols dosage:

PART 01:

The dosage of polyphenols in urine is determined by using Gallic acid calibration curve:

y=1.588x +0.0494 ; R^2 = 0.9981

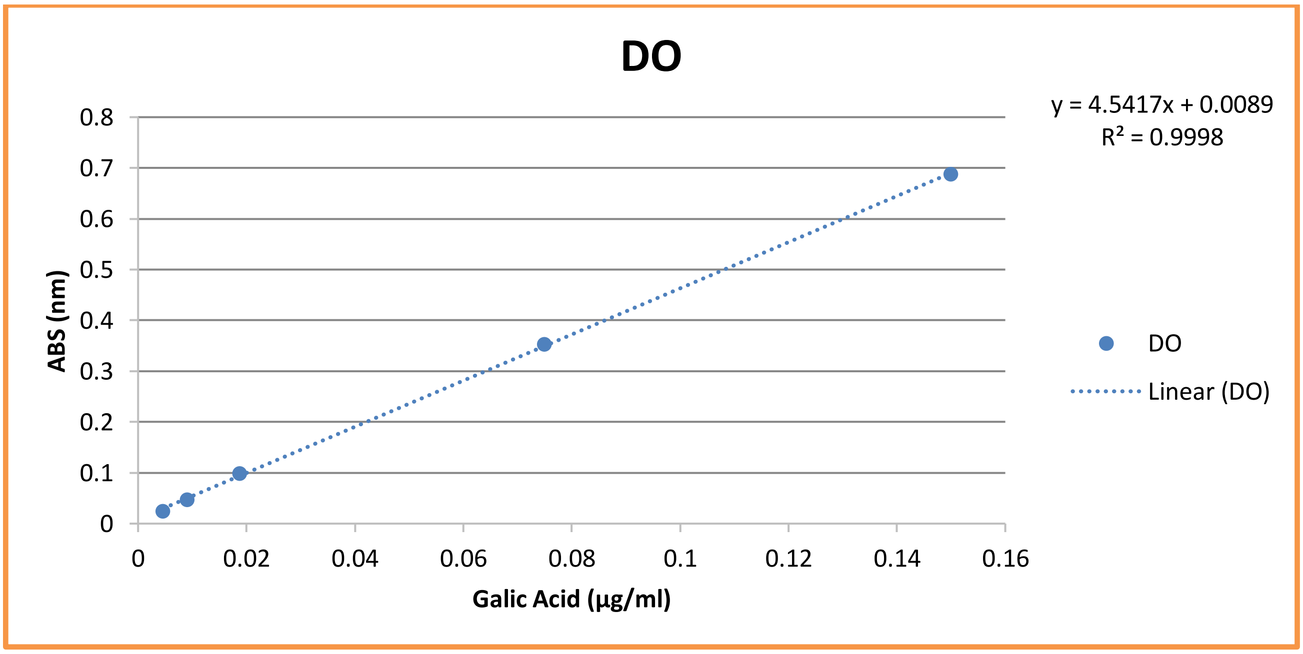


Figure 38: Calibration Curve of Gallic Acid (Original Figure)

The concentrations of polyphenols present in camel urine have been determined and classified in the following table:

Table 18 : the concentrations of polyphenols present in Urine

Samples	Absorbance in (1/8) dilution (Average)	Concentration (µg/ml) (Average)
Urine	1,214636364	2,1412 ± 0,6583

The table shows that the average of polyphenol concentration in urine is 2, 1412 µg/ml.

2, 1412 µg/ml = 2, 1412 mg/l

The concentration in urine is high than the Results of (Kuoidri and Chemla, 2020) (0,2054 ± 0,0365 mg EAG/lU.)

The variability in polyphenol content in the analyzed urine is related to the animal's physiological state and the phenolic composition of the ingested food, part of which is excreted primarily in the urine and bile.

PART 02:

The concentration of polyphenols in raw milk and his mixture with urine is determined first by using the extraction of total phenolic compounds then by Gallic acid calibration curve

Table 19 : the concentrations of polyphenols present in RM, RM+U

Samples	Absorbance	Concentration (µg/ml) (Average)
RM	1,1278125	0,2478 ± 0,11649
RM+U	0,283636364	1,1029± 0,6487

The results shows that the concentration of polyphenol in RM+U (1,1029 µg/ml) is significantly higher than RM (0,2478 µg/ml).

The result of polyphenols in milk is too lower than the results of (MERCHA et *al.*, 2019) (varied from 17.29 ± 0.61 to 22.65 ± 1.15 mg GAE/g , and in a another specific camel milk ranged from 10.74 ± 0.39 to 12.46 ± 0.33 mg GAE/g of DW.) and (Alhumaid et *al.*, 2010) (17.29 ± 0.61 mg GAE/g)

The results of (MERCHA et al., 2019) are determined first by the extraction of total phenolic compounds from camel milk according to the methods of (Alyaqoubi et *al.*, 2014).

PART 03:

The concentration of polyphenols in pasteurized milk and his mixture with urine

Table 20: the concentrations of polyphenols present in PM, PM+U

Samples	Absorbance	Concentration (µg/ml) (Average)
PM	1,6966	0,3733 ± 0,0489
PM+U	1,3266 DI	0,5826± 0,2016

The results shows that the concentration of polyphenol in PM+U (0,5826 µg/ml) is significantly higher than PM (0,3733 µg/ml).

THE COMPARISON OF THE RESULTS:

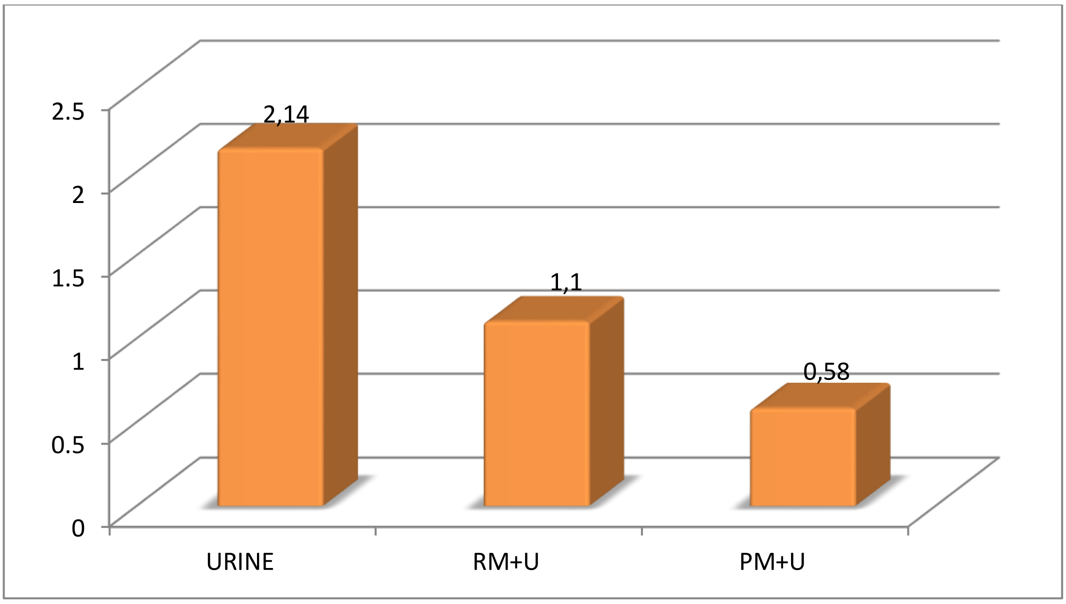


Figure 39: Comparison between polyphenols concentration in urine and his mixture with RM and PM (Original Figure)

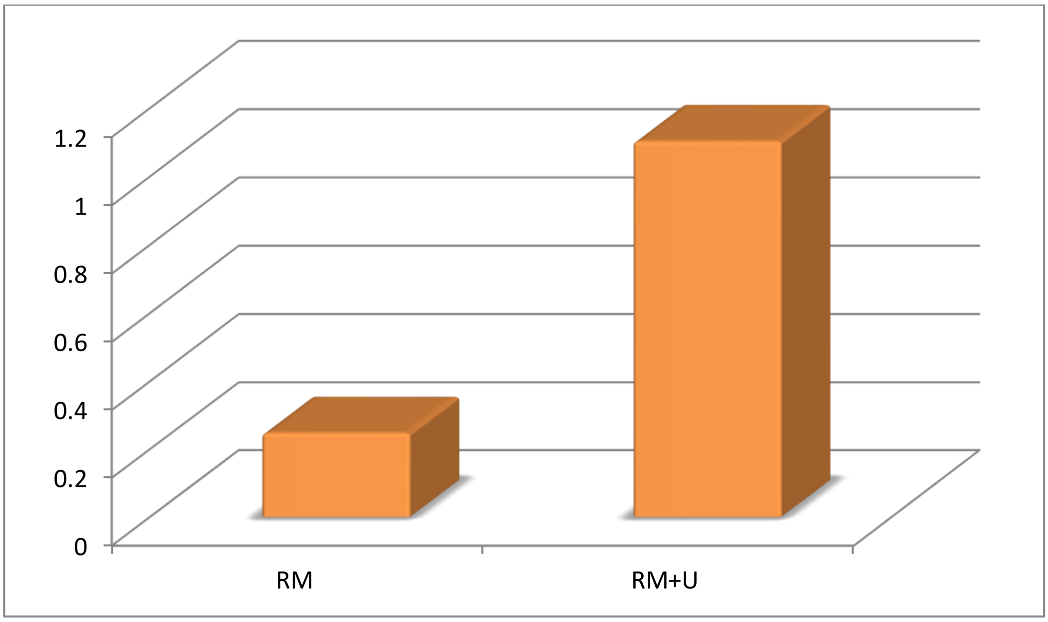


Figure 40: Comparison between polyphenols concentration in RM and his mixture urine (Original Figure)

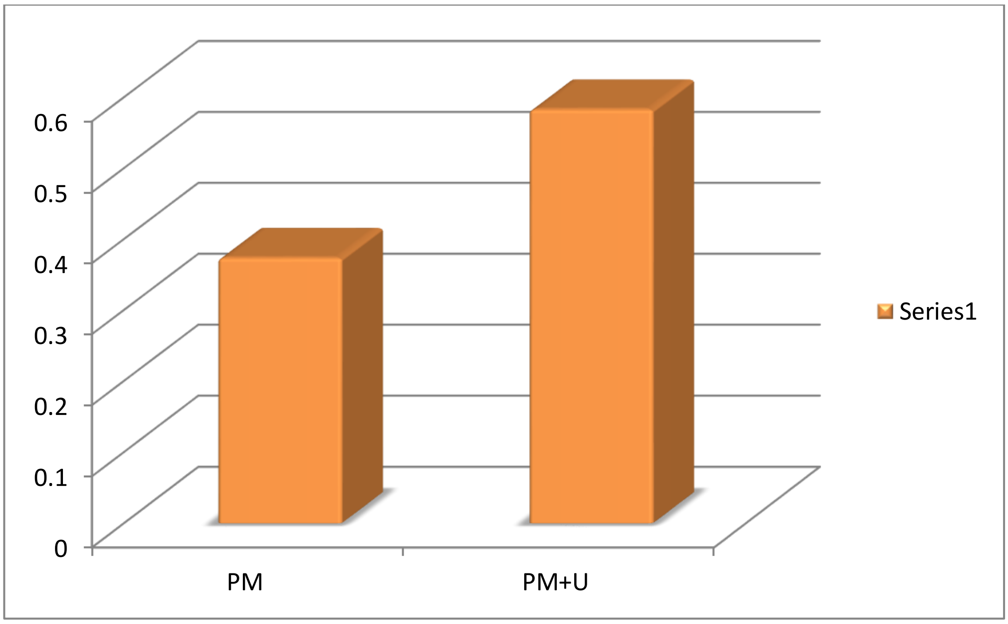


Figure 41: Comparison between polyphenols concentration in PM and his mixture urine (Original Figure)

The obtained results of the mixture RMU and PMU fall within the average range of concentrations of polyphenols present in milk and urine

According to (Mcgee et *al.*, 2023) the chemical mixture has a negative impact on the phenolic components of urine.

Polyphenols are esteemed for their advantageous effects on health and their promising role as functional components in dairy items. Milk proteins act as inherent vehicles for polyphenols, predominantly establishing non-covalent connections, while certain sources indicate the presence of covalent linkages. The nature and intensity of these connections are impacted by the structural attributes of both substances and external elements like pH and temperature. (Yildirim-Elikoglu, 2019)

II. 4. Antioxidant activity:

Scavenging of DPPH radical is considered the basis of DPPH antioxidant assay (Marmouzi et *al.*, 2015).

The antioxidant activity of raw camel milk, pasteurized milk, and urine was assessed using the extraction of antioxidant components, with the results expressed as a percentage of inhibition.

The results of the antioxidant activity in relation to lactation stage

Table 21: The results of the antioxidant activity of milk in relation to lactation stage

	Beginning Of Lactation(Colostrum)	Mid-Lactation	End Of Lactation
% inhibition	**51%**	**60% ± 0,1013**	**83% ± 0,08547**

The table shows that the antioxidant activity in the colostrum (51%) is significantly lower than the other stage of lactation, in the opposite the last stage of lactation have the highest percentage (83%).

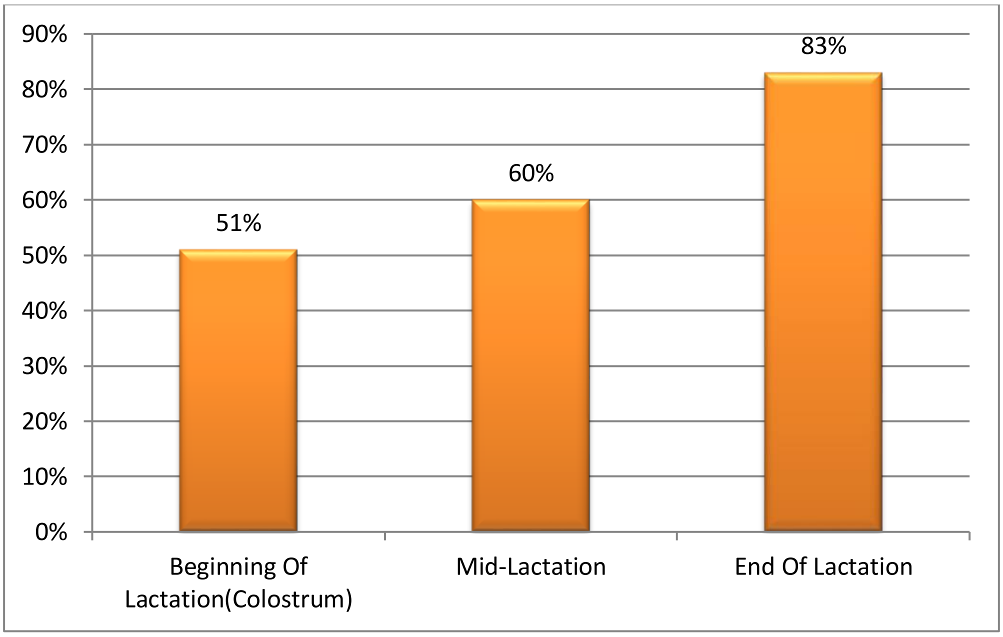


Figure 42: Comparison between the activity antioxidant of milk in relation to stage of lactation (Original Figure)

The results of the antioxydant activity of urine and his mixture with RM and PM

Table 22: the results of the antioxidant activity of urine, RM+U, PM+U

	Urine	RM+U	PM+U
% inhibition	**84%±0,162**	**83%± 0,068**	**43% ± 0,089**

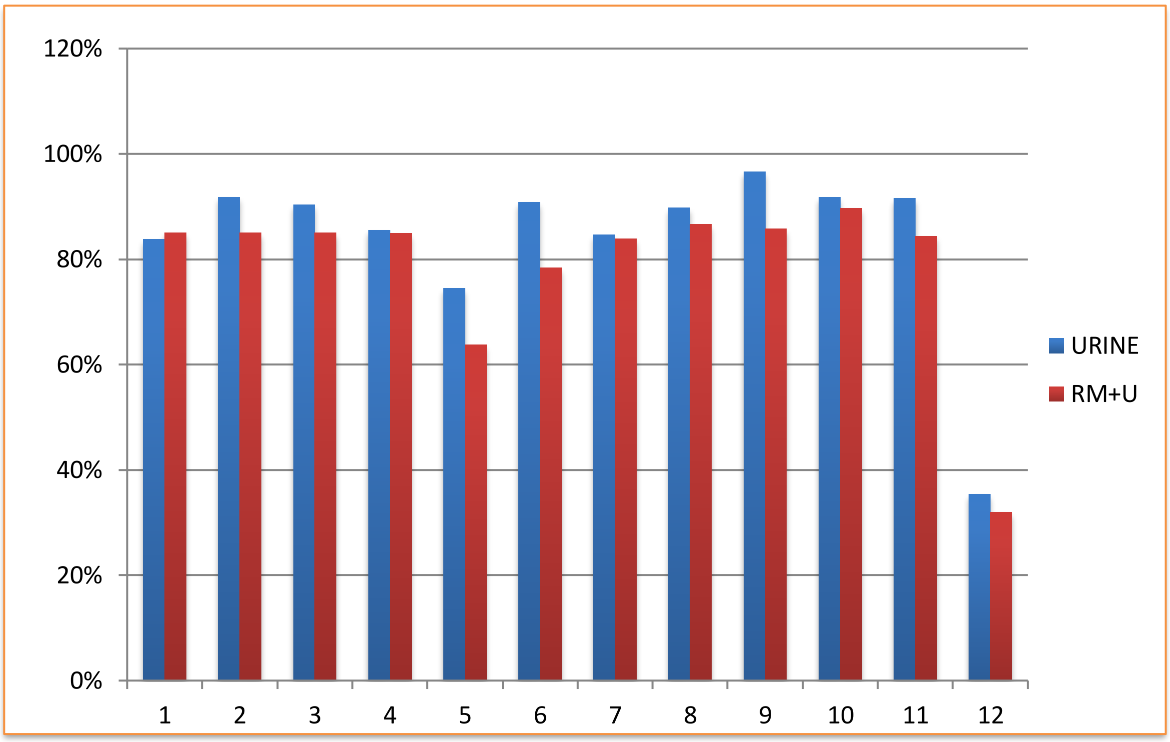


Figure 43: DPPH radical scavenging activity of urine and the mixture RM+U (Original Figure)

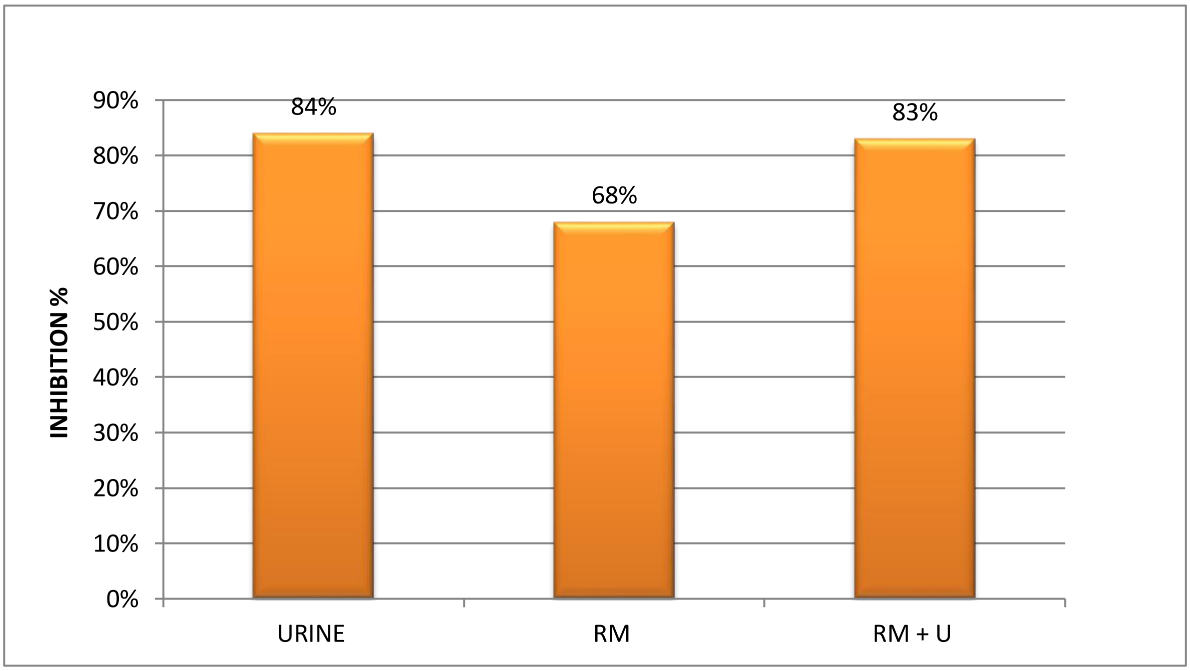


Figure 44: DPPH radical scavenging activity of raw milk, urine and their mixture (Original Figure)

The results of the antioxydant activity of the mixture of RM+U and PM+U

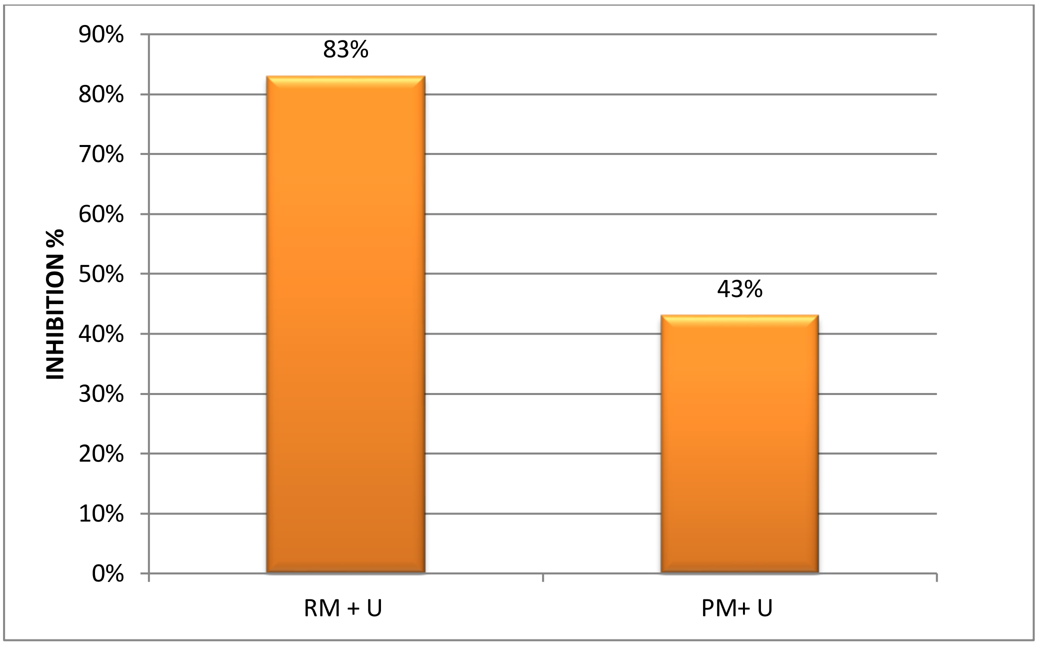


Figure 45: DPPH radical scavenging activity of the mixture of RM U and PM+U (Original Figure)

The Results of antioxidants activity of urine, RM, RM+U, PM+U are expressed as the percentage of inhibition, which indicates the efficacy of each sample in neutralizing free radicals

Urine and his mixture with raw milk show the highest antioxidant activity, with an inhibition rate of 84% and 83 % respectively. This indicates that they have a strong ability to neutralize free radicals, suggesting a high concentration of antioxidant compounds. Compared with raw milk (68%) and the mixture of pasteurized milk with urine (43%)

The mixture of pasteurized milk and urine has the lowest antioxidant activity. This suggests that pasteurization has a negative impact on the antioxidant properties of milk mixture with urine. Probably due to degradation of heat sensitive antioxidant compounds during pasteurisation.

The activity inhibition of raw milk (68%) falls within the range of these results (MERCHA et al., 2019) with an average inhibition between 74.22% - 63.44%, and (Balakrishnan and Agrawal, 2012) (64.4 ± 1.42%) for raw camel milk, as well as those concerning camel milk fermented with lactic acid bacteria (62.03%) (El Hatmi et *al.*, 2018).

Conclusion

This study underscores the significant therapeutic potential of camel-derived products, particularly milk and urine, which are deeply embedded in the traditional medicine practices of Saharan regions. Our comprehensive in vitro analysis revealed the physicochemical properties and chemical composition of these products, both individually and in combination, highlighting their nutritional and medicinal value.

Key findings include:

- The increase in Dornic acidity and slight changes in pH when urine is mixed with milk.
- Variations in protein, vitamin C, flavonoid, and polyphenol concentrations, indicating the complex interplay between these substances when combined.
- High antioxidant activity in camel urine and raw milk, with notable retention in their mixture, although pasteurization diminishes these benefits.

The results clearly demonstrate that raw camel milk and urine retain substantial therapeutic properties, including high antioxidant activity, attributable to their rich organic and inorganic molecule content. Conversely, pasteurization adversely affects these beneficial properties, suggesting that consuming these products in their raw form may maximize health benefits.

Given the observed properties and potential health benefits, further research is imperative to isolate and understand the specific compounds responsible for these effects. Further research is needed to identify the specific compounds responsible for these effects and their potential applications in medicine. Such studies could pave the way for new applications in health and medicine, harnessing the full potential of camel milk and urine in traditional and modern therapeutic practices.

Appendix

Appendix 01: The breeds studied:

Figure 46: Ouled sidi cheikh breed (Original Picture)

Figure 47: Rguibi breed (Original Picture)

Figure 48: Targui breed (Oridinal Picture)

Appendix 02: Collection of Milk and Urine:

Figure 49: Milking (Original Picture)

Figure 50: Milk and Urine samples (Original Picture)

Appendix 03: Equipment

Figure 51: Lactoscane ULTRASONIC MILKANALYZER (Original Picture)

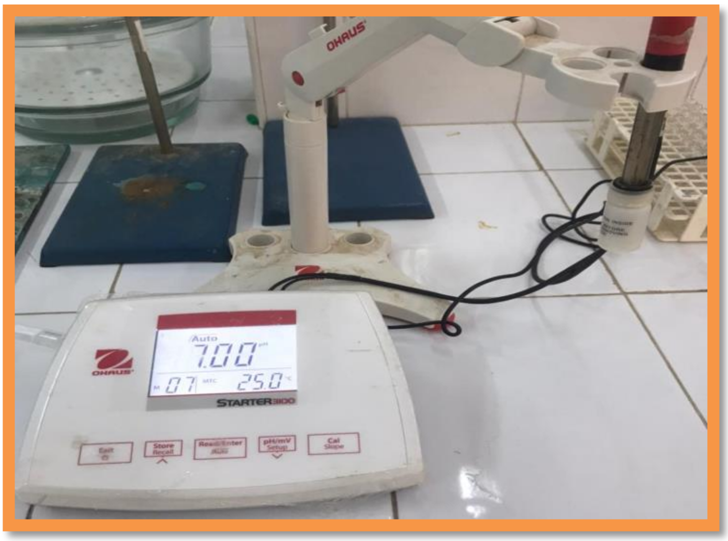
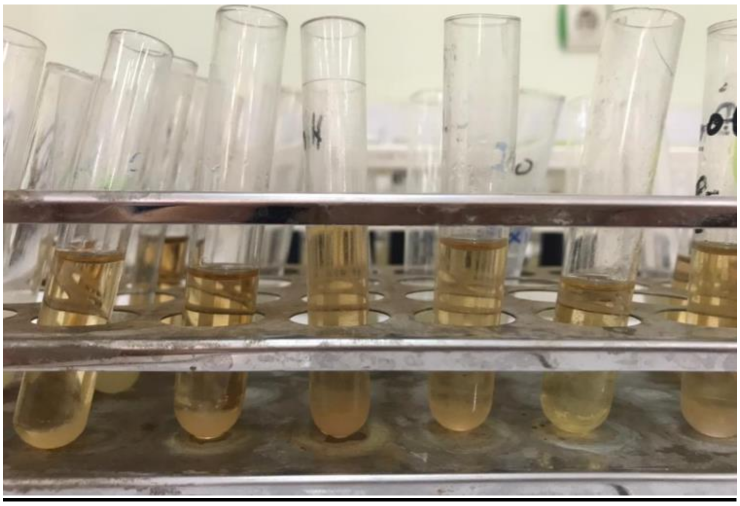


Figure 52: Bench-top pH meter starter 2100 (Original Picture)

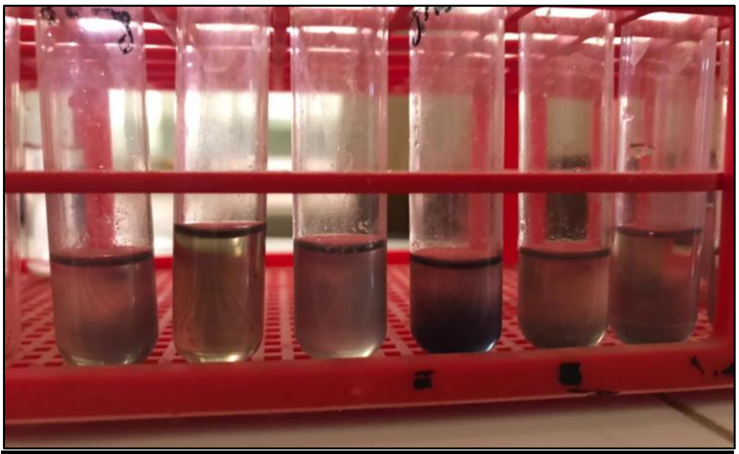
Figure 53: Centrifuge Sigma (Original Picture)

Appendix 04: polyphenols concentration

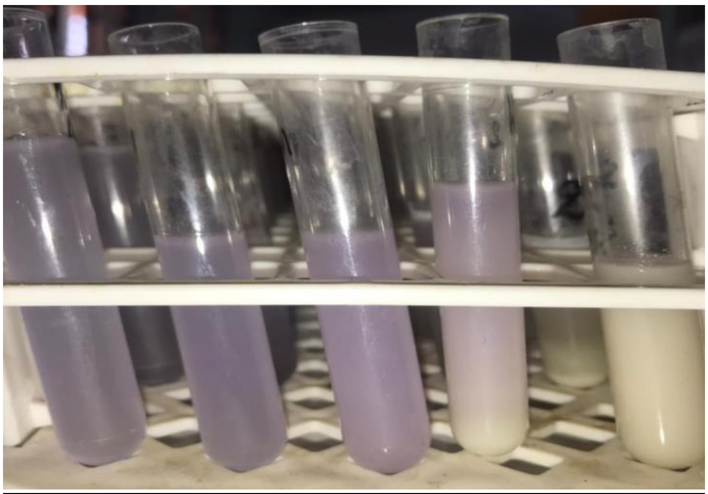
Appendix 05: Flavonoide concentration



Appendix 06: vitamin C concentration

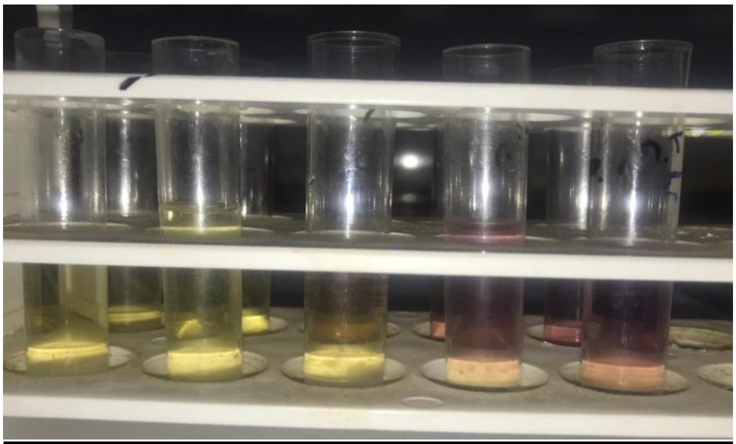


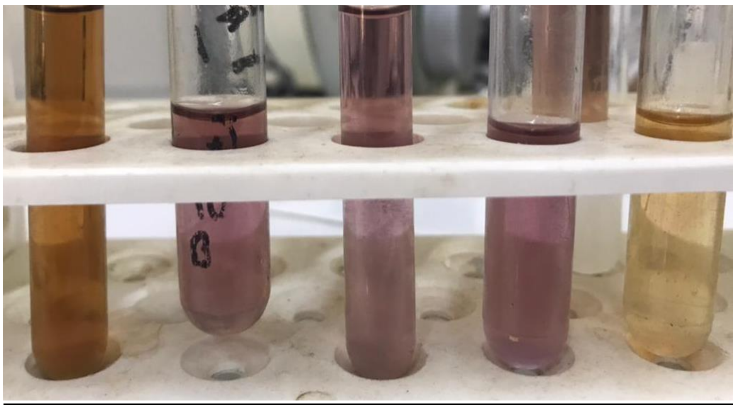
Appendix 07 : Protein concentration



Appendix 08: Extraction of total phenolic compounds

Appendix 09 : Antioxydant activity





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