EFFECT OF LACTATION STAGE, PARITY AND SUPPLEMENTARY FEEDING WITH BUFFELGRASS (*Cenchrus ciliaris*) AND OLD MAN SALTBUSH (*Atriplex nummularia L.*) ON YIELD, COMPOSITION, AND MICROBIAL PROPERTIES OF CAMEL MILK PRODUCED IN TSABONG

A Dissertation Submitted to the Department of Animal Sciences in Partial Fulfilment of the Requirements for the Degree of Master of Science (MSc) in Animal Science (Animal Nutrition)

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CERTIFICATION

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STATEMENT OF ORIGINALITY

The work contained in this dissertation was compiled by the author at the Botswana University of Agriculture and Natural Resources (BUAN), during the period of August 2019 to May 2022. It is my original work and all the sources that I have used or quoted have been indicated and acknowledged using complete references. It shall not be submitted for the award of any other degree or diploma from any other university.

Author's signature ..... Date 24.09.2024

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I am most grateful to my supervisor, Dr. Wame Boitumelo, and co-supervisors Professor Eyassu Seifu and Professor Ayana A. Abdeta for their time, patience, and guidance throughout this research. Their critical contributions were so fantastic and eye-opening towards writing publishable articles. This sharpened my research skills. I would like to thank them so dearly.

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DEDICATION

I dedicate this work to my daughter and son, Onneile Wadikgomo (*Fenah*) and Leatile Wadikgomo (*Shimi*), whose smiles gave me strength and inspiration throughout the straining period of this work. Daddy loves you, my children!!!

Table of Contents

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

%	Percentage
ADF	Acid Detergent Fibre
BGBB	Brilliant Green Bile Broth
Ca	Calcium
CFU	Colony forming units
cm	Centimetre
CP	Crude Protein
CRD	Completely Randomized Design
DM	Dry Matter
DMI	Dry Matter Intake
GDP	Gross Domestic Product
IVDMD	In-Vitro Dry Matter Degradability
K	Potassium
LS	Lactation Stage
Mg	Magnesium
MRD	Maximum Recovery Diluent
Na	Sodium
NDF	Neutral Detergent Fibre

NRC	National Research Council
P	Phosphorus
PA	Parity
SEM	Standard Error of the Means
VRBA	Violet Red Bile Agar

DECLARATION

The research described in this thesis was carried out in the Department of Animal Science and Production, Botswana University of Agriculture and Natural Resources, Gaborone, under the supervision of Dr. Wame Boitumelo, Professor Eyassu Seifu and Professor Ayana A. Abdeta.

This is to declare that this thesis is the result of my investigation and has not been presented in any previous application for a degree. All sources of information are shown in the listed references and all assistance by others has been duly acknowledged.

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Abstract

The study involved two experiments. The first experiment of the study evaluated the effects of lactation stages (LS) and parity (PA) on the composition and properties of camel milk from 24 dromedary camels reared in Tsabong Camel Park, after which the second experiment was a feeding trial done to investigate the effect of supplementary feeding with either or both Cenchrus ciliaris *and* Atriplex nummularia*, in order to evaluate their effect on the quality and quantity of Tsabong Dromedary camels' milk. In the first experiment, a factorial design in CRD was used where lactating camels were randomised into three lactation stages (early, mid and late) each of which fell into each of the two parity stages (primiparous and multiparous). For the second, feeding experiment, twenty-four (24) ear-tagged multiparous camels in mid-lactation, were separately given the supplementary feed used as treatment (None supplemented, as a control group,* Cenchrus ciliaris *alone,* Atriplex nummularia *alone,* Cenchrus ciliaris & Atriplex nummularia*). In the first experiment, the highest levels of total solids (TS) (9.785±0.697%) were observed in primiparous in early lactation camels. Total fat (3.675±0.442%) was highest in primiparous camels in early lactation camels. Multiparous camels in mid-lactation camels had the highest protein (1.910±0.188%). primiparous in late lactation camels had the highest content of solids-not-fat (SNF) (6.330±0.313%). Milk from primiparous in early lactation camels contained the highest free fatty acids (FFA) (0.580±0.057g/L) and multiparous in mid lactations camels had the highest casein (1.718±0.104%). Primiparous camels in late lactation produced the highest level of lactose, (3.568±0.227%). While Primiparous camels in their early lactation produced the highest levels of galactose and glucose, (0.685±0.066%) and (0.363±0.042%), respectively. In the second experiment, supplementary feeding with all three treatments positively affected milk protein,*

urea, and casein percentages. Also increased were milk components of fat, galactose, glucose, solids not fat (SNF), total solids (TS), as well as citric acid contents and density. Another improvement was found in daily milk yield being influenced greatly (P=0.0001) by the supplementary feeding with all three treatments, with the highest positive effect coming from supplementing with Atriplex nummularia*. However, supplementary feed sources significantly reduced (P<0.05) free fatty acids (FFA) content as well as the freezing point of camel milk. The highest concentrations of fat, galactose, glucose, protein, and total solids were 4.222%, 0.878%, 0.372%, 3.143% and 11.762%, respectively, were detected on milk from camels supplemented with* Atriplex nummularia *alone. Supplementing with* Cenchrus ciliaris *alone produced milk with the highest SNF content (7.458%), whilst a combined feed (*Cenchrus ciliaris *and* Atriplex nummularia*) significantly produced milk with the highest milk casein (2.473%). All treatment feeds insignificantly (P>0.05) reduced concentrations of lactose. The results of the study show that the composition of camel milk produced in Tsabong, from free-ranging dromedary camels, under the existing and unimproved feeding conditions, was affected by parity and stage of lactation. Generally, supplementing Tsabong dromedary camels could greatly improve camel milk yield and quality during the dry season. Milk processors could target specific levels of desired components of camel milk, based on milk obtained from camels at specific stages of lactation and parities. Those components that were not directly influenced by parity and lactation will have to be improved by the combined effect of supplementary feeding plus the effect of parity and stage of lactation. Tsabong raw camel milk, at the time of the study, was of good keeping quality. The low FFA content of the milk is an indicator that there was minimal milk straining, bacterial contamination and that the milk confers to good storage quality.*

CHAPTER ONE

GENERAL INTRODUCTION

1.1 Background

There are two different species of camels belonging to the genus Camelus, namely, the one-humped Dromedary camel (*Camelus dromedarius*) and the two-humped Bactrian camel (*Camelus bactrianus*) (Dioli, 2020). In arid and semi-arid areas where cattle are affected by the heat, and lack of water and feed, camels play a major role in supplying milk to the rural population (Yoganandi *et al*. 2015). Camels also play an important role in the livelihoods of desert dwellers and camel milk is a major source of protein and energy for them (Sawaya *et al.,* 1984). The one-humped camels were introduced to colonial Southern Africa in the late 19th and early 20th centuries for military work and to maintain law and order, for use in postal services and experiments in connection with rinderpest (Wilson, 2013a and Wilson, 2013b). In Botswana, although a few remain with the police at Tsabong, the majority were sold or given away at the turn of the 21st century with the expectation of using them for eco-tourism (Gitao and Field, 2012).

Tsabong, which is in the Kgalagadi arid regional district, falls on the south west part of Botswana. Camels kept there are currently used mainly as a tourist attraction (Wilson, 2013b; Seifu *et al*., 2018). However, elsewhere in the traditional camel-rearing regions, camels are mainly kept for milk production followed by transportation and meat production. The camels could be utilised to produce high-quality milk for people living in arid lands (Gitao and Field, 2012). Zeng and McGregor (2008) reported that, although camels contribute to the tourism

business, the number of animals required for tourism is small (150-200). The number of camels managed at the Tsabong Camel Park is currently 576 and this may suggest that the present number of camels is more than the number required for the ecotourism business at a single location. Therefore, concurrent with eco-tourism, there is a need to explore the use of camels for other productive businesses, apart from camel riding, which includes the investigation of the utilization of camel by-products as well as camel milk (BTO, 2012).

Results of a study by Nolte *et al.* (2005), indicated little difference exists between camels from Southern Africa and the Sudan camels, which according to Eisa and Mustafa (2011), yield up to 10 kg/day of milk in early lactation. Yields of up to 30 litres of milk daily have also been reported in Sudanese camels (Osman *et al.*, 2015). Reports indicate that well-fed camels can yield up to 15-20 litres of milk daily during a lactation period of up to 18 months (Razig *et al*., 2008; Faraz *et al*., 2013) or more, up to 24 months (Wernery, 2006). With good feeding, daily yields of 22 kg of milk have been recorded from Egyptian camels, a rise from 4 kg/day when feeding was unstable (Yagil, 1982). This could mean that Tsabong camels may have the potential to produce milk yields better than the annual 2497 litres of cow's milk, which translates to an average of 8 liters/day, as reported by (Moreki and Tsopito, 2013) from dairy cows kept in other parts of Botswana, even when fed. Whilst the free-ranging camels in Tsabong receive no extra feed to supplement them during the dry season but yet, according to Seifu *et al.* (2019), on average, they each can produce 1.7 Litres of milk per day, over 10 months of lactation.

Besides usually being not of the best quality, the majority of dairy cattle breeds used in Botswana, are hardly suitable for local climatic conditions (Moreki *et al*., 2011), hence their unsatisfactory performance. This is evidenced by the absence of dairy farms in the Kgalagadi district probably due to the high ambient temperatures prevalent in this arid area of the country.

Kgalagadi district favours beef and small stock production. However, the district is not suitable for production of fodder crops to feed dairy herds, due to its soils which are sandy and thus poor in plant nutrients (Moreki *et al*., 2011). For dairy animals, in Botswana, there is a decline in milk yield and reduced animal weight gain due mainly to high temperatures and inadequate feeds (Moreki and Tsopito, 2013).

Camels continue to produce milk during very dry periods when cattle and goats are barely surviving (Onjoro *et al.,* 2006). They can produce milk in drought areas where other domestic animals have very low production (Sisay and Awoke, 2015). According to Shawket and Ibrahem (2013), camels are reliable milk producers with a long lactation period (up to 18 months) and they maintain milk production throughout the long dry spells. Camels need only 1.9 kg of dry matter to produce a litre of milk, compared to 9.1 kg for cows (Raziq *et al*., 2008). These adaptations of camels to aridity, allow them to continue providing high volumes of milk that are suited for human consumption, and as such, camels should be reared for milk production.

The results of this study will encourage policymakers in deciding to diversify the economy further by encouraging climatically-disadvantaged Kgalagadi farmers in the arid regions of Botswana to venture into camel dairying, which is more suitable for their otherwise harsh environment, and less suitable for cattle dairy enterprises. The use of camel milk, with its anticipated nutritional and therapeutic benefits, could be the main source of nutrients in arid areas, including vitamin C, and may curb residents' expenditure on medical bills.

1.2 Problem Statement

There is clear evidence of the unsatisfactory performance of cattle dairy production in Botswana, due to the harsh climatic conditions associated with the aridity of Kgalagadi, Kweneng West, Gantsi, and Southern regions of the country, contributing to sandy soils that are poor in plant nutrients and high temperatures (Moreki *et. al*., 2011; Moreki and Tsopito, 2013). In these arid and semiarid areas, which are not suitable for crop production and less suitable for other livestock production, camels continue to live, making them superior to all other livestock in terms of food security (Gebreyohanes and Assen, 2017). Botswana has camels that have been thriving in those climatic conditions for almost a century, producing milk, but ignored as a potential alternative to the cattle dairy industry in those areas. With the anticipated effects of climate change likely to worsen the status of cattle dairying in arid lands, camels may provide an alternative protein source through milk supply.

The use of old man saltbush (*Atriplex nummularia* L) was tried in Bokspits in the Kgalagadi desert in Botswana to stabilize sand dunes and as a fodder crop with successful results (Aganga *et. al*., 2003), has not been fully explored as a means of a locally available but crucial source of perennial fodder for camels, yet research has shown that camels have a high preference for salty plants (Igbal and Khan, 2001) and fresh *Atriplex halimus* in the diet of camels increases the milk production (Shawket and Ibrahem, 2013). Farmers in the arid regions of Botswana need to utilize the niche opportunity derived from a Special Assistance Programme to Kgalagadi District, Gantsi, Kweneng West and Southern Districts with a package for salt-bush fields (LIMID, 2014), and use the fodder to feed and venture into camel dairy production enterprise. The Tsabong camel herd is underutilized and performing well below its potential. Nutritional problems evidenced by Seifu *et al*. (2019) are exacerbated by a lack of

supplementary feeding to improve on feed shortage in the park where camels are only reliant on natural grass and browse, which does not meet the carrying capacity as worsened by the absence of rotational grazing; poor condition of the vegetation in the park and continuous drought. These hamper the production potential of Tsabong camels.

1.3 Justification

The results from this research should enhance development of strategies to improve camel milk production and quality and also shift in perceptions towards carmel milk consumptions. It should also benefit people in the Kgalagadi area in several ways, which include:

a) Monetary- Communities can sell milk to outside markets. By-products such as spray-dried milk, pasteurised milk, butter (ghee) for cosmetics, and whey to sell by owners as a source of protein. According to Kgautlhe (2019), already, the demand for camel milk by visiting tourists is high and the camp has not been able to meet it. The customers, at Tsabong Camel Park, have to wait before they can get the milk as camels take time to produce enough.

b) Health-Boost- Nutritional status of communities will improve. Lactose-intolerant children will thrive better from the use of camel milk in place of the current bovine milk. The high Vitamin-C content of camel milk will meet the vitamin C requirements of people living in the arid Kgalagadi region where it is usually difficult to obtain green vegetables and fruits rich in vitamins.

1.4 Objectives

- ❖ The main objective of the **first study** was to determine, the effects of lactation stage and parity on the composition and microbial quality of camel milk produced in Tsabong, from free-ranging camels.

The specific objectives of this study were:

1. To assess the effects of parity and lactation stage on camel milk composition of free-ranging dromedary camels kept in Tsabong.
2. To determine the microbiological quality of camel milk produced in Tsabong.

- ❖ The main objective of the **Second study** is to establish whether supplementary feeding using Saltbush and Buffelgrass will improve milk yield and milk composition of free-ranging dromedary camels kept in Tsabong.

The specific objectives of the second study were:

1. To analyse the composition of feed/forage for their nutritive value by measuring the *in-vitro* dry matter digestibility (IVDMD) of *Atriplex nummularia* L (Old Man Saltbush.), *Cenchrus ciliaris* (Buffelgrass*)* and sample forages supplying the daily nutrition of Tsabong Camel Park dromedary camels.

2. To determine the changes in milk quality and quantity of foraging camels supplemented with either or both *Atriplex nummularia* and *Cenchrus ciliaris* feeds.

1.5 Hypothesis

1.5.1 On the composition and microbial quality of raw milk

H_O: Parity and lactation stages do not have effects on milk composition.

H_A: Parity and lactation stages have effects on milk composition.

1.5.2 On the milk yield and composition of forage-supplemented camels

H_O: There will not be changes in milk yield, composition and properties of free-ranging camels when supplemented with *Atriplex nummularia* (Saltbush) and *Cenchrus ciliaris* (Buffelgrass) to non-supplemented ones.

H_A: There will be changes in milk yield, composition and properties of free-ranging camels when supplemented with *Atriplex nummularia* (Saltbush) and *Cenchrus ciliaris* (Buffelgrass) compared to non-supplemented ones.

CHAPTER TWO

LITERATURE REVIEW

2.1 Supplementary feeding of camels and milk production

Several factors (i.e. feedstuffs, number of milking per day, health status, genetic makeup, and age and water availability) have been known to influence milk yield and composition (Al-Dobaib, 2009). The most influential factor remains the quantity and quality of the available feedstuffs. Supplementary feeding is a well-known and established concept. Its successful implementation depends on establishing available high-quality feed materials, their cost implications, and formulating rations that work (Noor, 2013).

Noor (2013) recorded a decline in wet-season milk off-take by 33% during the dry season and by 55% during severe drought. Supplementary feeding intervention is expected, from the nutritional point of view, to sustain and improve milk offtake on Tsabong natural browse forage by boosting the nutrient supply needed for maintenance and milk synthesis for potential Tsabong camel owners

Supplementary feed for camels has been provided in the form of pods of certain trees, such as *Acacia* trees (Noor, 2013; Sagala *et al.*, 2021). Other supplementary feeds like *Cenchrus ciliaris*, being highly palatable to camels (Ali *et al.*, 2009) and containing calcium and potassium content that is enough to meet ruminants' dietary supplements Alghamdi (2016), have also been used. *Atriplex nummularia* having high levels of nitrogen and phosphorus (Aganga *et al.*, 2003), characteristic nutrient elements involved in protein synthesis, has been used to supplement lactating camels with cobalt and phosphorus by Onjoro *et al.* (2006), where it significantly increased milk yield. Shawket and Ibrahem (2013) concluded that fresh *Atriplex*

nummularia in the diet of camels increased milk production, as the feed protein content directly affects milk protein (percentage) content and was responsible for increasing milk lactose (percentage) content.

2.2 Composition of Camel milk

On average, according to Brezovecki *et al.* (2015), camel milk contains 81.4-87% water, 10.4 % dry matter, 1.2-6.4 % milk fat, 2.15-4.90 % protein, 1.63-2.76 % casein, 0.65-0.80 % whey protein, 2.90-5.80 % lactose and 0.60-0.90 % ash. Soliman (2005) compared camel milk with buffalo, cow, and goat milk and found, among those species compared, the highest Fe, Zn, Na, and Cu content in Camel milk.

Camel milk differs from other mammal milk as its chemical composition has low cholesterol, low sugar (ranging between 3.3 to 5.80%), high minerals (0.60 to 1.0 percent), high vitamin C (mean value of 34.16 mg/L) and higher protective proteins like lactoferrin, lactoperoxidase, immunoglobulins and lysozyme but lacks B-lactoglobulin. β-lactoglobulin (Kula and Tegegne, 2016). Insulin in camel milk is safe and efficacious in improving long-term glycemic control in a diabetic patient. The milk has high levels of Lactoferrin, a protective protein, which has the ability to inhibit the proliferation of cancer cells. Camel milk is rich in magnesium and zinc and thus endowed with antiulcer properties. Camel milk has high α-hydroxyl acids, which are known to plump and smooth the skin (Kula and Tegegne, 2016).

Maximum whey proteins (0.80%±0.03%) were found in camel milk by Rafiq *et al.* (2016) from comparing nitrogen fractions and amino acids profile of milk from buffalo, cow, sheep, goat and camel. These whey proteins were also positively correlated with true proteins in all milk species.

The content of fatty acids, determined by Mohamed and Mustafa (2016), in milk from camel (*Camelus dromedarius*), on natural pasture, contained 85.7 mg/g of total fat, where, saturated fatty acids formed 63.8%, unsaturated fatty acids formed 36.2%, USFA/ SFA was 0.57, respectively. Monounsaturated acids (MUFA) formed 34.4%, and polyunsaturated acids (PUFA) formed 1.8%. Palmitic acid (C16:0) and Stearic acid (C18:0) were the major SFA, Oleic (C18:1n9c) and Palmitoleic (C16:1) acids were the major MUFA, Linoleic acid (C18:2n6c) was the major PUFA. Short-chain Caproic acid (C6:0) was 0.2%, medium chains (MCFA) was 10.7%, and long chains (LCFA) 89% in milk. Omega 3 and 6 (N-3/ N-6) ratio was 0.08. The content of fatty acids in the milk of *Camelus dromedarius* could be a good potential source of essential fatty acids able to provide the daily requirement of a healthy diet.

2.3 Nutritional values of camel milk

Camel milk is highly nutritious and is very suitable for human nutritional requirements (Al-Juboori *et al.*, 2013). Camels produce more milk of high nutritional quality and for a longer period than other species in an environment that may be rightly termed as hostile in terms of extreme temperature, drought and lack of pasture. Camel milk is rich in vitamin C, Camel milk and its products are a good nutritional source for the human diet Patel *et al.* (2016). The value of camel milk is to be found in the high concentrations of volatile acids and especially, linoleic acid and polyunsaturated acids, which are essential for human nutrition. Camel milk contains high whey proteins such as lactoferrin and immunoglobulin confer to it the high antimicrobial properties. Lactoferrin helps to establish a favourable microflora in the guts and consequently promotes the growth of bifidobacteria.

Camel milk is rich in vitamin C. This is important from the nutritional standpoint in areas where fruit and vegetables containing vitamin C are scarce. Vitamin C in camel milk has antioxidant and tissue repair protection activities. Vitamin C is an essential water-soluble vitamin that helps protect the immune system. According to Pullar *et al.* (2017), Vitamin C is necessary for the body to produce collagen, a protein that aids in the growth of cells and blood vessels and gives skin its firmness and strength. Collagen is found in the skin, joints and cartilage; by increasing the production of collagen. Vitamin C strengthens the structural support and resiliency of skin so helps repair it. Vitamin C is an antioxidant that slows the rate of free-radical damage which causes skin dryness and wrinkles. Vitamin C reverses skin ageing.

Magnesium slows down the ageing process in the skin because magnesium stabilizes DNA and RNA which are both negatively charged and are attracted to the positively charged magnesium. Magnesium is also needed for hair to grow properly because chronic stress can cause hair to fall out as a result of unstable blood sugar, chronic inflammation, not eating properly or getting enough sleep (Pullar *et al.*, 2017).

2.4 Keeping quality of camel milk

Raw camel milk, according to Omer and Eltinay (2009), can successfully be stored at 4^{o}C for 42 days (7days for cow), 7^{o}C for 15 days (70 hours for cow) and at room temperature for 3days (2days for cow), with no significant changes in fat, and protein but in pH, lactose and total solids. Generally, the authors found little change in camel milk during storage at a different temperature, the obvious being odour, and taste but did not coagulate. Shaking was enough to bring the milk back into its original form. El-Demerdash and Al-Otaibi (2012) also showed that pasteurization and refrigeration at 4^{o}C, of raw camel milk, improves the keeping quality

and extends the shelf life for 21 days while Gnan *et al.* (2013), on the other hand, discovered the shelve life in pasteurised camel milk to be longer with, 46 days compared to cow and goat of 36 days only.

2.5 Factors that affect the yield and composition of camel milk

Stage of lactation, parity and season of the year are non-genetic factors having significant effects on daily milk yield, the composition of fat, protein and dry matter (Zeleke, 2007). The study showed that with little decline in milk yield as a lactation stage advanced, camels beyond the fifth parities yielded a lower volume of milk and seasonal effects on milk production potential of camels had the highest daily milk yield recorded during the wet season as compared to the dry season. While the percentage composition of lactose remained unaffected by all variables considered, the percentage compositions of fat and protein were highest during the first 3 months of the lactation period. Similarly, the highest percentage compositions of protein, fat, and dry matter were recorded from camels of 3rd parity.

Babiker and El-Zubeir (2014) recorded the highest means of fat, SNF, protein, and lactose, for the milk of camels in semi-intensive farming during the early lactation stage and at parity number five, showing that husbandry systems, stage of lactation, and parity number have an impact on the chemical composition of camel milk

Factors such as the type of production system feeding strategies and breed differences, investigated by Aljumaah *et al*. (2012) also attribute to variations in camel milk composition. Like Aljumaah *et al*. (2012), Nagy *et al.* (2017) also observed, in a 5-year study, some milk composition variations among the 7 breeds tested, but none of the genotypes was found to be superior to the others in that respect. They also detected a significant, yet small calf sex-biased

difference in milk yield and composition. Mean fat, protein, SNF, and TS concentrations showed a high seasonal variation (9.5 to 28.7%), with the lowest and highest values measured during summer and winter, respectively.

In addition, Nagy *et al.* (2017), showed that dromedary camel milk quantity has a positive correlation with lactose and a negative correlation with all other measured components of fat, protein SNF and TS concentrations of the morning milk. On the other hand, parity exerted a strong effect on all milk parameters, with primiparous dromedaries producing less milk with higher concentrations of components than multiparous animals. Brezovecki *et al.* (2015) also found variations due to analytical methods, geographical area, nutrition conditions, and age among others.

2.6 Microbiological quality and safety of camel milk

Milk is an excellent culture medium for the growth of microorganisms. The rate of multiplication of microbes depends mainly on storage temperature and time, level of nutrients and handling conditions (Matofari *et al*., 2013). The external sources of microbes include the equipment, the personnel, and water. The ability of microorganisms to cause spoilage and disease depends upon the type present, the initial load of contamination of the milk, handling conditions, and the time-lapse from production before consumption. Since the milk from Tsabong camels might need to be transported to markets countrywide, including the processing units usually situated in towns (Gaborone, Phikwe and Francistown, etc.), handling procedures along the chain may predispose to contaminations and the possibility of occurrence of pathogens along different temperature gradients. Even though the results of the lactic acid development of camel milk, by the authors, may imply the self-preserving quality of the camel milk, spoilage at any level will contribute to food safety hazard to the consumers.

For standardisation of quality parameters in camel milk, grading according to levels of microorganism detection are set, at which milk would be either acceptable or rendered not fit for human consumption. Microbiological limits set by the Kenya Bureau of Standards and Botswana Bureau of standards for raw milk are displayed, for plates incubated for 48 hrs at 32^{o}C and 24 hrs at 37^{o}C, for Total Viable Count and Coliform Count respectively, in Table 2.11.1 and Table 2.11.2 below, for camel and cow raw milk, respectively.

Table 2.11.1: Grade per total viable count and coliform limits of camel milk

Grade	Total viable count (counts/ml)	Coliform counts (counts/ml)
I	<200 000	0 – 1 000
II	>200 000 – 1 000 000	1 000 – 50 000
III	>1 000 000 – 2 000 000	50 000 – 100 000

Adapted from: Kenya Bureau of Standards classification of raw camel milk KS (2016)

Table 2.11.2: Microbial requirements of Raw Cow's milk

Parameter	Counts (CFU/ml)
Total plate count	< 200 000
Total coliform count	<20
Escherichia coli	not detectable

Source: Botswana Bureau of Standards Raw Milk-Specifications BOS 64:2018

2.7 Contribution of Camel Milk to Pastoral Livelihoods in Arid Zones

2.7.1 Nutritional benefits

According to Brezovecki *et al.* (2015), camel milk could soon become the new superfood due to its high nutritional value, easy digestibility (suitable for lactose intolerant people), and low share of fat. Given the observed results of the physicochemical properties of the camel milk, Khaskheli *et al.*, (2005) concluded that camel produces nutritious milk for human consumption.

Camel milk is full of evenly balanced nutritional constituents and displays a wide variety of biological actions that influence the growth and development of particular body organs, metabolic responses towards nutrient absorption, digestion, and the fight against diseases (Abbas *et al*., 2013). Camel milk, in the arid region, where citrus fruits and vegetables containing Vitamin C can only be imported, may be an important source of essential components as it is three to five times higher in vitamin C (34.16 mg/L) than in cow's milk (Wernery, 2006; Sharma and Singh, 2014; Yadav *et al*., 2015; Seifu, 2022). Camel milk contains, also, vitamins A, E, D, and B group, though lower in vitamins A and B_2 than cow's milk (Farah *et al*., 2007; Brezovecki *et al* 2015). It is ten times higher in iron than cow's milk (Sharma and Singh, 2014). According to its chemical composition, camel milk is most similar to human milk (Brezovecki *et al* 2015). Compared to cow's milk, it's reported and to be having low cholesterol, low sugar, high minerals (sodium, potassium, iron, copper, zinc, manganese, and magnesium), (Sharma and Singh, 2014; Yadav *et al*, 2015; Seifu, 2022).

Though both camel and donkey milk are enriched with minerals and lactose (Al-haj and Al-Kanhal, 2010). The Ca, P, K, Na and Mg contents, of 109 mg/100 ml, 76 mg/100 ml, 179 mg/100 ml, 58 mg/100 ml, and 14 mg/100 ml, respectively of camel milk observed by Rathore

et al. (2011) were lower than that of donkey milk, which were reported to be 676.6 mg kg^{-1}, 487.0 mg kg^{-1}, 497.2 mg kg^{-1}, 218.3 mg kg^{-1}, and 37.3 mg kg^{-1} for Ca, P, K, Na and Mg, respectively. Also lower than the values 807.09 mg L^{-1}, 638.42 mg L^{-1}, 746.61 mg L^{-1}, 140.94 mg L^{-1} and 81.69 mg L^{-1} for Ca, P, K, Na and Mg, respectively reported by Fantuz *et al.* (2012) for donkey milk samples.

Camel milk is a good source of protein and Brezovecki *et al.*, (2015) referred to it as a "complete meal" because it contains enough nutrients for maintaining life and is often given to children suffering from malnutrition. Yadav *et al.* (2015) reported camel milk proteins to contain a satisfactory balance of essential amino acids. Ethiopian pastoralists gave reasons for their preference for camel milk over milk of other domestic animals (Sisay and Awoke, 2015), citing cow's milk to have a tendency of making people fat whilst that of camels gives strength, endurance, and stamina, and attributes that pastoralists need to pursue a nomadic lifestyle.

2.7.2 Medicinal benefits

Fresh and fermented camel milk products are thought to provide treatment for gastritis, asthmatics, and stomach discomfort, and alleviate symptoms associated with HIV, nausea, tuberculosis, fever, urinary problems, hepatitis, jaundice, common cold, diarrhoea (Asres and Yusuf, 2014).

Al-Haj and Al-Kanhal (2010) and Seifu (2022) found fresh and fermented camel milk, due to the presence of bioactive substances in milk (lactoferrin, lysozyme, lactoperoxidase, hydrogen peroxide and immunoglobulins), to provide various potential health benefits including angiotensin 1-converting enzyme-inhibitory activity, hypocholesterolaemia effect, hypoglycaemic effects, antimicrobial and hypoallergenic effects.

Other potential therapeutic properties such as anti-hypertensive, anti-diabetic and anti-carcinogenic were also reported (Yadav *et al*., 2015; Seifu, 2022). Camel milk also has the ability to reduce elevated level of bilirubin, globulin and granulocytes as contains disease-fighting immunoglobulins, which are small in size, allowing penetration of antigens and boosting the effectiveness of the immune system (Yadav *et al.*, 2015; Sharma and Singh (2014) reported camel milk in India being used therapeutically against dropsy, jaundice, problems of the spleen, tuberculosis, asthma, anaemia, and piles as well as reported slimming properties. In Ethiopia, additionally, camel milk is also used to provide treatment for a series of diseases such as malaria, and constipation, clean the stomach, post-partum care for women, and detoxifying snake venom and flatulence (Asres and Yusuf, 2014).

2.7.3 Economic opportunities

The dairy cattle sub-sector contributes insignificantly to the National Gross Domestic Product (GDP). However, the contribution of the dairy camel sub-sector can add to the National Economic Report, as has the camels contributed to the tourism industry over the past decade. Research is underway to investigate the use of camel milk as a supplement to mother's milk or as an alternative formula to provide nutritious fresh milk (Yagil *et al.,* 1994). Faye (2014) justified growing interest in camel milk from the urbanised populations of countries with desert environments. Indications from market-oriented smallholder camel dairying in east Africa, already far outweigh those from alternative traditional agricultural activities (Farah *et al*., 2007). This might be a possible and big market opportunity for Kgalagadi farmers to produce and sell camel milk to milk processors locally and internationally.

To extend the storage life of camel milk, for far markets like Gaborone and the rest of Botswana, value added products like cheese, butter and fermented milk (Brezovecki *et al.*, 2015) can be processed as niche opportunities. Some studies, including that of Ibrahem and El-Zubeir (2016), showed that through variations in camel and sheep milk, camel-sheep yoghurt mixtures produce a higher content of SNF, fat and protein compared to pure camel milk. This presents possibilities of processing and marketing it as the health benefits of camel milk and fermented products are well documented. Small scale mobile processing units may be established to make use of the valuable camel milk. For Kgalagadi farmers with many sheep, this may be a solution for proper utilization of resources which could improve food security and enhance rural development by including their milk in the production process.

CHAPTER THREE

EFFECT OF LACTATION STAGE AND PARITY ON CHEMICAL COMPOSITION AND THE MICROBIAL QUALITY OF RAW CAMEL MILK UNDER RANGE CONDITIONS IN TSABONG CAMEL PARK

3.1 Abstract

Effects of lactation stage (LS) and parity (PA) on composition and properties of camel milk were evaluated using 24 dromedary camels reared in Tsabong Ecotourism Camel Park. Primiparous and multiparous camels in their early, mid and late lactation stages were randomly selected for the experiment. Ten (10) milk samples were randomly selected and analysed for total coliform bacteria and total viable microorganisms. Coliforms were not detected in the milk samples. The total viable microorganism in the milk samples ranged between <1.00 X 10^2 CFU/ml and <14.20 X 10^2 CFU/ml. The composition of camel milk produced in Tsabong was affected by lactation stage as well as interaction of LS x PA. Interaction between lactation stage and parity had a significant reduced levels of fat, solids-not-fat (SNF), free fatty acids (FFA), galactose, and density. The lactation stage reduced galactose($p<0.05$) and glucose ($p<0.01$) levels. Casein, citric acid, freezing point, lactic acid, lactose, total solids (TS), protein and urea were not affected by both parity and lactation stages. The highest levels of TS (9.785±0.697%) and fat (3.675%±0.442%), FFA(0.580±0.057g/L), galactose (0.685±0.066%) and glucose (0.363±0.042%n), were all observed in primiparous camels in early lactation. Protein (1.910±0.188%) and casein (1.718±0.104%) both highest in multiparous camels in mid-lactation, while those of SNF (6.330±0.313%) and lactose, (3.568±0.227%), were highest in primiparous camels in late lactation. Lactation stage and interaction between lactation stage and parity increased ($P<0.05$) SNF, FFA, galactose, glucose, and density of the milk. Those components that were not positively influenced by parity and lactation stage will have to be improved by supplementary feeding and nutrition. Camel milk producers could target specific levels of desired components of camel

milk as may be required by prospective consumers and milk processors, based on milk obtained from camels at specific stages of lactation and parities.

3.2 INTRODUCTION

Camel milk is important to the human diet in many parts of the world (El-Demerdash and Al-Otaibi, 2012). The mean values of camel milk composition reported by Al-haj and Al-Kanhal (2010) were; 3.1%, 3.5%, 4.4%, 0.79 and 11.9% for protein, fat, lactose, ash and total solids, respectively.

The quality of milk is influenced by different bacteria present in the milk. The presence of coliforms in milk suggests the hygienic condition under which the milk was produced, and it also indicates the likelihood of presence of pathogenic bacteria in the milk. Camel milk was found to have high contents of proteins with antimicrobial properties (El-Demerdash and Al-Otaibi, 2012) and as a result it usually has longer shelf life as compared to bovine milk and milk from other species.

To date, no study has been conducted on the chemical composition and microbial quality of camel milk produced under Botswana arid climatic condition. It is known that changes in the environment have had a significant effect on the natural physiological function of animals, so it is very important to make such a study under Botswana's environment (ecology), hoping to give an understanding and explain some of the malnutrition problems in Botswana. Data from the study will also inform milk processing industries about the characteristics of camel milk produced in Botswana which will aid in processing techniques and equipment calibrations and adjustments.

3.3 MATERIALS AND METHODS

3.3.1 Study area

The study was conducted in Tsabong Ecotourism Camel Park, which is found in the Kgalagadi District in southern Botswana (Figure 1). The study site, Tsabong Ecotourism Camels Park (25º56'15.5" S, 22º27'30.5" E), is located at 520 km from the capital Gaborone and 10 km north of Tsabong town and comprises a fenced area of 3200 hectares. The area is characterized by poor and unreliable rainfall with annual precipitation of less than 250 mm and average ambient temperatures of above 35°C during summer and less than 2°C in winter (Kgaudi, 2014). With an annual average rainfall of about 200 mm, this area, like the rest of Kgalagadi South, is one of the driest in Botswana (Ditlhogo *et al*., 2020). The area has sparsely distributed vegetation dominated by *Acacia, Boscia, Grewia* and *Schmidtia* species and some species of grass. It is dominated by native species of thorn bushes that grow naturally in grass veld and thornbush savannah (Table 3.3.1.1).

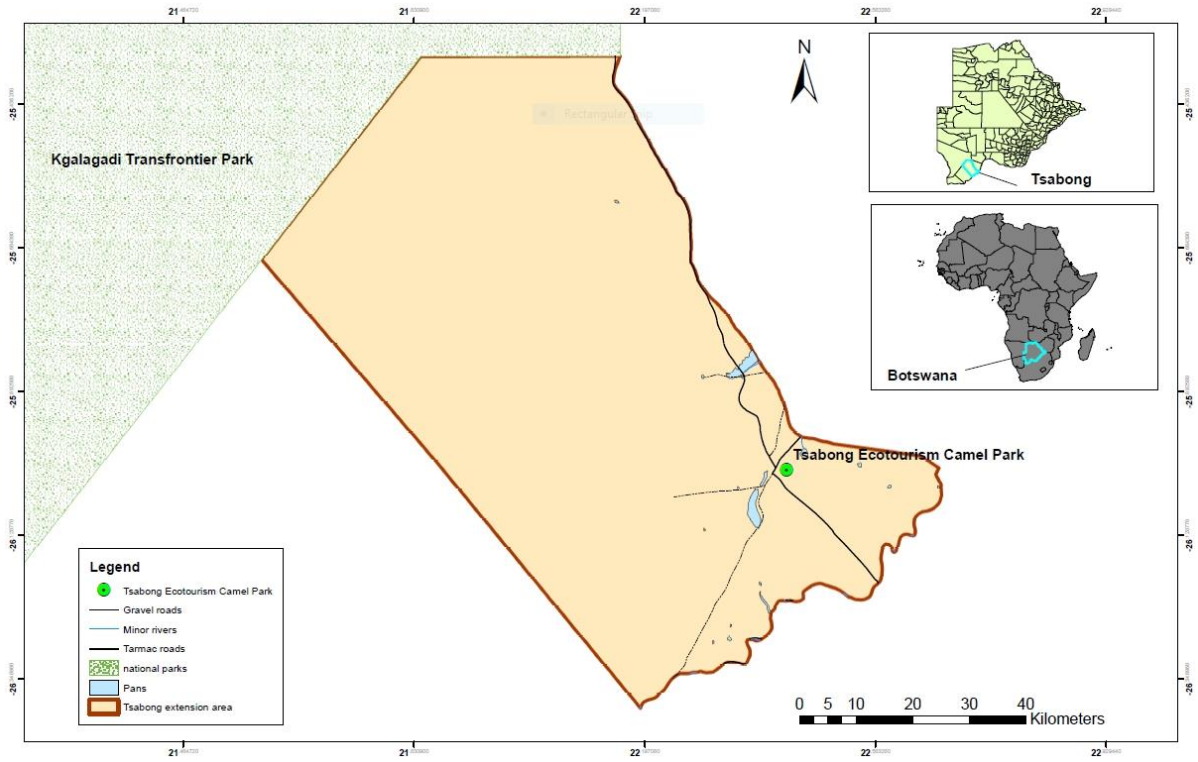

Figure 1: Location of Tsabong Ecotourism Camel Park

Table 3.3.1.1: Plant species browsed by camels in Tsabong Ecotourism Camel Park

Scientific name	Setswana name	English name
Vachellia erioloba E. Mey	Mogotlho	Camel Thorn
Senegalia galpinii Burtt Davy or A. luederitzii Engl.	Mokala	Monkey Thorn
Vachellia hebeclada DC.	Sekhi	Candle-pod Acacia
Vachellia karroo Hayne	Mokha	Sweet Thorn
Senegalia mellifera (Vahl) Benth.	Mongana	Black Thorn
Boscia albitrunca (Burch.) Gilg & Benedict	Motlopi	Shepherd's Tree
Grewia flava DC	Moretlwa	Brandy Bush
Rhus tenuinervis Engl	Modupaphiri	Kalahari currant
Rhigozum trichotomum Burch.	Mokurubane	Threethorn
Schmidtia kalahariensis Stent	-	Kalahari sour grass
Tribulus terrestris L.	Mosetlho	Devil's thorn

Source: Seifu *et al.* (2019)

3.3.2 Management of the camels

When this study was conducted in 2020, Tsabong Camel Park had over 470 camels, of which about 86 were lactating. Twenty-four (24) primiparous and multiparous camels in their early, mid and late lactation stages were randomly selected for the experiment and used for the experiment. All camels were managed and watered similarly. They were all free-ranging and not kraaled at night, except at nights before milking. Before milking, the teats of the camels were washed with clean tap water and dried with a single service towel. The health of the camels was checked regularly so that when necessary, they would be treated accordingly. The camels were identified by their already existing brands and the stage of lactation, parity, pregnancy, the season of calving, and the age of the camels were recorded.

3.3.3 Collection of milk samples

Milk samples for the determination of chemical composition were obtained from each of the 24 individual camels. Primiparous camels in their early, mid and late stages of lactation and Multiparous camels in each of those three stages of lactation were randomly selected for the experiment with four replicates of each combination for this study. Since Tsabong dromedary camels' lactation lasts for a year, lactation stages were determined as Early-lactation (up to 4 months), Mid-lactation (over 4 months, but less than 8 months) and Late-lactation (over 8 months). Calves were identified and captured using brands as per appendix AP3b. Milking was done straight into sample bottles for each camel (appendix AP1e). Milk samples for microbial analysis were obtained in duplicate, placed into sterile (autoclaved) containers (glass bottles) and transported to the laboratory immediately after collection by placing them in a cooler box and kept at 4°C overnight until laboratory analysis.

For the microbial analysis, ten (10) milk samples out of the 24 collected were randomly selected using R software version 3.6.0 (R Core Team, 2019) using the formula:

```
# r sample multiple times without replacement

sample (c (1:24), size =10, replace = F),
```

and the 10 samples were analysed for the total viable count, and coliform count and were also tested for the presence of the *Escherichia coli* O157:H7 pathogens.

3.3.4 Milk composition and properties

The percentages of: Lactose, Galactose, Glucose, Total Solids, Solids-Not-Fat, Fat, Protein, Casein, Urea, Free Fatty Acids, Lactic Acid and Citric Acid contents were analysed using an automatic milk analyser (MilkoScan FT 1; Foss A/S, Hillerød, Denmark). The physical properties of the milk samples: freezing point, and density (SG) were measured using the same milk analyser. The MilkoScan FT 1 User Manual 6004 5478/Rev.3, and Software Manual 6004 4622/Rev.11, were followed during the procedure, for the preparation of samples and analysis, respectively.

3.3.5 Microbiological Analysis

3.3.5.1 Total Viable Microorganisms

Total viable count (TVC), as a quantitative estimate of the concentration of microorganisms such as bacteria, yeast and moulds in milk samples, was used as a hygienic quality indicator to ensure consumer safety. The testing was done at Botswana National Veterinary Laboratory, following the work instructions for enumeration of total viable microorganisms in milk and

milk products by pour plate method (BNVL, 2020). One mL of raw milk sample was aseptically added to 9 mL of diluent (1% peptone water) and mixed thoroughly to produce a 10^{-1} dilution. 1 mL of the 10^{-1} dilution was then transferred to 9 mL of the diluent and mixed thoroughly to produce a 10^{-2} dilution. The 1 mL of the 10^{-2} dilution was further transferred to 9 mL of the diluent and mixed thoroughly to produce a 10^{-3} dilution. The procedure was repeated until the required 10^{-6} dilution was made. One mL of each dilution was aseptically transferred to the centre of the corresponding dish containing 12 to 15 mL of agar (PCA), cooled to 44-47^{o}C, and poured onto all plates. The solidified plates were, then overlaid with approximately 4 mL of PCA, inverted and aerobically incubated at 30^{o}C for 72 hours. Following the period of incubation, the controls were checked as per provided table for acceptability, that was provided in the (BNVL, 2020) manual. Plates with colonies ranging from 10 to 300 colony-forming units (CFU) were then selected for the determination of standard plate count. All spreading colonies were counted as a single colony. The number of microorganisms present in the test sample was calculated using the following equation:

$\mathbf{N} = \sum C / (V (n_1 + 0.1 n_2) d)$, (BNVL, 2020)

Where:

N is the number of colony-forming units/g or ml of sample

V is the volume of inoculum applied to each dish, in millilitres

$\sum$**C** is the sum of colonies counted on all the dishes retained

$\mathbf{n_1}$ is the number of dishes retained at the 1st dilution

$\mathbf{n_2}$ is the number of dishes retained at the 2nd dilution

d is the dilution factor corresponding to 1st dilution retained

To the total colony-forming units/mL of the sample obtained, the Uncertainty of Measurement value was calculated using the following formula:

$UM=1.96\sqrt{N}$ (BNVL, 2020)

Where:

UM is the Uncertainty of Measurement

$\sqrt{\mathbf{N}}$ is the square root of the total number of colony-forming units/mL

1.96 is used in this case assuming 95% confidence limits

The results were recorded as $X \times 10^{n}$cfu/ml

Where:

X is the number between 1.0 and 9.9

n is the appropriate power of 10

3.3.5.2 Total Coliform count

Total coliforms are indicator organisms used to monitor conditions in consumer products. In this method, coliforms rapidly ferment the lactose in violet red bile agar (VRBA) and so reduce the pH of the medium, producing purple colonies due to the inclusion of neutral red and crystal violet. Samples testing was done at Botswana National Veterinary Laboratory, following the instructions for the enumeration of total coliform organisms in milk and milk products by the pour plate method described by (BNVL, 2021).

Volumes (1 ml) of appropriate dilutions, up to 10^{-5}, were plated by the pour plate technique in duplicate using Violet Red Bile Agar. Culturing was done as for the total viable microorganism above. Then approximately 12-15 ml of VRBA cooled to 44-47^{o}C was poured onto each plate, including control plates, covering the entire bottom of the plates. After allowing the plate to solidify, about 4 ml of the plating medium was distributed as an overlay. This was done to ensure anaerobic conditions which suppress the growth of non-fermentative Gram-negative bacteria, encourage the fermentation of lactose which favours the formation of clearly visible purple colonies surrounded by a purple halo, and inhibit surface colony formation. The plates were then inverted and incubated aerobically at 30^{o}C for 26 hours. Following the incubation period, controls were checked as per the acceptance criteria for test results (provided by BNVL).

After incubation, typically dark red or purplish-red colonies appearing on the plates were counted as coliforms. For the confirmatory test, five to ten typical colonies from each plate were transferred into tubes containing 2% Brilliant Green Bile Broth (BGBB) containing a Durham tube (each colony into a separate tube) and incubated at 38˚C for 26 hours. The presence of gas in the inverted Durham tube of effervescence within the incubation period was considered sufficient evidence for the presence of coliforms. Plates with 15 to 150 colonies were used. Since at counting no atypical colonies were observed, the total coliform count was calculated in the test samples using the formula used in section *3.3.5.1* above, for calculating the number of total viable microorganisms present in the test sample. There was no need for confirmation by further inoculating colonies in BGBB.

3.4 Statistical Analysis

The chemical composition of milk samples was analysed using the General Linear Model procedure of the Statistical Analysis System (2006). The design of the experiment was a Factorial in Completely Randomized Design (CRD) and Duncan's multiple range tests (P = 0.05) were used to determine significant differences between means.

The statistical model used: $Y_{ijk} = \mu + \alpha_i + \beta j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$

Where

Y_{ijk} = chemical composition of j^{th} milk from an i^{th} animal

μ = the overall mean

$\alpha_i = j^{th}$ (1, 2) effects of parity (Primiparous and Multiparous)

$\beta j = i^{th}$ (1, 2….3) effects of Lactation stage (Early, Mid and Late)

$(\alpha\beta)_{ij}$ = interaction between Parity and Stage of Lactation

ε_{ijk} = milk compositional error

A CRD involves two factors with different levels for each: Lactation stage Levels (early, mid and Late) and Parity Levels (Primiparous and Multiparous) applied to the camels. The response variables were measurements of various milk quality parameters being; total solids, protein, fat, solids-not-fat, free fatty acids, casein, lactose, galactose, glucose, freezing point, lactic acid, citric acid, density, and urea contents of raw camel milk.

Experimental design

Factor	Stage of Lactation A=S level					
Parity B = P level	Level	a_1=Early	a_2=Mid	a_3=Late	Mean (a_{bi})	a_3-a_1
	b_1= Primiparous	PE	PM	PL	Mean (PE:PL)	(se A,b_1)
	b_2=Multiparous	ME	MM	ML	Mean (ME:ML)	(*se* A,b_2)
	Mean (a_ib)	Mean (PE:ME)	Mean (PM:MM)	Mean (PL:ML)		(me A)
	b_2-b_1	(*se* B,a_1)	(*se* B,a_2)	(*se* B,a_3)	(me B)	

Where: S=Stage of lactation, P=Parity, PE=Primiparous in Early Lactation, PM=Primiparous in Mid lactation, PL=Primiparous in Late lactation, ME=Multiparous in Early Lactation, MM=Multiparous in Mid Lactation, ML=Multiparous in Late Lactation, *se*=standard error, me=mean error.

General ANOVA table for the two-way CRD factorial experiment

Source	Degrees of Freedom	SS	MS	F
Lactation	2	SS Lactation	MS Lactation	MS Lactation /MSE
Parity	1	SS Parity	MS Parity	MS Parity /MSE
Lactation x Parity	2	SS Lactation x Parity	MS Lactation x Parity	MS Lactation x Parity/MSE
Error	19	SSE	MSE	
Total	23	TSS		

Where: SS=Sum of Squares, MS=Mean of Squares, F=F-value, MSE-Error Mean of Squares, SSE=Error Sum of Squares.

3.5 RESULTS

The findings of this research are presented in Tables 3.5.1 to 3.5.4. Tables (3.5.1-3.5.3) compared milk composition amongst camels within combinations of three lactation stages and two parity stages. Table 3.5.4 presented the microbial quality of milk samples randomly sampled from 10 camels found at Tsabong Camel Park. Significant differences were tested at (P=0.05). It was found that interaction between lactation stage and parity (P<0.05) affected levels of fat, SNF, FFA, galactose, and density. Glucose was highly affected (P<0.01) by both lactation stage (P=0.0013) and interaction between the lactation stage and parity (P=0.0012). Parity alone did not show any significant effect (NS) on all components.

Table 3.5.1 shows the chemical composition of Tsabong dromedary camel's milk as influenced by parity and lactation stages, and the means of the components measured. The highest total solids, fats and FFA in primiparous camels were found in early lactation, whilst the highest protein SNF and casein were found in the late lactation stage. Except for SNF and casein, all other components of the milk were higher than those recorded for multiparous at the same lactation stages. Multiparous camels produced the highest total solids, protein, SNF and casein during mid lactation, and those of fat and FFA during early lactation stages. Except for fat, all other components exceeded the levels found in primiparous at the same stages of lactation. Milk Fat and FFA components in multiparous, as well as casein in primiparous camels all decreased with advancement in lactation stages. All other milk components fluctuated between lactation stages. The means of total solids (TS), fat, and protein, solids-not-fat (SNF), free fatty acids (FFA) and casein were 8.546±0.402%, 2.585±0.255%, 1.791±0.108%, 5.811±0.181%, 0.485±0.033g/L, 1.632±0.060%, respectively.

There were no significant changes in the levels of TS, protein and casein that were influenced by lactation stage, parity or interaction between lactation stage and parity

Table 3.5.1: Means of chemical composition of dromedary camels' milk at Tsabong Camel Park

Parameter		Parities			Sign.	
		Primiparous	Multiparous	LS	PA	LSxPA
	Lactation Stage	LSM±SEM	LSM±SEM			
Total solids (%)	Early	9.785±0.697^{A}	8.293±0.697^{A}			
	Mid	7.880±0.697^{A}	8.370±0.697^{A}			
	Late	8.976±0.697^{A}	7.970±0.697^{A}			
	Mean[1]	8.881±0.402^{A}	8.211±0.402^{A}	NS	NS	NS
Fat (%)	Early	3.675±0.442^{A}	2.653±0.442^{A}			
	Mid	2.355±0.442^{B}	2.170±0.442^{B}			
	Late	2.678±0.442^{AB}	1.980±0.442^{B}			
	Mean[1]	2.903±0.255^{A}	2.268±0.255^{AB}	NS	NS	*
Protein (%)	Early	1.860±0.188^{A}	1.600±0.188^{A}			
	Mid	1.647±0.188^{A}	1.910±0.188^{A}			
	Late	1.863±0.188^{A}	1.865±0.188^{A}			
	Mean[1]	1.790±0.108^{A}	1.792±0.108^{A}	NS	NS	NS
SNF (%)	Early	5.903±0.313^{AB}	5.525±0.313^{AB}			
	Mid	5.323±0.313^{B}	6.000±0.313^{AB}			
	Late	6.330±0.313^{A}	5.785±0.313^{AB}			
	Mean[1]	5.852±0.181^{A}	5.770±0.181^{A}	NS	NS	*
FFA (g/L)	Early	0.580±0.057^{A}	0.516±0.057^{AB}			
	Mid	0.407±0.057^{B}	0.457±0.057^{AB}			
	Late	0.539±0.057^{AB}	0.413±0.057^{AB}			
	Mean[1]	0.509±0.033^{A}	0.462±0.033^{A}	NS	NS	*
Casein (%)	Early	1.583±0.104^{A}	1.513±0.104^{A}			
	Mid	1.580±0.104^{A}	1.718±0.104^{A}			
	Late	1.683±0.104^{A}	1.715±0.104^{A}			
	Mean[1]	1.615±0.060^{A}	1.648±0.060^{A}	NS	NS	NS

Values in the same columns or rows, except mean[1], with different superscripts $^{A, B,}$ were significantly different ($P \leq 0.05$). [1]Mean values in the same rows with different superscripts $^{A, B}$ were significantly different ($P \leq 0.05$). LS= lactation stage, PA=parity, NS=No significant difference among the means, *=significant ($P<0.05$) **=highly significant ($P<0.01$) ***= highly significant ($P<0.001$), SNF=solids-not-fat, FFA=free fatty acids, LSM=least square mean and SEM=standard error of mean.

The Sugar fractions of Tsabong dromedary camel's milk as influenced by parity and lactation stages and the means of the components measured are shown in Table 3.5.2. The means of lactose, galactose and glucose were 3.332±0.131%, 0.498±0.038% and 0.216±0.024%, respectively. Lactose values were the same ($P>0.05$) across all stages of lactation and parities, while milk galactose levels decreased ($P<0.01$) by 61.9% from early lactation to mid lactation

in primiparous camels. Galactose concentration decrease was observed between primiparous in early lactation and multiparous in late lactation, where a 36.5% decrease ($P<0.05$) was recorded. Generally, there was an interaction effect of lactation stage and parity on milk sugar content, with the highest effect ($P\leq0.01$) observed for glucose concentration. Galactose decreased ($P\leq0.05$) with advancement in lactation for the multiparous camels. While Primiparous camels in their early lactation produced the highest levels of galactose and glucose, (0.685±0.066%) and (0.363±0.042%), respectively. Primiparous camels in late lactation produced the highest levels of lactose (3.568±0.227%).

Table 3.5.2: Means of sugar fractions of dromedary camel milk at Tsabong Camel Park

Parameter		Parities			Sign.	
		Primiparous	Multiparous	LS	PA	LSxPA
	Lactation Stage	LSM±SEM	LSM±SEM			
Lactose (%)	Early	3.498±0.227^{A}	3.230±0.227^{A}			
	Mid	2.990±0.227^{A}	3.448±0.227^{A}			
	Late	3.568±0.227^{A}	3.258±0.227^{A}			
	Mean[1]	3.352±0.131^{A}	3.312±0.131^{A}	NS	NS	NS
Galactose (%)	Early	0.685±0.066^{A}	0.530±0.066^{AB}			
	Mid	0.438±0.066^{B}	0.445±0.066^{B}			
	Late	0.455±0.066^{B}	0.435±0.066^{B}			
	Mean[1]	0.526±0.038^{A}	0.470±0.038^{A}	*	NS	*
Glucose (%)	Early	0.363±0.042^{A}	0.265±0.042^{AB}			
	Mid	0.138±0.042^{C}	0.175±0.042^{BC}			
	Late	0.195±0.042^{BC}	0.163±0.042^{BC}			
	Mean[1]	0.232±0.024^{A}	0.201±0.024^{A}	**	NS	**

Values in the same columns or rows, except mean[1], with different superscripts $^{A, B, C}$ were significantly different ($P\leq0.01$).[1]Mean values in the same rows with different superscripts $^{A, B}$ were significantly different ($P\leq0.05$).
LS= lactation stage, PA=parity, NS=No significant difference among the means, *=significant ($P<0.05$)
=highly significant ($P<0.01$) *=highly significant ($P<0.001$), LSM=least square mean and SEM=standard error of mean.

Table 3.5.3 shows the physical properties of Tsabong dromedary camels' milk as influenced by parity and lactation stages, and the means of the components measured. Means for freezing point, lactic acid, citric acid, density and urea were -0.408±0.016^{o}C, 0.109±0.003%, 0.142±0.006%, 1023.904 ±0.735 and 193.9±10.439mg/L, respectively. Freezing point, lactic

acid, urea and citric acid mean values remained constant (P≥0.05) across all levels of parity and lactation stages. Lactic acid and citric acid showed insignificant increases with advancement of lactation stage in primiparous camels. The difference in milk density was observed highest between primiparous camels in mid and late lactations

Table 3.5.3. Properties of dromedary camel milk at Tsabong Camel Park

Parameter		Parities			Sign.	
		Primiparous	Multiparous	LS	PA	LSxPA
	Lactation Stage	LSM±SEM	LSM±SEM			
Freezing point (ºC)	Early	-0.435±0.027^{A}	-0.408±0.027^{A}			
	Mid	-0.365±0.027^{A}	-0.410±0.027^{A}			
	Late	-0.438±0.027^{A}	-0.390±0.027^{A}			
	Mean[1]	-0.413±0.016^{A}	-0.403±0.016^{A}	NS	NS	NS
Lactic acid (%)	Early	0.104±0.005^{A}	0.104±0.005^{A}			
	Mid	0.107±0.005^{A}	0.114±0.005^{A}			
	Late	0.114±0.005^{A}	0.110±0.005^{A}			
	Mean[1]	0.108±0.003^{A}	0.109±0.003^{A}	NS	NS	NS
Citric acid (%)	Early	0.135±0.011^{A}	0.138±0.011^{A}			
	Mid	0.143±0.011^{A}	0.145±0.011^{A}			
	Late	0.150±0.011^{A}	0.143±0.011^{A}			
	Mean[1]	0.143±0.006^{A}	0.142±0.006^{A}	NS	NS	NS
Density (SG)	Early	1023.850±1.272^{AB}	1022.425±1.272^{AB}			
	Mid	1021.975±1.272^{B}	1024.800±1.272^{AB}			
	Late	1025.975±1.272^{A}	1024.400±1.272^{AB}			
	Mean[1]	1023.933±0.735^{A}	1023.875±0.735^{A}	NS	NS	*
Urea (mg/L)	Early	180.708±18.081^{A}	193.883±18.081^{A}			
	Mid	168.495±18.081^{A}	207.304±18.081^{A}			
	Late	208.813±18.081^{A}	203.965±18.081^{A}			
	Early	186.007±10.439^{A}	201.717±10.439^{A}	NS	NS	NS

Values in the same columns or rows, except mean[1], with different superscripts $^{A, B,}$ were significantly different (P≤0.05).[1]Mean values in the same rows with different superscripts $^{A, B}$ were significantly different (P≤0.05).
LS= lactation stage, PA=parity, NS=No significant difference among the means, *=significant (P<0.05) **=highly significant (P<0.01) ***=highly significant (P<0.001), LSM=least square mean and SEM=standard error of mean.

Table 3.5.4., below, shows the results of ten milk samples randomly selected and tested for coliform bacteria and total viable microorganisms. Coliforms were not detected in the milk samples. The total viable microorganism in the milk samples ranged between <1.00 X 10^2 CFU/ml and <14.20 X 10^2 CFU/ml.

Table 3.5.4. Microbial quality of dromedary camels' milk samples collected from Tsabong Camel Park

Sample	Detection and enumeration of Coliforms/ml	Enumeration of Total Viable Microorganisms
1	<1.0	<4.00 X 10^2 cfu/ml
2	<1.0	<3.70 X 10^2 cfu/ml
3	<1.0	<14.20 X 10^2 cfu/ml
4	<1.0	<4.25 X 10^2 cfu/ml
5	<1.0	<5.95 X 10^2 cfu/ml
6	<1.0	<10.85 X 10^2 cfu/ml
7	<1.0	<2.00 X 10^2 cfu/ml
8	<1.0	<1.00 X 10^2 cfu/ml
9	<1.0	<4.15 X 10^2 cfu/ml
10	<1.0	<9.50 X 10^2 cfu/ml

Where cfu=coliform units and ml=millilitre

3.6 DISCUSSION

Lactation stage and parity did not have significant effects on all milk components, but their interaction effects showed differences for fat, solids-not-fat and free fatty acid concentrations (Table 3.5.1).

The results were in disagreement with the observations reported by Mal *et al*. (2007) and Mustafa *et al.* (2021) who reported higher protein, casein, fat, lactose, and total solids contents in the late phase of lactation, and Zeleke (2007) and Babiker and El-Zubeir (2014) who reported fat and protein percentages being highest in the early months of the lactation period. Idrees *et*

al. (2016) reported significant effects of lactation on total solids and fat, both highest in the late stages of lactation, and no significant changes in the levels of protein and lactose contents of camel milk.

A similar observation to ours was that made by Alwen and Zwaik (2014) who found no significant effects of lactation stages on fat and protein concentrations on camels reared under desert and farm conditions. Our results show a non-significant decline in concentrations of fat towards the late stage of lactation, for both primiparous and multiparous camels.

For free ranging camels reared under conditions of similar arid environments like Tsabong camels and Libyan Maghrebi camels (Alwen and Zwaik, 2014), the level of nutrition and supplementary feeding appear to directly influence the lactation stage's effect. This is as evidenced by the results of those studies where management involving supplementary feeding with concentrates, roughages etc., (Mal *et al*., 2007; Mustafa *et al.*, 2021; Babiker and El-Zubeir, 2014 and Idrees *et al.* 2016) detected some influences on protein, casein, lactose, total solids in both early and late stages of lactation.

Similarly, on the effect of parity, Mustafa *et al.* (2021) found no significant difference between primiparous and multiparous camels on lactose and fat contents in camel milk, but significant differences were observed between lactation stages, for protein, SNF and density. Besides fluctuation of density values, fat, lactose, protein and SNF increased significantly with the advancement of the lactation stage in their study.

Except for galactose, lowest in multiparous camels in late lactation, the lowest sugar concentrations of lactose and glucose recorded in primiparous camels, were found in early and mid-lactations. While the results are in agreement with Zeleke (2007) who observed no effect on the composition of camel milk by the lactation stage, significant effects in Tsabong camels

milk were observed for galactose ($P<0.05$) and for Glucose ($P<0.01$) as shown in Table 3.5.2, as affected by stage of lactation.

Babiker and El-Zubeir (2014) found the highest means of fat protein, lactose and SNF in camel milk during the early stage of lactation, which was partly in agreement with this study's finding of fat (3.675±0.442%) and SNF (6.33±0.313%) both also observed in early lactation. Their results however differ from our observation of means of protein (1.910±0.188%) and lactose (3.568±0.227%) contents, which were highest during mid and late lactations, respectively. However, their observation of the highest means of protein, lactose and SNF being recorded in multiparous camels were all contrary to our results as all those means were highest in primiparous camels from Tsabong Camel Park (Table 3.5.1 and Table 3.5.2). Dereje and Ud'en (2005) reported that in the dry season, both young male and young female camels spent more time browsing than adult male and adult females. Because for this study the majority of primiparous camels consisted of young females, they could have spent more time feeding than the otherwise all-adults multiparous females. Small animals generally require more feed per unit of body weight for maintenance and general functions than larger animals, and this effect is greater when young (smaller) animals are compared with larger adults within a species (Van Soest, 1994).

Contrary to Brezovecki *et al.* (2015), who reported primiparous dromedaries to have produced milk with higher concentrations of components than did multiparous animals, our Tsabong dromedaries did not show differences ($P>0.05$) in milk components being affected by parity. There were no significant differences ($P>0.05$) in the means of protein and fat values for primiparous (1.790±0.108%) and (2.903±0.255%), and multiparous (1.792±0.108%) (2.268±0.255%) camels, respectively, as shown in (Table 3.5.1), while Mustafa *et al.* (2021) observed primiparous milk having been richer in fat and multiparous richer in protein.

Similarly, the findings of Mohamed and El-Zubeir (2020) showed parity orders significantly affect camel milk composition, as samples from camels at the second parity revealed higher total solids, fat, protein, lactose and density, compared to those found for camels at the third parity.

The mean value of urea concentration reported by Faye *et al.* (2010) was 81.6±60.4mg/L with a range of 0–290.5 mg/L. Those values changed significantly ($P < 0.001$) according to season, the highest concentration was observed in spring when the grass contained the highest soluble nitrogen. The milk urea was positively correlated to the total protein concentration in milk as reported by Faye *et al.* (2010). This relationship of milk urea to milk protein was evidenced by the unchanged low protein and casein levels as to that of urea (Tables 3.5.1 and 3.5.3) from results which show no significant differences ($P>0.05$) at all levels of parity and lactation stages, for both urea and protein concentrations in the Tsabong dromedary camels' milk. Generally, the lower milk urea values in Tsabong dromedaries were encountered in case of limited availability of degradable nitrogen from the pasture (5.88 CP) (Table 4.4.4.2).

At the time of this study Tsabong dromedary camels' milk, under unimproved conditions, as recorded in Table 3.5.1 to 3.5.3; had protein, (1.791±0.108%), similar to the (1.8±0.19% protein) previously reported by Makgoeng *et al.* (2019), but lower than those reported by (Al-haj and Al-Kahn, 2010, Babiker and El-Zubeir, 2014, Brezovecki *et al*., 2015, Faraz A., 2020 and Karaman *et al*., 2022), which all fell in the rage of 2.36% and 4.9%. Fat content (2.585±0.255%), on the other hand, was within the range of 2.46 % and 4.01% reported by (Babiker and El-Zubeir, 2014, Faraz A., 2020 and Karaman *et al*., 2022) and higher than the 2.0 % pre-reported from the same Tsabong camels by Makgoeng *et al.* (2019). Values for lactose, SNF, density and total solids; 3.332±0.131%, 5.811±0.181%, 1023.904 ±0.735 and 8.546±0.402%, respectively, were all lower than the lowest reported by Makgoeng *et al.*

(2019), of 4.59%, 8.49%, 10.29g/mL and 10.00%, for lactose, SNF, density and total solids, respectively. Hence, Milk from camels in Tsabong is higher in glucose content (0.216±0.024%) as compared to the 0.19± 0.07 % reported by Babiker and El-Zubeir, (2014) and Karaman *et al*. (2022).

The differences in variations between our observations and other authors could be in relation to a number of factors. Babiker and El-Zubeir (2014), for instance, evaluated the effects of parity and lactation stages on camels reared under different physiological, nutritional and environmental factors. Their camels were reared under intensive, semi intensive and the genetically improved (grazing+supplement group), with feeding systems involving supplementary feeding with alfalfa, groundnut, groundnuts cake, and concentrates within the production systems. The high crude protein, and concentrates sources in their feed may have contributed to the higher values if protein and lactose percentages. Tsabong free-ranging camels in our study were on the other hand, evaluated for effects of stages of lactation and parity being the main sources of variation under unimproved conditions during the dry season. No supplementary feeding was provided to the camels under this study at the time of the experiment.

Total viable microorganisms (Table 3.5.4) were only in the ranges of $<1.00 \times 10^2$ CFU/ml to $<14.20 \times 10^2$ CFU/ml, which under the Kenya Bureau of Standards (KEBS) for raw whole camel milk is regarded as good when the total viable counts (TVC) are between $0\text{-}5 \times 10^5$ CFU/ml for grade I and II (KBS, 2016).The milk had lactic acid concentration (0.1080±003%), that is lower than the 0.6-1.2% lactic acid range found in fermented milk (Alm, 1982) which gives milk its self-preserving quality (Motofari *et al*., 2013). This low titratable acidity (% lactic acid) implies that the milk was fresh and of good quality at the time of milking. It had, the milk had no coliforms detected, conferring it high hygienic status.

With low protein content of 1.791±0.108% found in Tsabong dromedaries' milk, the milk still falls within Grade I, as determined by the Kenya Bureau of Standard, for TVC and coliform count, as stated in (Table 3.5.4). The absence of coliform bacteria indicates the good hygienic conditions in which Tsabong camel milk was produced (Abera *et al.* 2016). In a previous study on the same Tsabong camels, Makgoeng *et al.* (2019) discovered the average $\log_{10}$ (CFU/ml) total plate and coliform counts of the milk samples to be 3.1±0.97 and 3.9±1.46, respectively, showing that if handled well and kept from contaminants, Tsabong camel milk collected at the time of the study was hygienic. At the time of the experiment, the results from the milk microbial tests conferred the milk to a high safety standard when compared to the more contaminated Moroccan camels reported by Ismaili *et al.* (2016), which had total coliforms averaging counts of 1.82×10^7 CFU mL^{-1}.

Free fatty acids (FFA), according to Cardak *et al*. (2003), are usually present in milk in at low levels and give an idea of the amounts of unsynthesised fatty acids in fresh milk, whereas, in stored milk, they express lipolytic activity (Cardak *et al*., 2003). As such, the concentration of free fatty acids (FFA) in milk is an indicator of dairy animals' nutrition, bacterial contamination, and storage quality (Hanus *et al.*, 2008). Fluctuations in FFA content in primiparous camels and no changes in multiparous camels with advancement in lactations (Table 3.5.1) are in contradiction with the findings of Cardak *et al*., (2003) that FFA increases during lactation. FFA responsible for milk flavour varies between 1.15 µmol/mL and 9.55 µmol/mL (Shihata *et al.*, 2000; as cited by Cardak *et al*., (2003). Hence, Tsabong camel (0.485±0.033 g/L, being 1.054 µmol/mL) milk falls within acceptable limits for normal flavour.

These low levels of bacterial counts observed in the milk could be attributed to the fact that the milk samples were stripped directly into the cup, limiting any contamination from the hand

milking. Like Yagil *et al*. (1994) observed, the detected bacteria in the milk could be those normally found in the mouths of calves, proving that bacteria mostly come from the hands of a camel herder who helps in the milking. Where large amounts of milk are to be collected, the use of sterile milking machines would be appropriate in order to maintain low levels of bacterial contamination. In addition, the low FFA content found in the milk from dromedary camels reared in Tsabong is an indicator that there was minimal bacterial contamination and that the milk confers to good storage quality.

3.7 CONCLUSION

The results of the study show that the composition of camel milk produced in Tsabong, from dromedary camels, under the existing and unimproved feeding conditions, is affected by lactation stage and interaction between parity and lactation stage. Interaction between lactation stage and parity reduces levels of fat, SNF, FFA, galactose and density with increase in parity. Galactose and glucose were the only components significantly affected by the lactation stage. There was a reduction in milk galactose concentration from primiparous-early-lactation camels to those in multiparous-late-lactation stages of up to 36.50%. Another significant decrease was observed in primiparous camels where glucose concentration lowered by 61.98% from early lactation to mid lactation stages. Parity alone did not have significant effects on any measured components. Parity alone did not show any significant effect on all components. Casein, citric acid, freezing point, lactic acid, lactose, total solids, protein and urea were not affected by both parity and lactation stages. Consumers and milk processors may have an option to target specific levels of desired components of camel milk, based on milk obtained from camels at specific stages of lactation and parities. Supplementary feeding with protein and energy sources

during the dry periods may help improve those components that were not highly influenced by parity and lactation

The microbiological quality and safety of camel milk produced in Tsabong at the time of conducting the research conformed to the Kenya Bureau of Standards for raw camel milk and Botswana Bureau of Standards for raw cow milk. There were no detections of coliforms, in the milk. The total counts of coliform bacteria were below the tolerable limits for camel's milk after milking. Though it is highly recommended that milk prior to storage, should be pasteurised to increase shelve-life, these properties, found in Tsabong dromedary camel milk, confer the milk safe for drinking by tourists who frequently visit the park for camel riding and subsequently camel milk tasting and drinking especially those who prefer to have it raw and unpasteurized.

3.8 RECOMMENDATIONS

The current study was conducted during the dry season and so studying the effects of lactation and parity on Tsabong dromedaries' camel milk extended into the wet season, when forage nutrition is at its highest, could give insight into which group of camels will need specific attention at feeding, to influence levels of desired milk components. Fatty acid and amino acid profiles of Tsabong dromedaries' milk should be studied to assess if it would be influenced by parity and lactation stage as well. The use of milking machines should be introduced at Tsabong Camel Park to minimise possible contamination through hand milking, to maintain the hygienic qualities of the milk.

CHAPTER FOUR

EFFECT OF SUPPLEMENTARY FEEDING WITH BUFFELGRASS (*Cenchrus ciliaris)* AND OLD MAN SALTBUSH (*Atriplex nummularia L.*) ON CAMEL MILK QUANTITY AND QUALITY

4.1 Abstract

This study investigated supplementary feeding with either or both Cenchrus ciliaris *and* Atriplex nummularia *to evaluate their effect on the quality and quantity of milk obtained from dromedary camel kept at Tsabong Ecotourism Camel Park. Twenty-four (24) ear-tagged primiparous camels in mid-lactation, which browse within the paddock, were separately given the supplementary feed used as treatment (None supplemented-control group,* Cenchrus ciliaris *alone,* Atriplex nummularia *alone,* Cenchrus ciliaris *plus* Atriplex nummularia*). Supplementary feeding with high CP, sources of* Cenchrus ciliaris *and* Atriplex nummularia*, in the advent of low crude protein in the daily nutrition of free-ranging Tsabong camels, during winter, greatly influenced (P<0.05) camel milk composition. Supplementary feeding with all three treatments, i.e.* Cenchrus ciliaris *alone,* Atriplex nummularia *alone,* Cenchrus ciliaris *plus* Atriplex nummularia*, positively affected milk protein, urea, and casein percentages. Also improved were milk components of fat, galactose, glucose, solids-not-fat (SNF), total solids (TS), as well as citric acid contents and density. Another improvement was found in daily milk yield being influenced greatly (P=0.0001) by the supplementary feeding with all three treatments, with the highest effect coming from supplementing with* Atriplex nummularia*. However, supplementary feed sources significantly reduced (P<0.05) free fatty acids (FFA) content as well as the freezing point of camel milk. The highest concentrations of fat, galactose, glucose, protein and total solids were 4.222%,*

0.878%, 0.372%, 3.143% and 11.762%, respectively, which were detected on milk from camels supplemented with Atriplex nummularia *alone. Supplementing with* Cenchrus ciliaris *alone produced milk with the highest SNF content (7.458%), whilst a combined feed (*Cenchrus ciliaris *plus* Atriplex nummularia) *significantly produced milk with the highest casein (2.473%) content. All treatment feeds insignificantly (P>0.05) reduced concentrations of lactose. Generally, supplementing Tsabong dromedary camels could greatly improve camel milk yield and composition during the dry season.*

4.2 INTRODUCTION

Supplementary feeding of camels has the effect of improving milk yield. Utilization of *Cenchrus ciliaris* and *Atriplex nummularia L,* (Old-man Saltbush) in numerous studies have proved highly advantageous in improving milk of camel herds. *Cenchrus ciliaris* is a good grazing grass and is highly palatable to camels (Ali *et al*., 2009). Because its CP drastically decreases as the plant matures (Walker, 2013), there is need to complement it with sources of high CP like saltbush. Salt is another important factor in the passage of water and urea in the gut and the kidneys and when inadequate in the diet will lead to less milk production in camels, which becomes even more important when drinking water is restricted (Yagil, 1982). Since camels need about six to eight times as much salt as other animals, they need to regularly graze on halophytic plants to remain healthy (Ali *et. al*. 2009). Since the high mineral content of saltbush can have a negative impact on animal performance, its use should be in combination with feeds such as high-quality grass hay (Walker, 2013). Camels grazing *Atriplex nummularia* responded in a similar way to the hay clover-fed, supplementation at 50% ad-lib level mates, though less in magnitude and efficiency, promoted reasonable weight gain and efficiency of ME and DCP utilization (Abdel-Wahed, 2014).

4.1.1 *Atriplex nummularia L,* (Old-man Saltbush)

Inadequate feed resources for camels is one factor which is affecting milk yield and composition of foraging camels. Supplementary feeding with Saltbush is one amelioratory strategy and inclusion of the different browse species in the camel diets have potential to improve milk yield and quality. Saltbush as a fodder crop is highly digestible and contains high mineral value and crude protein (Aganga *et. al*., 2003) and can be a crucial source of perennial fodder for camels. Yagil *et al*. (1994) highlighted salt as one limiting factor in camel health. There is some information on the use of this fodder as a diet in camels. A review by Igbal and Khan (2001) revealed that in summer, camels have a high preference for salty plants and shrubs. Supplementation of lactating camels with cobalt and phosphorus significantly increased milk yield (Onjoro *et al.*, 2006). *Atriplex nummularia* is known to have high levels of nitrogen and phosphorus (Aganga *et. al.*, 2003), characteristic nutrient elements involved in protein synthesis. Shawket and Ibrahem (2013) concluded that fresh *Atriplex nummularia* in the diet of camels increased milk production, as the feed protein content directly affects milk protein (%) content and is also responsible for increasing milk lactose (%) content. Assessment of the impact of long-term feeding *Atriplex nummularia* on camel's milk production under arid conditions, by Shawket and Ibrahem (2013), revealed that camels can produce milk under prolonged feeding of *Atriplex nummularia* (saltbush) with a suitable source of energy supplementation without changing either milk chemical composition or milk physical properties. This system of nutrition successfully provided more than the protein and energy needed for both maintenance requirements and milk production. *Atriplex nummularia* is much valued for its ability to provide all-year grazing of green feed by extending feed availability into dry periods (NSW, 2010).

Less milk production in camels was observed when salt in the diet is inadequate (FAO, 2017); hence the need for the inclusion of saltbush to supplement the Kgalagadi range feed resources.

4.1.2 *Cenchrus ciliaris* (Foxtail grass, Buffelgrass)

One other beneficial fodder crop used for supplementing to improve milk yield and composition is *Cenchrus ciliaris* (Buffelgrass). It has several benefits, which include crude protein of about 9.6 percent and *in-vitro* dry matter digestibility (IVDMD) and crude protein (CP) digestibility ranging from 50-60 percent (Belgacem and Louhaichi, 2015).

In Kgalagadi, during the dry season (May-September), perennial grasses get replaced by less nutritious annual grasses. Vast areas are then covered by Sour Grass (*Suir grass* or *Schmidtia kalahariensis*) that provides livestock with the nutrition only for a short time when it is still green after the rains. Because their roots are less substantial, they do little to hold the soil together in the dry season (Reed *et. al*., 2008). This scenario could be improved by farmers sowing and feeding the high yielding buffelgrass as a management practice. *Cenchrus ciliaris* will do well in Kgalagadi arid area as it is the most drought tolerant of the commonly sown grasses. Buffelgrass, prefers a sandy and sandy loam soils, does well in summer growing season (when high temperatures coincide with high rainfall), even establishes in regions that receive less than 250 mm rainfall annually (Marshall *et al*., 2012). These are the climatic conditions found in Tsabong. Giving better dry matter yields and being highly competitive than most grasses (Mganga *et. al.,* 2010), *Cenchrus ciliaris* can withstand strong winds, low annual rainfall, an acute erosion and a nutrient-depleted soil profile (Ashraf *et. al.,* 2013)

4.3 MATERIALS AND METHODS

4.3.1 Feed sources

Cenchrus ciliaris used in this study was sourced from that growing at the Department of Agricultural Research (DAR) fields in Sebele and baled by BUAN. It was harvested at its late stage of growth. The hay was baled into 100 block bales, weighing approximately 22 kilograms each, using a tractor pulled baler and immediately stored under shade. It was transported immediately and at Tsabong, it was kept in a closed bunker. *Atriplex nummularia* being planted widely in Tsabong by the Department of Animal Production and piloted to farmers with successful results. The Departmental office awarded the use of prunes from their office and most were purchased from farmers for the feeding trial. The supplementary feeds values are presented in table 4.4.2. Both supplementary feeds were air-died into hay for easy handling, weighing and feeding (see appendix AP2g-k).

4.3.2 Evaluation of the forage sources

An assessment of relative forage preference of lactating camels during the dry season was used to assess the forage feed quality supplied from the available pasture, to compare the digestibility and quality of the forage to that supplied by the supplementary feed, prior to the feeding trial.

4.3.2.1 **Study Area**

The study area was located within the perimeter fence of the Tsabong Ecotourism Camel Park (25º56ˈ15.5ˮ S, 22º27’30.5” E), described in section 3.3.1 above.

4.3.2.2 **The animals**

Twenty-four (24) lactating, multiparous camels in the mid lactation were used in the study. The camels grazed with the 3200 hectares fenced area and, except for the day before sampling, were not kraaled at night because of the scarcity of forage on the farm. They were provided with water *ad-libitum*, at two (2) water points, 2 kilometres apart. One borehole feeds both water points through reticulation. One point is in the holding kraal and the other within a small saltpan from where the camels normally drink during the rainy seasons, from rainwater.

4.3.2.3 Data collection

In this experiment, scan sampling, adapted from Alkali *et.al*. (2017), was used to determine the forages preferred during the dry season. During that time, for four hours in the morning (0800 -1200 h), and three hours in the afternoon (1500 -1800 h), the animals were followed and observed as they grazed or browsed. The observations were made for five consecutive days. Each camel was observed using binoculars, to avoid interrupting their feeding (see appendix AP2a-d3) and monitored during feeding to ensure accurate identification of the plant consumed at an interval of 5 minutes. The time spent by the camel on each forage was thus recorded in minutes per interval. A list of available forage species in the park was used to identify and record the time the camels selectively foraged on plant species (See appendix AP3a).

4.3.2.4 **Forage-feed sample collection and preparation**

After the animals had moved onto a different location, a pruning knife was used to clip the similar portion of the plants to that which was browsed (appendix AP2e) and collected into labelled sample paper bags. The samples were immediately taken to the nearest laboratory

(Tsabong Unified Secondary School science laboratory) for initial weighing. On arrival at BUAN Biochemistry laboratory, samples of the most highly preferred forage were chopped, and oven-dried at 70^{o}C for 48 hours. About 200 grams of each sample was ground to pass through a 2 mm screen and the samples, together with those of experimental feed samples, were subjected to the same chemical analysis.

An additional sample to individual feed samples prepared for analysis was a composite feed prepared through the estimation of proportionate forage-feed samples as daily intake ratios. Each forage feed type sample was represented, in percentage volume, a proportion calculated from the preference percentage (see appendix AP2f).

4.3.3 Chemical analysis of feed and forage samples

Prior to feeding, the experimental feeds and forage samples were first analysed for digestibility and composition. *In-vitro* (incubation) technique using camel's faecal liquor (Golshani *et. al.*, 2014) as an alternative microbial inoculum source to estimate digestibility of those feeds was done. Also analysis was done for phosphorus (P), crude protein (CP), acid detergent fibre (ADF) and neutral detergent fibre (NDF) contents using; DAISY^{II} Incubator, Shimadzu UV Visible Spectrophotometer UVMini-1240, Kjeldahl Technique (AOAC, 1996) and Ankom Fibre Analyser (ANKOM^{220}), respectively. The analysis of results indicated the digestibility and quantities of different nutrients in the feed and were used to determine the quantities of feed to use in the trial as well as limitations as may be required.

To appreciate the positive or negative impact that the experimental feed could have on the milk quality and quantity, samples from available pasture were collected and analysed for quality to

check if the experimental feed would have an addition or dilution effect on the already available forage quality.

4.3.3.1 Using camel faecal material as a microbial inoculum source for an *in vitro* technique to estimate the dry matter digestibility of fodder species to be fed to camels (adapted from Laudadio *et. al.*, (2009)

4.3.3.1.1 Scope

Different feeds are utilised to varying extents by livestock. It is therefore necessary to have information on how a feed is utilised when such feed is to be fed to particular classes of livestock. *In vitro* digestibility is an attempt to reproduce in the laboratory, the reactions which take place in the alimentary tract of animals. Since the camels used for the research were not cannulated or fistulated, it was not possible to try to collect rumen fluid for the inoculation. As such, fresh faecal material was used as the source of inoculum for this experiment guided by the procedure of Laudadio *et al*. (2009)

4.3.3.1.2 Procedure

In vitro fermentation was conducted for 48 h using the Daisy^{II} incubator following the method of ANKOM (1998), modified using faeces as a source of inoculum by the method of Laudadio *et. al.* (2009). The complete unit consists of 4 incubation vessels with a capacity of 2 L each. Each vessel contained 1.6 L of buffer solution, 400 mL of faecal liquor as the inoculum, and 22 nylon bags. Collected plant samples were ground to pass through a 1-mm screen. Each of the feeds was digested in duplicate for each source of inoculum. Nylon filter bags (Ankom F57,

ANKOM Tech., Fairport, NY) were rinsed in acetone and allowed to air dry before drying at 100^{o}C for 24 h, after which dry bag weights were recorded. For each feed sample, 0.5 g of ground sample were added to 20 nylon bags, heat-sealed and the dry sample plus bag weight were recorded. Duplicate nylon bags for each feed type were randomly allocated to one of the four digestion vessels, and therefore to one of the four inoculum treatments.

The microbial inoculum was prepared from fresh faeces collected directly from camels' rectums. Fresh faeces samples from camels mentioned above were taken before morning feeding and transferred to the pre-warmed heater. After collection, the faecal samples were placed in an air-tight container and transported to the laboratory at 39°C in a car-powered cooler/heater fridge (Appendix AP2l-n). At the lab, the faecal liquor inoculum was prepared by homogenizing 40 g of faeces with 360 mL of warm, distilled water for 2 minutes under CO_2 and then filtered through a double-layered cheesecloth directly into the pre-warmed digestion vessels. Each digestion vessel contained 400 mL of inoculum and 1.6 L of buffer solution. The buffer solution consisted of 1.33 L Buffer Solution A; (KH_2PO_4, 10.0 g/L; $MgSO_4.7H_2O$, 0.5 g/L; NaCl, 0.5 g/L; $CaCl_2.2H_2O$, 0.1 g/L; and Urea (reagent grade) 0.5 g/L) and 266 mL of Buffer Solution B; (Na_2CO_3, 15.0g/L and $Na_2S.7H_2O$, 1.0 g/L), mixed in each digestion vessel and the pH were adjusted to 6.8 at 39^{o}C. The faecal liquor prepared from camel faeces inoculum was then added to the buffer solution in separate digestion vessels after which CO_2 was purged for 30 seconds and then sealed. The sealed digestion vessels were placed into the pre-warmed Daisy^{II} incubator. The incubator maintained a constant temperature of 40°C throughout the incubation while the digestion vessels were continuously agitated. The digestion vessels were removed after 48 h and the filter bags were immediately rinsed for 30 minutes with cold water to stop the microbial activity. The rinsed bags were then placed in the fibre analyser and the procedure for determining NDF was followed (ANKOM, 1998). The post *in vitro* NDF weight

was then recorded as NDFγ for the formula below. The NDF analysis removes microbial debris and any remaining soluble fractions.

The % IVDMD was calculated using the following formula (ANKOM, 1998):

$$\% \text{ IVDMD} = 100 - (((\text{NDF}\gamma - (W_1 \times C_1)) \times 100/W_2))$$

Where:

W_1 = Bag tare weight

W_2 = Sample weight

NDFγ = Final bag weight after the NDF determination

C_1 = Blank bag correction (final oven-dried weight / original blank weight)

4.3.3.2 Analysis of feed and forage for Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF)

4.3.3.2.1 Scope

Analysis for neutral detergent fibre entails first boiling the ground feed samples for 1 hour in a neutral detergent solution of sodium laurel sulphate and ethylene di-methyl tetra–acetic acid (EDTA). This detergent removes lipids, sugars, organic acids and other water-soluble materials. Other compounds removed by NDF solution are pectin (a fibrous carbohydrate), non-protein nitrogenous compounds, water-soluble proteins, some tannins and silica. The insoluble residual material is known as the Neutral Detergent Fibre NDF. This material contains major cell wall constituents such as hemicellulose, cellulose and lignin. Some major amounts of some cell wall contents like protein, bound nitrogen, minerals and cuticle may also be included in the NDF.

Acid Detergent Fibre (ADF), on the other hand, is the residue after refluxing ground feed samples for 1 hour in acetyl tri-methyl ammonium bromide dissolved in 0.5 M sulphuric acid. This technique extracts those components of the feed which though insoluble in a neutral detergent, are readily dissolved in an acidic detergent. These components include hemicelluloses and cell wall proteins leaving behind a residue consisting of cellulose, lignin, lignified nitrogen (which is indigestible), cutin, silica and some pectins.

The NDF and ADF determinations were done in duplicates. After empty fiber bags were weighed, a sample amount of 0.5g was placed in each of the two bags per treatment and weighed. Two empty fiber bags were included to determine the NDF and ADF in the blank bags for correction. The bags were heat sealed. The NDF and ADF were then determined according to the AOAC procedures of 1996 and Van Soest's proximate analysis (1994b)

4.3.3.3 Analysis of feed and forage for protein

4.3.3.3.1 Scope

Proteins are the main nitrogen-containing components. Thus, by measuring the nitrogen content of the feed, it is possible to derive its protein content. The procedure that was used to determine the crude protein – the Kjeldahl technique – involves first digestion (for up to 8 hours) of feed samples to be analysed in concentrated sulphuric acid (90% H_2SO_4). This converts the nitrogen to ammonium sulphate – $(NH_4)_2SO_4$. After digestion, the mixture is then cooled, diluted with water and neutralised with sodium hydroxide (NaOH), which changes the nitrogen into a form of ionised ammonium. The solution is then distilled, and the distillate

containing the ammonium is titrated with 0.01 N sulphuric acid. The amount of acid used in titrating indicates the amount of nitrogen present in the feed sample.

4.3.3.3.2 Procedure

The method followed the procedure set out in the AOAC (1996) for the determination of crude protein in livestock feeds. About 1.25 grams of the dried ground samples were accurately weighed onto lens tissue on a tarred analytical balance in duplicates, wrapped and placed in the digestion (Kjeltec) tubes, including a blank sample containing only lens tissue. To each tube, 20 ml of 72% sulphuric acid (H_2SO_4) was added using a graduated liquid dispenser. The sulphuric acid contained selenium as a catalyst. The digestion tubes were placed in a test-tube rack and put on a digestion block in a fume cupboard, with the heater and the suction pumps switched on. The first program was set such that the start temperature of the block was 150^oC and the samples boiled in acid for 1 hour, increased to 250^oC and the digestion continued for another hour followed by 2 hours at 330^oC, after which the heater was then turned off and the samples lifted off the block to allow them to cool overnight. Using a graduated liquid dispenser, 4 ml of hydrogen peroxide (H_2O_2) was added to each tube and the tubes were lowered onto the digestion block and resumed digestion at 330^oC for a further 2 hrs as set in Program 2. After digestion was complete the heater was switched off and allowed to cool overnight. The contents of the digestion tubes were then transferred into labelled 250 ml volumetric flasks. The tubes were then rinsed with distilled water and the water used for rinsing was emptied into the respective flasks and filled up with distilled water to the mark.

The nitrogen content of each sample was then determined by first distilling 25 ml of solution with sodium hydroxide over 1% boric acid and then titrated against 0.01 N sulphuric acid using

the Kjeldahl titration unit. The amount of nitrogen in the solution was calculated based on the amount of acid required to neutralise the nitrogen. From the nitrogen content, the crude protein content (dry matter basis) was calculated as:

% Crude protein = % Nitrogen x 6.25 (AOAC, 1996)

Where the nitrogen (N) content of each sample was determined based on:

1 ml of 0.1 N H_2SO_4 = 0.014 g nitrogen

N % of sample = ((Volume of acid and digest/Aliquot used in titration) x 0.014 x Molarity of acid x (Sample titration - Blank titration) x 100) / Sample weight

4.3.3.4 Analysis of supplementary feed and forage samples for Phosphorus

4.3.3.4.1 Scope

Phosphorus is the second most required mineral in an animal's diet but has more functions in the body compared to any other mineral. Phosphorus plays a major role in energy utilisation, transfer and metabolism. Phosphorus compounds are involved in all major functions such as protein synthesis. Phosphorus is also involved in the transport of fatty acids and the exchange of amino acids. As such it is essential for milk production.

4.3.3.4.2 Procedure

From the samples, digested in preparation for crude protein analysis, Phosphorus (P) was detected using Shimadzu UV Visible Spectrophotometer UVMini-1240, Shimadzu Corporation, Japan). And absorbance was detected at 670 nm wavelength following molybdenum blue method of Dickman and Bray (1940).

4.3.4 Computation of captured data

Data for relative forage preference of lactating camels during the dry season was computed using Microsoft Excel (Microsoft Office 2016) and displayed as descriptive statistics.

4.3.5 Experimental Animals (Conditioning & Feeding)

The feeding trial was conducted at Tsabong Ecotourism Camels Park (25°56'15.5" S, 22°27'30.5" E), as described in section 3.3.1 above. Twenty-four (24) ear-tagged multiparous camels in mid-lactation, within three to five years, which browsed within the paddock, were separately fed the supplementary feed used as treatment (none supplemented group, *Cenchrus ciliaris* alone, *Atriplex nummularia* alone, *Cenchrus ciliaris* plus *Atriplex nummularia*), 6 camels per treatment. The camels were ear-tagged using M10 sized ear-tags for easy visibility from long ranges (appendix AP2o-p). Each group of the six camels given a feed treatment was tagged using a specific-colour ear tag. *Cenchrus ciliaris*-Supplemented Group were tagged GREEN, *Atriplex nummularia*-Supplemented Group-BLUE, *Cenchrus ciliaris/Atriplex nummularia*-Supplemented Group-RED and the None-Supplemented Group-YELLOW. This was to make sure that during a gathering of the camels, the herders do not leave behind those selected for the experiment, for each treatment group. The selected animals were given the supplementary feed twice a week for two weeks to allow for them to get accustomed to the

new feed, as they had never tasted *Cenchrus ciliaris* nor *Atriplex nummularia* before. The camels were group-fed and were only released to pasture graze after all the supplementary feed was finished from the troughs. All the six camels in each group were fed together from a single trough. Data were collected on the third week of feeding, as the animals took longer than expected to get consume the hay without hesitation.

Based on the female camel's BW= (300 – 540 kg), AVG (420kg), the maximum supplemental feed amount was calculated based on the DMI values for camels grazing natural pastures, which have been estimated to be 1.6-3.8 kg (AVG 2.74kg) DM per 100 kg LW (Richard, 1989, as cited by Hashi and Kamoun. 1995). Wardeh (2004) found lactating camels to consume 38.8% more DM than dry ones, even more (57%) when the camels are individually fed. As such the animals were not going to be over supplemented, since the author estimates the dry matter requirements of the dromedary for maintenance at 2.5% of the body weight.

Therefore 11.508 kg were the estimated daily DM requirements per a 420 kg camel. Blackwood and Clayton (2007) recommend supplementation, of dairy animals, with good quality hay to be 30% of the daily DMI, at a frequency of 2-3 times a week. Since supplemental feed will comprise only up to 30% of the daily feed requirement, 3.45 kg of feed were given in the morning and the animals were given a chance to finish before they were released with the rest of the herd.

4.3.6 Camel-milk collection and testing

The camels were kraaled the day before milking and feeding. A night before milking days, the lactating camels were kept together in an open kraal. The calves were kept in a separate pen adjacent to their dams throughout the night and only let out one by one to suckle for a few

seconds to stimulate milk let-down (appendix AP1a-e), and after hand milking was completed, they were fully released to join their mothers. The camels were milked and supplemented twice a week, in the mornings before they were let out to graze. Milk collection for compositional analysis was done at the end of the feeding trial. Milk samples were collected straight from the teats, by hand milking, into sterile (autoclaved) 250 ml bottles, re-sealed and stored in cooler boxes preserved in ice for the duration of transportation to the lab where they were stored in a refrigerator at 4^{o}C overnight. All the milk samples were sent to Botswana National Veterinary Laboratory, where they were analysed, the next morning, using the automatic milk analyser (MilkoScan FT 1; Foss A/S, Hillerød, Denmark). 40 mL of each sample was subjected to the analyser where a probe attached auto sampled 20 mL for analysis, and the results displayed in the attached computer having the devices software. The procedure followed was derived from the user's manual (MilkoScan FT 1 user manual 6004 5478). Samples were analysed for levels of Lactose (%), Galactose (%), Glucose (%), Total Solids, Solids-Not-Fat (%), Fat (%), Protein, Casein (%), Urea, Free Fatty Acids, Lactic Acid and Citric Acid (%). Milk properties; Freezing Point, and Density (SG) were also measured on the milk samples using the same milk analyser, following the procedure used for section 3.3.4 for milk composition and properties.

4.3.7 Monitoring of milk yield

The milk yield of the selected primiparous dromedary camels was monitored during their mid-lactation for a period of 90 days. The milk yield of each camel was measured using a graduated cylinder. The milk yield of each camel was recorded in a logbook (appendix AP3c). Camels were milked twice a week and the average yields were used to estimate the weekly milk yield.

4.3.8 Statistical analysis

To assess the effect of supplementary feeding on milk yield and milk composition changes, data from milk samples tested were analysed using the General Linear Model procedure of Statistical Analysis System (2006). The experiment was a Randomized Complete Design (RCD) and Duncan's multiple range tests ($p<0.05$) were used to determine the least square means.

The model for a Randomized Complete Design is:

$y_{ij} = \mu + \tau_i + \varepsilon_{ij}$

where:

y_{ij} = an observation in treatment i and block j

μ = the overall mean

τ_i = the effect of treatment (Feed Supplement Type) i = 1..., a; (No supplement, Old Man Saltbush (OMB), Buffel Grass (BG) and OMB+BG))

ε_{ij} = random error of the j^{th} observation from the i^{th} treatment.

We assume ε_{ij} *IID N* $(0, \sigma^2)$, assuming all effects are fixed.

4.4 RESULTS

Forage species preference of Tsabong dromedary camels during the dry period were assessed and displayed in Table 4.4.1. At the time of the study, camels tended to maximize gut fill by opting for Kalahari sour grass (72.51%) of their feeding time. Camel thorn and Black thorn

species attracted camels more than the least preferred dryer leaves of the Brandy bush shrub (1.89%) of the camels' feeding time.

Table 4.4.1. Relative forage species preference of Tsabong dromedary camels during the dry period

Scientific name	Vernacular name (English)	Vernacular name (Tswana)	Plant type	Potion of the plant eaten	Relative forage species preference (%)
Vachellia erioloba E. Mey	Camel Thorn	*Mogotlho*	Tree	Leaves	8.53
Vachellia erioloba E. Mey	Camel Thorn	*Mogotlho*	Tree	Pods	7.58
Senegalia mellifera (Vahl) Benth.	Black Thorn	*Mongana*	Shrub	Twigs	6.16
Boscia albitrunca (Burch.) Gilg & Benedict	Shepherd's Tree	*Motlopi*	Tree	Leaves	3.32
Grewia flava DC	Brandy Bush	*Moretlwa*	Shrub	Leaves	1.89
Schmidtia kalahariensis Stent	Kalahari sour grass	*Segwane*	Grass	Leaves/Stem	72.51

Table 4.4.2 below displays the chemical composition and qualities of the forage feed and supplementary feed ingredients. Grass species (Kalahari sour grass and Buffelgrass) had the highest Neutral Detergent Fibre (NDF) content, 75.43%, and 72.84%, respectively, and the lowest NDF was found in Saltbush. The least digestible forage was Kalahari sour grass, IVDMD (27.32%) with the highest ADF (46.38%). Of all the forage samples analysed, camel thorn had the highest crude protein content (9.24%), which was highly diluted when combined with the rest of the forage to an overall CP (5.88%). That made all supplementary feeds superior in crude protein contents, thereby expected to elicit a positive response in terms of protein supply to the supplemented animals.

Table 4.4.2. Means of chemical composition and quality of the forage feed and supplementary feed ingredients

	DM Composition (% DM)				
Forage Feed	NDF	ADF	IVDMD	CP	P
Vachellia erioloba E. Mey (leaves)	40.90	37.27	58.52	9.24	0.0017
Vachellia erioloba E. Mey (pods)	41.99	36.94	61.60	7.56	0.0024
Senegalia mellifera (Vahl) Benth (twigs)	62.27	44.82	38.86	6.30	0.0025
Boscia albitrunca (Burch.) Gilg & Benedict (leaves)	32.48	23.35	71.05	7.98	0.0020
Grewia flava DC (leaves)	38.67	36.13	61.54	6.72	0.0023
Schmidtia kalahariensis Stent (leaves/stem)	75.43	46.38	27.32	1.68	0.0022
Combined feed forage sample	62.76	42.49	39.04	5.88	0.0021
Supplementary Feed					
Cenchrus ciliaris (Buffelgrass)	72.84	42.44	29.78	7.14	0.0024
Atriplex nummularia (Saltbush)	31.54	18.52	79.56	13.02	0.0022
Buffelgrass + Saltbush (50/50)	53.67	31.06	52.55	9.24	0.0019

NDF=neutral detergent fiber, ADF=acid detergent fiber, IVDMD=in-vitro dry matter degradability, CP=crude protein, P=phosphorus

The effects of supplementary feeding of *Atriplex nummularia* (Saltbush) and *Cenchrus ciliaris* (Buffelgrass) on the chemical composition of camel's milk are displayed in Table 4.4.3. Supplementary feeding with Saltbush more than doubled concentrations of milk fat, galactose, glucose and increased concentrations of protein and total solids by 70.63% and 44.85%, respectively. The combined feed supplement on the other hand increased casein concentration by 45.47% and lowered lactose concentration by 6.63%. The highest concentrations (%) of fat, galactose, glucose, protein, and total solids, 4.222, 0.878, 0.372, 3.143, and 11.762, respectively were detected on milk from camels supplemented with *Atriplex nummularia* alone. Supplementing with *Cenchrus ciliaris* alone produced milk with the highest SNF content (7.458%), whilst a combined feed (*Cenchrus ciliaris*/*Atriplex nummularia*) significantly produced milk with the highest milk casein (2.473%), increasing casein concentration by

26.99%. No significant difference in lactose content was observed between all the treatment groups including the control.

Table 4.4.3. Means of chemical composition of camel's milk due to effect of supplementary feeding of camels with *Atriplex nummularia* (Saltbush), *Cenchrus ciliaris* (Buffelgrass) and their blends

	Experimental diets					
Chemical composition	***Atriplex nummularia***	***Cenchrus ciliaris***	**Buffelgrass /Saltbush (50/50)**	**Control**	**SE**	**P-Value**
Casein (%)	2.445^{A}	2.472^{A}	2.473^{A}	1.700^{B}	0.114	0.0002
Fat (%)	4.222^{A}	3.257^{AB}	3.360^{AB}	2.108^{B}	0.481	0.0434
Galactose (%)	0.878^{A}	0.737^{A}	0.780^{A}	0.413^{B}	0.094	0.0135
Glucose (%)	0.372^{A}	0.318^{AB}	0.347^{A}	0.147^{B}	0.061	0.0651
Lactose (%)	3.143^{A}	3.103^{A}	3.083^{A}	3.302^{A}	0.176	0.8135
Protein (%)	3.143^{A}	3.103^{A}	3.083^{A}	1.842^{B}	0.200	0.0003
SNF (%)	7.195^{A}	7.458^{A}	7.133^{A}	5.873^{B}	0.254	0.0012
TS (%)	11.762^{A}	11.047^{A}	10.790^{A}	8.120^{B}	0.638	0.0036

Values in the rows, with different superscripts $^{A, B}$ were significantly different ($P \leq 0.05$). Where SNF=solids-not-fat, TS=total solids, SE=standard error, P – value=probability

The effect of supplementary feeding of *Atriplex nummularia* (Saltbush) and *Cenchrus ciliaris* (Buffelgrass) on the properties of camel's milk is displayed in Table 4.4.4. Lactic acid was not affected ($P>0.05$) by supplementary feeding. Supplementary feed with the combined saltbush/buffelgrass increased ($P \leq 0.05$) the milk citric acid by 55.24%. Milk from camels supplemented with Buffelgrass alone had density increased by 0.59%. FFA of the milk from saltbush supplemented camels decreased by 52.29%, while milk urea was more than doubled to 147.38% higher in concentration. *Atriplex nummularia* supplement significantly produced the lowest concentration of milk FFA (0.219 g/L) and the highest concentration of milk Urea (490.765mg/L). Supplementing with *Cenchrus ciliaris* significantly produced milk with density of (1030.217g/mL), the lowest freezing point (-0.455^{o}C), and supplementing with the

combined feed (*Cenchrus ciliaris*/*Atriplex nummularia*) significantly increased citric acid (%) to 0.222 content of camel milk.

Table 4.4.4. Means of physical properties of camel milk due to the effects of supplementary feeding of camels with *Atriplex nummularia* (Saltbush), *Cenchrus ciliaris* (Buffelgrass) and their mixture on properties of camel's milk

Physical Property	**Experimental diets**				**SE**	**P-Value**
	Atriplex nummularia	***Cenchrus ciliaris***	**Buffelgrass /Saltbush (50/50)**	**Control**		
Citric Acid (%)	0.200^{A}	0.202^{A}	0.222^{A}	0.143^{B}	0.011	0.0003
Density (SG)	1028.683^{A}	1030.217^{A}	1028.233^{A}	1024.100^{B}	1.083	0.0050
FFA (g/L)	0.219^{C}	0.303^{BC}	0.381^{AB}	0.459^{A}	0.039	0.0021
Freezing Point (°C)	-0.441^{B}	-0.455^{B}	-0.412^{AB}	-0.395^{A}	0.015	0.0394
Lactic Acid (%)	0.127^{A}	0.125^{A}	0.119^{A}	0.116^{A}	0.005	0.3074
Urea (mg/L)	490.765^{A}	436.818^{A}	451.781^{A}	198.387^{B}	20.460	<0.0001

Values in the rows, with different superscripts $^{A, B, C}$ were significantly different ($P \leq 0.05$). Where; FFA=free fatty acids, SE=standard error, P – value=probability

The effects of supplementary feeding with *Atriplex nummularia* (Saltbush) and *Cenchrus ciliaris* (Buffelgrass) on the daily yield of camel's milk are displayed in Table 4.4.5. Overall supplementary feeding greatly (P<0.0001) influenced, positively, milk yield. *Atriplex nummularia* supplementation had the highest effect (p<0.05) on milk yield by increasing yield by 60.25%, followed by a combined feed (*Atriplex nummularia*/*Cenchrus ciliaris*) which increased yield by 46.05%, while the least effect detected on supplementing with *Cenchrus ciliaris* alone increasing milk yield only by 36.71%, when compared to the non-supplemented group.

Table 4.4.5. Means of milk yield from supplementary feeding of camels with *Atriplex nummularia* (Saltbush), *Cenchrus ciliaris* (Buffelgrass) and their blends on daily milk yield

Item	**Experimental diets**				**SE**	**P-Value**
	Atriplex nummularia	***Cenchrus ciliaris***	**Buffelgrass /Saltbush (50/50)**	**Control**		
Milk yield (mL/day)	1500.694^{A}	1280.208^{B}	1367.708^{AB}	936.458^{C}	57.741	<0.0001

Values in the rows, with different superscripts $^{A, B, C}$ were significantly different (P≤0.05). Where; SE=standard error, P – value=probability

Figure 2 below shows variations in milk yield during the three (3) months of supplementary feeding of Tsabong dromedary camels with *Atriplex nummularia* (Saltbush), and *Cenchrus ciliaris* (Buffelgrass) and Buffelgrass + Saltbush (50/50) on average daily milk yield of camels. Saltbush supplementary feeding dominated milk yield from week 1 to week 6 and from week 9 to week 12, while the combined feed of Saltbush and Buffelgrass dominated milk yield during week 7 and week 8. The non-supplemented, control group generally produced the same milk yield, with a slight increase at the last, 12^{th}, week. Overall milk yield, during the 12 weeks' period, increased towards the 8^{th} week, after which it slightly declined into the rainy season towards week 12, The highest mean milk yield (2104.17±191.06 mL/day) recorded was in

week 8 from camels supplemented with Buffelgrass + Saltbush (50/50), and the lowest (695.83±191.06 mL/day) in week 1 from the Non-Supplemented (Control) group.

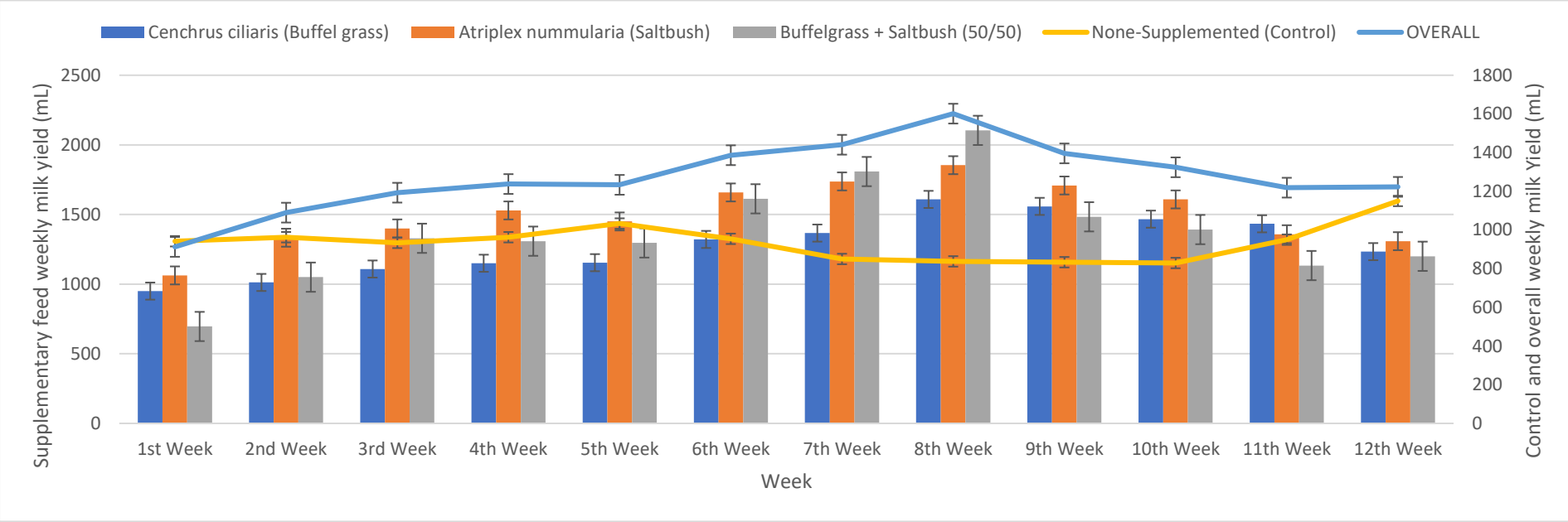

Figure 2 : Weekly variations in milk yield (mL/day), per treatment, during the three months experimental period

4.5 DISCUSSION

In their natural habitat, dromedary camels prefer to browse plants of great nutritional value for most of the year in arid zones compared with grasses (Igbal and Khan, 2001)). The preference of camels for some plant species is geared towards the presence of high crude protein content, according to Igbal (1999). In the present study, preference was limited by the unavailability of species for choice. The most preferred, *Schmidtia kalahariensis* Stent, had the lowest CP (1.68%), (Table 4.4.1). The results of this study support Faraz *et al.* (2022) that, in the dry seasons when forage abundance declines, camels widen their dietary acceptance range by resorting to eating more grasses, leaves, litter, vines, and lignified twigs. Lu *et al*. (2012) observed uninterrupted foraging camels show a positive correlation between the percentage cover of forage species eaten by camels and the proportion of the species in the camel's diet. Although the correlation was significant in the wet season and not significant in the long and dry winter season, Tsabong camels consumed more of the available forage as there was a limited choice, having tree and shrub leaves being mostly dried out and out of reach for most female camels. However, for this study, like Igbal and Khan (2001) it was observed that due to limitations in the availability of high protein forages, in winter, camels widen their dietary acceptance range apparently to compensate for the declining forage abundance by eating more grass, litter leaves, and twigs. Kalahari sourgrass was the most preferred above *Grewia flava* and *Boscia albitrunca* leaves which were less consumed due to its highly sparse distribution. Tsabong Camel Park's pasture is dominated by sourgrass, which provides the most available forage when compared to trees and shrubs, which would otherwise be preferred by camels, during the dry season. Kenyan camels studied by Kuria *et. al*. (2012), concentrated mainly on evergreen shrubs as well as collecting dry leaves of the usually preferred shrubs from the ground as grasses and herbaceous grazing materials were hardly available. The leaves and

twigs on the studied Kenyan pasture occupied 71% to 100% of their grazing time, while in Tsabong camel Park those occupied only 19.9% of camels feeding time.

Of the forage feed samples collected, *Vachellia erioloba* E. Mey leaves had the highest CP (9.24%) (Table 4.4.2), making it the forage containing the highest CP in the area, even in the wet season as recorded by Ditlhogo *et al.* (2020). *Schmidtia kalahariensis Stent* had the lowest CP (1.68%). Advance from the rainy season towards the dry period has been found to significantly reduce crude protein, ADF, and P in the range plants by Tegene *et al*. (2010). Generally, the CP contents were lower than those recorded by Ditlhogo *et al.* (2020) for *Vachellia erioloba* E. Mey pods (11.92%), *Senegalia mellifera* leaves (26.77%) *Boscia albitrunca* (20.63%) collected from the same catchment area, in the wet season. The decline in CP during the dry season, worsened by the depletion of alternative forages, poses the need for supplementing lactating camels with forages of higher CP such as saltbush. At the time of the experiment, though in the dry season, *C. ciliaris*' CP was within the average recorded by Aganga *et al.* (2011), whose value in the wet season has a crude protein of 10.2 % which declined during the dry season to as low as 4.2%.

.

The phosphorus contents of forages were very low. This could be related to the generally low soil levels of phosphorus in the country, with Tsabong measurements of phosphorus found to be as low as 0.8 ppm (0.00008%) recorded by Pule-Meulenberg *et al.* (2005).

Our data (Table 4.4.2) is the first report about the *in vitro* dry matter digestibility of forage species available to camels at Tsabong Camel Park, during the dry season. The highest and the lowest IVDMD% were observed for Shepherd's Tree leaves (*Boscia albitrunca (Burch.)* and Kalahari sour grass leaves/stem (*Schmidtia kalahariensis Stent*), at 71.05% and 27.32%,

respectively, which is inversely correlated to the fiber content (both NDF and ADF) of the two species. The results of the analysis showed a strong negative correlation between digestibility and both NDF and ADF with r = -0.90 and r = -0.99, respectively. The IVDMD of natural pasture was also found to be negatively correlated with NDF by Tegene *et al.* (2010).

Grass species *Schmidtia kalahariensis Stent* had the highest NDF (75.43%), which may lower their nutritive quality, and thus can be classified as a low-quality feed as increased NDF concentration of the diet corresponds to a linear decrease in milk production (Beauchemin, 1991). This plant species remains the most important and preferred in the dry season (Table 4.4.1) even though lowest in digestibility (Table 4.4.2). This indicates that lactating camels selected this plant in the absence of good quality feed, rendering it important to supplement lactating camels in the dry season, as they have no chance to select for better feed than the larger bulls that can otherwise reach much higher branches for tree leaves.

The NDF content of (53.67%), is not expected to limit the nutritive value of the combined supplementary feed (Buffelgrass + Saltbush (50/50)), as it is lower than the critical value of 55-60% (Van Soest and Wine, 1967), and lower than the NDF of combined forage feed (62.67%). Therefore, the supplementary feed is not expected to elicit a dilution effect when fed, but rather an improvement in the forage feed quality. Buffelgrass alone, though high in NDF (72.84%) at this time of the year, was very palatable to the camels at feeding. That could have been due to the camels feeding preference being related to foraging quality, in particular to protein content and digestibility (Shwarts *et. al*. 2012), as the bulk (72.1%) of the available forage at Tsabong Camel Park, at the time of the study, consisted of a less digestible, (ADF (46.38% Vs 42.44%)) and low protein (CP (7.14 Vs 1.68)) of *Schmidtia kalahariensis* Stent forage grass compared to *Cenchrus ciliaris* (Table 4.4.1 and Table 4.4.2).

Compared with the control (non-supplemented) group, supplementing with *Atriplex nummularia* has shown to greatly increase fat, galactose, glucose, protein and total solids contents of camel milk. Shawket and Ibrahem (2013) concluded that the feed protein content in fresh *Atriplex nummularia* added to the diet of camels, increased milk protein (%) content and is also responsible for increasing milk lactose (%) content and total solids. It was however not true for the lactose part, where the results of this study showed a rather slight, though insignificant decrease in the lactose content of milk from supplemented camels. This could have been that the glycemic-propionic acid, which was supposed to be the main substrate for lactose synthesis was produced from degrading Atriplex. The resultant energy, however, could have then been used to provide energy to the microbes in the gut, which degraded the long retained mature grass from the grazing pasture, in an effort to derive as much energy from the bulk feed mostly containing *Schmidtia kalahariensis* Stent) (Table 4.4.1).

The results of this study agree with Shawket and Ibrahem (2013), as total solids (TS) content in the milk greatly increased ($P<0.05$) through supplementing with *Atriplex nummularia* of Tsabong dromedary camels.

Solids-not-fat consists of all solids in milk other than fat, making it valuable to the consumer for its flavour and nutritional value and to the manufacturer of milk products, especially relating to cheese yield. Supplementing with *Cenchrus ciliaris* most significantly increased SNF ($P=0.0003$). It was however, expected that supplementing with grass hay would normally produce higher acetate and less butyrate and propionate, when compared to feeding dicotyledonous roughages like saltbush. Acetate in this matter would be the primary substrate for lipis sysnthesis. However, variations in milk components have previously been discovered to be influenced by other factors such as husbandry and production systems, calf sex-biased

differences, breed, time of milking, analytical methods, geographical area, seasonal influences, nutrition conditions, and age among others (Aljumaah *et. al*., 2012; Babiker and El-Zubeir, 2014; Brezovecki *et al.*, 2015 and Nagy *et al.*, 2017). Slightly lower contents of fat and protein were also observed for camels reared under desert conditions than those reared under farm conditions, in dromedaries (Alwan and Zwaik, 2014), which could have influenced less fat production by Tsabong dromedaries, as Cenchrus supplementation added to the already 73% grass bulk in the feed provided by grazing on *Schmidtia kalahariensis Stent*) (Table 4.4.1)

A comparison of dicotyledons and monocotyledon plant sources when fed to ruminant animals, by Medjekal *et al.* (2018), revealed a higher concentration of propionate and butyrate produced from feeding *Atriplex nummularia* species, than grass-fed ruminants. On the other hand, grass-fed ruminants would yield more acetate than the *Atriplex*-fed ones. Propionic acid, being glycogenic, would be expected to promote lactose synthesis, but not to be used for synthesizing milk fat in ruminants. Acetate, on the other hand, is a primary substrate for lipid synthesis in milk along with butyrate. In this study, where most milk-fat and protein came as a result of feeding Atriplex, may imply that our feed contained a high propionate: acetate ratio, which increased milk protein output and the high efficiency of digestion by dromedary camels contributed to the high energy content derived from the feed. As a result, the camels may have stored excess energy, produced in those short spells, in the form of fat, which then was excreted to through milk as milk fat.

Feeding high protein diets negatively affected the value of fat content and positively affected that of the protein content of camel milk, in an experiment by Shuiep *et.al*. (2008). However, in our study, although protein increased with the protein content of the feed, fat also increased, indicating the camels' efficiency in extracting energy from the supplementary feed as compared to the control group. The highly digestible saltbush could have provided energy to

the gut microbes, which in turn efficiently degraded the grass substrate to produce high yields of acetate, which was used for lipid synthesis in milk.

The recommendation by Walker (2013), to use saltbush in combination with feeds such as high-quality grass hay; to curb the negative impact due to its high mineral content was proved by the results of combining saltbush with *Cenchrus ciliaris*, which slightly (P=0.0434) decreased the fat content of camel milk (Table 4.4.3).

Contrary to Nagy *et al.* (2017) that dromedary camel milk quantity has a positive correlation with lactose and a negative correlation with all other measured components of fat, protein SNF and TS concentrations of the milk, Tsabong camel milk yield increased with supplementary feeding of sources that improved all components except lactose, (Table 4.4.1 and Table 4.4.5), which remained unchanged (P>0.05).

Milk urea level is an approximate indicator of the amount of crude protein in camel's diet. Milk urea is formed by the metabolism of absorbed amino acids protein. The positive increase in milk urea content of the supplemented camels therefore, indicates an improvement in the protein supply in the camels' diet. Supplementary feeding with *Atriplex nummularia* greatly increased (P<0.0001) urea and reduced (P=0.0003) FFA concentrations. The high protein content of Atriplex feed, as a supply of adequate amino acids, may have provided to the property of camel milk usually known to contain high whey proteins such as lactoferrin and immunoglobulin (Patel et al. 2016). That may have contributed to the low levels of FFA signifying low bacterial invasion. On the other hand, supplementing with *Cenchrus ciliaris* alone reduced the freezing point (P=0.0102). This may be to do with the high SNF content of the milk, as higher SNF content depresses the freezing point of the milk, as much as it elevates the boiling point, and increased the milk density (P=0.0007), while supplementing with the

combined feed (Buffelgrass/Saltbush (50/50) had the most positive effect ($P<0.0001$) on citric-acid concentration.

All supplementary feeds greatly increased ($P<0.05$) urea concentration of Tsabong camel milk, to levels that exceeded the mean value of urea concentration reported by Faye *et al.* (2010) of 81.6±60.4 mg/L with a range of 0–290.5 mg/L. Those could have been attributed to increased levels of soluble nitrogen in the feeds as shown in Table 4.4.2, as illustrated by the high protein content of the feed compared to the none supplemented group that only depended on an average low crude protein content (5.88%) from combined feed forages. The urea content of Tsabong camel milk was positively correlated to the corresponding total protein concentration in milk by supplementary feed type, just as was reported by Faye *et al.* (2010). Generally, the lower milk urea values in the non-supplemented Tsabong dromedaries' group were encountered due to the limited availability of degradable nitrogen from the pasture (5.88 CP) (Table 4.4.4.2). This correlation in milk urea to milk protein was evidenced by the increased protein and casein levels with an increase in milk urea values (Tables 4.4.3 and Table 4.4.4), results of which all show significant differences ($P<0.05$) at all levels of supplementation.

Milk from supplemented camel groups increased steadily towards week 8 then yield decreased with an increase in available succulent new growth forage species, as the rainy season advanced from week 7. That could have been the effect of energy dilution effect as the animals' feed intake consisted of less dry matter and therefore less energy to go into production. While on the other hand, the control group (none-supplemented), made a steady increase in milk yield as the rainy season progressed beyond week 10, and new shoots emerged in the forage, which possibly increased intake due to an increase in more palatable fiber, and the energy density of the forage feed was possibly reduced by the high mineral content of Saltbush feeding

4.6 CONCLUSION

It is concluded that supplementary feeding with *Cenchrus ciliaris* and *Atriplex nummularia* influenced camel milk composition. Supplementary feeding with all three treatments increased levels of milk urea, citric acid, density, casein, fat, galactose, glucose, protein and total solids. Reduced by the effect of supplementary feeding were concentrations of milk free fatty acids, lactose and solids-not-fat (SNF). Another improvement was found in the weekly milk yield which was generally influenced positively by the supplementary feeding with all three treatments, during the dry period with the most effects coming from supplementing with *Atriplex nummularia*. The lactic acid (%) content of Tsabong camel milk, was not affected by supplementary feeding. Highly significantly affected ($p<0.01$) were SNF, total solids, density, and FFA. Very highly significantly affected ($P<0.001$) were casein, protein, citric, and urea concentrations. Supplementary feeding with Saltbush (*Atriplex nummularia*) more than doubled the concentrations of milk urea, fat, galactose, glucose. It also increased milk concentrations of protein and total solids by 70.63% and 44.85%, respectively. However, saltbush supplementary feeding reduced concentrations of free fatty acids in milk by 52.29%. Supplementing with Buffelgrass (*Cenchrus ciliaris*) increased camel mil density by 0.59% and reduced concentrations of solids-not-fat by 26.99%. Supplementary feeding with the combined *Cenchrus* and *Atriplex* feed was found to increase milk citric acid and casein by 55.24% and 45.47%, respectively, but lowered concentrations of lactose by 6.63%.

4.7 RECOMMENDATIONS

The use of *Cenchrus ciliaris* and *Atriplex nummularia* to supplement camels during the dry periods to keep up with the future demand for camel milk, will be helpful to Tsabong Camel Park management, as the research has proved their worth in improving general milk quality and quantity. The existing sour-grass may also be directly supplemented with saltbush. The sour-grass may have to be harvested and hay baled while at its vegetative state, when it is most likely to be nutritious. Readily available energy sources like molasses meal which is sold in the in the area, as well as di-calcium phosphate may be used to cover for the very low phosphorus currently supplied by the grazing pasture.

CHAPTER FIVE

GENERAL CONCLUSIONS AND SCOPE FOR FUTURE RESEARCH

5.1 General conclusion

The overall study proved that lactation stage, parity and supplementary feeding are parameters, which have selective influences on concentrations of camel milk compositional components. Galactose and glucose were the only components significantly affected by the lactation stage. There was a reduction in milk galactose concentration from primiparous-early-lactation camels to those in multiparous-late-lactation stages of up to 36.50%. Another significant decrease was observed in primiparous camels where glucose concentration lowered by 61.98% from early lactation to mid lactation stages. Parity alone did not have significant effects on any measured components. The significant effects of the supplementary feeds elicited different responses. Significant effects were observed on fat, galactose, and freezing point. Highly significantly affected ($p<0.01$) were SNF, total solids, density, and FFA. Very highly significantly affected ($P<0.001$) were casein, protein, citric, and urea percentages. Glucose, lactose, and lactic acid components remained unchanged by the supplementary feed sources. Supplementary feeding with Saltbush (*Atriplex nummularia*) more than doubled the concentrations of milk urea, fat, galactose, glucose. It also increased milk concentrations of protein and total solids by 70.63% and 44.85%, respectively. However, saltbush supplementary feeding reduced concentrations of free fatty acids in milk by 52.29%. Supplementing with Buffelgrass (*Cenchrus ciliaris*) increased camel mil density by 0.59% and reduced concentrations of solids-not-fat by 26.99%. Supplementary feeding with the combined *Cenchrus* and *Atriplex* feed was found to increase milk citric acid and casein by 55.24% and 45.47%, respectively, but lowered concentrations of lactose by 6.63%.

Another improvement was found in daily milk yield being improved by the supplementary feeding with all three feed supplement treatments, with the highest effect coming from supplementing with *Atriplex nummularia.* However, supplementary feed sources significantly reduced ($P<0.05$) free fatty acids (FFA) content as well as the freezing point of camel milk.

The microbiological quality of camel milk produced in Tsabong at the time of the research was satisfactory according to the Kenya Bureau of Standards for raw camel milk and Botswana Bureau of Standards row raw cow milk. The microbial content related to total counts of coliform bacteria was below the tolerable limits for camel's milk after milking. This suggests that the milk was produced and handled under relatively good sanitary conditions. These findings are a useful contribution to the limited information available regarding the chemistry and microbiological properties of camel's milk in Tsabong.

5.2 Observations and recommendations for future research

Forage preference studies during the mating period makes it rather difficult to follow up with the animals during grazing, as the bulls mostly confine the females and their calves to specific territories and do not allow the females to wander out to browse/graze. These social organizations of the mating groups adversely affect foraging, especially of the females with their calves. Alternatively, one should confine the bulls away from the females to allow females to forage freely and get the maximum out of the veld. The study should be repeated during the summer period to fully investigate the effect of seasonal variations on milk parameters, as influenced by available forage nutrients, including minerals, to make informed decisions on what to supplement camels according to season. Further studies on milk composition involving fatty acids and amino acid profiles and mineral content of camel milk produced in Tsabong and

the influence of selected feeds towards its improvement are also recommended. A study of a combined effect of lactation stage, parity and type of feed supplement, on milk yield and composition is highly recommended for the Tsabong camels, in order to evaluate the nutritional requirements of these camels for feed formulation standards. Energy values of forage feed and supplementary feeds should also be evaluated, since energy and protein are the main factors influencing milk output in livestock. These findings are a useful contribution to the limited information available regarding the chemistry and microbiological properties of camel's milk in Tsabong. However, further studies on genetic variations and husbandry methods are needed in this field to support and enhance the production and utilization of this valuable food.

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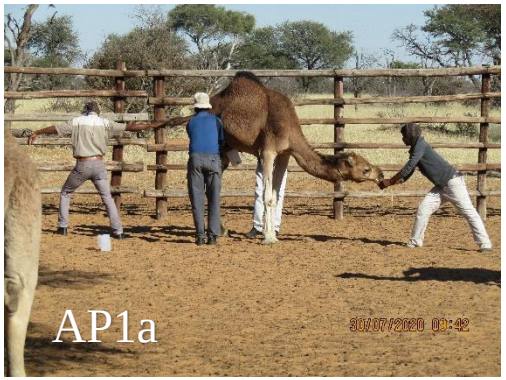
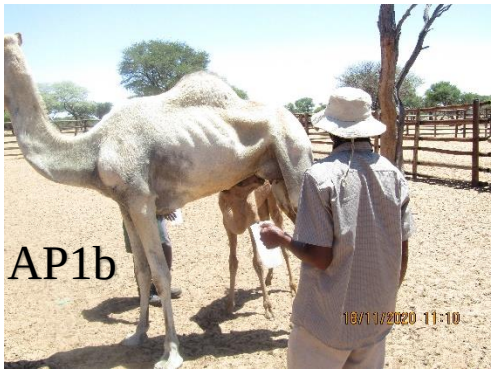
APPENDICES

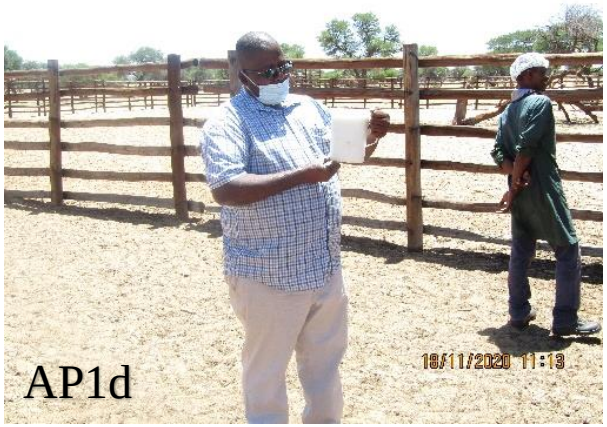
Appendix 1

Experiment 1. Milk collection from camels for analysis (AP1a-e)





(a) Restraining Primiparous camel in early lactation (b) Camel calf allowed to suckle first to facilitate milk let-down, (c) followed by hand milking into calibrated containers.



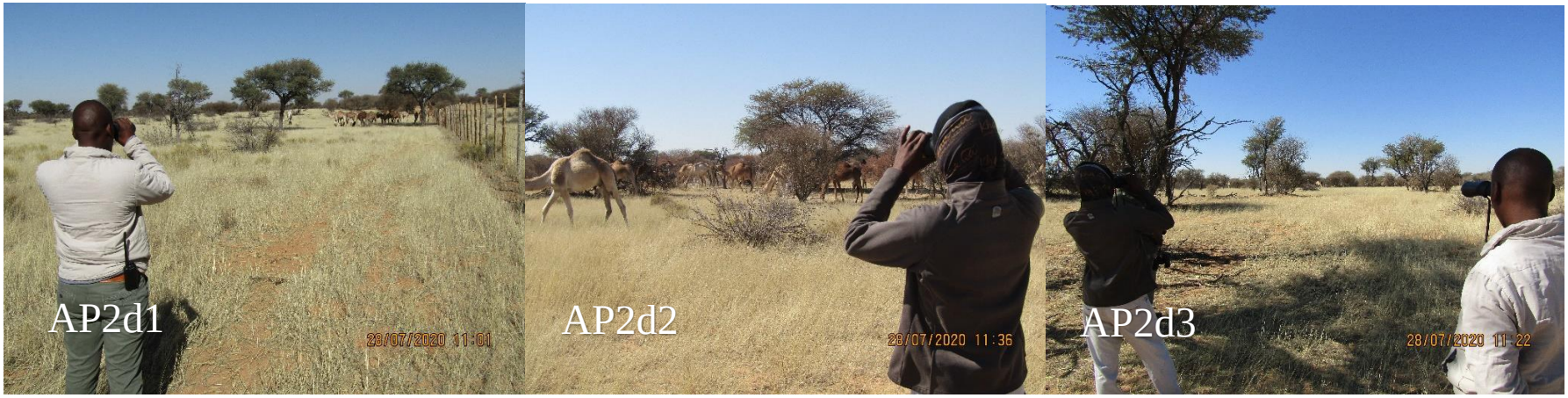
(d) Reading of the milk level on the calibrated scales on the containers

(e) Milk collection for testing done by stripping right into a sterile container

Appendix 2

Experiment 2. Camels forage preference analysis (AP2a-d3)

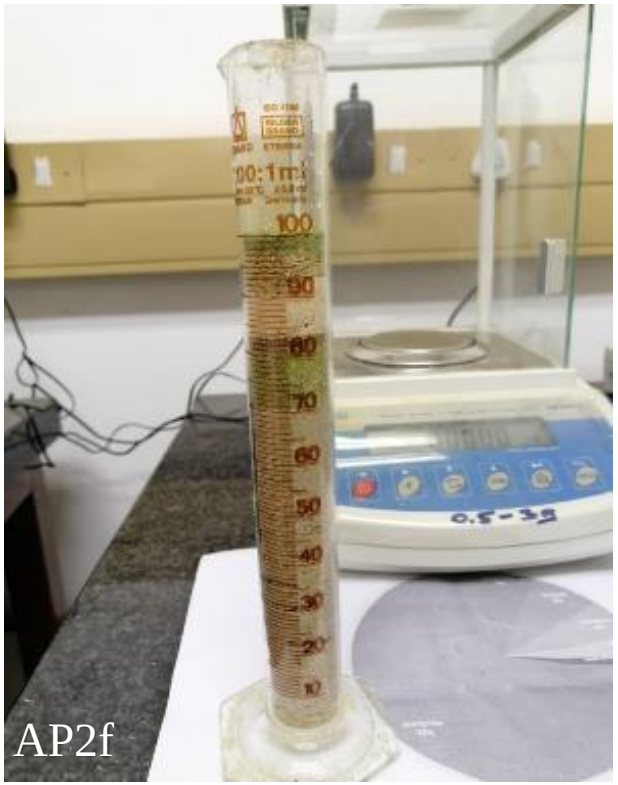
(A) Camel browsing on twigs and (B) grass while (C) Duration spent on browsing/grazing each species were observed and recorded



(d1-d3) Tracking and observing camels foraging preferences through binoculars to avoid disturbing the camels during their normal feeding

Experiment 2. Camels forage samples collection and preparation for analysis (AP2e-f)

(e) Clipping of forage within the browse line of camels



(f) Proportional representation of the forages as per feeding time

Experiment 2. Camel's feed and feeding (AP2g-k)

(g) Bales of *Cenchrus ciliaris* hay transported from BUAN to Tsabong

(h) Selected experimental camels group-fed.

(i) Saltbush harvesting

(j) Saltbush air-dried into hay

(k) Bagged hay weighing before feeding

Experiment 2. Preparation and handling of camel feces used for IVDMD (AP2l-n)

(l) Camel faeces vacuum packed immediately after collection, (m) Preserved during transport into a car electric heater/cooler, and (n) Temperature checked timely during transportation.

Experiment 2. Identification and preparation of camels for feeding and milking (AP2o-p)

(o) Camels used for the feeding trials ear tagged with varying tag colours according to the treatment assigned to experimental groups (p) Bi-weekly fetching of camels from the veld for milk sampling and supplementary feeding as per treatment

Appendix 3

Field data capture forms (AP3a-c)

AP3a: Forage feeding preference data collection sheet

Camel ID: _____ Date collected_____ Morning Data ☐ Afternoon Data ☐

Time	Forage species											
	Scientific Name	*Vachellia erioloba* E. Mey	*Senegalia galpinii Burtt Davy or A. luederitzii Engl.*	*Vachellia hebeclada* DC.	*Vachellia karroo Hayne*	*Senegalia mellifera (Vahl) Benth.*	*Boscia albitrunca (Burch.) Gilg & Benedict*	*Grewia flava* DC	*Rhus tenuinervis Engl*	*Rhigozum trichotomum Burch.*	*Schmidtia kalahariensis Stent*	*Tribulus terrestris* L.
	English Name	Camel Thorn	Monkey Thorn	Candle-pod Acacia	Sweet Thorn	Black Thorn	Shepherd's Tree	Brandy Bush	Kalahari currant	Threethorn	Kalahari sour grass	Devil's thorn
	Tswana Name	*Mogotlho*	*Mokala*	*Sekhi*	*Mokha*	*Mongana*	*Motlopi*	*Moretlwa*	*Modupaphiri*	*Mokurubane*	-	*Mosetlho*
5min												
10min												
15min												
20min												
25min												
30min												
35min												
40min												
45min												
50min												
55min												
60min												

AP3b: Milk collection data capture for experiment 1 camels

Date Sampled _____ Time Sampled _____ Place _____

Animal Number	**Animal ID** (Brand No. / Tag No.)	**Sample ID**	**Age**	**Parity**					
				Primiparous			Multiparous		
				Stage of Lactation			**Stage of Lactation**		
				Early	Mid	Late	Early	Mid	Late
1									
2									
3									
4									
5									
6									
7									
8									
9									
10									
11									
12									
13									
14									
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17									
18									
19									
20									
21									
22									
23									
24									

Preservative used _____

AP3c: Camels' weekly milk yield recording sheet

Week_____ Milk Yield Recording **(*Cenchrus ciliaris*-Supplemented Group) RED**

Animal ID	Day1 yield (Litres)	Day2 yield (Litres)	Day3 yield (Litres)	Week average
Total				

NB: The camels are to be milked on the day and just before feeding.

Week_____ Milk Yield Recording **(*Atriplex nummularia*-Supplemented Group) BLUE**

Animal ID	Day1 yield (Litres)	Day2 yield (Litres)	Day3 yield (Litres)	Week average
Total				

NB: The camels are to be milked on the day and just before feeding.

Week_____Milk Yield Recording **(*Cenchrus ciliaris/Atriplex nummularia* - Supplemented Group) GREEN**

Animal ID	Day1 yield (Litres)	Day2 yield (Litres)	Day3 yield (Litres)	Week average
Total				

NB: The camels are to be milked on the day and just before feeding.

Week_____ Milk Yield Recording **(None-Supplemented Group) YELLOW**

Animal ID	Day1 yield (Litres)	Day2 yield (Litres)	Day3 yield (Litres)	Week average
Total				

NB: The camels are to be milked on the day and just before feeding.