Sultan Qaboos University
&
United Arab Emirates University

Characterization of Camel Milk Protein Isolates as Nutraceutical and Functional Ingredients

Collaborative Research Project SQU/UAEU
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Contents

Executive Summary

The project consists of six objectives distributed between UAEU and SQU.

This report summarizes the results and the deliverable obtained for each of the six objectives.

Five (5) breeds of Emirati and two (2) Saudi breeds of milking camels have been selected. A total of 98 milk samples (both colustrum and raw camel milk) after parturition have been collected over a period of 10 months from Al Ain Dairy. Skimmed milk was prepared by centrifugation to remove the fat. The whole milk protein, mainly the casein was obtained by precipitation at pH 4.6, followed by centrifugation to obtain the supernatant of whey proteins. Both the casein proteins and the whey proteins were suspended in buffer. Each protein aliquot was subjected to chromatography on HPLC representing 196 samples being analyzed.

Preliminary fractions of proteins chromatograms were obtained and subjected to electrophoresis to determine their molecular weights and characteristics. The electrophoresis of the obtained various fractions are being subjected to SDS-PAGE electrophoresis.

Raw camel milk was obtained from Al Ain Dairy. The whole milk was subjected to evaporation and concentration prior the spray drying.

Process development was conducted using the following various parameters: Set temperature, Inlet temperature, outlet temperature, product flow rate, and the direction of the product versus the drying air.

Based on these processing parameters, the best conditions were selected to produce excellent camel milk powder based on the cow milk powder quality standards such as solubility, flowability, color and moisture content. Additional analyses were conducted such as functional properties, heat stability and accelerated shelf life of the camel milk powder.

A few batches of camel milk soft cheeses were produced with some limited success using some commercial enzymes. Over runs were conducted to test other commercial enzymes. A Master student worked on the extraction of plant enzymes; these enzymes were used successfully to produce soft curd camel milk cheese. In addition a PhD student has isolated chymosin from camel abommasum. Again soft curd cheese was produced.

Milk was fractionated into its four components: fat (pure fat and cream), casein, whey and lactose. The cream was separated by centrifugation and out of the cream the fat was separated by solvent extraction (*Rose-Gottlieb* method). Casein was precipitated by bringing the pH to 4.3. The whey was precipitated by saturating the casein and fat - free milk with ammonium sulphate. Finally, the lactose was precipitated by adding ethanol to the milk. A Master student thesis reported that two glass transition of casein proteins were obtained and the whey protein extracted by ammonium sulfate more stable than whey protein extracted by ethanol because it has high glass transition temperature than the other.

During the last three year periods of the collaborative research project, four master students have been trained and two PhD have partially worked on some objectives of the project.

Overall this project has demonstrated the richness of camel milk protein fractions/isolates, their molecular weights, their amino acids content and some of their functional properties as Nutraceuticals and as functional ingredients.

In addition a number of publications are in progress of submission and the principal investigators have disseminated their findings during their respective “research day” or research activities.

Introduction:

There are over 300,000 milking camels in both Oman and UAE with an annual production of 38,000 MT of milk in UAE (UAE, Municipality of Agriculture and Fishery, 2003). The production of camel milk has significantly increased during the last few years with now pasteurized fresh camel milk in the super market.

The potential application and the use of camel milk proteins, camel milk protein isolates and camel whey powder as functional foods and nutraceutical ingredients have not been established. Functional foods are defined as food substances with demonstrated physiological benefits and are designed to lower the risk or delay the onset of certain diseases. A nutraceutical is any substance that is a food or a part of a food and provides medical or health benefits, including the prevention and treatment of disease. Such products may range from isolated nutrients, dietary supplements, etc.

Camel milk is somehow different from cow milk in its chemical composition but it contains all essential nutrients as cow milk (Elagamy, 1988), also its high whey proteins such as lactoferrin and immunoglobulin confer to it the high antimicrobial properties. In average, camel milk contains more proteins and whey protein than cow milk (Farah, Z. 1993; Walstra *et al.*, 1999).

Casein fractions have been isolated in camel milk and found to be homologous with bovine casein. The balance between the different casein fractions is very different, however, and chiefly identified by a low amount of kappa casein of only about 5 percent of the total casein, compared with about 13.6 percent in bovine casein (Ramet, J. P.). This major difference in kappa casein content has shown difficulties in cheese making (Mohamed, M. A. 1990; Laleye, L. *et al.*, 2005, unpublished data). There is little information available on the ability of camel milk to undergo enzymatic coagulation. It has been reported that high dosage of calf rennet is necessary to obtain detectable coagulation (Chapman, 1985).

The quantity of whey proteins is higher in camel milk than cow's milk, at 0.9 to 1.0 percent and 0.7 to 0.8 percent, respectively (Mohamed, 1990). This is primarily due to the higher content of albumin and lactoferrin (Farah, 1993).

In an attempt to compare the functional properties of bovine milk proteins with camel milk proteins, these proteins have been separated and characterized (Beg *et al.*, 1987);

it was found an important thermodynamic property related to the heat stability. The camel milk whey proteins were found to be considerably more heat stable than cow's milk (Farah and Atkins, 1992). However, this heat stability has not been investigated in term of functional properties such as gelification properties of camel milk whey proteins.

Our current research on the functional properties of camel milk proteins and rennet-whey proteins demonstrated that using Differential Scanning Calorimeter, unlike the case in cow whey, the denaturation peak appeared at a much higher temperature (200ºC). The appearance of the peak at this high temperature indicated that camel milk whey is more stable than cow milk. Laleye *et al.* 2006 (unpublished data).

The whey produced during cheese making contains approximately 20% of all milk proteins. Due to their beneficial functional properties, whey proteins are used as ingredients in many food products (Cheftel and Lorient, 1982).

Studies of heat denaturation of major whey proteins (cow milk) either in separated purified forms, or in forms present in fresh industrial whey indicated significant differences in their denaturation (Bertand-Harb *et al.,* 2002)

Functional properties such as thermal stability (Allain, et al, 1999; Subirade et al, 1998), emulsifying (Lefevre and Subirade, 2003; Lefevre and Subirade, 2001; Lefevre and Subirade, 2000; Dufour et al, 1999) gelling (Lefevre et al, 2005; Remondetto et al, 2004; Remondetto and Subirade, 2003; Remondetto et al, 2002; Line et al, 2005; Gilbert et al, 2005; Lefevre and Subirade, 2000; Allain et al, 1999; Subirade et al, 1998) and foaming properties have been thoroughly studied and reported on cow milk whey proteins. However, there is dearth of report on camel milk casein proteins and whey proteins.

In addition, very little information is available on thermal transition characteristics and water sorption behavior of camel milk protein isolates. Water activity has traditionally been used to describe microbial and chemical stability of food products. During the last two decades, attempts have also been made to apply glass transition concept to explain the physical, chemical and microbial stability of food, bioactive compounds and pharmaceutical products. In the glassy state, reactions that depend on molecular diffusion, such as chemical and enzymatic changes, are reduced significantly. Despite of the research efforts, the significance and the mechanism of

the glass transition as an indicator of chemical and biochemical stability is still not clear. Due to the interrelation among factors (i.e. temperature, water activity and pH) influencing chemical reactions, it is difficult to interpret the results. A few studies have compared both criteria (i.e. water activity and glass transition) from the viewpoint of product stability. Information on stability of bioactive compounds in food materials such as fruits, vegetables, and dairy products and neutraceuticals formulations as influenced by water activity and glass transition is scarce. One of the objectives of this study will be to generate glass transition and water sorption data of camel milk protein isolates. Storage stability studies with such protein isolates may also help to increase our understanding of molecular mobility. This would enhance the knowledge of the necessary requirements for proper storage conditions and chemically stable formulations

Unlike cow milk, it was found that camel milk can be preserved for a longer time at 30ºC and most importantly the camel milk can be kept at 4ºC for more that three months without any visible change (Yagil, *et al.* 1984)

It has been reported that camel milk has a higher amount of lactoferrin compared to cow milk. The bacteriostatic activity of lactoferrin is well documented (Law, B. A., and B. Reiter, 1977). In addition, lactoferrin helps to establish a favourable microflora in the guts and consequently promotes growth of bifidobacteria. This functional property will be valuable as a neutraceutical agent in functional food products.

The ability of camel milk to inhibit growth of pathogenic bacteria and its relation to whey lysozyme has been demonstrated by Barbour et al (1984).

It has also been reported that camel milk is higher in α-lactalbumin, as it is in human milk compared with cow milk. Unpublished commercial data reported that some infant formula contains high level of α-lactalbumin in replacement to breast feed milk (Dr. Carver J., Khaleej Times, September 24, 2005).

More recently, Agrawal *et al.* (2003, 2005) have reported a unique camel milk health benefit in diabetic patients. These researchers have demonstrated that using camel milk as an adjunct to insulin therapy has improved the long-term glycemic control and led to reduction in doses of insulin in patient with type-1 diabetes.

Objective Distribution (SQU/UAEU):

No.	Objectives	UAEU/SQU
1.	To characterize the low molecular weight peptides present in camel milk proteins and identify the major components.	UAEU
2.	To determine the percentage content of the amino acids in the major proteins present in camel milk and correlate that with sequence of insulin.	UAEU
3.	To produce spray dried and freeze dried camel milk powder and determine the drying parameters and physico-chemical properties;	UAEU/SQU
4.	To develop a new processing cheese making technology from camel milk.	UAEU/SQU
5.	To determine and characterize the functional properties of camel milk proteins isolates in relation with their molecular structural changes and to study the thermal transition characteristics and water sorption behavior of these isolates.	SQU
6.	To incorporate camel milk powder in food formulations such as ice cream as fat replacer, flavour enhancer, texturizer, emulsifier and stabilizer and to conduct the sensory evaluation of the new formulated ice cream products.	UAEU/SQU

Research Team

Year	UAEU	SQU
2008	• Dr. Louis Laleye, P.I. • Dr. Salman Ashraf, Co-Inv. • Dr. Hanan Afifi, Co-Inv. • Dr. Oya Sipahioglu, Co-Inv.	• Dr. Ahmed Ali Al Alawi, P.I. • Dr. Shafiur Rahman , Co-Inv. • Dr. Nejib Guizani , Co-Inv.
2009	• Dr. Louis Laleye, P.I. • Dr. Salman Ashraf, Co-Inv. • Dr. Hanan Afifi, Co-Inv.	• Dr. Ahmed Ali Al Alawi, P.I. • Dr. Shafiur Rahman , Co-Inv. • Dr. Nejib Guizani , Co-Inv.
2010	• Dr. Louis Laleye, P.I. • Dr. Salman Ashraf, Co-Inv.	• Dr. Ahmed Ali Al Alawi, P.I. • Dr. Shafiur Rahman , Co-Inv. • Dr. Nejib Guizani , Co-Inv.

Research Assistant Employment:

Year	UAEU	SQU
2008	1. Ms. Amel Sboui, Ph.D. student	1. Mr. Mohammed Al-Raziqi (On request) 2. Mr. Salium Al-Abri (On request) 3. Ms. Bushra Al-Gafri, Undergraduate student
2009	1. Ms. Hina Kamal, Research Assistant, Full time 2. Mr. Osama G. El Amin, Master student 3. Ms. Amna Mohammed, and Ms. Badreya Ali, Undergraduate students	1. Ms. Siham Al-Hanai, Full time 2. Ms. Siham Al-Hanai, On request
2010	1. Ms.Hina Kamal , Research Assistant, Full time 2. Mr. Osama G. El Amin, Master student	1. Ms. Samaya Al-Tobi, On request

Involvement of Graduate Students

Year	UAEU	SQU
2008	1. Amel Sboui, PhD. University of Tunis	1. Tina (visiting student from India, MSc.)
2009	1. Osama G. El Amin, Master student, Al Ghezira University, Soudan	1.
2010	1. Osama G. El Amin, Master student, Al Ghezira University, Soudan 2. Rim Trabelsi, Master student, INAT, Tunis 3. Saliha Boudjena, University of Algeria, Algeria.	1. Hala Al-Hakmani, Master student

Coordination meetings and communication

- On April 5-7, 2010, Dr. Louis Laleye and his team (Mr. Ismail and Mr. Osama) conducted some pilot plant trails at SQU. The purpose of the pilot trials was to concentrate the milk using SQU concentrator facility and then produce the powder at UAEU.
  The team also used the cream separator facility to produce camel milk cream.
- On May 12, 2009, Dr. Louis Laleye and his team (Dr. Oya, Mr. Ismail and Mr. Osama) visited SQU for one day. The purpose of the visit was to set up the SQU concentrator facility and evaporator facility and to conduct "water" run. This was done successfully. The milk evaporator is very important equipment in the project, but SQU dairy plant staffs have no experience on running the machine.
- On April 14, 2009, Dr. Al Alawi received some camel powders from UAEU team to conduct FTIR analysis.
- On March 25 and 26, 2009, Dr. Al Alawi and the SQU documenting team visited, interviewed and filmed the research activities at the UAEU research laboratories. The documenting team also filmed the camel milk processing plant and the camel farm in Al Ain Farm
- Ahmed Al Alawi, PI from the Department of Food Science and Nutrition, Sultan Qaboos University met with Dr. Louis Laleye, PI, UAEU, and the UAEU research team on October 22, 2008, to discuss the project status and updates.

Annual Meetings

- On April 4-6, 2011, Annual SQU/UAEU steering committee meeting and technical presentation on the progress of the project held at SQU. Dr. Louis laleye and Dr. Ahmed Al Alawi attended the meeting.
- On June 2, 2010, Dr. Ahmed Al Alawi and Dr. Louis Laleye attended the annual meeting of SQU/UAEU research meeting to present the progress in the project. The meeting was held at UAE University.
- On December 6, 2009, Dr. Ahmed Al Alawi and Dr. Louis Laleye attended the annual meeting of SQU/UAEU research meeting to present the progress in the project. The meeting was held at SQU University.
- On April 13 and 14, 2009, Dr. Ahmed Al Alawi and Dr. Louis Laleye attended the annual meeting of SQU/UAEU research meeting to present the progress in the project. The meeting was held at UAEU University.
- On April 13-15, 2008, Dr. Ahmed Al Alawi and Dr. Louis Laleye attended the annual meeting of SQU/UAEU joint research committee meeting to discuss the project's proposal with the committee members. The meeting was held at Rotana Hotel.

Field Trip

On April 16-18th, 2008, Dr. Ahmed Al-Alawi visited Dhofar region (Oman) to see the current situation of camel milk industry in the region and to discuss with camel farmers for possible collaboration.

On January 23ed, 2009, Dr. Ahmed Al-Alawi visited Thamrate Willayat (Dhofar region) to attend the traditional camel milking contest.

Training

On the period from 22 September 2008 to 4 October 2008, Mr. Sulayem Al Abri (research assistant) was sent to Holland for training on Cheese making.

Collaborators

- Royal Court Affairs, Barka, Oman
- Al Ain Dairy, Al Ain, UAE

Objective One:

To characterize the low molecular weight peptides present in camel milk proteins and identify the major components

Abstract

The aim of the present study is to characterize ‘‘Camel's milk protein” using RP-HPLC. Furthermore, the content α-casein, β-casein, κ-casein, α-lactalbumin, β-lactoglobulin was determined by a reversed-phase HPLC method and the samples were prepared in triplicate. The present study showed. Individual standards injected in HPLC yielded the information about the retention time. Whereas the injection of mix standards helped us to develop calibration curve. Camel milk protein contains low amount of κ-casein and β-lactoglobulin compared to cow’s milk. Variant of α-casein fraction found in camel and cow milk was not same. Amount of α_{S2} casein was more in camel milk compared to cow milk. High amount of α-Lactalbumin found in camel milk requires confirmation by mass spectroscopy or capillary electrophoresis.

Introduction:

The overall composition of camel milk is similar to cow's milk; some differences exist in the molecular composition of proteins and lipids and in the mineral balance.

Protein. The average main composition of protein and nitrogen fractions of camel milk is generally similar to those of cow's milk. The average values for the casein and whey protein contents vary from 1.9 to 2.3 percent and 0.7 to 1.0 percent, respectively. The nitrogen content of casein is a little lower than cow's milk, reaching 71 to 79 percent compared with 77 to 82 percent in cow’s milk

Casein fractions being isolated in camel milk were found to be homologous with bovine casein. However, the balance between the different casein fractions is very different; for example, the amount of kappa casein is only about 5 percent of the total casein in camel milk compared with about 13.6 percent in bovine casein. Also the molecular weights and amino acid composition of casein fractions differ from those of cow's milk.

The state of the casein micelle structure has seldom been investigated. Most results, however, conclude that the size distribution of casein particles in camel milk is significantly broader than in cow's milk, exhibiting a greater number of large particles. The average micelle diameter of camel milk was found to be about double that of cow's milk at 320 nm and 160 nm, respectively.

The quantity of whey proteins is higher in camel milk than cow's milk, at 0.9-1.0% and 0.7-0.8% respectively. Individual fractions have been identified according to chromatographic and electrophoretic mobility and to the primary sequence of their amino acid chains. Two types of α-lactalbumin similar to bovine milk have been isolated. β-lactoglobulin has not been clearly identified (Conti *et al.*, 1985; Beg *et al.*, 1987; Farah, 1986). Two novel camel whey proteins, unlike any known bovine milk whey proteins, have been separated and characterized. The heat stability of camel milk whey proteins was found to be considerably higher than in cow's milk.

Lactose The average lactose content of camel milk is slightly lower (4.62%) than cow's milk (4.80%). It seems, however, that the variability is higher, with extreme values between 2.90 to 5.80 percent in camel milk compared with 4.40 to 5.80 percent in cow's milk.

Characteristics of camel milk

Close analysis of camel milk does show some medicinal potential. The milk protein lactoferrin, which present in large quantities in camel milk (ten times higher than in cow milk), does have some anti-viral and anti-bacterial properties. Fermented camel milk is high in lactic bacteria, which have been shown to be effective against pathogens including *Bacillus*, *Staphylococcus*, *Salmonella* and *Escherichia*. Furthermore, vitamin C content in camel milk is generally double that in cow's milk. In Russia, Kazakhstan and India as much as a Liter a day being prescribed to hospital patients to aid recovery from tuberculosis, Crohn's disease and diabetes.

A natural component of cow and human milk, lactoferrin is also found throughout the human body; it occurs in all secretions that bathe mucous membranes, such as saliva, tears, bronchial and nasal secretions, hepatic bile and pancreatic fluids. Exactly how lactoferrin functions is not entirely clear, but it is known to enhance the immune response, both directly and indirectly (passively) in reaction to a wide range of immune challenges and is an essential factor in the immune response in humans.

The health-promoting properties of camel milk are a strong boost for sales and, in certain regions such as the Middle East; they are the driver for intensification of camel dairying. According to Ulrich Wernery of the Central Veterinary Research Laboratory

in Dubai, it is time for camel to be managed in the some of the ways now well established and successful with milk cows. "I'm convinced that where there's money, such as in the United Arab Emirates, there will be dairy camel operations in the future, just like the world has now with dairy cows. Maybe there will even be high-tech rotary milking parlours" says Wernery. Trials are also proceeding to increase milk yields through intensification and breeding. "We are looking at solar systems to power small-scale milk units. And, we're looking at the genetic potential of the animal too because, in two generations, we will design and breed a camel to suit an automatic system," he claims.

Beneficial effects

Carmel's milk have been consumed for thousands of years in Africa and the Middle East, it's medical benefits toward modern diseases were not known until recently.
In 1986, an insulin-like protein has been detected in camel milk. Following clinical trials in human diabetes type I have shown that the daily consumption of 0.5 litre camel milk reduces the need for insulin medication by an average of **30%**. The anti-diabetic properties of camel milk have been demonstrated in several other studies.
Camel milk has positive effects in controlling high blood pressure and helps in the management of Arteriosclerosis and Osteoporosis. Research has demonstrated the presence of potent anti-bacterial and anti-viral factors in camel milk. Clinical trials showed that recovery from infectious disease (e.g. Tuberculosis) was significantly faster in patients consuming camel milk regularly.
Think about it, nature designed camel's milk to help baby camels grow up in some of the world's roughest environments - deserts and steppes. That helps explain why it is 3 times as rich in Vitamin C, 10 times as rich in iron as cow's milk, as well as containing many medicinal compounds.

Camel Milk is Useful in the Treatment of Diabetes

The intake of camel milk reduced the excessive need for insulin as it contains high levels of insulin or insulin like protein, which can pass through the stomach easily without getting destroyed. Stomach acidity would normally destroy the insulin taken, but one can take camel milk to avoid this. Oral insulin is worth the try. Long term

effects of camel milk are yet to be researched, but yet it is considered to be useful for controlling the glucose levels in the blood as of now.

Camels are generally looked upon as animals to travel upon in the deserts of Rajasthan, but now one has become more aware of the importance of camel milk in the control of Diabetes. The milk contains high insulin and insulin-like protein, which can help in regulating the blood glucose levels. This was so in the case of type I Diabetes and it was observed that drinking a pint of camel milk daily helped to improve the glucose levels.

Camel milk is an adjunct to insulin therapy and improves the long term glycaemic control and reduces the intake of insulin in Type I Diabetic patient. Research is on by the Indian Council of Medical Research (ICMR) to find out why the Rajasthani tribe, Raika does not suffer from Diabetes. Incidentally India is likely to become the epicenter of Diabetes by 2025. This race showed immense tolerance to glucose. Whether it is genetic or due to camel milk it needs to be further ascertained.

A one-month study conducted in Britain of Type I Diabetes patients has shown that a pint of camel milk daily does improve the blood sugar levels.

Camel milk does not form coagulum in acidic environment, which allows the camel milk to pass quickly through the stomach with the specific insulin and remain in the intestine for absorption. The radio immuno assay levels of camel milk are on the higher side.

Camel milk is highly nutritious, contains lower fat and lactose, higher levels of potassium, iron and vitamin C and large amounts of insulin like protein. An Austrian entrepreneur Johan is planning to produce camel milk chocolates, yoghurt and butter.

The solution to a Diabetic problem could lie in having more of camel’s milk. The milk may not be tasty but has ingredients that help a Diabetic to find solutions to his insulin problem.

Camel milk protein fractions

The concentrations of individual casein and whey proteins in camel milk differ markedly to respective protein concentrations in bovine milk. The ratio of β-casein to κ-casein is considerably higher in camel milk.

β-Lactoglobulin is absent, but whey acidic protein and peptidoglycan recognition protein have been detected.

Table 1. Average amount [mg l^{-1}] of some casein and whey proteins in mature milk from different species. nd = not detected. ↓ indicates a downregulation of gene expression between colostral and mid-lactational milk. ↑ indicates an upregulation in case of mastitis. Data from (Aguirre et al., 1998; Cals et al., 1994; Cuilliere et al., 1997; Hennighausen et al., 1994; Kappeler et al., 1999b; Ragona et al., 2000).

Milk protein	Camel	Cow	Human	Rodents	Function
α_{S1}-Casein	5000	12,000	minute	1600	Formation of casein micelle
α_{S2}-Casein	2200	3000	minute	nd	Formation of casein micelle
β-Casein	15,000	10,000	4670	4500	Formation of casein micelle
κ-Casein	800	3500	minute	nd	Formation and rennet coagulation of casein micelles
α-Lactalbumin	3500	1260	3400	nd	Regulatory subunit of lactose synthetase
β-Lactoglobulin	nd	3500	nd	nd	Binding of fatty acids and retinol
Whey acidic protein	157	nd	nd	1500	Probably an epithelial growth regulator, similar to WDNM1
Lactophorin (PP3 component)	950	300	nd	nd	Lipolysis inhibition
Lactoferrin	95 ↓↑	140 ↓↑	565 ↓↑	nd	Anti-inflammational, nutritive, iron uptake, regulative
Lactoperoxidase	nd	30	6 ↓	465	Anti-inflammational, bacteriolytic activity
Peptidoglycan recognition protein	107 ↑	nd	nd	nd	Anti-inflammational
Lysozyme C	nd	~100 ↓↑	274 ↓	nd	Bacteriolytic activity, N-acetylmuramidase

Objective:

The aim of the present study is to characterize the various fractions of Camel's milk proteins using RP-HPLC.

MATERIALS AND METHODS

Chemicals

Bis-tris, Tris (hydroxymethyl) aminomethane (trizma base), dithiotreitol (DTT), acetonitnle, trifloro acetic acid, α-casein, β-casein, κ-casein, α-lactalbumin, β-lactoglobulin and other reagents were from Sigma Chemical Co. (St. Louis, MO, USA). Reversed-phase column Symmetry C_4, 150 × 4.6 mm, 300-A˚ pore size, 5-µ particle size, was from (Waters Corp. USA).

Method to isolate individual camel caseins:

The following steps are illustrated in the reference literature* to isolate individual camel caseins:

- The milk was skimmed at 1000g at 4 °C for 15 min.
- The casein fraction was isolated by acid precipitation at 37 °C for 20 min with acetic acid (1:10, v/v, 100 ml/l) followed by addition of 1 m-sodium acetate pH 8.0 (1:10, v/v) and centrifugation at 4000g for 5 min.
- Supernatant and pellet were frozen and stored at - 28 °C.
- For crude preparation of an alpha and a beta - CN fraction, the casein pellet from 1 L fresh milk was dissolved in 200 ml 10 M-urea, diluted with 460 ml double distilled water and the pH adjusted to 7.5 with 1 M-NaOH. The solution was then diluted with 200 ml double distilled water and adjusted to pH 5.0 with 1 M-HCl. The precipitate consisted mainly of alpha and kappa - CN.
- After centrifugation at 600g for 5 min, the supernatant was saturated with ammonium sulphate for precipitation of beta - CN.
- Both precipitates were lyophilized.
- Individual caseins were separated by HPLC on an analytical reversed-phase C18 column.

- Before analysis, 1 g acid precipitated casein was dissolved in 5 ml 10 M-urea, 140 mM sodium citrate, 35 mM-1,3-bis[tris(hydroxymethyl)-methylamino]propane, 780 mM b-mercaptoethanol, 200 mM-Tris-HCl buffer, pH 8.0, stirred for 1 h and passed through a hydrophilic 0.45 μM fliter.
- Solvent A was trifluoroacetic acid (TFA, 1 ml/l) in water.
- Solvent B was TFA (1 ml/l) in acetonitrile.
- After injection of 10-50μl filtrate, elution was performed by a linear gradient from 0 to 350 ml solvent B/l over 15 min, followed by a linear gradient from 350 to 450 ml B/l over 35 min, a linear gradient from 450 to 1000 ml B/l over 5 min, a 3 min hold at pure B, then from 1000 to 0 ml B/l over 2 min.
- The flow rate was 1 ml/min and runs were performed at room temperature.
- The column effluent was monitored with a diode array detector from 200 to300 nm.
- Proteins eluted were collected manually and lyophilized.

NOTE: For large scale isolation of individual caseins, a semi-preparative column was used to separate the proteins of the crude alpha and beta – CN fractions. After injection of 1ml filtrate, elution was performed by a 9min hold with pure solvent A, followed by a linear gradient from 0-400ml solvent B/l over 3 min and a linear gradient from 400-430ml solvent B/l over 28min. The flow rate was 9.5ml/min and runs were performed at 30*C.

Stefan Kappeler, Zakaria Farah and Zdenko Puhan. Sequence analysis of Camelus dromedaries milk caseins. *Journal of Dairy Research (1998) 65 209-222.

Preparation of samples

Samples were prepared as per the method suggested by Moatsou et.al (2008). Pasteurized sample of camel and cow milk obtained from the market (Al-Ain Dairy) were used in this study. Milk samples were first defatted to remove fat by centrifugation at 3000g for 30 min at 15 ºC. Samples for HPLC injection were prepared as follows: 0.5ml of defatted milk was dissolved in 1ml Tris-buffer, pH 7 (100mM Tris-HCl, 8 M urea, 13g/l trisodium

citrate, 20mM dithiothreitol). After incubation for 1 h at 37ºC, 10ml of solvent A containing 6M urea was added to the sample solution and the pH was adjusted to 2.1-2.2 by the addition of 0.5ml of TFA solution (100ml/l). After filtration through 0.45µm filter (Millipore Corporation, Bedford, MA 01730, USA), 100 µl of sample was injected on HPLC. Each sample was analyzed in triplicate..

Preparation of standards

Standard were prepared by weighing individual pure protein standards of α-casein, β-casein, κ-casein, α-lactalbumin, β-lactoglobulin, from 15 to 75mg. Standards were dissolved in 1ml Tris-buffer pH 7 followed by incubation and dilution with solvent A and filtration as mentioned in sample preparation. Individual protein standards were mixed in equal amount to produce mix standards for all the five proteins and diluted to achieve different concentration level. Concentrations of individual and mix standards were produced as shown in table 1 and table 2 respectively

Table 1 Concentration of individual milk protein standards

Milk Protein	Concentration mg/ml
α-casein	6.25
β-casein	6.36
κ-casein	1.45
α-Lactalbumin	1.46
β-Lactglobulin	1.45

Table 2 Concentration of mix protein standard (mg/ml)

Standard level	α-casein	β-casein	κ-casein	α-Lactalbumin	β-Lactglobulin
Mix standard 1	1.25	1.27	0.29	0.29	0.29
Mix standard 2	0.62	0.63	0.14	0.14	0.14
Mix standard 3	0.31	0.31	0.07	0.07	0.07

Reversed phase high performance liquid chromatography

The complete chromatographic system from Waters corp. (USA) was used in the analysis. It consisted of the following: a HPLC pump model 1525, an auto sampler model 717 and a Dual Absorbance UV detector 2487. The temperature of the column was maintained by a HPLC column thermostat model SCH. All data were treated by the chromatographic system software Breeze version 3.30 SPA. The sample vials were kept at constant low temperature (7ºC) by refrigeration system provided with auto sampler.

The chromatographic separation was performed on a reversed-phase analytical column Symmetry C_4, 150 × 4.6 mm, 300-A˚ pore size, and 5µm particle size. with the following programme suggested by Bordin et at (2001), It consisted of a linear gradient from 26.5 to 28.6% B in 7 min (0.30% B min), then from 28.6 to 30.6% B in 10 min (0.20% B 21 min) and from 30.6 to 36.1% B in 11 min (0.50% B min), followed by an isocratic elution at 36.1% B during 10 min and a final increase to 43.3% B in 18 min (0.40% B min), at a flow-rate of 1 ml min , where solvent A is composed of 10% (v/v) acetonitrile and 0.1% TFA in ultrapure water and solvent B composed of 10% water and 0.1% TFA in acetonitrile. The column temperature was kept at 40ºC and injection volume was 100 µl. Chromatogram was recorded at UV 214 nm and 280 nm wavelength.

RESULTS AND DISCUSSION

Identification of protein and calibration

Initially chromatogram for samples and standards was developed by the method suggested by Moatsou et al (2008) where a linear gradient of solvent B from 35 to 62% in 54 minutes was used. This resulted in a chromatogram with unresolved peak for some of the variants of α-casein, β-casein and α-Lactalbumin. Modifications were made to mobile phase and the method suggested by Bordin et al (2001) with some modification as mentioned above was used. To identify the individual protein in the mix standard and samples; individual protein standards were injected and retention time of each protein was determined. Table 3 shows the retention time range for each protein and number of peaks exhibited in the chromatogram for variants of each protein.

Table 3 Retention time range and number of variant peak for each milk protein.

Milk protein	Retention time range (min)	Number of peak
κ-casein	11.77 to 23.75	3
α-casein	25 to 32.71	2
β-casein	41.5 to 46.6	3
α-Lactalbumin	50.5	1
β-Lactglobulin	51 to 53.44	2

Mix standards of milk protein viz. α-casein, β-casein, κ-casein, α-lactalbumin, and β-lactoglobulin were injected at three different concentrations in duplicate and calibration

curves were plot for each protein The calibration curve for each protein was linear passing through zero with coefficient of regression above 0.93, which is considered as very good calibration. Figure 1 to 3 shows the chromatogram of standard mix and Table 4 shows the results of response and regression coefficient for each protein.

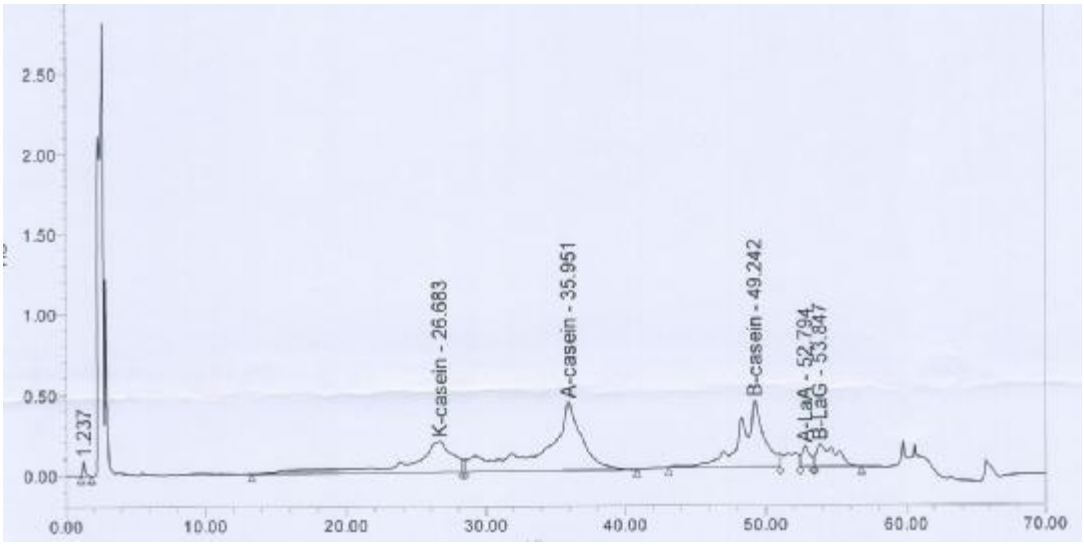

Figure 1 S1: Mix of standards (α- casein, β- casein, Κ- casein, a-lactalbumin, b-lactoglobulin)

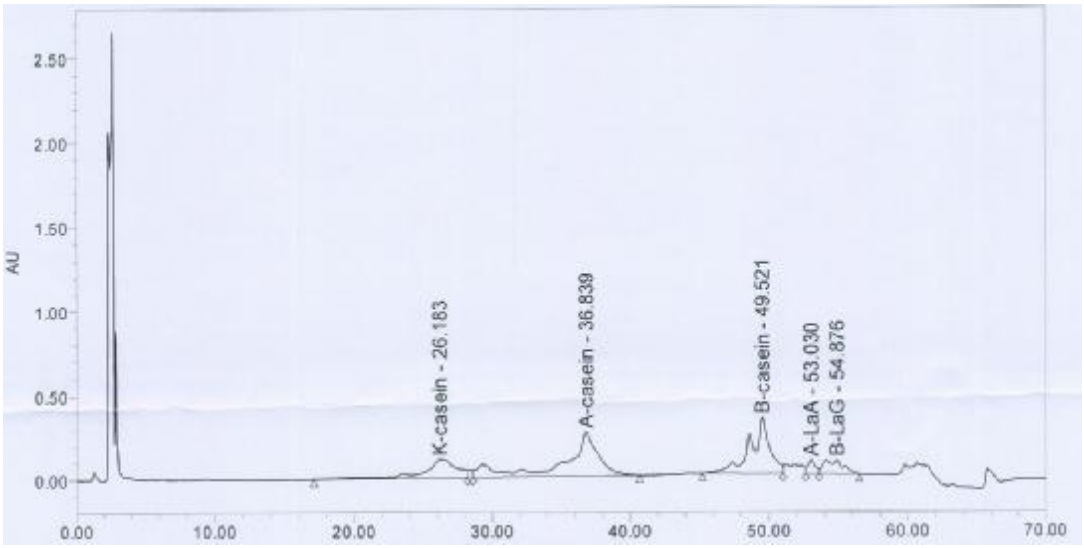

Figure 2 S2: Mix of standards (α- casein, β- casein, Κ- casein, a-lactalbumin, b-lactoglobulin)

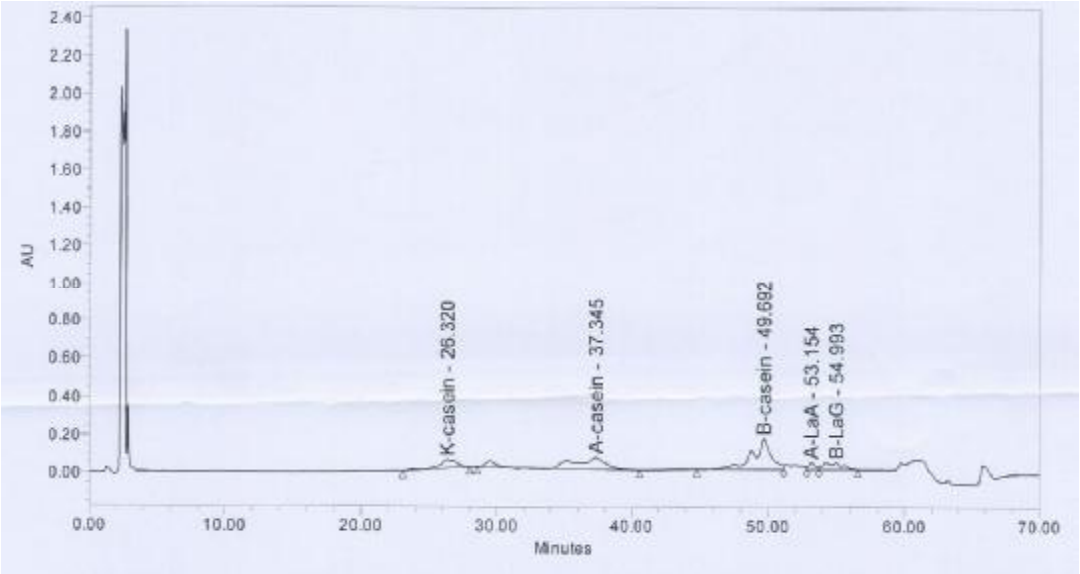

Figure 3 S3: Mix of standards (α-casein, β-casein, κ-casein, a-Lactalbumin, β-lactoglobulin)

Table 4 Response area and regression coefficient for each milk protein

Milk protein	Average Response for standard (area, µV*sec)			Regression Coefficient
	Mix Std 1	Mix std 2	Mix std 3	
κ-casein	41300000	17810000	3704000	0.97
α-casein	78760000	45120000	11570000	0.94
β-casein	47590000	31790000	14970000	0.93
α-Lactalbumin	4948000	2934000	1272000	0.98
β-Lactoglobulin	13140000	7393000	3528000	0.99

Following the calibration, samples of camel milk and cow milk were injected on HPLC. Figure 4 and 5 shows the chromatogram of camel milk and cow milk respectively.

Chromatograms were integrated using breeze software and quantification was done for each protein present in milk samples. Table 5, depicts the amount of each protein found in both camel milk and cow milk.

Table 5 shows the amount of different protein found in camel milk. It is clear that the amount of β-casein in cow milk was the highest whereas, for camel milk protein β-casein has low amount compared to α-Lactalbumin In total, camel milk protein contained higher amount of protein and this explain the appears of two peaks of β-casein and one big peak α-LaA in camel milk chromatogram in figure 4 compared with cow milk which has three peaks for β- casein (fig. 5.).

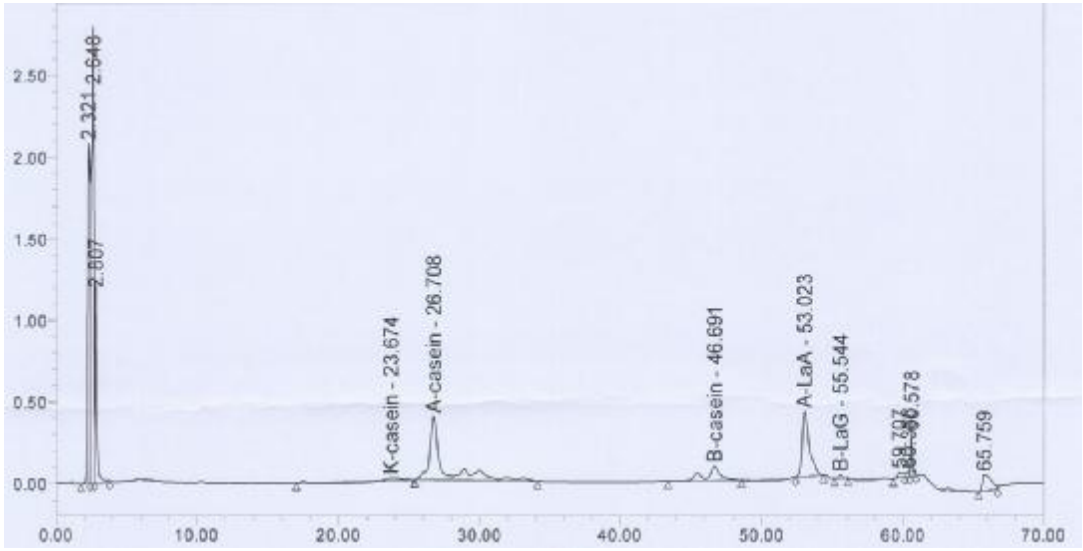

Figure 4 Camel milk sample

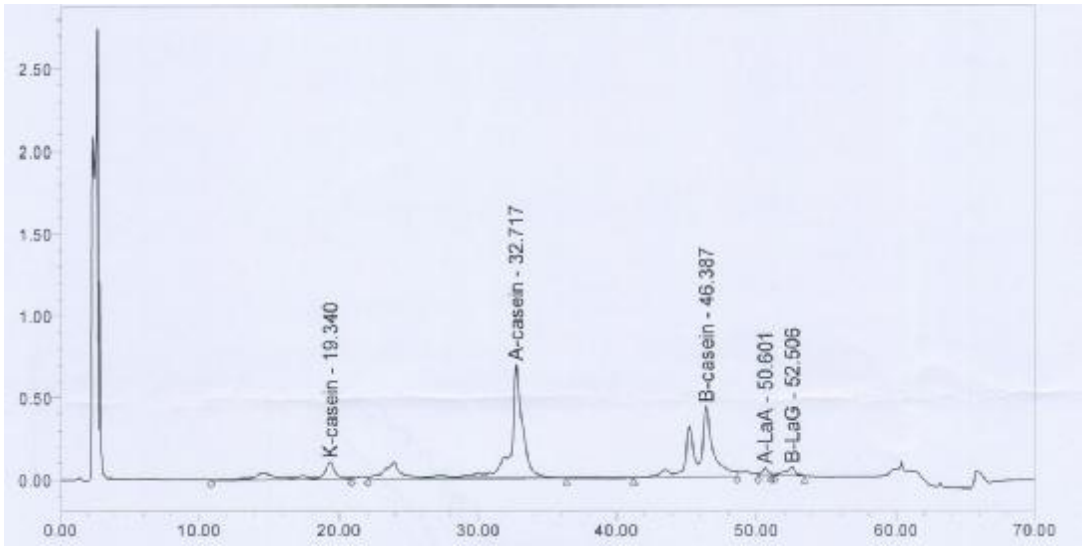

Figure 5 Cow milk sample

Table 5 Amount of different proteins found in camel milk and cow milk

Protein	Cow milk (mg/L)		camel milk (mg/L)	
	Pasteurized market milk	Literature value*	Pasteurized market milk	Literature value
α-casein	15,210.67	15,000.00	6,944.00	7,200.00
β-casein	20,914.67	10,000.00	6,944.00	15,000.00
κ-casein	3,554.67	3,500.00	744.00	800.00
α-Lactalbumin	2,810.67	1,260.00	23,312.00	3,500.00
β-Lactglobulin	2,397.33	3,500.00	248.00	zero
Total protein	44,888.00	33,260.00	61,173.33	26,500.00

A chromatogram showing the separation of a mixture of standard proteins in three different concentrations (α-CN, β-CN, κ-CN, α-La, β-Lag) is shown in Fig. 1 to 3.

The standards κ-CN and β-CN chromatogram has three main isomers that is why it has three main peaks and it is clear it had three different concentrations (fig. 1 to 3). Also, standards α-CN and β-Lag each has two peaks and the standard α-La has one peak (fig. 1 to 3).

From Table 4 it is clear that the values of the retention time decreased as the concentration of the standard decreased, so it is dependent on the amount of protein.

Conclusion

The present study has increased our knowledge about the ‘‘camel milk” protein. Through Reversed phase high performance liquid chromatography followed by electrophoresis. In the beginning the separation of protein of camel and cow milk samples was difficult because the peaks weren't separate nicely also for the standards we faced the same problem, some isomers for the standards wasn't appearing. To solve this problem the injection done three times with some change in conditions to get good results.

On the basis of the results obtained in our studies, we found that casein protein in camel milk is less than that in the cow milk but it has all essential nutrients. On the other hand, camel milk contained more whey protein than cow milk. This variation is primarily due to the higher content of albumin in camel milk.

Annex to the objective of camel milk protein characterization

REVERSED PHASE HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (RP-HPLC) METHODOLOGY FOR CAMEL MILK PROTEINS

1. Introduction

Sometimes you may wonder who was the first to make an experimental procedure for a specific analysis. The route from an idea to an actual standard operating procedure (SOP) is not easy. In the early days it was a long period of trial and error to establish a document where common users could benefit from, but with modern technology such is not the case now. To abridge, this document will try to explain some of the basic jargon and glossary concerning RP-HPLC methodology for camel milk proteins.

2. Materials and Methods

Reversed Phase High Performance Liquid Chromatography (RP-HPLC) for camel milk proteins includes the following basic steps, such as:

1. Skimming of camel milk,
2. Ultra filtration of skimmed camel milk
3. Acid precipitation of skimmed camel milk,
4. Lypophilization of camel milk casein and whey proteins,
5. Quality evaluation of C4 column,
6. Preparation of required RP-HPLC grade chemicals,
7. Preparation of camel milk protein samples anent RP-HPLC protocol,
8. Injection of camel milk protein samples and
9. Integration and Quantification of results.

2.1 Skimming of camel milk

Skimmed camel milk was prepared from raw fresh milk by centrifugation at 3000g for 15minutes at 4°C.[1] This is perhaps the most essential step in sample preparation, because if the milk is not skimmed, fat particles will block the filtration units, thus resulting into loss of sample prepared.

2.2 Ultra filtration of skimmed camel milk

Once the camel milk is skimmed, it is suggested to remove sugar (lactose) and minerals present in the skimmed milk by ultra filtration. The prime objective of ultra filtration is to remove all other impurities, other than the camel milk protein, in order the get define peaks once the samples are injected. Ultra filtration requires 10ml of skimmed camel milk to be placed into the centrifugal filter units (with a molecular weight cut off at 30K Da). The skimmed camel milk placed in the centrifugal filter devices was then simply centrifuged at 5000g for 20minutes at 6°C (these conditions were in accordance to the user guide manual provided with the centrifugal filter devices).

2.3 Acid precipitation of skimmed camel milk

After the removal of fat and impurities in the first two steps, i.e., 2.1 and 2.2, skimmed milk was subjected to acid precipitation for the separation of casein and whey proteins, such that to the measured volume of skimmed milk, for e.g., 20ml, 30ml etc, 10% (v/v) acetic acid and 1M Sodium acetate was added, to the point where the pH of the skimmed

milk was lowered close to the iso-electric point, i.e., 4.6 and once the desired pH of 4.6 was obtained, skimmed milk sample was centrifuged at 5000g for 30minutes at 15°C.[1] Once the centrifugation is concluded, separation of casein and whey proteins is quite vivid, such that the white casein pellet rests at the bottom of the centrifugation tube, whereas the milky whey (supernatant) floats at the top.

2.4 Lypophilization of camel milk casein and whey proteins

The acid precipitated camel milk casein and whey proteins are then lyophilized (freeze dried) using a laboratory scale freeze dryer at -75°C for almost 6-24 hours, here one cannot limit the time required for the completion of freeze drying as it is dependent on the quantity of sample placed for freeze drying such that in order to freeze dry a 1-2g sample roughly 6 hours are required.

2.5 Quality evaluation of C4 column

It is advised to check the quality of the column, if it is not used too often, before the injection of any samples for RP-HPLC analysis. The quality check is always in accordance to the manual guide provided by the supplier of the column, such that in order to evaluate the quality of C4 column, following is the test method, as mentioned in the brief provided by the supplier:

Mobile Phase	50/50 Acetonitrile/Water
Sample	Acetone 6ul/ml
Flow rate	0.9ml/min
Detection	254nm
Temperature	Ambient
Injection Volume	3.0ul

Note: The quality check performed over the C4 column came negative, such that with acetone as the sample injected, it was expected to get a single define peak, but for every 5 runs that we conducted in order to confirm the quality of the C4 column showed results with multiple and distorted peaks, thus concluding that either the column is at waste now or there is some problem with the entire unit, which needs to be checked out thoroughly.

2.6 Preparation of required RP-HPLC grade chemicals

Sample preparation for RP-HPLC analysis requires the preparation of certain HPLC grade chemicals, such that:

2.6.1 Mobile phases

Mobile Phase "A" was 1.06 ml/l trifluoroacetic acid (TFA) in ultra pure water and Mobile Phase "B" was 1 ml TFA, 800 ml acetonitrile and 200 ml ultra pure water. The flow rate for each of the mobile phase was 1 ml/min, the analyses were carried out at 40 °C and the eluent was monitored at 214 nm. A linear gradient from 350 to 620 ml/l Solvent B, within 54 min, was applied.[2]

2.6.2 RP-HPLC grade chemicals

Other than the mobile phases, RP-HPLC analysis requires the preparation of

- Tris Buffer, pH 7.0 (100 mM Tris–HCl, 8 M urea, 13 g/l trisodium citrate, 20 mM dithiothreitol).
- Solvent "A" containing 6 M urea was added to the sample solution (mobile phase "A") and the pH was then adjusted to 2.1–2.2 by the addition of 0.5 ml of a TFA solution (100 ml/l).

2.7 Preparation of camel milk protein samples anent RP-HPLC protocol

60mg of freeze dried camel milk casein was dissolved in 1 ml buffer, pH 7.0 (100 mM Tris–HCl, 8 M urea, 13 g/l trisodium citrate, 20 mM dithiothreitol). After 1 h at 37 °C, 10 ml of Solvent "A" containing 6 M urea was added to the sample solution and the pH was adjusted to 2.1–2.2 by the addition of 0.5 ml of a TFA solution (100 ml/l). After filtration through 0.45 µm filter (Millipore Corporation, Bedford, MA 01730, USA), 50 µl of sample was injected. The same procedure is adopted for the camel whey proteins.[2]

Note: Here it is important to mention the fact that initially when we first injected our first batch of camel milk casein and why samples for analysis we used to quantify only 40mg of sample either casein or whey proteins and based on this initial weight (40mg) of casein and whey samples measured, we calculated the results obtained after the quantification and integration of the samples injected for RP-HPLC analysis, but later on the concentration was increased from 40mg to 60mg and finally to a maximum of 100mg, this increment in the concentration of camel milk protein (casein and whey) was conducted to maximize the concentration of protein in the RP-HPLC sample prepared, so that a more define peak is obtained. Moreover, the other objective was to (a) collect fractions of the camel milk proteins (casein and whey), (b) freeze dry the fraction collected, (c) preparation and re-injection of the same fraction collected freeze dried camel milk proteins (casein and whey) anent RP-HPLC protocol, (d) again fraction collection followed by freeze drying, preparation, re-injection, in short this particular step was to be carried out to a minimum of 5 times, and if similar and define peaks are obtained for each re-injection, it was to confirm that protein as the standard for the camel milk, because earlier we used to compare the camel milk proteins with bovine standards as a reference.

2.8 Injection of camel milk protein samples

Before the injection of the samples, the method adopted for analysis, or in other words the conditions required for analysis, such as absorbance wavelength, injection volume, time required etc are all equilibrated for a minimum of one run of 30minutes. As for the camel milk protein samples, i.e., casein and whey, the conditions required were already designed and saved in the system as we used the same method earlier even, so the method was just selected from the list of methods and equilibrated.

2.9 Integration and Quantification of results

Note: ***Not yet carried out for the current fraction collection analysis of camel milk proteins as the results obtained had multiple and distorted peaks! Also for the results of***

the first batch, the data obtained was compared with bovine standards as reference, hence the values obtained are not correct, because apart from the similarities between cow and camel milk, still there is a huge difference between the structure and physical properties of camel and cow milk proteins.

3. References:

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Objective two:

To determine the percentage content of the amino acids in the major proteins present in camel milk and correlate that with sequence of insulin.

Introduction

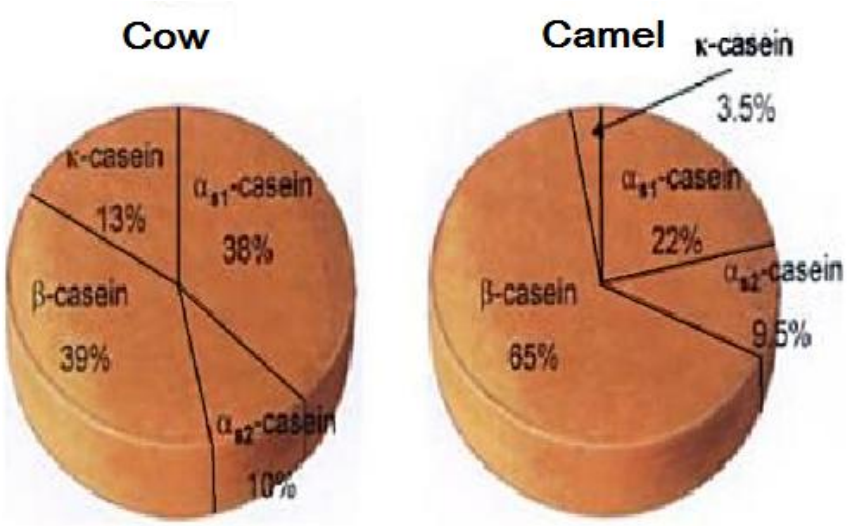
The results of the objective one related to the various fractions of camel proteins confirmed that camel milk proteins contained two major fractions: the casein proteins and the whey proteins. The casein proteins consisted of α-casein and β-casein in almost equal amount (6,944 mg/L) and a small amount of κ-casein (744 mg/L). The whey proteins consisted of mainly α-lactalbumin. From the RP-HPLC there were several fractions unidentified. This is due to the limitation of the available standard. It should be pointed out that we have been using commercial standards of protein fractions from bovine milk. Ideally we should use protein standards from camel; unfortunately these standards were not commercially available. We have attempted to separate these fractions chemically. The chemical separation was not done completely due to non availability of all chemicals. Partial separation will be presented in this report.

Camel milk casein

Camel caseins, indeed, have been studied by many authors subsequently handling with different properties such as micelle aspect, the diameter of the micelles and its distribution (Mehaia and Cheryan, 1983; Gouda *et al.*, 1984; Ali and Robinson, 1985; Farah and Farah-Riesen, 1985; Mehaia, 1987 a, b and c; Farah and Ruegg, 1989; Jardali and Ramet, 1991; Ochirkhuyag *et al.*, 1997; Kappeler *et al.*, 1998; Attia *et al.*, 2000; Kherouatou *et al.*, 2003)

Camel caseins are representing the most abundant protein fraction of camel milk amounted between 73 and 81% of total protein against an average of 83% in bovine milk (Sood and Sidhu, 1979; Mehaia *et al*, 1995). Most results, however, conclude that the size distribution of casein particles in camel milk is significantly broader than in cow's milk, exhibiting a greater number of large particles. The average micelle diameter of camel milk was found to be about double that of cow's milk at 320 nm and 160 nm respectively (Ramet, 2001). It is estimated to be around 260-300 nm; well above average in cow's milk (100-140 nm) according to Ali and Robinson (1985), Farah and Ruegg (1989) and Jardali and Ramet (1991). Camel milk casein and their fractions were found to be poor in crude protein when compared with cow milk (Pant and Chandra, 1980; Attia *et al.*, 2001). Proteins revealed pronounced differences in quantitative distribution of

casein and whey proteins. β-casein was found in higher concentration than in bovine milk, whereas κ-casein amounted to only 3.5% of the casein fraction, which presented a rate of three times lower than in milk of 36 cows (Kappeler *et al.*, 1998;). The whey proteins mainly consisted of α-lactalbumin, serum albumin and lactophorin (Proteose Peptone component 3). β-lactoglobulin, the main whey protein of bovine milk, was not found (Kappeler, 1998; El Hatmi, 2006). Figure 2 illustrates the relative amounts of different casein types to both camel and cow milk according to Farah *et al. (*2004). Attia *et al.* (2000) and Kherouatou *et al.* (2003) studied the state of casein micelles during acidification of milk and concluded that the maximum micelle demineralization occurs at pH 4.3 lower than that of bovine casein (4.6). Similar results were reported earlier by Wangoh *et al.* (1998).



Relative amounts of caseins in cow and camel milk (Farah *et al.*, 2004)

Amino acid composition

Caseins are highly digestible in the intestine and are a high quality source of amino acids whose composition is appropriate for growth and development of the nursing young. Indeed, the amino acid composition of camel milk proteins is very similar to that reported for the cow milk (Sawaya *et al.*, 1984; Mehaia and Alkanhal, 1989). Table 7 shows the different amino acid composition in both dromedary and bovine milk.

Table 6 Amino acid composition of camel and cow milk in g/100g of protein

Amino acid	*Camel milk (Shamsia, 2009)*	*Cow milk (Renner, 1991)*
Alanine	3.3	3.5
Arginine	5.1	3.7
Aspartic Acid	7.2	7.9
Cystine	1.5	21.8
Glutamic Acid	21.1	2.1
Glycine	1.2	2.8
Histidine	2.9	6.4
Isoleucine	4.9	10.4
Leucine	9	8.3
Lysine	6.6	2.7
Methionine	2.6	5.2
Phenylalanine	3.7	10.0
Proline	13	5.6
Serine	3.0	5.1
Threonine	5.3	1.4
Tyrosin	3.0	6.8
Tryptophan	1.8	5.3
Valine	4.8	-

The amino acids content of the major fractions of camel milk protein

Camel milk proteins have been isolated, purified and characterized by many researchers (Farah et Farah-Riesen, 1985; Larson-Razikiewicz et Mohamed, 1986; Ochirkhuyag *et al.*, 1997; Kappeler *et al* 1998). Other researchers reported the amino acids composition and their primary sequence (Farah et Farah-Riesen, 1985; Kappeler *et al.*, 1998).

α_{S1}-Casein

This is the most abundant casein representing 22% of the total casein. Alpha S-1 casein of camel milk contains 215 amino acids with a molecular weight 25,773 Da with an iso electrical pH of 4.4 (Kappeler *et al.*, 1998).

α_{S2}-Casein

This fraction is present in the camel milk at an average of 2.6g/l (Ribadeau-Dumas Grappin, 1989). The primary structure contains 207 amino acid residues (Grosclaude *et al.*, 1989) from which 11 residues of phosphorilated serine and two cysteine residues that will bring the disulfide bridge. The molecular weight of this fraction is approximately 21,266 Da.

β-Casein

The β-casein is composed of 217 amino acids with a molecular weight of 24,651 Da and an isoelectric point at pH 4.76 (Kappeler *et al.,* 1998).

κ-Casein

This is the most studied protein due to its role in milk coagulation by rennet (Cayot et Lorient, 1998). The κ casein from camel milk is composed of 162 amino acids and its molecular weight is 22,500 Da. κ casein is hydrolyzed by the chymosin resulting into fractions: the para-casein kappa and the caseinomacropeptide which is responsible for milk coagulation (Mercier *et al.,* 1976).

The soluble proteins

The quantitative and qualitative distribution of the soluble whey proteins is unique and has a particularity in camel milk compared to other milks. The following soluble proteins are present in camel milk: α-lactalbumine, Serum albumin, Lactophorine, Lactoferrine, Lactoperoxydase and Immunoglobulines.

MATERIALS AND METHODS

Electrophoresis

In polyacrylamide gel electrophoresis, proteins migrate in response to an electrical field through pores in a polyacrylamide gel matrix; pore size decreases with increasing acrylamide concentration. The combination of pore size and protein charge, size, and shape determines the migration rate of protein (Gallagher, 2008)**.** In this study, **s**odium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) was performed using a Mini- Protean III dual slab cell (Bio-Rad Laboratories, Watford, UK).

Gels preparation

The stacking gel was 4% and the resolving gel was 13 and 15%. All solutions were stored at 4 °C. The electrophoresis was performed in a Mini- Protean III dual slab cell (Bio-Rad Laboratories, Watford, UK). The gel mixtures were gently poured in the casting modules. After filling, the resolving gel (5 cm deep) was carefully overlaid with 1-2 mm deep layer of butanol to allow a truly flat surface and protect the top of the gel mixture from atmospheric O_2. After polymerization, the alcohol was replaced by the stacking gel (1 cm

deep). After polymerization, gels were ready to run. Detailed dosages of reagents are mentioned in the laboratory manual.

- ***Sample preparation***

Variation of the samples in protein content was the limiting factor for the electrophoresis work that is why many attempts were made to get good separation on the electrophoresis gel. Actually, we needed to concentrate the proteins by precipitation by trichloroacetic acid TCA 97% (0.45 ml of extract/0.06 ml of TCA). The mixture was left for 30 min on ice and then was centrifuged for 10 to 15 min at 15000g.

After throwing the liquid, we added ethanol 0.5ml and put in the freezer for 5-10min. and then we centrifuged again and threw the ethanol in excess. Samples were kept at room temperature for overnight then, when dry, 1x SDS sample buffer is added.

Loading quantities were as follows: 0.02 ml of protein marker, 0.04ml of all the samples except the some protein fraction ones which were loaded by 0.01 ml.

- ***Run conditions***

Electrophoresis was performed at room temperature using a voltage stepped procedure: voltage was kept constant (130 V) until the tracking dye reached the bottom of the gel.

- ***Fixing, staining and destaining***

Immediately after ending electrophoresis, gels were removed from the plates and placed in a staining solution containing 100mg of coomassie brilliant blue, 50ml of 95% ethanol and 100ml of 85% phosphoric acid, all diluted to 1l of distilled water. Destaining of gel was accomplished after 2h of immersing in a destaining buffer containing 40% methanol and 10% acetic acid

Amino acid analysis

The amino acid analysis in both camel milk and bovine milk were conducted using the procedure developed by the Department of Nutrition & health UAEU. The samples were then filtered using a Millipore membrane (0.45 μm) and injected in the Ultra Performance Liquid Chromatography system, UPLC (Waters® ACQUITY UPLC® system).

Protein Fractionation

Five (5) breeds of Emirati and two (2) Saudi breeds of milking camels have been selected. A total of 98 milk samples (both colustrum and raw camel milk) after parturition have been collected over a period of 10 months from Al Ain Dairy. Skimmed

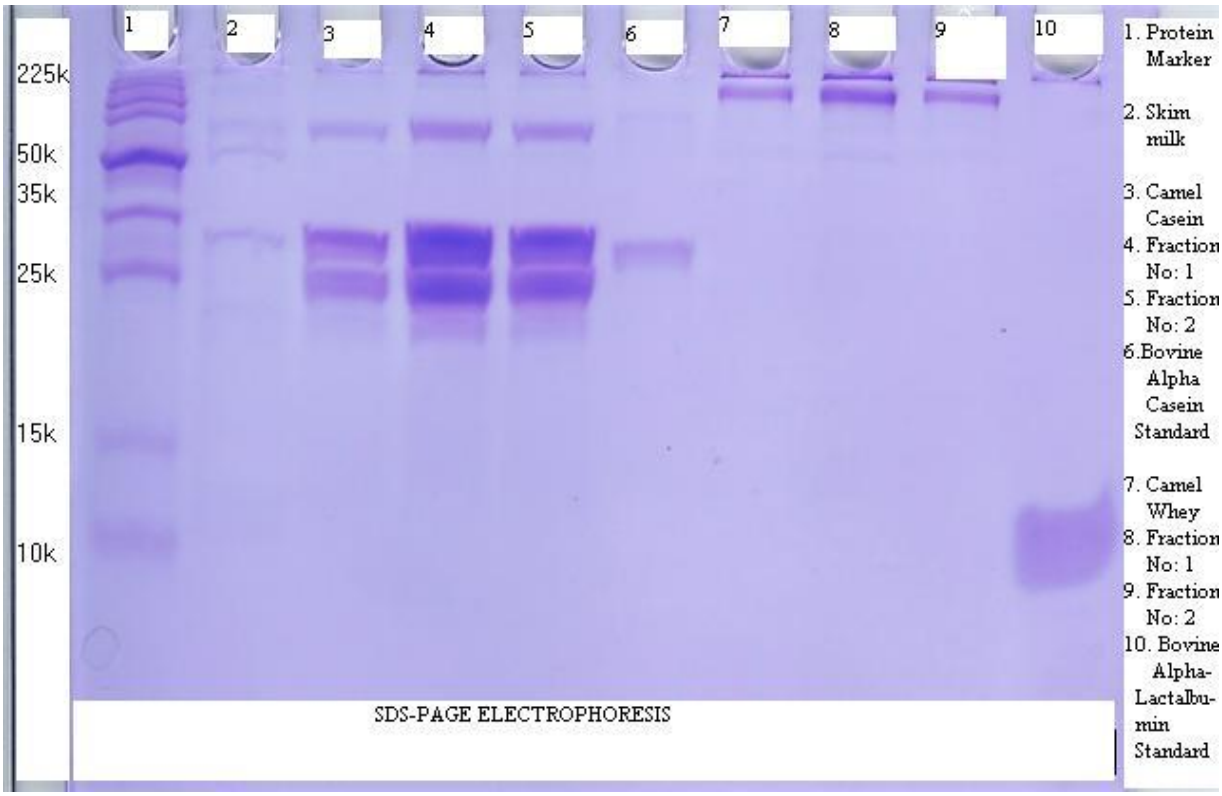
milk was prepared by centrifugation to remove the fat. The whole milk protein, mainly the casein was obtained by precipitation at pH 4.6, followed by centrifugation to obtain the supernatant of whey proteins. Both the casein proteins and the whey proteins were suspended in buffer. Each protein aliquot was subjected to chromatography on HPLC representing 196 samples being analyzed.

Preliminary fractions of proteins chromatograms were obtained and subjected to electrophoresis to determine their molecular weights and characteristics.

Results and Discussion

Electrophoresis

Various fractions of camel milk proteins (casein, whey proteins) and other unknown fractions were collected from HPLC and ran in electrophoresis gel. Unknown fractions 1 and 2 (collected from camel casein) corresponding to the well 4 & 5 showed low molecular weight of (~25 KD). However, the fractions obtained from camel whey proteins were not properly separated and identified.



Amino acids Composition

The amino acids composition of camel milk and bovine milk is shown on the below table. The amino acid composition of the species seems very heterogenous. The camel milk showed high amount of gluatamic acid (4 times), leucine (2 times) lysine (2 times), histidin (2 times) arginine (3 times) while the bovine milk is rich in phenylalanine and serine.

Overall, camel milk is particularly rich in essential amino acids, which explain the health benefit of camel milk for human nutrition.

Table 7 Comparison of amino acids composition of camel and bovine milk (Amino acid/100 g of milk)

	Camel milk	Bovine milk
Histidine	2. 9	1.4
Serine	2.2	5.9
Cystéine	0.3	-
Arginine	10.6	3.9
Glycine	2.2	2.3
Acide aspartique	10.2	10.7
Acide Glutamique	29.2	7.4
Thréonine	8	2.7
Alanine	3.1	5.1
Proline	17.8	14.0
Lysine	10	4.7
Tyrosine	0.9	0.3
Méthionine	3.7	1.1
Valine	10.3	8.1
Phenylalanine	4.1 7	7.9
Isoleucine	8.9	7.3
Leucine	31.3	19

Objective three:

To produce spray dried and freeze dried camel milk powder and determine the drying parameters and physico-chemical properties

ABSTRACT

In the present study, fresh raw camel milk was concentrated to 20 and 30% total solids. The camel milk concentrate was then dried using a pilot spray dryer, where different drying parameters were employed. The physicochemical characteristics of the various powders such as moisture content, water activity, color, Hausner ratio, angle of repose and insolubility index were compared between the camel milk powders and cow milk powders. Hausner results of camel milk powder 1 and camel milk powder 2 were found to have good flowability of 1.21 and 1.22 respectively. Insolubility index for all camel milk powders was found in a range of 0.3-0.5, with camel milk powder 1 and 2 were at 0.3 and 0.45; camel milk powders 3, 4 and 5 showing the least merits of insolubility. The physiochemical properties of the resultant camel milk powders illustrated that each functional property is dependent on some variables and drying parameters.

1. Introduction

Camel milk is consumed as a major staple food, mainly by the desert nomad tribes because it is one of the most readily available raw materials, which contains all the needful nutrients required in the dry conditions of the desert. Moreover, camel milk like any other human consumable milk consists of fat, proteins (soluble proteins and caseins) and one major carbohydrate (lactose) as major components (Farah & Fischer, 2004). It also contains minerals and vitamins as minor components, to abridge; camel milk can be titled as a nutritious source of all the required essentials of a complete diet, not just for the desert nomads, but also for general usage.

Milk is considered as a perishable food commodity and hence the same rule applies to camel milk, like any other milk origin. The first process adopted to extend the shelf life of camel milk was fermentation, by the manufacturing of fermented dairy beverages, soft cheese, etc (Farah & Fischer, 2004), but with the increase in milk production and consumption, spray drying can be applied to camel milk, in order to produce any milk powders (Nijidam & Langrish, 2005). The development of the processing technology in producing camel milk powder will prolong the shelf-life and to facilitate storage and handling. However, the heat processing conditions may affect the physicochemical and

functional properties (e.g. flowability, reconstitution properties, particle caking, Maillard and enzymatic reactions, emulsifying and foaming properties) of the powders (Thomas, Scher, Desobry-Banon, & Desobry, 2004). Other factors may affect the flowability of food powder such as the relative humidity, temperature, moisture content, particle size, etc (Kim, Chen, & Pearce, 2009a, 2009b, 2009c; Teunou & Fitzpatrick, 1999, Teunou, Fitzpatrick, & Synnott, 1999).

Spray drying process is not new and it is the most commercial method used for drying food and particularly milk. The process involves a very short time of heat contact and the high rate of evaporation give high quality product with relatively low cost compared to other drying processes (Jinapong, Suphantharika, & Jamnong, 2008; Liu, 1997; Schuck, 2002; Thomas, Scher, Desobry-Banon, & Desobry, 2004).

A dry powder product is highly desirable since it not only possesses long shelf life, but also requires relatively low transportation cost and storage capacity and the product can be used as food security for camel milk producer countries. Thus, a process for producing a dried camel milk powder that is soluble and without loss of nutritive value is highly desirable.

A spray drying system for cow milk powder has been characterized by various factors such as inlet air temperature, feed rate, atomizer speed, out let air temperature product temperature, thermal and evaporative efficiencies (Kim, Chen, & Pearce, 2009c; Perez–Munoz & Flores , 1997; Schuck, 2002). However, there is little report on the processing conditions of the production of camel milk powder.

Spray dry processing of camel milk powder is similar to the processing of cow milk powder, but the effects of drying on the physiochemical properties of both milk powders are different, but comparable (Maury & Murphy, 2005). Due to the difference in heat stability of camel milk proteins compared to bovine milk proteins, several researchers are now focusing on the functional properties of the camel milk proteins and heat coagulation of camel milk (Farah & Atkins, 1992; Laleye, Jobe & Wasesa, 2008). Camel milk powder can be used as food ingredients, mainly because of the functional properties of its proteins which are different from bovine milk proteins (Al-Saleh, 1996; El-Agamy, 2000; Laleye, Jobe & Wasesa, 2008; Wernery, 2006).

Milk powders obtained from cow milk are commonly being used today in a wide range of products such as baked goods, snacks, soups, chocolates and confectionary e.g., milk chocolate, ice cream, infant formulae, nutritional products for athletes, hospital use etc., recombined milks and other liquid beverages (Kim & Chen, 2005); therefore the purpose of this study is to investigate the effect of drying conditions on the physiochemical properties of camel milk powders and the potential of camel milk powder as food ingredients.

Materials and Methods

Materials

Raw, fresh cow milk and camel milk were supplied by Al Ain Dairy, Al Ain, UAE.

Methods

Evaporation

Fresh raw camel and cow milk was concentrated to 20 and 30% total solids by using a modified concentrator, i.e., the milk to be concentrated was filled up in the gourmet ice cream gelato equipment (Carpigrani's technology, pastomaster RTX, Italty) and was heated at 80°C with constant stirring, this equipment was sealed then attached to a rotary evaporator (Rotavapor RII, Buchi, Switzerland) in order to create a vacuum, where the evaporated water was collected in a round bottom flask, this excess water was measured using volumetric flasks to analyze the concentration of the milk.

Spray drying

Different camel milk concentrates (20 and 30% total solids) were dried using a spray dryer (FT 80 Tall, Spray Dryer, Armfield LTD., UK). The spray dried runs were cycled in two sets of different varying conditions, i.e., (a) drying set temperature (T_{SET}), % total solids and liquid feed volumetric flow rate (V_{LF}) were kept constant, but the direction of

feed was changed; (b) % total solids and liquid feed volumetric flow rate (V_{LF}) and the direction of feed were kept constant, but drying set temperature (T_{SET}) was varied. Moreover, for comparison cow and camel milk concentrates were cycled too, the process included liquid feed volumetric flow rate (V_{LF}), direction of feed and % total solids to be constant, with varying drying set temperatures (T_{SET}). The experiments are the mean of two repetitions using separate raw milk supplies with the same processing parameters as indicated above.

Water activity

Water activity of the different spray dried powders was measured using a water activity analyzer (Rotronic SW with hydrolyte VD sensor, Rotronic Instrument Corp., Huntington, NY)

Moisture content

Spray dried milk powders were dried in a vacuum oven at 70°C until constant weight was obtained, in order to calculate the moisture content (AOAC,1990).

Color

In this study, measurements of color parameters of milk powders were performed by using a MOM-color colorimeter (Hunter Lab) and the results were expressed in the CIE, L, a, b, where "L" value is an indication of lightness, "a" measures redness and "b" value is an indication of yellowness (AOAC, 1990).

Hausner ratio

Hausner ratio is the ratio of un-tapped bulk density and tapped bulk density. Un-tapped bulk density was determined by sifting milk powder into a 100 ml cylinder and then weighing. Tapped bulk density was determined by reading the volume after tapping the cylinder 100 times (AOAC, 1990).

Insolubility index

10 g of whole milk powder was mixed with 100 ml of water at 24°C in a mixer at high speed for 90 sec. The milk was then left for 15 minutes, after which it was stirred with a spatula. Then, 50 ml of the reconstituted milk was placed into a graduated centrifuge glass tube with conically graduated bottom. The tube was centrifuged for 5 min and then the sediment free liquid was drained off. Water addition process was repeated once more. The sediment was expressed in ml and is termed as insolubility index (AOAC, 1990; Supplee & Bellis, 1924).

Angle of repose

Angle of repose was determined by the fixed base method. Approximately 100 g powder was sifted over a funnel through which the powder flowed; the tip of the funnel was 11 cm from the base of the collection dish. The collection dish was 8.85 cm in diameter with side walls 1.40 cm tall. When the powder pile at its peak height, the height was measured, excluding the height of the dish. The peak height was the height of powder pile just prior to its collapse. To obtain the peak height, the powder pile was measured and more powder was added to the pile, alternatively. The last measurement before the powder pile collapsed was considered the peak height. Powder was not allowed to build up beyond the edges of the dish. The angle of repose of the powder pile was calculated using the radius and height of the pile (AOAC, 1990; Kim & Chen, 2005; Teunou, Fitzpatrick & Synnott, 1999).

Results and discussion

Effect of direction of feed on camel milk powders

The effect of change in direction of feed, co-current and counter current, on the physiochemical properties (water activity, flowability, color and in-solubility index) of camel milk powders obtained from the laboratory scale spray dryer is presented in Table 8.

Table 8 Spray-dried process parameters of camel milk powders. The experiments are the mean of two repetitions using separate raw milk supplies with the same processing parameters.

Experiments	T_{SET} ^0C	% Total solids	T_{INLET} ^0C	T_{OUTLET} ^0C	% Rh	Direction of feed
Powder 1	200	30	200-201	85-86	4.2-4.5	Co-current
Powder 2	210	30	209-212	89-91	4.2-4.9	Co-current
Powder 3	210	30	209-211	95-105	2.2-3.3	Counter current
Powder 4	210	20	209-212	84-90	4.2-5.8	Counter current
Powder 5	220	20	218-219	89-93	4.1-5.8	Counter current

In this study, two spray dried runs, i.e., Camel milk powders 2 and 3 were compared for analysis. Table 8 explains the details of the process for each run, such that for camel milk powder 2, was obtained by spraying the milk concentrate as very fine droplets by a nozzle into a hot dry air stream, where the direction of feed (milk concentrate) was co-current (same) to the direction of hot air, however for camel milk powder 3, the order was reversed, i.e., direction of feed was counter current (opposite) to the direction of hot air. Table 9 provided a compendium of the different physiochemical tests carried out for each run, which concluded that when the direction of feed is co-current, i.e., in direction with the hot dry air, % water activity is less, than when the direction of feed is counter current, even when the temperature is kept constant at 210°C, mainly because the contact of hot dry air with the feed is more thus resulting into more removal of water.

Color intensity for each run was measured using MOM-color colorimeter Hunter "L", "a", "b" (AOAC, 1990). The results of the colorimetric tests were mentioned in Table 2 and Figure 2, which explained the phenomenon of lightness and yellowness of milk

powders and its correlation, such that, camel milk powder 2 had a higher value of "L" (94.37), but a lower value of "b" as compared to camel milk powder 3, which clearly defined the co-relation between lightness and yellowness of milk powder, i.e., as the lightness of milk powders increased, there was a decrease in the yellowness or vice versa. An increase in yellowness as observed in camel milk powder 3; this could be a result of burning of feed during spray drying or Maillard reaction (Tonon & Brabet, 2008).

Table 9 Physico-chemical measurements of spray dried camel milk powders.

Experiments	% Moisture	% Water Activity	In-solubilty index	Angle of Repose °	Hausner Ratio	Color L, a, b
Powder 1	1.22	0.176	0.3	51.73	1.21	L = 96.73 a = – 0.77 b = 8.54
Powder 2	1.21	0.193	0.45	47.24	1.22	L = 94.37 a = 0.11 b = 11.05
Powder 3	1.24	0.204	0.5	49.12	1.27	L = 90.24 a = 2.93 b = 17.99
Powder 4	1.94	0.208	0.5	49.95	1.29	L = 94.03 a = 0.88 b = 13.32
Powder 5	1.20	0.21	0.45	47.37	1.3	L = 93.82 a = 1.06 b = 12.93

Solubility is an important feature in judging the physical characteristics of milk powder. The term solubility used herein refers to the ability of desiccated milk when mixed with water to form a solution, suspension or emulsion which will simulate the physical characteristics of natural milk, which is measured as in-solubility index (Fitzpatrick *et al*., 2007). Table 9 depicted that in-solubility index for both powders 2 and 3 was over the required limit of 0.2 ml, but even then camel 2 had a slightly lower in-solubility index than camel milk powder 3, due to less % moisture content (1.24%), induced by the increase in contact between the feed and the hot dry air. The camel milk powder 4 had higher moisture content than camel milk powders 1. 2 and 3 at 1.94% with high solubility index of 0.5. These data from camel milk powder 4 may affect the shelf life, during

storage; this is an agreement with the findings of Fitzpatrick *et al*., (2007) and Thomas, Scher, Desobry-Banon, & Desobry, (2004).

Powder flow properties are important in handling and processing operations. The flow functions for camel milk powder 2 and 3 were measured using the Hausner ratio and Angle of repose and the results of the experiments are presented in Table 9, which states that camel milk powder 2 had a good flowability with 1.22 (Hausner ratio) and 47.24° (Angle of repose), while camel milk powder 1 had 1.21 for Hausner ratio but 51.73 for the Angle of repose; whereas Hausner ratio for camel milk powder 3 was slightly above the desired limit of 1.25, thus resulting into poor flowability. Hausner ratio and angle of repose are not intrinsic properties of the powders, they mainly depend on the methodology used (Kim & Chen, 2005).

Effect of drying temperatures on camel milk powders

In this study, two spray dried runs, i.e., Camel milk powder 4 and 5 were compared for analysis. Table 8 explains the details of the process of the experiments and drying temperatures at 210 and 220°C. Water activity is a dimensionless quantity used to represent the energy status of the water in a system and is very temperature dependent (Fitzpatrick *et al.*, 2007). Table 9 explains the effect of change in the drying temperatures on the water activity of both camel milk powders, such that at 210°C, camel milk powder 4 had a water activity of 0.208% at a relative humidity (Rh) of 4.2-5.8%, which increased approximately by 0.01% for camel milk powder 5 at 220°C, thus verifying the fact that water activity is temperature dependent and the less the water activity the longer the shelf life (Thomas, Scher, Desobry-Banon, & Desobry, 2004). Temperature changes water activity due to changes in water binding, dissociation of water, solubility of solutes in water, or the state of the matrix. Although solubility of solutes can be a controlling factor, but control is usually from the state of the matrix. Since the state of the matrix (e.g. glassy vs. rubbery state) is dependent on temperature, one should not be surprised that temperature affects the water activity of the food (Fitzpatrick, 2007).

In this study the flowability of both camel milk powders was determined by (a) Hausner ratio and (b) Angle of repose. According to the results of Hausner ratio, mentioned in

Table 9, all camel milk powders 4 and 5 had a value greater than 1.25, indicating the merits of poor flowability. However, Nijdam & Langrish (2005) reported that milk powder produced in small scale had fine powders subjected to cohesion and their flowability is more difficult, therefore, particle size is one of the most important physical properties which affects the flowability of powders (Kim, Chen, & Pearce, 2009b; Teunou, Fitzpatrick, & Synnott, 1999), this parameter was not reported in this study, but this laboratory-scale spray dryer produces small particle size of milk powders (Nijdam & Langrish, 2005). Athough Hausner ratio is a number that is co-related to the flowability of a powder, but it is not an absolute property of a material, its value can vary depending on the methodology used to determine it (Kim & Chen, 2005). Angle of repose is a characteristic related to the inter-particulate friction or resistance to movement between particles (Kim & Chen, 2005; Teunou & Fitzpatrick, 1999). In comparison camel milk powder 5 (47.37°) dried at 220°C had a fairly good flowability than camel milk powder 4 (49.95°), due to high temperature, 220°C compared to 210°C, and low % moisture content (1.20%), thus resulting into less segregation of material and consolidation as the cone was formed, measured by the Angle of repose.

Temperature had a pronounced effect on the in-solubility index of milk powders, also depicted in Figure 6, such that an increase in temperature during evaporation or spray drying will cause (a) casein to denature, these denatured caseins form complex combinations of casein-whey-lactose, (b) increased temperature of the concentrate during evaporation will cause a pronounced age thickening resulting an increase in viscosity and bad atomization, thus contributing to an increase in temperature during drying (Teunou & Fitzpatrick, 1999), all these factors ultimately resulted in the increase of in-solubility index as observed in both the camel milk powders 4 (0. 5) and 5 (0.45). Other reasons than the ones mentioned above for increase values of in-solubility index could be the bad quality of milk with increased in lactic acid as it was also observed that powders with high lactose content such as baby foods would practically never get an increase in-solubility index as lactose protected the protein from denaturation (Kim, Chen, & Pearce, 2009a, 2009b, 2009c; Teunou & Fitzpatrick, 1999).

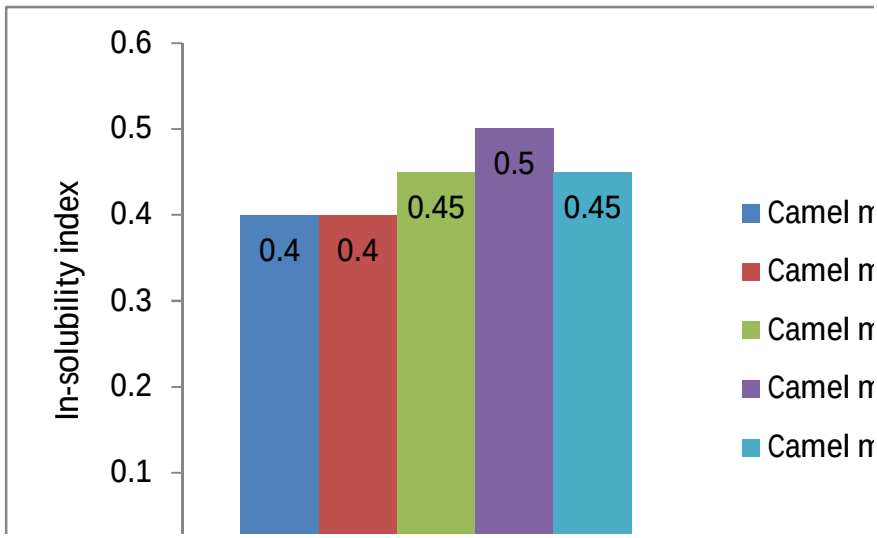


Figure 6 In-solubility index of various spray dried camel milk powders

Comparison between bovine and camel milk powders

When developing and identifying the best process conditions for the production of a spray dried camel milk powder it is usual that it is compared with bovine milk powder, as bovine milk spray dried powders are common ingredients in many food and dairy products. For this reason comparison between bovine and camel milk concentrates have been reported using a laboratory scale mini spray dryer, since the development experiments were routinely performed on a laboratory scale mini spray dryer (FT 80 Tall, Spray Dryer, Armfield LTD., UK). The following process conditions were kept constant: liquid feed volumetric flow rate (V_{LF}), direction of feed and % total solids, but drying set temperatures (T_{SET}) varied.

The effects of various spray drying conditions on the physiochemical properties, both primary powder properties (water activity, tapped bulk density, un-tapped bulk density, color) and secondary powder properties (in-solubility index, flowability) of the laboratory spray dried camel milk powder 4 and 5 along with bovine milk powder 1 and 2 milk were investigated.

As discussed earlier in section 3.2, that water activity is very temperature dependent; the same phenomenon is used here to explain the relation between water activity and temperature. Table 10 depicts the effect of change in temperature on the water activity of both bovine milk and camel milk powders, such that at 210°C bovine milk powder 1 had a water activity of 0.29% at a relative humidity (Rh) of 3.1-4.4%, which decreased approximately 0.035% for bovine milk powder 2 at 220°C, thus verifying the fact that water activity is temperature dependent. However, when the water activity for both bovine and camel milk powders were compared, it was observed that camel milk powders 4 and 5 at constant liquid feed volumetric flow rate (V_{LF}), direction of feed and % total solids had less water activity than bovine milk powders, thus promoting the idea of a longer shelf life (Fitzpatrick, *et al.*, 2007).

In this study the flowability of both camel and bovine milk powders was determined like earlier studies of section 3.1 and 3.2 by (a) Hausner ratio and (b) Angle of repose. According to the results of Hausner ratio, mentioned in Table 10, camel milk powders 3 to 5 had a value greater than 1.25, indicating the merits of poor flowability, but here it could also be rightly said that in comparison, camel milk powders had a better flowability than bovine milk powders, mainly because the values for both camel milk powders 4 and 5 were lower than bovine milk powders 1 and 2 at 1.47 and 1.48 respectively (Hausner ratio).

Table 10 Spray-dried process parameters of bovine and camel milk powders. The experiments are the mean of two repetitions using separate raw milk supplies with the same processing parameters.

Experiments	% Total Solids	T_{SET} °C	T_{INLET} °C	T_{OUTLET} °C	% Rh	Direction of feed
Bovine milk powder 1	20	210	209-212	88-90	3.1-4.4	Counter current
Bovine milk powder 2	20	220	218-221	92-104	2-4	Counter current
Camel milk powder 4	20	210	209-212	84-90	4.2-5.8	Counter current
Camel milk powder 5	20	220	218-219	89-93	4.1-5.8	Counter current

The extent of browning may also be linked to the lactose content of camel milk. It well known that camel milk contains more lactose than bovine milk (Farah & Fisher, 2004), suggesting that camel milk powder will tend to be more "browner". In milk powders the

“L” value indicating the lightness and the “b” value representing the yellowness are determined (Thomas, Scher, Desobry-Banon, & Desobry, 2004). From Table 11, it can be found that bovine milk powders had their “L” value higher than the camel milk powder, an average value of 95 compared to 94 respectively, but the “b” values are higher for camel milk powder compared to bovine milk. The phenomenon of higher trend of lightness "L" in both the bovine milk powder samples 1 and 2 could be due to the susceptibility of the cow proteins denaturation during concentration and the heat drying resulting into browning. Figure 7 describes the different merits of lightness for each camel milk powders 4 and 5, and also for both the spray dried bovine milk samples. The variability of the “L” values as shown in Figure 7 was due to the different parameters of the drying conditions of powders. This finding could support the complexity of the non enzymatic browning and that the tristimulus method may not be the more adapted one, as the initial color of milk powders can be very different. However, other authors performed colorimetric measurement parameters to calculate a browning index, which was found to be an adequate measure of non enzymatic browning reactions (Schebor, Buera, Karel & Chirife, 1999).

Table 11 Physico-chemical properties of bovine milk and camel milk powders. The experiments are the mean of two repetitions using separate raw milk supplies with the same processing parameters.

Experiments	% Moisture	% Water Activity	In-solubility index	Angle of repose°	Hausner Ratio	Color L, a, b
Cow milk powder 1	1.59	0.29	0.5	42.93	1.47	L = 95.60 a = – 0.58 b =10.54
Cow milk powder 2	2.15	0.255	0.8	50.75	1.48	L = 95.78 a = – 0.26 b = 10.29
Camel milk powder 4	1.94	0.208	0.5	49.95	1.29	L = 94.03 a = 0.88 b = 13.32
Camel milk powder 5	1.20	0.21	0.45	47.37	1.3	L = 93.82 a = 1.06 b = 12.93

As mentioned earlier in section 3.1 and 3.2, that the main reason for the increase in in-solubility index was the denaturation of casein and elevated temperature, this effect was observed in bovine milk powder 2. Table 11 showed that this trend was also demonstred

in the other three samples, i.e., bovine milk powder 1 (0.5), camel milk powder 4 (0.5) and camel milk powder 5 (0.45), but in comparison both the bovine milk powders had a higher in-solubility index than camel milk powder samples, at the same conditions, thus reducing the ability of formation of solution, suspension or emulsion which would stimulate the physical characteristics of natural milk. This relative high insolubility obtained for the bovine milk powders could be explained by the "high" denaturation of proteins in particular the whey proteins compared to camel milk proteins (El-Agamy, 2000; Laleye, Jobe & Wasesa, 2008) and thus the insolubility would increase during the storage which would be affected by the moisture content of the bovine milk powders at 1.59 and 2.15% respectively for bovine milk powder1 and 2. This is in agreement with the findings of Fitzpatrick *et al.*, (2007).

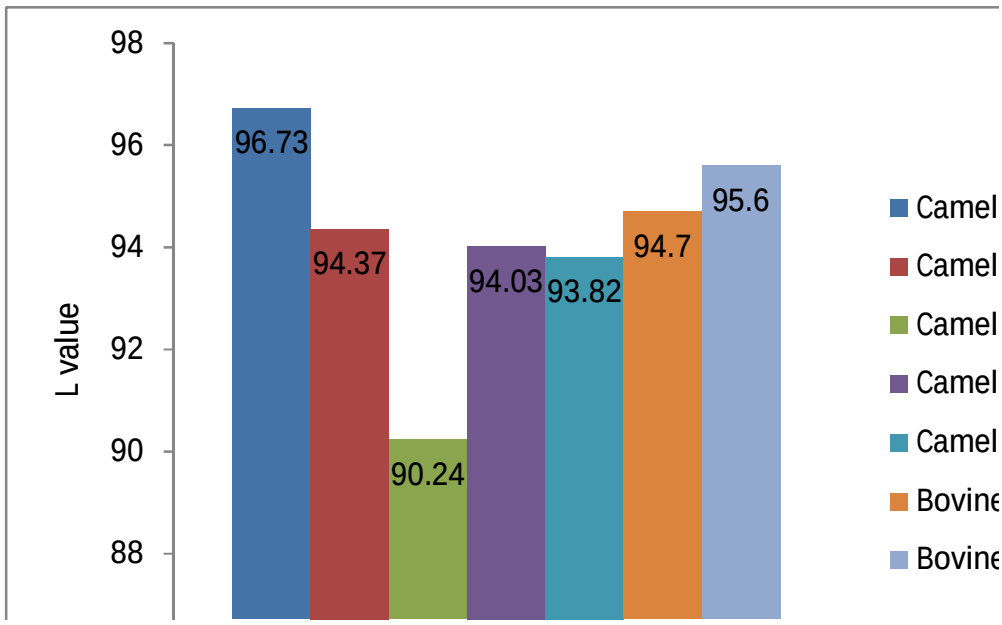


Figure 7 Color (lightness) measurements of various spray dried milk powders

Conclusions

This work investigated the effects of spray drying on the physiochemical properties, both primary powder properties (water activity, tapped bulk density, un-tapped bulk density, color) and secondary powder properties (in-solubility index, flowability) of the laboratory spray dried camel milk powders. Moreover, for comparison bovine and camel milk powders were also reported. The dominant factor affecting the primary and secondary properties of the milk powders was the high temperature and the direction of milk concentrate (feed).

With an increase in temperature, a decrease in % water activity was observed for each of the five camel spray dried samples to as low as 0.176% (Camel milk powder 1), thus indicating the possibility of a longer shelf life. It was also observed that an increase in temperature resulted in higher values of in-solubility index mainly due to the denaturation of proteins; this irreversible change could limit the usage of these camel milk powders in the form of re-combined milk. The flowability of all the camel milk powders were below 50°, thus promoting a fairly good flow, however this trend was also observed in bovine spray dried powders, but only in bovine milk powder 1 (42.93) which was dried that a $210^{\circ}C$, slightly lower than bovine milk powder 2. Color of each spray dried camel milk powders was lighter than yellow.

Direction of milk concentrate (feed) was also significant and resulted into change in physiochemical properties if the direction of feed was co-current or counter current. Experiments carried out concluded that a laboratory scale spray-dryer demonstrated the use of the counter current mode of feeding where the feed and the direction of hot dry air are in opposite direction so as to avoid burning when the direction of feed and hot dry air is co-current.

Moreover, in comparison camel milk powders showed a lower water activity, in-solubility index, yellowness, angle of repose than bovine milk powders, thus suggesting the prospects of usage of camel milk powders in snacks, chocolates, ice cream, infant formulae but surely further insight investigations are required to validate the prospects of camel milk powder on an industrial scale.

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Objective Four:

To develop a new processing cheese making technology from camel milk. This objective consists of three sub objectives:

- Selection and development of appropriate starter culture in order to acidify the camel milk in a short period of time and to prepare the milk for coagulation

- To test various commercial chymosin enzymes as well as the development of alternatives sources of enzymes for camel milk coagulation such as plant enzymes, chymosin extracted from the abomassa of young and old camels.

- The development of an appropriate cheese making process technology

1) Selection and development of appropriate starter culture in order to acidy the camel milk in a short period of time and to prepare the milk for coagulation

2) To test various commercial chymosin enzymes as well as the development of alternatives sources of enzymes for camel milk coagulation such as plant enzymes, chymosin extracted from the abomassa of young and old camels.

Work accomplished:

Camel Milk Cheese Methodology

Before experimenting on making cheese from camel milk, cheese was made on a trial basis, using cow milk, but all the protocols required were followed as mentioned in the literature, this was carried out so, in order to know the creditability of the process and the different disciplines indulged in the process. After the successful trial, the cheese that was made, had a structure in between cottage cheese and cheddar cheese, it was called in between the two cheese categories, because it neither possessed the complete characteristics of cottage cheese nor cheddar cheese, to abridge it was a combination of both, such that it's structure was neither too soft like cottage cheese nor firm like cheddar cheese, it was more of a slightly firm structure, with a rubbery taste. The cheese made was portioned into two, one portion was vacuum packed (99.9%) and the other was immersed in a 22% brine solution. This process of cheese making was carried out again, but successful results were not obtained, i.e., the second trial produced results till coagulation, but while cooking, the pH didn't drop as it was required, so it was discarded. However, the methodology adopted (to date) for the processing of camel milk cheese is discussed as follows:

A- Processing of Camel Milk Cheese using the following materials:

- **0.02% Calcium Chloride**
- **20ml/10L RST 744**
- **1.5/10L CHYMAX M (liquid)**
- **0.25g Coagulant stick (rennet)**

Results:

Observations concluded as follows:

- Starter culture was unable to drop the pH from 5.9.
- Coagulation started after a time frame of 3 hours.
- The coagulum was very fragile and settled at the bottom of the cheese vat, leaving the milky whey to float at the top.
- Ever since the coagulation started, the state of coagulum didn't change, it remained fragile as it was in the starting of the process.
- Ultimately, after 4 hours of coagulation period, no profound coagulation was obtained.

B- Processing of Camel Milk Cheese using the following:

- **0.03% Calcium Chloride**
- **30ml/10L RST 744 + R704**
- **1.5g/10L CHYMAX (powder)**

Results:

Observations concluded as follows:

- Starter culture was unable to drop the pH from 5.9.
- Coagulation started after a time frame of 3 hours.
- The coagulum was very fragile and settled at the bottom of the cheese vat, leaving the milky whey to float at the top.
- Ever since the coagulation started, the state of coagulum didn't change, it remained fragile as it was in the starting of the process.
- Ultimately, after 5 hours of coagulation period, no profound coagulation was obtained.

C- Culture Activity Test of R704 and RST 744 (blend)

- Starter culture was prepared by emptying the sachets, i.e., RST 744 and R704 in 1L of distilled water.

Results:

Observations concluded as follows:

- pH dropped from 6.50 to 5.44, depicting the activity of the starter culture.
- Lastly, since the calculated titratable acidity was 0.468%, this also depicts that the starter culture is active, since the starter culture with a titratable acidity of over 0.34% is regarded as active, according to the literature.

D- Determination of the optimum concentration of calcium chloride and kappa carrageenan individually for the coagulation of camel milk.

Results:

All the samples depicted fragile coagulation for each concentration of Calcium Chloride and κ-Carrageenan, but in comparison to Calcium Chloride, κ-Carrageenan, produced better results

E- Determination of coagulation time using camel milk in combination with freeze dried camel milk powder, skimmed cow milk powder and full fat cow milk powder.

Results:

The observations concluded as follows:

- Best results in regards to coagulation time and coagulum firmness, in ascending order, it was camel milk + 6% freeze dried camel milk powder then camel milk + 6% skimmed cow milk powder, but best results were produced by camel milk + 6% full fat cow milk powder.

F- Determination of coagulation time of different concentrations of Camifloc 3, using the following:

- **Camel milk,**
- **6% skimmed cow milk powder,**
- **Calcium chloride and**
- **Kappa carrageenan.**

Results:

The observations concluded as follows:

- Best results with respect to coagulation and coagulum firmness was produced by Beaker C and Beaker D, both the contents in the two beakers coagulated after 45minutes and depicted a firm coagulation, quite similar to the reference beaker, whose content percentages were used earlier to make cheese.

§ The results of this experiment concludes that, like earlier cheese made from 25% Camifloc 3 (object 11) can be made, similar in texture mainly, by using exactly the same proportions of all the contents for e.g., camel milk, 6% skimmed cow milk, 0.05% calcium chloride, 0.02% kappa carrageenan, but instead of using 25% Camifloc 3, either 15% or 20% Camifloc 3, can be used to produce same results.

Development of the use of plant as an alternative to chymosin

Summary of the research work

Background

Cheese making involves a number of main stages that are common to most types of cheese. The basic principle step implicated in making all natural cheese is to coagulate or curdle the milk so that it goes into curds and whey. Milk coagulation, the first step in cheese-making, begins when *Chymosin* splits κ-casein into two parts: Para-κ-casein and glycol-macro-peptide CMP. Chymosin has been, for a long time, the conventional milk-clotting enzyme obtained from the fourth stomach of calves and the most widely used coagulant cheese manufacturing all over the world. With no doubt, this enzyme has proved its efficiency with milk of reference: bovine milk but also has shown greater specificity for it than for the other species especially camel milk.

Most attempts to make cheese from camel milk have revealed major difficulties in getting the milk to coagulate. Studies comparing effect of rennet on both of camel and cow milk led to that, with the same amount of *Chymosin*, clotting time for camel milk is two to three folds that of cow's milk and desired coagulation time is consequently achieved by higher concentration. The rennet also causes the milk to curdle in small flakes without a firm coagulation. This inconveniency is due not only to the non specific interaction of the protease with camel kappa-caseins which gives the bovine *Chymosin* a very low catalytic rate for their proteolysis, but also to the low ratio of this type of caseins to whole caseins comparing with cow milk ratio, estimated at 3.5% in camel milk against 13% for cow milk. Over again, the poor rennetability may be generated by the difference in the size distribution of casein micelles in both milks (El Agamy, 2006).

As the Enzymes traditionally used in the manufacture of cheese generally are *Chymosin* and pepsin, the history of milk clotting was performed using the complex enzymes extracted from stomach of animals that can be either goat, lamb or cow and more recently camel. But the main contribution in the supply of animal enzymes does not exclude the use of plant as fig tree, cardoon and artichoke, pineapple, papaya, silk tree, etc (Roseiro *et*

al., 2003). With their specific enzymes, these plants also provide ways of variable actions that generate parallel individual tastes.

The main purpose of this study is to find alternative plant enzymes to *Chymosin* able of coagulating camel milk and having consequently a good yield and better quality of the camel milk cheese. In this report, we will begin by a literature review presenting camel milk, its composition and its particularity. Then the methodology of the project will be treated and finally discussion and interpretations will take part.

Major findings

The main purpose of this study is to find a substitute of *Chymosin* which is capable of coagulating camel milk and improving camel cheese quality.

To achieve this goal, the study was divided into 3 parts. As a preliminary step, the choice of the enzymes studied is of a particular interest. As mentioned before, it was based on their availability and also because they are known for their clotting and proteolytic activity with cow milk. 3 plants; *C. procera*, *S. dubium* and *C. scolymus* were extracted with different buffers at different pH.

Calotropis procera leaves and latex were tested for their proteolytic and coagulant activities and an extra sample was added to see the efficiency of the dialysis. Results showed that latex extract are more active than leaves and the dialysis step is too important especially when ultra-filtration is the next step noting that it removes all the rubber present in the latex solution.

Besides, S. dubium expressed a high clotting activity towards cow milk but any activity for camel one. Regarding the proteolytic activity, mainly, all the enzymes tested proved higher proteolytic activities for camel milk than for bovine one even for *S. dubium.* When compared to Chymosin, all the enzymes seems to have higher proteolytic and coagulant activities In fact, clotting time was greater with Chymosin even if this latter is concentrated and supposed to clot camel milk rapidly.

During partial purification, we noticed that both the specific clotting and specific proteolytic activities are increasing. Proteolysis increase is probably du e to the concentration of non specific proteins after ultrafiltration. This is can be confirmed by the electrophoresis work that gathered all fractions issued from the purification. These fractions were run into a gel where ultra-filtrated fractions generally presented a halo in the lane indicating not only the concentration of the solution but also the migration of bands according the molecular weight of the ultra-filtration tubes used.

The second part of the project studied the effect of the enzyme concentration, temperature and pH on clotting time. *C. procera*, *C. scolymus* and *Chymosin* have proved an overall clotting activity towards camel milk and demonstrated an activity in a wide range of pH (5-6.8). Moreover, plant enzymes established high thermo-stability at 70°C for Calotropis and 65°C for Cynara unlike Chymosin that was denaturized at 45°C. Concerning effect of the concentration, the plant enzymes were able to clot camel milk even at very low level of concentration. Calotropis procera extract has a very high coagulant activity that is why the extract was diluted to 10 times. C. scolymus was put by 0.5% while Calotropis was put by 0.05%.

According to these parameters (concentration and temperature), we proceeded by the manufacture of laboratory scale cheeses and then characterized the curd and whey which consists in the third part of the study.

Approximate analysis of the whey revealed that Chymosin registered the biggest loss of the protein fractions and ash content followed by C. procera whose whey is rich with soluble nitrogen other nitrogenous compounds. However, *C. scolymus* curd presented a slight higher mineralization than C. procera. These significant differences in the loss of the constituents are closely related to

On one hand the rheological parameters of the curd and on the other hand the microstructure of the gel formed. Obviously, Chymosin generated the weakest gel microstructure that is why the majority of the components are drained in the whey. SEM analysis can confirm this interpretation by taking a look at the internal gel microstructure,

the most porous and fragile network belongs to chymosin. On the contrary, C. scolymus generated the firmer gel with a consistent network.

C. procera showed also high porosity into the gel structure with non homogenous and non consistant network affirming the incidence of the high proteolytic activity on the excessive hydrolysis of the curd proteins leading by the way to the loss of the amino acids; Valine and then nutritional value.

Texture profile analysis of the curds proved that the C. procera curd is chewier than Chymosin and C. scolymus at the same time more adhesive than *C. scolymus*.

During draining, *C. scolymus* revealed a lower syneresis capacity and also the lowest cheese yield but previous results affirm that the extra weight measured is probably due to the high moisture content which was confirmed by assessment of the dray matter content.

As a conclusion, Calotropis procera and Cynara scolymus proved insuring a greater specificity towards camel milk and better quality of camel curd with preference for Cynara scolymus as alternative to *Chymosin* because excessive proteolytic activity of Calotropis is a limiting factor in cheese processing.

Development of the use of chymosin extracted from camels' abomassum

Summary of the research work

Comparative study of milk clotting activity of crude gastric enzymes extracted from camels' abomasum at different ages and commercial enzymes (rennet and pepsin) on bovine and camel milk

Abstract

Coagulation of camel milk for the production of cheese has been proved to be difficult by the use of the available commercial rennets, chymosin and pepsin. Therefore this study focused on the use of crude gastric enzymes extracted from the abomasum of camels (*Camelus dromedarius*) at different ages (1, 3 and 9 years old). The non-purified gastric enzyme extracts from camels (GEC) at different ages were characterized for their protein content, clotting and proteolytic activities and were also compared with those of bovine rennet and pepsin. The conditions of milk clotting by the GEC were optimized at different pHs 5.8, 6.0, 6.3, 6.6 and temperatures 30, 37 and 42°C. The flocculation time of bovine and camel milk by all the enzymes studied were reported. The data showed that the GEC from the older camels gave the best results significantly ($P \leq 0.05$) for both milk clotting activity and flocculation time of both bovine and camel milk compared with the other tested enzyme preparations. The optimum flocculation time was obtained at pH 5.8 and 42°C for the camel milk and at pH 6.0 and 37°C for bovine milk.

Major findings

The crude gastric enzyme preparations from camels (GEC) obtained from the older camels showed better coagulation activity in both milks. Flocculation time data showed that the GECs and bovine pepsin had good specificity towards bovine casein and camel casein. In addition, the short flocculation time obtained for GEC 9 (older camels) at an optimum temperature of 42°C and a pH of 5.8 thus encouraging the fact that older camels are more available for slaughter in Algeria. Therefore the production of GEC from older camels could be an excellent substitute for the commercial chymosin for cheese making using either bovine or camel milk.

This study focused primarily on the coagulation step on making cheese curd that represents a key step in cheese making. It is recommended that additional research be conducted to purify the extract, to characterize the extract using electrophoreses and finally for the production of various types of cheeses from camel milk.

Objective Five:

To determine and characterize the functional properties of camel milk proteins isolates in relation with their molecular structural changes and to study the thermal transition characteristics and water sorption behavior of these isolates.

Thermal Characteristics of Different Components of Camel Milk

Introduction

Milk defined as the normal secretion of the mammary glands of mammals. Milk is the nature designed food for the young and adults. It's a complex mixture that supplies human with carbohydrate (lactose), fat, protein, calcium, essential minerals and vitamins. Milk plays an important role in the dietary intake as it helps to improve bone and dental health and possibly protect against hypertension and colon cancer (Haug et al, 2007), for these benefits human being consumes milk from different animal species such as cow, goat and camel. In many industrialized countries, raw milk is processed before it is consumed. During processing the fat content of the milk is adjusted, various vitamins are added, and potentially harmful bacteria are killed. In addition to being consumed as a beverage, milk is also used to make butter, cream, yogurt, cheese, and a variety of other products when milk production increased (O'Connor, 1995).

MILK PRODUCTION

Milk production is low and seasonal and it fluctuates according to pasture accessibility, feed quality and the proportion of lactating cows (Debrah *et al.*, 1995; Sall, 2003). According to the recent statistics by the Food and Agriculture Organization (FAO), the world cow's milk production in 2008 stood at over 578 million tonnes, with the top ten producing countries accounting for 55.4% of production. The USA is the largest cow's milk producer in the world accounting for 14.9% of world production, producing over 86 million tonnes in 2008, an increase of 2.4% when compared to 2007. India is the second largest cow's milk producer, accounting for 7.6% of world production and producing over 44 million tonnes in 2008. The UK is the 9th largest producer in the world producing over 13 million tonnes in 2008 and accounting for 2.4% of world cow's milk production. (FAO, 2008)

NUTRITIONAL BENEFITS OF MILK

Chamberlain (1990) reported that the most important nutrients in milk to human were proteins, calcium, potassium, phosphorous, other trace elements and vitamins such as A, and B complex. The nutritional value of milk for infants was very clear as it was usually the chief source of complete protein, calcium, vitamins, essential fatty acids and energy for the rapidly developing child (Gamall, 1999). Chamberlain (1990) stated that the whole milk was particularly important for babies less than a year old if breast milk is not available also milk for children is a very important weaning food. O'connor (1995) found that the chief function of lactose in milk was to supply energy for muscular activity and maintenance of body temperature, besides it has certain therapeutic properties and ability to enhance the intestinal absorption of calcium and phosphorus.

MILK POWDER (DRIED MILK)

Rosenthal (1991) reported that the milk powder is a product of lower water activity and better keeping qualities; which can be produced in large scale in modern plants. The powder produced can be stored for long period of time without significant deterioration of taste or nutritive value and can be dried into skim milk powder to obtain a shelf life of about two years, while whole milk powder can be stored for only six months.

According to Thompson (1996) that there were three types of milk powder which are whole milk powder, skim milk powder and partially skimmed milk powder. Whole milk powder is a soluble powder made by spray drying of fresh whole milk, and no other drying ingredient comes as close to the composition of fresh milk as whole milk powder. It was usually obtained by removing water from pasteurized, homogenized whole milk (USDEC, 2006). Skim milk powder obtained by means of whole milk and can be found in two forms, regular and instant, but both are made from milk by a spray drying process, both types have the same nutrient composition, but the regular type was found to be more compact and required less storage space than the instant type, however, it is more difficult to reconstitute. On the other hand, the partially skimmed milk powder is a powder product of milk originally obtained by means of skimming, concentration and drying of milk.

USES OF POWDERED MILK

Milk powders contribute nutritionally, functionally and economically to a variety of food formulations including bakery, confectionery, dairy, recombined milk, meat, nutritional beverages, and prepared foods. Milk powders provide many functional benefits as food ingredients. The major components in milk powder (proteins, lactose and milk fat) affect the way in which milk powders perform and their suitability for each type of application.
In most drying conditions a significant amount of the dried product remains in an amorphous state, mainly due to the insufficient time for crystallization to occur at the given drying conditions. Depending upon the rate of drying, the dried product obtained can also constitute some crystalline material. This will be however, influenced by the processing conditions, composition and property of the individual ingredients present (Roos, 1995)

Thermal Properties of Milk Powder

There is a large quantity and variety of materials produced industrially in powder form and there is a need for information about their handling and processing characteristics. A variety of those contain amorphous (metastable) materials that, for the most part, are matrices of carbohydrates and/or proteins. Examples include: skim/whole milk powders, coffee, instant soups/toppings, coffee whiteners, and cereals, to name a few. The storage stability of such systems is affected by the storage conditions (temperature, relative vapor pressure [RVP], oxygen availability), but it is also dependent on their composition (White *et al*, 1966; Peleg and Mannhei, 1977&Roos, 1995). The quality of milk powders during storage may also be decreased by fat oxidation (Shimada *et al*., 1991) and nonenzymatic browning (Saltmarch *et al*., 1981). Changes in the amorphous state lead to various time-dependent structural transformations such as stickiness, collapse (loss of structure, decreased volume), and crystallization during processing and storage (Schucka *et al*., 2007& Roos, 1990). Dehydrated sugar-rich products are particularly sensitive because of their high hygroscopicity and tendency to crystallize (Chirife *et al.*, 1973; Tsourouflis *et al.*, 1976; To & Flink, 1978).

Thermal properties aid in product process control, the prediction of storage characteristics and in alimentation. Differential Scanning Calorimetry (DSC) is used widely to characterize biological materials. Typical observed transitions include the glass transition of the amorphous phase, melting and crystallization processes, denaturation, free and bound water, onset of oxidation, and heat capacity.(Roos,2002 & Jouppila and Roos 1994).

The milk composition of dairy animals has been widely studied throughout the world and thousands of references are available especially with regard to milk consumed by humans. The literature data mainly concerns cow milk, which represents 85% of the milk consumed in the world and, to a lesser extent, goat and sheep milk. Studies on other dairy animals (buffalo, yak, mare, and camel) are rather scarce, in spite of their nutritional interest and medicinal properties. In addition, unlike other milk-producing animals, camels can thrive under extreme hostile conditions of temperature, drought, and lack of pasture, and still produce milk (Yagil and Etzion, 1980). For that in this context, thermal characteristic of camel milk and cow milk need to be further investigated in order to have more information about the biological value of heat-treated camel milk.

MATERIALS & METHODS

Milk Samples

Pasteurized camel milk was provided kindly by Royal Court Affairs (Barka - Oman) whereas the cow milk was purchased from local super market in Muscat – Oman. Both milks were stored at -60° until used for the experiments. Milk powder was produced at the dairy plant of Sultan Qaboos University, Muscat.

THERMAL ANALYSES

Sample Preparation

- **Preparation of Cream**

Four hundred ml of camel milk was centrifuged at 5000 rpm for 30 min at 4ºC. The top layer (cream) was removed manually from the supernatant and was divided into two equal halves. One half was freeze dried and the other half kept for fat analysis**.** The supernatant (skimmed milk) was kept for protein analysis **(**KARRAY *et al*, 2004).

- **Preparation of Fat**

Fat was extracted from cream by heating the cream at 25ºC. One liter of water was added and stirred well. The solution was then centrifuged at 5000 rpm at 4ºC for 30 min **(**KARRAY *et al*, 2004).

- **Preparation of Casein Protein**

10% acetic acid (v/v) was added to the skimmed milk and allowed to stand for 30 min at 35ºC. Then, 10% 1M Sodium acetate (NaOAc) was added to the acidified milk; pH adjusted to 4.3 with 6 M HCl. The mixture was allowed to stand for 30 min and then centrifuged at 20,000 rpm at 5ºC for 30 min. The supernatant (the liquid whey) was decanted in a separate beaker and kept for further analysis. The precipitate (the casein) was washed twice with distilled water (pH adjusted to 6.8) to remove any whey residue in the casein sediment. After every washing step, the casein is obtained by centrifugation at 5000rpm for 30 min at 10 ◦C. the collected supernatant from the washing steps was returned to the liquid whey supernatant collected previously. The washed casein was then freeze dried and stored at –20 °C. (Wangoha, 1998)

- **Preparation of Whey Protein**

Whey protein obtained by acid precipitation of caseins at pH 4.6, was split into two halves. One half was treated with ammonium sulphate (60% saturation) to precipitate the whey. The sample was kept in the fridge (4ºC) over night and then centrifuged at 10,000 rpm at 15ºC for 15 min. The supernatant was discarded and the sediment was collected, freeze-dried and stored at -20^{o}C.

Ethanol 2.5/1.0 (ethanol/supernatant) was added at 25ºC to 40% whey supernatant to make final concentration of 70% to precipitate the whey protein. It was centrifuged immediately at 5000 rpm for 5 min at 25ºC. The supernatant was removed and whey protein was separated. Collected whey protein was freeze dried.

- **Isolation of Lactose**

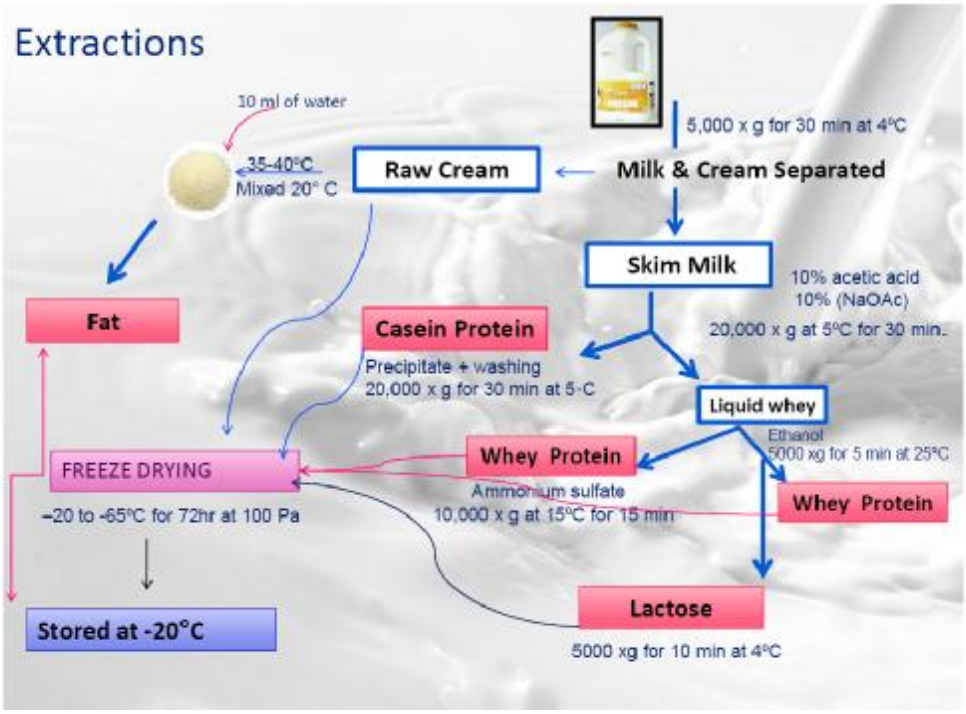
Lactose was crystallized by keeping the supernatant in the fridge overnight. It was centrifuged at 5000 rpm for 10 min at 4ºC. Manually remove the supernatant from the lactose. Finally freeze dried the lactose and kept at -20ºC (Bund, 2007).

Ø Preparation of model systems

The amorphous systems were obtained by freeze drying solutions containing 11 g/100 g of the different model systems. Aliquots of 1mL of the solution were placed in 5mL capacity vials, frozen 24 h at –26ºC, immersed in liquid nitrogen and freeze-dried. The freeze drying process lasted 48 h. A Heto–Holten A/S, cooling trap model CT110 freeze-dryer (Heto Lab Equipment, Denmark) was used which operated at –110ºC and at a chamber pressure of 4×10 4 mbar (Fernáandez, 2003). The freeze-dried samples were transferred into desiccators and kept at 26ºC over saturated salt solution that provided constant 11% of relative humidity (RH) (Greenspan, 1977).

Determination of total water content

The water content of the humidified samples was determined (in duplicate samples) by difference in weight before and after drying in a vacuum oven at 70ºC during 48 h in the presence of desiccant.



Differential scanning calorimetry (DSC and modulated DSC)

Differential scanning calorimetry with and without modulation (DSC Q10, MDSC Q1000, TA Instruments, New Castle, Delware) measured the glass transition of freeze camel milk samples. Details of operation and calibration procedure of the DSC and MDSC instruments were obtained by followed other papers (Rahman, 2004, 2007) with manner change in the procedure. About 5–10 mg of powder sample was weighed into a 50 µl DSC aluminium pan and press sealed with a lid using a DSC sample press. Thermal scanning of the samples were carried out using and sealed empty pan as a reference in four steps: (1) isothermal at 25 ºC for 1 min; (2) heating from 20 to 5 ºC at 10 ºC/min; (3) cooling from 5 to -90 ºC at 5 ºC/min; and (4) heating from 10 to 200 ºC at 10 ºC/min. Expect lactose was done at twice run with small change on heating from 20 to 155±250 ºC at 10 ºC/5min .The Tg values were analyzed from the second heat scanned curve using the DSC software at onset, midpoint and endset points. The analysis was carried out in triplicate. In the case of MDSC, samples were scanned from 10 to 200ºC at a constant rate within 3– 10 ºC/min with a modulation of ±0.50 ºC amplitude and 40 s period of modulation. Thermograms were analyzed from its total, reversible and non-reversible heat flow. The average values and standard deviations of 3–6 replicates were obtained

Results & Discussion

Freeze-Dried Whole Milk Powders (Cow and Camel)

Initially thermal characteristics of cow and camel milk (i.e. whole and skimmed) were performed before comparing the characteristics of separated components of camel milk. Figures 8 shows DSC thermograms of freeze-dried whole cow and camel milk powders. The thermogram for cow milk shows three endothermic peaks (marked as A_1, A_2 and B in Figure 8) and three shifts two at low temperature (marked as G_1 and G_2) and another one (marked as C) before non- fat solids melting.

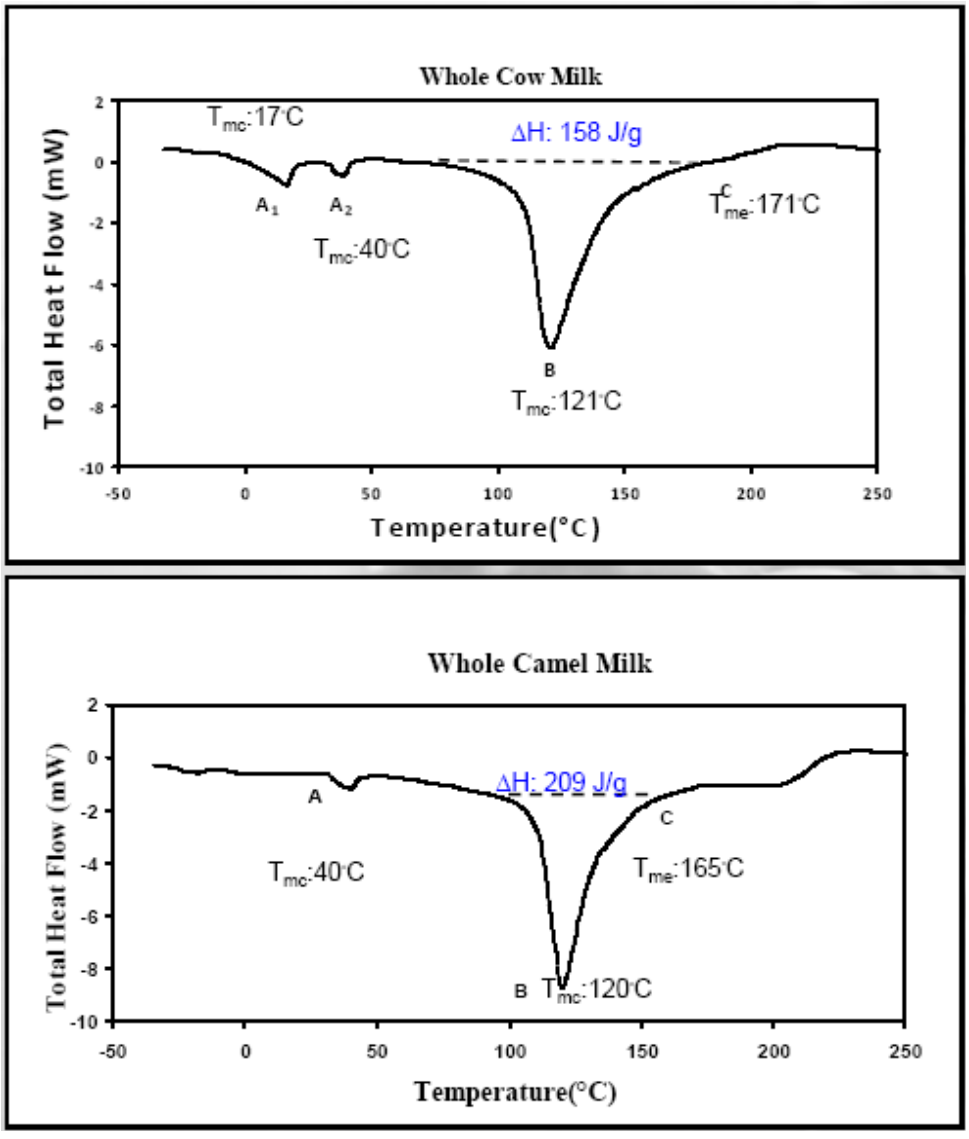


Figure 8 DSC thermogram of whole Camel and Cow milk

The two peaks at low temperature were due to the melting of fat (as it was evident later from the DSC thermogram for pure fat) and the larger peaks indicated the melting of non-fat solids in dried milk. It is important to know the melting point of fat since milk storage above melting could initiate undesirable agglomeration and caking Adhikari *et al.*, 2001). The shift at C indicated the formation of more ordered structure before melting of solids. Similar characteristics were also observed in the case of whole camel milk powder except higher temperature shift (marked as C) observed after melting of solids (Figure 8). Two shifts at low temperature were also observed in the cases of whole cow and camel milk, this could be the glass transitions. However it was difficult to identify which components in the milk was providing these low temperature transitions. It was difficult to trace the glass transition of different components of the whole or skimmed milk powder due to its complex interactions between the components. This behavior was also reported in the literature. Jouppila and Roos (1994) reported that glass transition of whole cow milk powder was 62.0°C stored at 11.5% relative humidity (RH) and they reported that it was difficult to trace using DSC curves for fat containing milk powders stored within RH 23.9 to 44.4%. They argued that the dominant fat melting endotherm in the same temperature range could be the reason.

Fernandez *et al.* (2003) measured the onset glass transition temperature at 61.0°C for whole cow milk powder stored at 11 % relative humidity, which was close to those obtained for lactose alone. They also agreed with Jouppila and Roos (1994) that whole cow milk products showed a fat melting endotherm that hindered to trace the glass transition. For this reason, milk powders were previously defatted before thermal analysis (Jouppila and Roos, 1994; Fernandez *et al.*, 2003; Silalai *et al.*, 2010). However the thermal characteristics of different components of camel milk were not reported in the literature.

Freeze-Dried Skimmed Milk Powders (Cow and Camel)

Figures 9 shows typical DSC thermograms of freeze-dried skim cow and camel milk powders. The thermogram for skim cow milk showed similar thermograms as whole milk, except the shift at higher temperature moved after solids melting. However in the

case of camel milk, the melting peaks for fat did not observe. The second glass transition for cow milk was observed as 54°C. Jouppila and Roos (1994) determined experimental glass transition of skim cow milk powder decreased as water content increased which started &om 58.0oe at 11.5% RH. They found that experimental glass transition of skim cow milk powder stored at 85.8% RH could not be determined due to rapid crystallization of lactose.

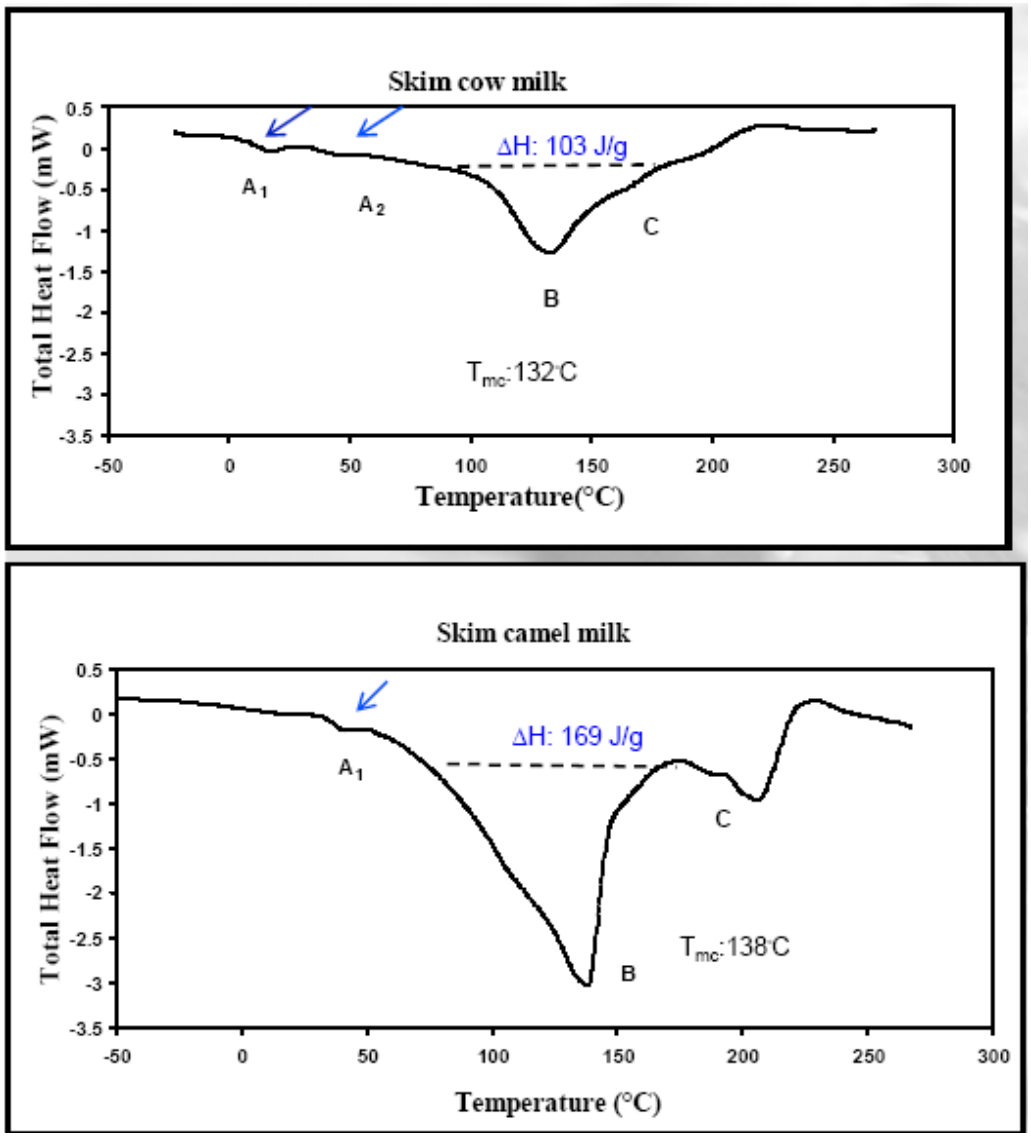


Figure 9 DSC thermogram of skim Camel and Cow milk

Fernandez *et al.* (2003) measured the onset value of glass transition temperature of skim cow milk powder at 11 % relative humidity was 62°C, which was slightly higher than whole milk powder. Fitzpatrick *et al.* (2007) observed glass transition properties of skim cow milk powder stored at 76.0% relative humidity and 200e atmosphere (20°C) for various exposure times. They found lowering and finally disappearance of the glass transition temperature with storage time. In addition the change in specific heat at the transition decreased and disappeared. They argued that this could be due to the crystallization of lactose over time. Furthermore, Hu *et al.* (2009) measured the DSC thermograms of dried cow milk powders with different levels of fat content (0.1-35 g/100 g milk) and found that there was no trace of glass transition or melting of fat. However, melting endotherms were observed with higher fat levels with increasing enthalpy as a function of fat content.

Thermal Transition of Fat and Cream of Camel Milk

Figures 10 and 11 show typical DSC thermograms of fat and cream of camel milk powders. The thermogram for camel milk fat shows two endothermic peaks (marked as Bl and B2 in Figure 4.5). The wider peak at low temperature was due to the melting of different fractions of fatty acid and the sharper peak indicated the melting of a specific fatty acid)it higher contents in the milk. Similar characteristics were also observed in case of camel milk cream except on wider endothermic peak show overlapping of several endothermic peaks (marked as B1 and B2 in Figure 4.6). The melting of fat in cream started at lower temperature -12°C as compared to pure fat at -5 °C. This decrease in melting temperature could be due to the effects of protein content in cream.

Karray *et al.* (2004) also observed similar characteristics of one wider peak and another sharp peak at higher temperature. They pointed that the wider endothermic peak showed overlapping of several endothermic peaks for different fatty acids or firmly bind between fat and protein in the case of protein. Table 4.3 shows characteristic melting temperatures and change in enthalpy (ΔH) for fat and cream of camel milk powder. The melting of fat started at -5°C and ended at 52°C.

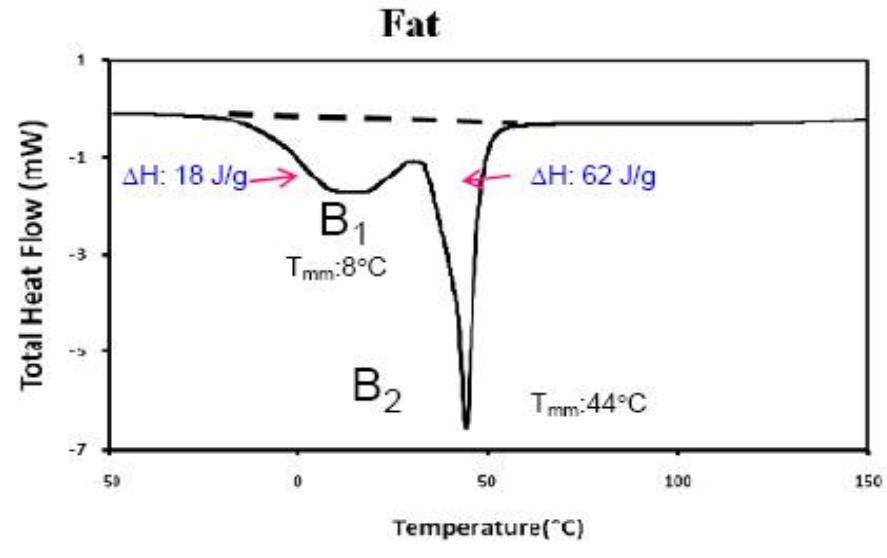


Figure 10 DSC thermogram of Camel milk fat

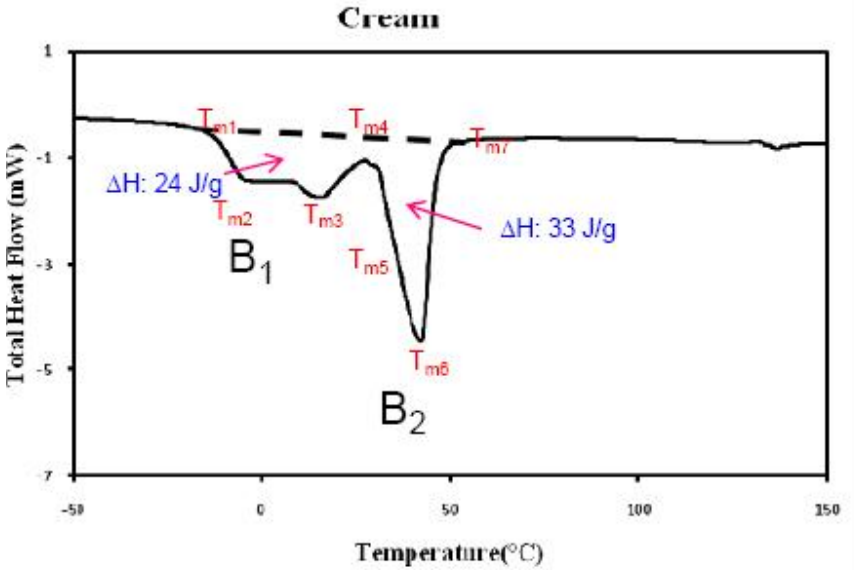


Figure 11 DSC thermogram of Camel milk cream

Ruegg and Farah (1991) found that melting of camel milk fat started around -26°C and completed at about 43°C; and the average heat of fusion was 79.2 J/g. Attia *et al.* (2000) found that final melting occurs at about 44°C for camel milk fat, whereas 39°C was observed for bovine milk fat. The different range of fat melting could be due to the different compositions of fatty acids (Ruegg and Farah, 1991; Attia *et al.*, 2000).

Lopez *et al.* (2005) stated that milk fat was mainly composed of triacylglycerols, which vary in chain length and degree of saturation was responsible for the broad melting range of milk fat that spans from about -40 to 40°C. Moreover, the polymorphism (identified are α, β', and β in increasing order of stability) associated with each triacylglycerols increased the complexity of the structural and thermal behaviors of milk fat (Karray *et al.* 2004; 2005). In addition cooling rate 3°C/min produced two endotherms as compared to one endotherm at cooling rate 0.1°C/min (Lopez *et al.*, 2005). The melting of fat in cream started at lower temperature as compared to the pure fat. This lowing could be due to the effects of protein content in the cream.

Thermal Transition of Casein Protein of Camel Milk

Figure 12 shows DSC thermograms of freeze dried of casein precipitated from camel milk. It shows one endothermic peak (marked as B in Figure 12) and two shifts (marked as G_1 and G_2), one just before melting of casein (marked as G_2). The endothermic peak was due to the melting of casein and the first shift at G_1) indicated first glass transition temperature and G_2 is the second glass transition temperature.

Figure 12 shows thermal transitions of casein including two glass transition temperatures, specific heat changes at the transitions and melting temperature with its enthalpy. The first glass transition started at 38°C, second glass transition started at 77°C and melting started at 95°C, respectively.

Hernandez *et al.* (2011) found the onset glass transition temperature of commercial β-casein equilibrated at 11 % RH (X_w: 3.3) as 112°C. However they did not observe the first glass transition as observed in this study. Similar value as 100°C was also observed for the β-casein at 5.0% (wet basis) moisture content (Mizuno *et al.*, 1999). Bengoechea *et al.* (2007) presented mid glass transition as 140°C for casein at 5% (wet basis) moisture content. The lower value of the second glass transition in this study could due to the source and types of casein. Bengoechea *et al.* (2007) observed 3 transitions as -64, -9 and 50°C when measured by DMTA (X_w: 12.9%, wet basis).

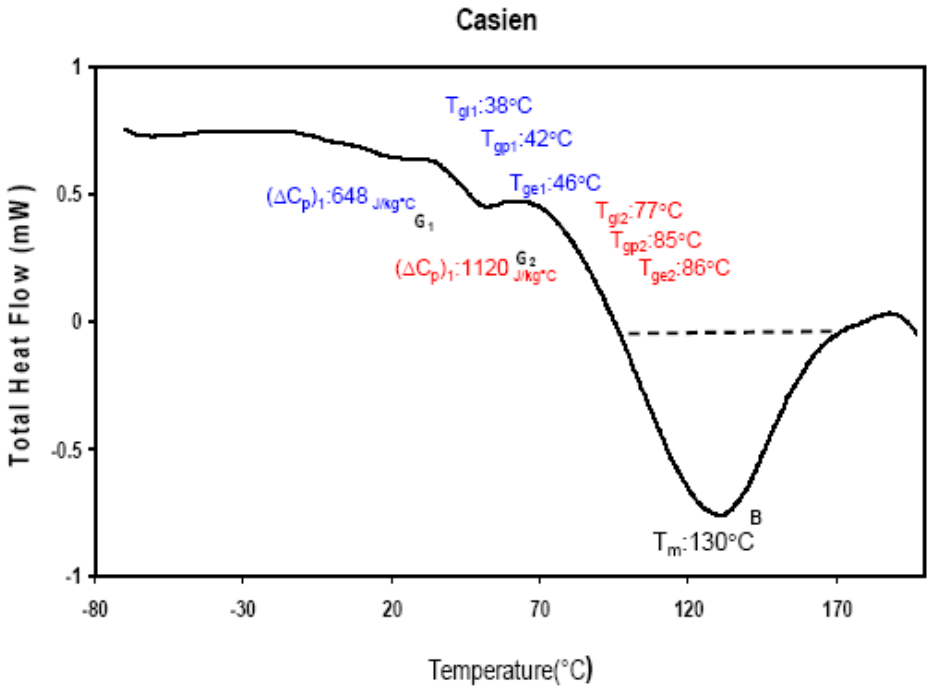


Figure 12 DSC thermogram of Camel milk casien

Rahman (2003) pointed that the two transitions occurred could be due to the backbone of large polymer or less mobile component and the other due to the less mobile or side chains. Mizuno *et al.* (2000) determined that the state of .the secondary structure (i.e. β-structure) of a protein was the key determining factor for its glass transition.

Thermal Transition of Whey Protein of Camel Milk

Figures 13 and 14 show DSC thermograms of freeze dried of whey protein (ammonium sulfate) and whey protein (ethanol) of camel milk powders. The thermogram for whey protein (ammonium sulfate) of camel milk shows one endothermic 'peak (marked as B in Figure 13) and two shifts one at low temperature (marked as G_1) and another one just before solids melting endotherm (marked as G_2).

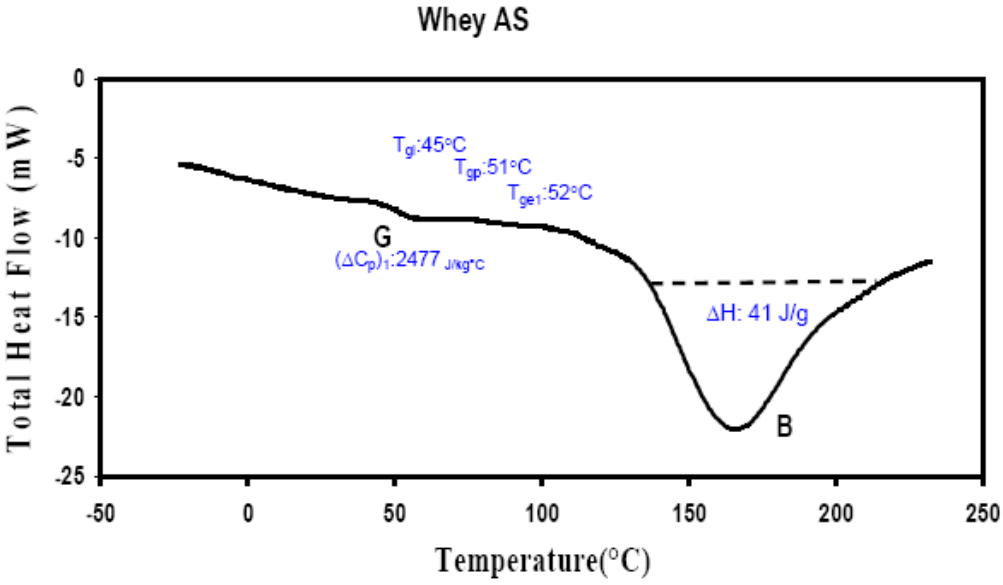


Figure 13 DSC thermogram of Camel milk whey extracted by Ammonium Sulfate precipitation

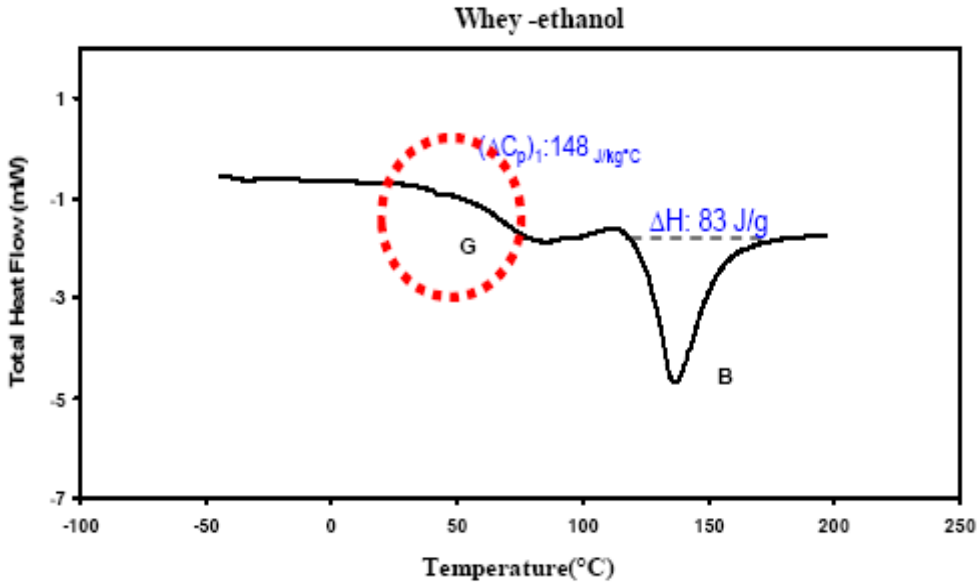


Figure 14 DSC thermogram of Camel milk whey extracted by ethanol precipitation

One endothermic peak at high temperature was due to the melting of whey protein and the shift at G_1 indicated first glass transition. In the case of ethanol extracted whey protein, the first glass transition started at -43°C, second glass transition started at 39°C and melting started at 67°C, respectively. However in the case of ammonium sulfate precipitated whey protein first glass transition started at 47°C, second one at 95°C and

melting started at 121°C, respectively. It was observed that ethanol precipitation lowered the first glass transition, which may be due to the denaturation of whey protein (Marshall, 1982).

In addition, ethanol precipitated whey protein decreased the onset of melting at 67°C with higher enthalpy 80 kJ/kg as compared to ammonium sulfate precipitated one at 121^{o}C and 41 kJ/kg, respectively. Zhou and Labuza (2007) measured glass transition of three commercial whey proteins (whey protein isolate (WPI), beta-lactoglobulin (BLG) and whey protein hydrolysate (WPH). In the case of equilibrated sample at 11.0% relative humidity, they did not trace the glass transition whereas WPH showed glass transition at 99°C (mid-point). Jara *et al.* (2009) measured the glass transition of dry commercial whey protein concentrate and observed onset glass transition at 48°C and end at 110°C, respectively.

Thermal Transition of Lactose from Camel milk and Commercial Lactose

First scan of camel milk lactose showed two endothermic melting peaks and did not show any trace of glass transition. For this reason, samples were annealed at 140°C for 0.1 minutes followed by a second scan. Second scan showed two glass transitions (marked as G_1 and G_2) and two endothermic melting peaks (Figure 15).

DSC thermograms of commercial lactose as presented in Figure 16 shows two endothermic peaks without any trace of glass transition. For this reasons, second scan was performed after annealing. However annealing at different temperatures (130, 140 and 150°C) for 0.1 to 5 minutes did not show any trace of glass transition. Figure 17 shows thermogram of commercial lactose with annealing at 150°C for 1 minutes. The two endothermic peaks at high temperature were due to the melting of lactose (as α and β forms) (Jouppila and Roos, 1994). Omar and Roos (2007) measured the onset glass transition of a-lactose equilibrated at 11.6% RH (X_w: 3.1%) as 62.2°C by DSC method. Similarly Shrestha *et al.* (2007) presented the mid as 82.2°C for the spray dried lactose equilibrated sample at 11.2% RH. The first glass transition value observed in this study was close to the reported literature values as mentioned above.

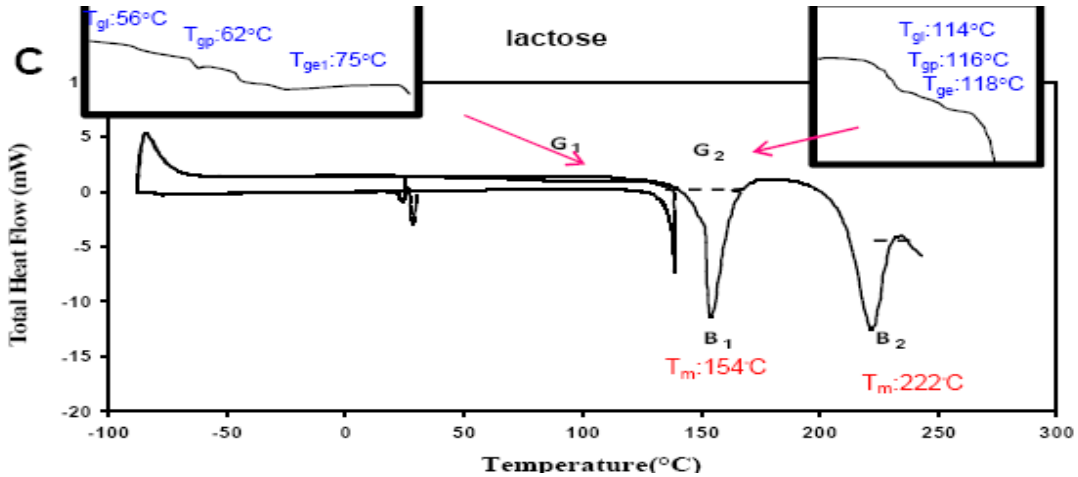


Figure 15 DSC thermogram of Camel milk lactose

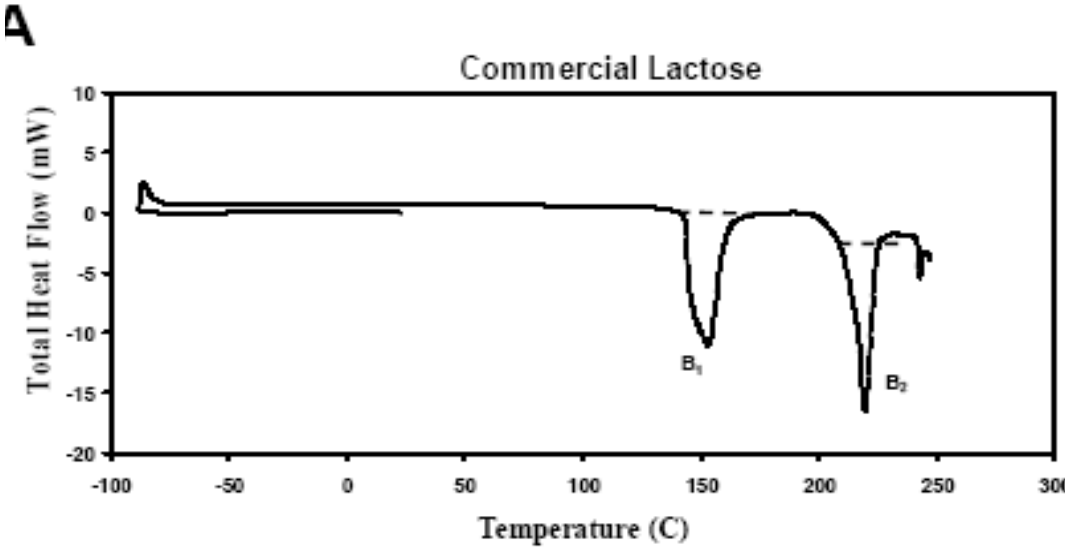


Figure 16 DSC thermogram of commercial lactose

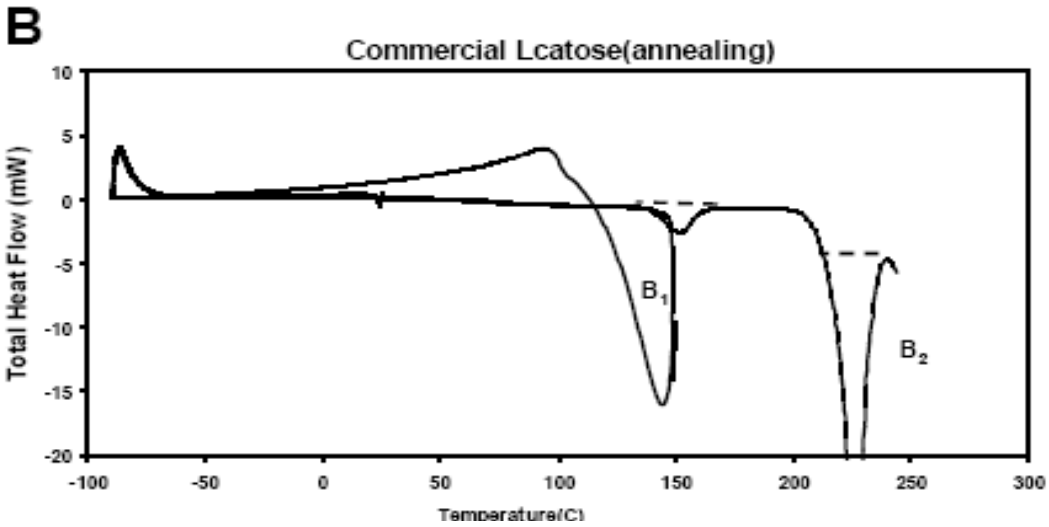


Figure 17 DSC thermogram of commercial lactose with annealing

The low temperature shift (G_1: -16°C) in the whole camel milk could be due to the glass transition of whey protein as extracted by ethanol (i.e. -43°C), the second shift (G_2: 31°C) could be due to the ftrst glass transition of whey protein extracted by ammonium sulfate (i.e. 47°C), second glass transition of whey protein extracted by ethanol (i.e. 39°C), and first glass transition of casein (i.e. 38°C). Two endothermic peaks in the whole camel milk at low temperature (i.e. 17 and 41°C) were related with the melting of fat, which were 8 and 44°C, respectively as shown in Figure 4.5. The wider endothermic peak at higher temperature (i.e. within l35-210°C, and peak 149°C) was due to the melting of casein (i.e. BO°C), whey proteins (114 and 150°C), and lactose (154 and 222°C). It was difficult to trace the melting of each solid component due to overlapping each other. The higher temperature shift (marked as C in Figure 8) could be due to the interactions of solids components after melting. This type of comparison between components and whole milk could provide relevant information on the interactions of components in the whole milk.

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Objective Six:

To incorporate camel milk powder in food formulations such as ice cream as fat replacer, flavour enhancer, texturizer, emulsifier and stabilizer and to conduct the sensory evaluation of the new formulated ice cream products.

Status:

- Camel milk powder was produced.
- The UAEU Department of Food Science does not have a milk separator to produce the cream
- Fresh raw camel milk was transported to SQU's pilot plant to produce cream from camel milk, however, the cream crystallized during the transportation to UAEU Food Science Department
- Additional work is required to conduct this objective

Appendix A:

LIST OF RESULTING PUBLICATIONS AND PRESENTATION AT CONFERENCE

[1] **Louis C. Laleye, Osama El Amin and Hina Kamel.** Effect of Drying Conditions on the Physiochemical Properties of Camel Milk Powders 11th Annual Research Conference, UAEU, Al-Ain, 26-28 April, 2010.

[2] **Louis C. Laleye, Baboucar Jobe and Abdulkadar Wasesa**. Heat stability and functionality of Camel milk proteins 2nd ISOCARD Conference, Djerba, Tunis, March 11-14, 2009

[3] **Louis C. Laleye, Baboucar Jobe, Abdulkadar Wasesa and Salman Ashraf**. Comparative study of the characterization of camel and bovine milk proteins 10th Annual Research Conference, UAEU, Al-Ain, 13-16 April, 2009

[4] **Louis C. Laleye, Baboucar Jobe and Abdulkadar Wasesa.** Comparative study on heat stability and functionality of camel and cow's milk whey proteins. Journal of Dairy Science, 91:1-8, 2008.

[5] **Louis C. Laleye, Baboucar Jobe and Abdulkadar Wasesa**. Heat stability and functional properties of Camel milk whey proteins. 9th Annual Research Conference, UAEU, Al-Ain, 21-23 April, 2008**.**

[6] **Louis C. Laleye, Osama El Amin and Hina Kamel.** Effect of Drying Conditions on the Physiochemical Properties of Camel Milk Powders International Dairy Journal (Submitted)

APPENDIX B: Dissemination of Information:

Characterization of Camel Milk Protein Isolates as Nutraceutical and Functional Ingredients

Collaborative Research SQU/UAEU
CL/SQU-UAEU/01/08
Dr. Ahmed Al-Alawi, Department of Food Science & Nutrition, SQU
Dr. Louis Laleye, Department of Food Science, UAEU

Introduction

There are over 300,000 milking camels in both Oman and UAE with an annual production of 38,000 MT of milk in UAE (UAE, Municipality of Agriculture and Fishery, 2005). The production of camel milk has significantly increased during the last few years with new pasteurized fresh camel milk in the super market.

The potential application and the use of camel milk proteins, camel milk protein isolates and camel whey powder as functional foods and nutraceutical ingredients have not been established.

Table 1 – Bioactive peptides derived from milk proteins (Cow) *

Bioactive peptides	Protein precursors	Bioactivity
1. Casomorphins	1. α$_1$, β-Casein	1. Opioid agonist
2. α-Lactorphin	2. α-Lactalbumin	2. Opioid agonist
3. β-Lactorphin	3. β-Lactoglobulin	3. Opioid antagonist
4. Casokinins	4. α$_1$, β-Casein	4. ACE-inhibitory**
5. Immunopeptides	5. α$_1$, β-Casein	5. Immunomodulatory
6. Lactoferricin	6. Lactoferrin	6. Antimicrobial
7. Phosphopeptides	7. α$_1$, β-Casein	7. Mineral binding

* H. Meisel, 1997

** ACE: Angiotensin I-Converting Enzyme

Camel milk is somehow different from cow milk in its chemical composition but it contains all essential nutrients as cow milk (Elagamy, 1988). In average, camel milk contains more proteins and whey protein than cow milk (Farah, Z. 1993; Walstra et al., 1999).

Table 2 – Approximate composition of camel, cow and goat milk (%)

Animal	Casein	Fat	Lactose	Albumin	Ash	Water
Camel	3.5	3.0	5.5	1.7	1.5	87
Cow	3.0	3.75	4.75	0.4	0.75	87
Goat	3.3	6.0	4.6	0.7	0.84	87

Casein fractions have some common features but are unique to be heterogenous with bovine casein. The balance between the different casein fractions is very different, however, and chiefly identified by a low amount of kappa casein of only about 5% of the total casein, compared with about 15-16 % in bovine casein (Ramet, J. P. 2001). This major difference in kappa casein content has shown difficulties in cheese making (Mohamed, M. A. 1995; Laleye, L. et al., 2005, unpublished data).

Table 3 – Protein fractions in milk

Casein	Whey Protein	Immunoglobulins
1. α$_{s1}$- Casein	1. β-Lactoglobulin	
2. α$_{s2}$- Casein	2. α-Lactalbumin	Lactoferrin
3. β- Casein	3. Serum Albumin	
4. κ – Casein		Proteose-Peptones
5. γ - Casein		

In an attempt to compare the functional properties of camel milk proteins with those of cow milk proteins, these proteins have been separated and characterized (Beg et al., 1987); it was found as important thermodynamic property related to the heat stability. The camel milk whey proteins were found to be considerably more heat stable than cow's milk (Farah and Atkins, 1992). However, this heat stability has not been investigated in term of functional properties such as gelification properties of camel milk proteins.

Our current research on the functional properties of camel milk proteins and camel whey protein demonstrated that using Differential Scanning Calorimeter, unlike the case in cow whey, the denaturation peak appeared at a much higher temperature (110°C). The appearance of the peak at this high temperature indicated that camel milk whey is more stable than cow milk. Laleye et al. 2006 (unpublished data). Functional properties such as thermal stability (Allain, et al, 1999; Subirade et al, 1998), emulsifying (Lefevre and Subirade, 2003; Lefevre and Subirade, 2001; Lefevre and Subirade, 2000; Dufour et al, 1999) gelling (Lefevre et al, 2003; Remondetto et al, 2004; Remondetto and Subirade, 2003; Remondetto et al, 2002; Line et al, 2005; Gilbert et al, 2003; Lefevre and Subirade, 2000; Allain et al, 1999; Subirade et al, 1998) and foaming properties have been thoroughly studied and reported on cow milk whey proteins. However, there is dearth of report on camel milk casein proteins and whey proteins.

In addition, very little information is available on thermal transition characteristics and water sorption behavior of camel milk protein isolates. Water activity has traditionally been used to describe microbial and chemical stability of food products. During the last two decades, attempts have also been made to apply glass transition concept to explain the physical, chemical and microbial stability of food, bioactive compounds and pharmaceutical products.

In the glassy state, reactions that depend on molecular diffusion, such as chemical and enzymatic changes, are reduced significantly. Despite of the research efforts, the significance and the mechanism of the glass transition as an indicator of chemical and biochemical stability is still not clear. Due to the interrelation among factors (i.e. temperature, water activity and pH) influencing chemical reactions, it is difficult to interpret the results.

A few studies have compared both criteria (i.e. water activity and glass transition) from the viewpoint of product stability. Information on stability of bioactive compounds in food materials such as fruits, vegetables, and dairy products and neutraceuticals formulations as influenced by water activity and glass transition is scarce. One of the objectives of this study will be to generate glass transition and water sorption data of camel milk protein isolates. Storage stability studies with such protein isolates may also help to increase our understanding of molecular mobility. This would enhance the knowledge of the necessary requirements for proper storage conditions and chemically stable formulations

More recently, Agrawal et al. (2005, 2007) have reported a unique camel milk health benefit in diabetic patients. These researchers have demonstrated that using camel milk as an adjunct to insulin therapy has improved the long-term glycemic control and led to reduction in doses of insulin in patient with type-1 diabetes. The isolation and the elucidation of the functional and biochemical properties of Camel milk proteins have enormous potential practical applications in nutraceuticals for food and in pharmaceutical fields.

Objectives

- **Overall Objectives:**
  To elucidate the high valued functional and nutraceutical properties of camel milk proteins
- **Specific Objective – Scientific (Basic):**
  1. To characterize the low molecular weight peptides present in camel milk proteins and identify the major components.
  2. To determine and characterize the functional properties of camel milk protein isolates in relation with their molecular structural changes and to study the thermal transition characteristics and water sorption behavior of these isolates;
  3. To determine the percentage content of the amino acids in the major proteins present in camel milk and correlate that with sequence of insulin.
- **Specific Objective – Applied**
  1. To produce spray dried and freeze dried camel milk powder and determine the drying parameters and physico-chemical properties;
  2. To develop a new processing cheese making technology from camel milk.
  3. To incorporate camel milk powder in food formulations such as ice cream as fat replacer, flavour enhancer, texturizer, emulsifier and stabilizer and to conduct the sensory evaluation of the new formulated ice cream products(s)

Practical Significance of Outcomes:

1. This is an exciting project, and a new field of applied research as it related to camel milk protein isolates. The use of cow milk proteins as food ingredients has been a growing field of research and beneficial to the dairy industry.
2. The current project addresses the isolation of camel milk proteins and the production of whey proteins to be used as food ingredients in food formulations (macro-application, i.e. functional ingredients) and the isolation of low weight molecular peptides to understand their function (micro-application, i.e., nutraceutical, pharmaceutical), disease prevention or curing especially diabetes which has abnormally high incidence rate in UAE and Oman populations.
3. From the industrial point of view, this project is important to find application of the use of camel milk for the dairy industry and in particular, the development of a process for the production of camel whey powder. Camel milk proteins and camel whey powder as food ingredients can be used in producing camel milk chocolate, and the formulation of new dairy products based on the unique functional properties of camel milk proteins.

Experimental Approach

UAEU

جامعة الإمارات العربية المتحدة
United Arab Emirates University

Development of Cappuccino Camel Milk Powder

The Issue

It has been said that camel milk is the "white gold" of the desert. UAE is a camel milk producing country with more than 300,000 milking camels producing annually 40,000 MT of camel milk in UAE .
The production of camel milk has been steadily increasing during the last few years.
The issue is that there no new usage of the potentiality of camel milk. Its highly foaming properties can be used in Cappuccino Coffee

The field research conducted by UAEU

The Department of Food Science, Faculty of Food and Agriculture, has developed a proprietary processing technology to produce Camel milk Cappuccino powder.
The consumer sensory survey has shown that the Cappucinno Camel milk powder had a similar sensory properties when compared with a regular Cappuccino coffee. Regular consumers of Cappuccino (males and Females) liked Camel milk Cappuccino. The product acceptance can be linked to more stable foam and more creamer taste.

Benefit to Abu Dhabi and the Nation

The research will help the camel milk farmers and the UAE Dairy Industry to find new applications and new products development from camel milk.

Investing in Knowledge for the Nation's Future -

Research and Education at UAEU

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UAEU

جامعة الإمارات العربية المتحدة
United Arab Emirates University

Camel Milk
Treating Diabetes

The Issue

It has been said that camel milk is the "white gold" of the desert. Camel milk is widely used in the UAE and in the Middle East. Studies have shown that Camel milk has several therapeutic benefits.

The issue is there is little or no research on the functional properties and the bioactivities of camel milk proteins.

The field research conducted by UAEU

The Department of Food Science, Faculty of Food and Agriculture, is currently isolating and testing various peptides derived from camel milk proteins.
Preliminary study with rats fed with camel milk has shown that camel milk significantly reduced blood glucose level in diabetic compared to untreated diabetic rats. Glucose handling was more efficient in treated diabetic group compared to untreated diabetics.
The number of insulin-producing cells increased significantly in the treated diabetic group. In conclusion, Camel milk reduced the signs of diabetes by increasing the number of insulin-positive cells.

Benefit to Abu Dhabi and the Nation

The research will prove the health significance of camel milk in treating diabetes and it will come up with recommendations to help controlling this disease using "Camel milk-diet".

This research will demonstrate the richness of camel milk in providing a good source of high-value "bioactive peptides" as an ingredients for pharmacology.

Investing in Knowledge for the Nation's Future -

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