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A laboratory investigation of cow and camel whey proteins deposition under different heat treatments

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ABSTRACT

Using a developed laboratory scale device, different heat treatment conditions were applied on camel and cow wheys. After each deposition experiment, photos of stainless steel plates were taken and dry deposit weights were determined. Proteins denaturation was studied by electrophoresis (SDS-PAGE) and differential scanning calorimetry (DSC). The obtained results have shown that heating both cow and camel wheys at 60 °C does not generate deposit. Furthermore, the heat treatment at above 70 °C was found to cause a severe fouling of stainless steel plate. The electrophoresis patterns indicated that heating at 90 °C caused camel serum albumin's (CSA) band disappearance for both rennet and acid wheys. However, α-lactalbumin's (α-la) concentration decreased versus temperature and heating time. DSC thermograms showed that denaturation temperatures were 73.8 °C for camel rennet whey, 60.5 °C for camel acid whey, 70.5 °C for cow rennet whey and 63.9 °C for cow acid whey. Taken into the count the absence of β-lg in camel milk and based on the obtained results several hypotheses were advanced to explain camel milk fouling during heat treatment.

1. Introduction

Cow milk is very important in the human diet thanks to its balanced basic nutrient composition (proteins, carbohydrates and fats) and high vitamins and mineral contents, including calcium (Haug et al., 2007). In order to ensure a better microbiological quality of milk and increase the duration of its storage for human consumption, dairy industrials have usually applied specific heat treatments, such as pasteurization and sterilization. However, these treatments have a direct influence on the nutritional, biological and functional properties of milk proteins (Changani et al., 1997) and heat exchangers performances (Bansal and Chen, 2006).

Camel milk is one of the main food resources for arid populations. It contains all nutrient components (protein, fat, lactose, minerals) and has a highly biological value due to the higher contents of antimicrobial factors such as lysozyme, lactoferrin and immunoglobulin (El-Agamy et al., 1998). Most of camel milk is consumed in the fresh state. Despite the low production's percentage of camel milk in the world compared to cow milk, the former's preservation has a real importance to avoid its microbial degradation owing to its nutritional specificities. The preservation of camel milk can be achieved using heat treatments such as pasteurization and sterilization processes. However, there are few studies pertaining to the camel milk behavior during heat treatment operations.

Otherwise, the effects of heating bovine milk were intensively studied with special interest in milk microbiological, biochemical and nutritional qualities (Changani et al., 1997), process performances and heat exchanger fouling (Lalande

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et al., 1984; Ayadi et al., 2008). Thus, during the heat treatment of bovine milk, various technological problems, caused by fouling mechanism such as the formation of deposits on heat exchangers surface could happen, inducing the formation of undesirable compounds. Therefore, the reduction of this phenomenon has always been a scientific and technological challenge. Previous studies showed the existence of a great correlation between whey protein denaturation and fouling mechanism (Lametti et al., 1997). One of the proteins responsible for this phenomenon of bovine milk is β-lactoglobulin (β-lg) (Lalande et al., 1984). The unfolded β-lg, an intermediate of the denaturation reaction, is able to aggregate with other proteins or is adsorbed at the deposit layer (Lalande et al., 1984; de Jong, 1997). However, previous studies showed that camel milk is devoid of β-lg (Farah, 1986). To the best of our knowledge, only few studies about the effect of heat treatments on camel milk composition are available in the literature (Farah, 1986; El-Agamy, 2000) and heat exchangers performances. Therefore, the understanding of an eventual fouling phenomenon during camel milk heating is of a great scientific and technological importance. Actually, the detection of potential fouling phenomena and understanding its mechanism during camel milk heating is of a major importance for its commercialization.

The objective of this paper is, therefore, to highlight the existence of a potential deposit formation during the heat treatments of camel rennet and acid wheys, in comparison with cow wheys. The description of this phenomenon was followed by the determination of the whey proteins denaturation degrees in comparison with whey proteins from cow samples.

2. Materials and methods

2.1. Milk samples

Fresh raw camel milk was obtained from an experimental station located in the south of Tunisia (Douz region in Kébili governorate). Fresh cow milk was purchased from a local breeding located in Sfax region (Sfax governorate). Once arrived to the laboratory at 4 °C, a pH (Metrohm pH meter) determination was realized. Then, both milks were skimmed by centrifugation at 3000 *g* during 20 min at 4 °C (Gyrozen 1580MGR, Multi-purpose Centrifuge, Daejeon, Korea). Rennet-wheys were obtained after rennet coagulation of fresh milks at 36 °C in the presence of 1.4 mL L^{-1} of microbial rennet (*M. miehei*, strength = 1:10,000, Laboratories Arrazi, Parachimic, Sfax, Tunisia). Acid-wheys were obtained after the acidification (HCl, 6 M) of fresh milks until pH = 4.6 and 4.3 for cow and camel milks, respectively; the wheys were recovered by centrifugation at 3000 *g* for 20 min at 4 °C (Gyrozen 1580MGR, Multi-purpose Centrifuge, Daejeon, Korea).

2.2. Heat treatment and deposition experiments

An experimental apparatus, at laboratory scale, was conceived to conduct deposition experiments during the heat treatment of wheys from camel and cow milks (200 mL) (Fig. 1). Deposition experiments were realized in inox container (total volume = 500 mL) containing rectangular dismantled plates (types 316 L; 20 mm × 60 mm), placed at the bottom of the recipient, under different heat conditions (durations: 60 and 120 min; temperatures: 60, 70, 80 and 90 °C), and reproduced at least 3 times. Heat treatment consisted in heating over a hot plate without agitation. After each experiment, the photos of

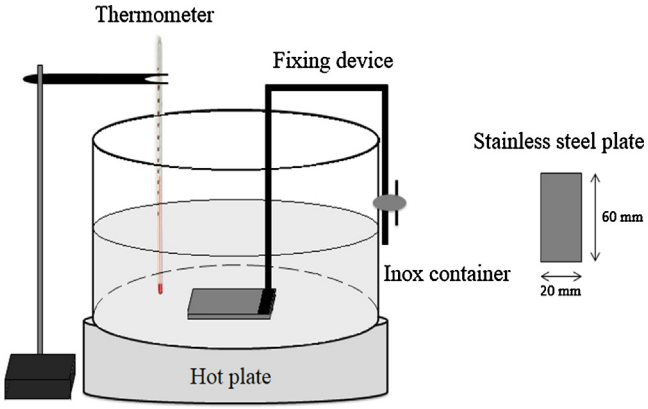

Fig. 1 – Schematic representation of the experimental apparatus to follow deposit generation during heat treatment of camel and cow wheys.

fouled plates were systematically taken, using a digital camera (Samsung EC-ES80, 12 MP, 5× Optical Zoom, USA) performing with no magnification, and dry stainless steel plates were weighed (drying condition: 105 °C/8 h). The results describing deposition are expressed in g of dry matter/cm^2.

2.3. Free thiols content

To quantify free thiols, 158 mg (2 mM) of 5,5′-dithio-bis (2-nitrobenzoic acid) (DTNB) was added to 820 mg of a 50 mM sodium acetate solution, dissolved in 200 mL ultrapure water and refrigerated before use. For assay, 1 mL quartz curvets were filled with 840 μL of ultrapure H_2O, 50 μL of the DTNB sodium acetate solution, 100 μL of Tris buffer (1 M, pH = 8.0), and 10 μL whey samples heated at different heat treatments. The curvets were incubated at 37 °C for 5 min and the optical density was measured at 412 nm (Ellmann, 1959). The absorbance values for each sample were divided by the molar extinction coefficient of the DTNB mixed disulfide complex (13,600 M^{-1} cm^{-1}), resulting in a concentration of thiols in solution.

2.4. SDS-PAGE electrophoresis

Electrophoresis experiments (sodium dodecyl sulphate polyacrylamide SDS-PAGE) were carried out using a Bio-Rad apparatus (Mini-Protean Tetra Cell) of gels in vertical blocks. The concentration of acrylamide gel was 15%. Electrophoresis was run at 120 mA until the change of the marker color (bromophenol blue) was at 0.5 cm from the anode end of the block (approximately 3 h). SDS-PAGE experiments were realized according to the procedure of Laemmli (1970). The molecular weights of the different protein fractions were estimated by comparing their electrophoretic mobilities with those of marker proteins having molecular weights known.

2.5. Thermal properties: differential scanning calorimetry (DSC) experiments

To improve the signal quality of the DSC analyses, fresh bovine and camel whey samples were previously concentrated using vivaspin 20 tubes (10,000 Da Molecular Weight Cut Off, sactoriusstedim biotech, GmbH, Germany) in a centrifuge unit at 1800 *g* at 20 °C for 2 and 4 h for rennet and acid wheys, respectively. By taking into account the reduction of volume of the retentate, this protocol allowed the concentration of proteins

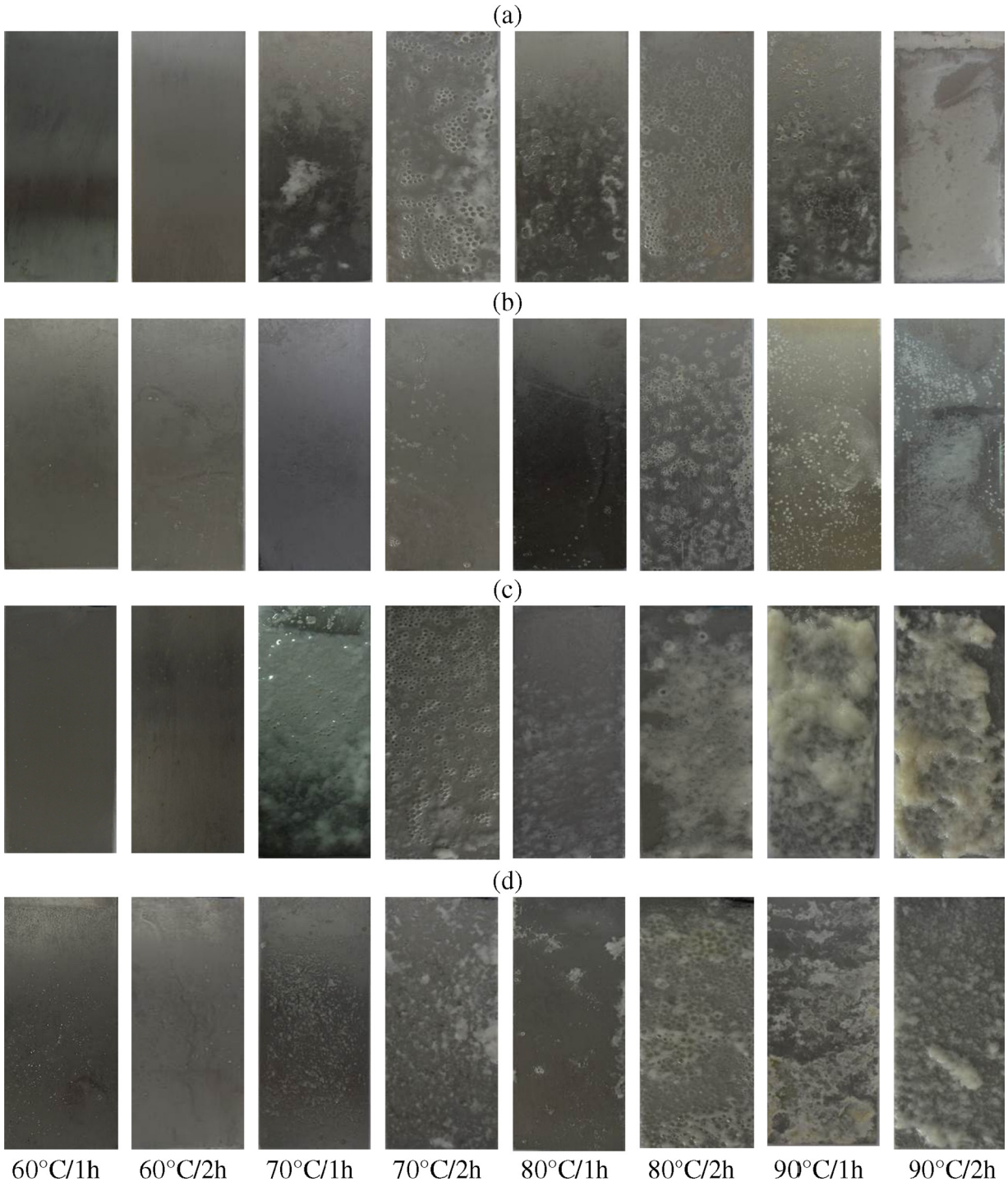

Fig. 2 – Photos of protein deposition generated by camel rennet (a) and acid (b) wheys and cow rennet (c) and acid (d) wheys as a function of heat treatments parameters.

for the bovine and camel samples, respectively. The samples were stored at 4 °C before DSC experiments.

The thermal properties of concentrated whey proteins were monitored by DSC using a TA Q-1000 calorimeter (TA Instruments, New Castle, DE). Calibration was made with an indium standard (melting point = 156.66 °C, ΔH melting = 28.41 J/g). About 90 mg of samples was accurately weighed in a hermetic stainless steel pan of 100 μL. A pan with 90 mg of distilled water was used as a reference, and both pans were hermetically sealed. The samples were tempered at 20 °C for 5 min, and heated from 20 to 160 °C at 5 °C/min. The measurements were performed5 times with independent samples. The baselines were not adjusted.

3. Results

3.1. *Deposit evolution during acid and rennet camel and cow wheys heating*

Fig. 2 shows the photos of wet plates after different heat treatment conditions of camel and cow wheys. The obtained deposit corresponds to that of type "A" described by Burton (1968), i.e., it is voluminous, spongy and of a whitish color. For both camel rennet and acid wheys, the deposit was absent at 60 °C for both processing times (1–2 h). Fouled plates after 1 h heating at 70 °C showed the formation of an initial deposit and an accentuation of this deposit after 2 h-treatment for

Table 1 – Dry deposit mass evolution (g/cm^2) versus temperature during 60 and 120 min: (a) Camel rennet whey; (b) Camel acid whey; (c) Cow rennet whey; (d) Cow acid whey.

Samples	Time of heating (min)	Temperature (°C)			
		60	70	80	90
(a)	60	0.000 ± 0.01^a	0.002 ± 0.00^b	0.002 ± 0.01^c	0.003 ± 0.01^d
	120	0.001 ± 0.00^a	0.003 ± 0.02^b	0.003 ± 0.01^c	0.004 ± 0.04^d
(b)	60	0.001 ± 0.00^b	0.000 ± 0.00^a	0.001 ± 0.00^b	0.001 ± 0.01^b
	120	0.001 ± 0.00^a	0.001 ± 0.00^a	0.002 ± 0.00^b	0.002 ± 0.01^b
(c)	60	0.001 ± 0.01^a	0.002 ± 0.00^b	0.003 ± 0.01^c	0.010 ± 0.02^d
	120	0.003 ± 0.03^b	0.002 ± 0.01^a	0.005 ± 0.02^c	0.013 ± 0.06^d
(d)	60	0.000 ± 0.00^a	0.001 ± 0.01^b	0.002 ± 0.01^c	0.003 ± 0.03^d
	120	0.000 ± 0.01^a	0.001 ± 0.03^b	0.002 ± 0.02^c	0.005 ± 0.02^d

Averages ± standard deviation (SD) of three replicates. $^{a\text{-}b\text{-}c\text{-}d}$Values within the same row with different superscripts differed significantly by Duncan's multiple-range test ($p < 0.05$).

camel rennet whey. However, for camel acid whey, the deposit appears only after 2 h processing. It is worthwhile to note that with the increase in temperature to 80 °C, the deposit formation of camel rennet whey increased while that of camel acid whey just began. Heating camel whey at 80 °C for 2 h, the plate became completely covered by a fouling deposit in both cases of camel wheys and this observation continues at 90 °C for 1 and 2 h processing.

Fig. 2(d) shows that after the heat treatment of cow acid whey at 60 °C, the deposit was formed after 1 h heating and continued after 2 h. In contrast to this observation, rennet whey (Fig. 2(c)) does not foul the plates under the same heating condition. Increasing temperature to 70 °C, the deposit formation became more important. At 80 and 90 °C, plates were completely covered by the deposit.

The visual comparison between the photos of camel and cow whey samples shows that the deposit is more important after cow wheys heating than after camel heating. In fact, the deposit formation started earlier for cow wheys at 60 °C while the first deposit layer appears after heating camel whey at 70 °C. The photos of the plates after heating both camel and cow wheys up to 80 °C show a change in the deposit aspect (appearance of a local over-heated zone and the presence of tiny bubbles).

The dry deposit mass evolution on the plate surfaces after deposit generation experiments is shown in Table 1. It can be seen that the deposit is more abundant in plates after heating cow samples than after heating camel wheys. Indeed, after heating camel rennet and acid wheys at 90 °C for 2 h, the deposit amount was 0.004 g/cm^2 and 0.002 g/cm^2, respectively. The deposit quantity after heating cow rennet and acid wheys at 90 °C for 2 h was 0.013 g/cm^2 and 0.005 g/cm^2, respectively. The dry deposit mass evolution (Table 1) clearly demonstrates that deposit formation was relatively lower after 1 and 2 h deposition experiments at 60 and 70 °C and became much important for an operating temperature higher than 80 °C for both camel and cow whey samples.

The deposit evolution for cow samples as well as for camel acid whey is described as follow: a first period with low deposit quantities (60–70 °C) followed by an increase in the deposit formation with increasing temperature and time of heating. In fact, during the first period of the run, the deposit process starts and the deposit layer thickness is insufficient to be detected. After this step, the deposit build-up starts, and continues to increase with temperature and time. On the other hand, Table 1 indicated that the dry deposit mass evolution of the heated camel rennet whey increased immediately with temperature and time of heating. The deposit generation after heating cow milk is mainly caused by the denaturation and adherence of β-lg on heat surfaces (Lalande et al., 1984; de Jong, 1997). It is also well known that camel milk is devoid of β-lg (Farah, 1986; Levieux et al., 2006). The results presented in Fig. 2 and Table 1 highlight the existence of deposit phenomenon during camel wheys heating in spite of the absence of β-lg. So the questions raised at this level are: What is the element responsible for the deposit phenomenon? Since the deposits obtained after heating camel wheys are of Type "A", then which protein or protein complex altering from 60 °C is responsible for deposit formation after heating camel whey?

3.2. *Free thiol contents and electrophoresis patterns*

The evolution of free thiols concentrations for camel and cow rennet and acid wheys under different heat treatment conditions is shown in Fig. 3. Free-SH groups are good indicators of proteins denaturation (Hoffmann and van Mil, 1997). Furthermore, the longer the heating times are, the more the number of available thiol groups decreases. The free -SH concentrations of cow rennet and acid wheys are much lower than those of camel rennet and acid wheys (Fig. 3).

Fig. 3(a) shows that after heating camel rennet whey at 70 °C for all durations, free thiol groups were not detected. Heated camel rennet whey at 70 °C has a free thiol content equal to that of raw camel rennet whey. This means that the camel whey proteins denaturation has not started at 70 °C yet (Farah, 1986; El-Agamy, 2000). The free thiol group curves of 80 and 90 °C have a similar evolution. Indeed, the initial free thiol concentrations detected were 0.26 and 0.35 mM. Just after heating camel rennet wheys at 80 and 90 °C, free SH concentrations reached their maximum, which could be explained by the proteins denaturation. Afterwards, they started to decline to 0.20 and 0.22 mM, respectively, due to the aggregation and adherence reactions on the heat surfaces, which started to appear after 30 min of heating (Lalande et al., 1984).

Similarly, Fig. 3(b) shows that no denaturation reaction was observed during camel acid wheys heating at 70 °C. The heat treatment at 80 and 90 °C indicated that free thiol concentrations increased after heating camel acid whey for 30 min. This result has confirmed that the denaturation reaction started after 30 min for both heat treatment (80 and 90 °C) then

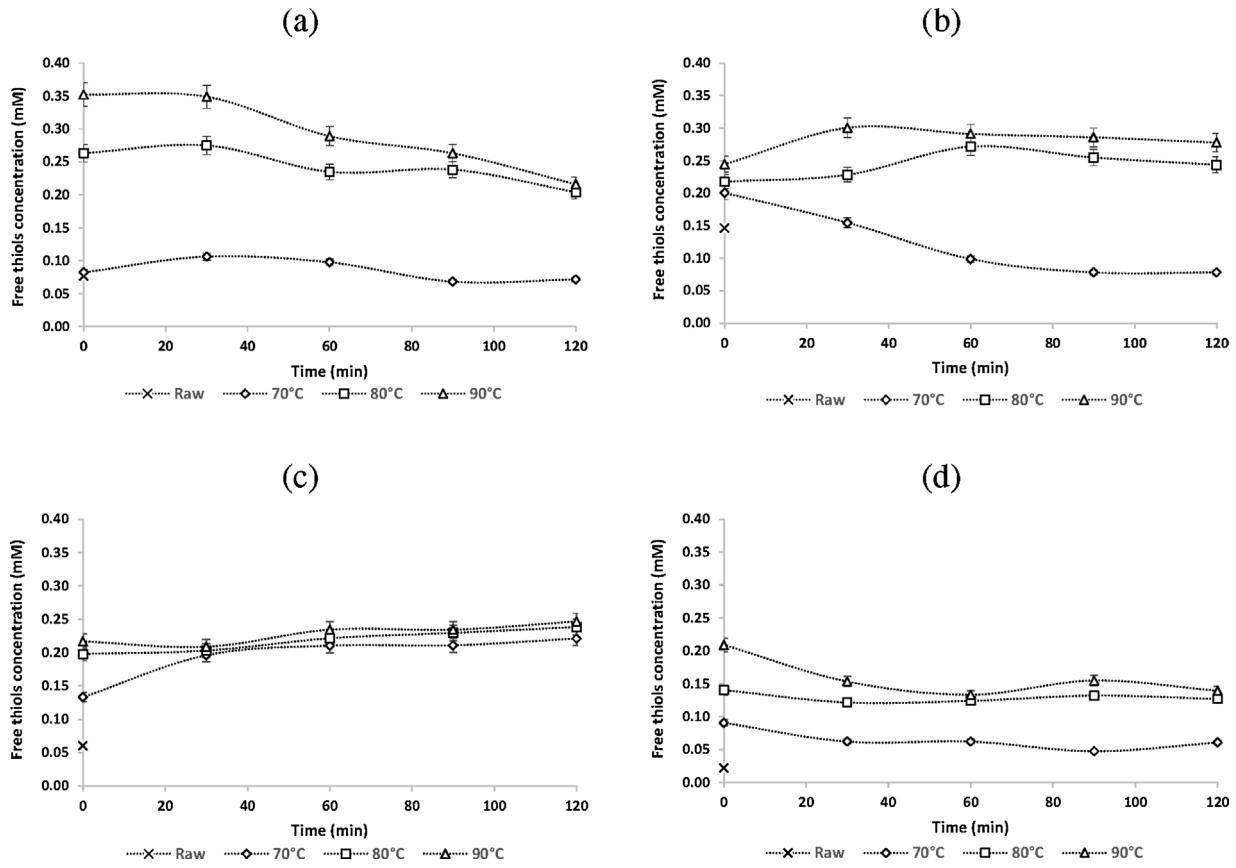

Fig. 3 – Evolution of free thiols concentration of camel rennet (a), acid (b), cow rennet (c) and acid cow wheys (d) after different heat treatment conditions (time and temperature).

remained constant. This could be explained by the structural changes of α-la, major protein in camel whey, due to pH effect. Yang et al. (2006) noticed that apo-α-la at pH 4.0–4.5 exists as an equilibrium mixture of the dimer and the native monomer (the native state). At neutral pH, the thermal stability of apo-α-la decreased.

Fig. 3(c) and (d) indicated that the magnitude order is the same for cow rennet and acid wheys. Proteins denaturation started after heating cow whey for 30 min for all temperatures. The whole phenomenon happened during 30 min of heating. An exhaustion of cow whey proteins was deduced after heating cow wheys at different temperatures for 30 min. This result may be explained by the thermal denaturation of β-lg.

A comparison between the evolution of the free thiol groups of camel and cow rennet and acid wheys revealed that cow wheys present lower initial free thiol concentrations compared to camel wheys. On the other hand, camel proteins denaturation was faster in comparison with cow samples. The thermal sensitivity of camel whey proteins was higher than their bovine counterparts.

The gel electrophoretic patterns of fresh and treated camel and cow wheys at different heat treatment conditions are shown in Fig. 4 and differences in electrophoretic patterns of camel and cow wheys were found. The major camel whey protein bands correspond to α-lactalbumin (α-la) and to camel serum albumin (CSA). On the other hand, the most abundant protein in cow whey is β-lg, whereas it was not detected in camel whey. This result reinforces previous data reported by Farah (1986) and El-Agamy (2000).

When camel rennet whey was heated at 90 °C, α-la and CSA bands disappeared (Fig. 4). For raw camel acid whey (Fig. 4(b), L1), four bands were observed: α-la, CSA and two other bands with about 20 and 30 kDa as molecular weights. These bands could be attributed to whey proteins complex or to caseins traces. The heat treatment of camel acid whey at 80 °C induced the immediate disappearance of CSA band, and that of camel α-la band after 90 min. On the other hand, when cow rennet whey was heated at 90 °C, β-lg, α-la and bovine serum albumin (BSA) bands disappeared. However, BSA band disappeared from the gel patterns when the cow acid whey was heated at 70 °C for 30 min. Both β-lg and α-la bands disappeared when cow acid whey was heated at 90 °C for 30 min. No effect was recorded on camel and cow whey proteins heated at 63 °C for 30 min (Farah, 1986). Similar results were obtained by El-Agamy (2000). This author noticed no change in whey proteins electrophoretic patterns when camel and cow wheys were heated at 65 and 75 °C for 10, 20 or 30 min except for β-lg which was affected during cow milk heating. At 85 °C, BSA band intensity decreased in cow whey (El-Agamy, 2000).

The disappearance of the electrophoretic bands could be the result of proteins denaturation and/or aggregation. Previous studies reported that β-lg denaturation is involved in surfaces fouling during cow whey heating (Lalande et al., 1984; de Jong, 1997).

3.3. *Thermal properties of wheys characterized by DSC*

Fig. 5 shows the DSC curves recorded of concentrated rennet (Fig. 5(a)) and acid wheys (Fig. 5(b)) with a comparison between camel and cow wheys. In all cases, the recorded thermal phenomena were endothermic and attributed to unfolding of globular proteins during heat treatment, which requires a heat absorption to break the intramolecular bonds (non-covalent, disulfide bonds, etc.). Hence, the endotherms recorded

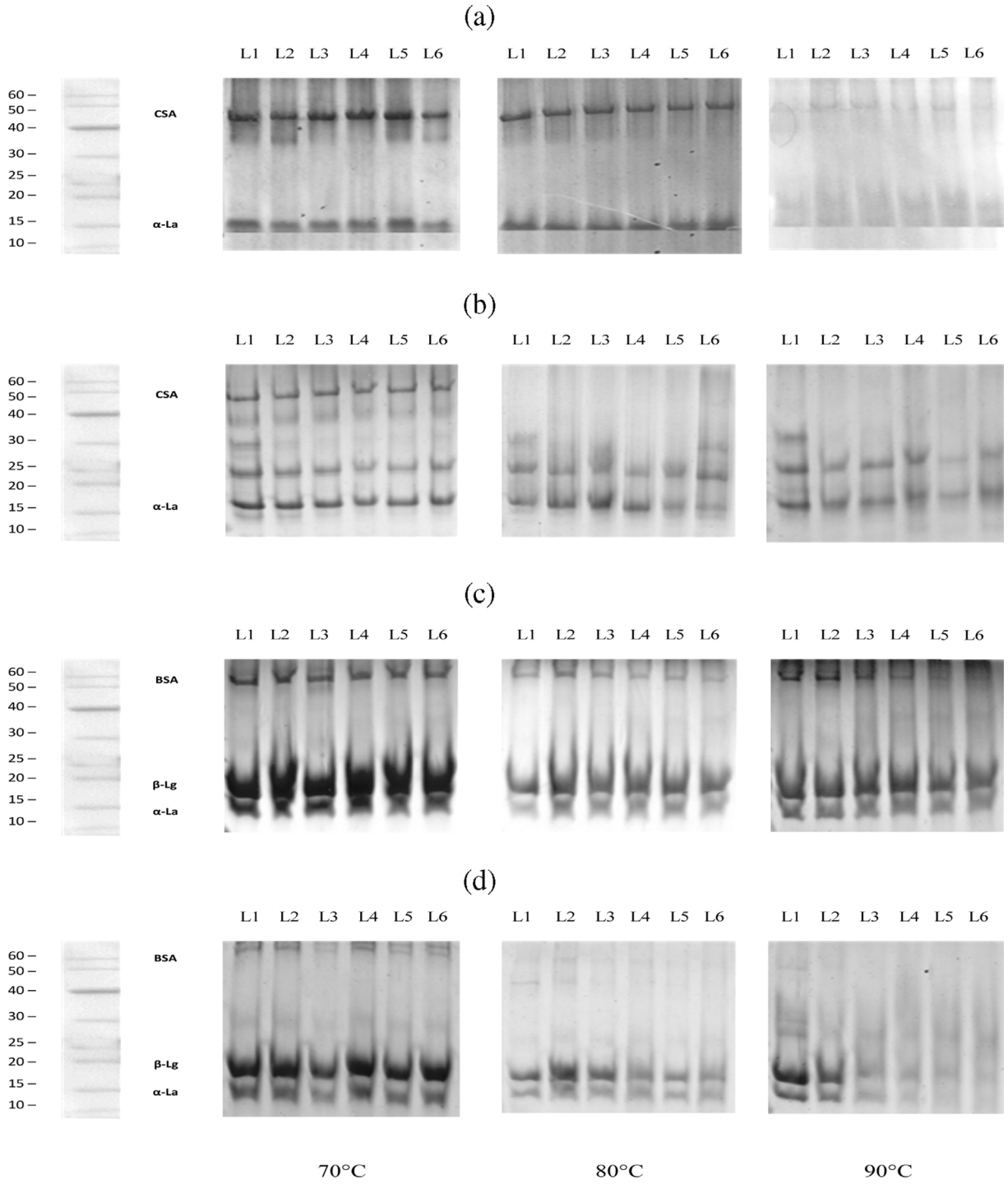

Fig. 4 – SDS-PAGE electrophoresis patterns of camel rennet (a), acid (b) and cow rennet (c) and acid (d) whey proteins heated at 70, 80 and 90 °C for different durations. On the left of this figure, markers of molecular weights. L1: Unheated whey, L2: heated whey (0 min), L3: 30 min, L4: 60 min, L5: 90 min, L6: 120 min.

during the heating of the wheys were mainly interpreted as the denaturation of the proteins contained in the wheys.

The thermogram of concentrated camel rennet whey proteins (Fig. 5(a)) reveals a broad endothermic event peak corresponding to the overlapping of two main endotherms, accredited to the denaturation of camel α-la at 73.8 °C and to that of CSA at 91.1 °C (Fig. 5(a)(1)). The DSC curve recorded for cow rennet whey (Fig. 5(a)(2)) displays a broad thermal event that may correspond to three overlapped endothermic peaks, interpreted as the successive thermal denaturations of α-la (around 70.5 °C), β-lg at 79.6 °C and BSA at 94.9 °C. This broad endotherm could also correspond to the formation of a whey proteins complex. The comparison of these results with those described in the literature shows some similarities and some differences. In fact, El-Agamy (2000) has reported that camel whey proteins, isolated from liquid camel milk, are relatively heat stable and this stability is due to the absence of β-lg. As for Levieux et al. (2006), they have confirmed that purified immunoglobulins, CSA and α-la are denatured at 67.2, 73 and 77.5 °C, respectively. With respect to Ruegg et al. (1977), they have demonstrated that the denaturation temperature of whey proteins in simulated cow milk ultrafiltrate were 62.2, 65.2 and 72.8 °C for BSA, α-la and β-lg, respectively. Regarding cow whey proteins, Paulsson et al. (1985) have reported the

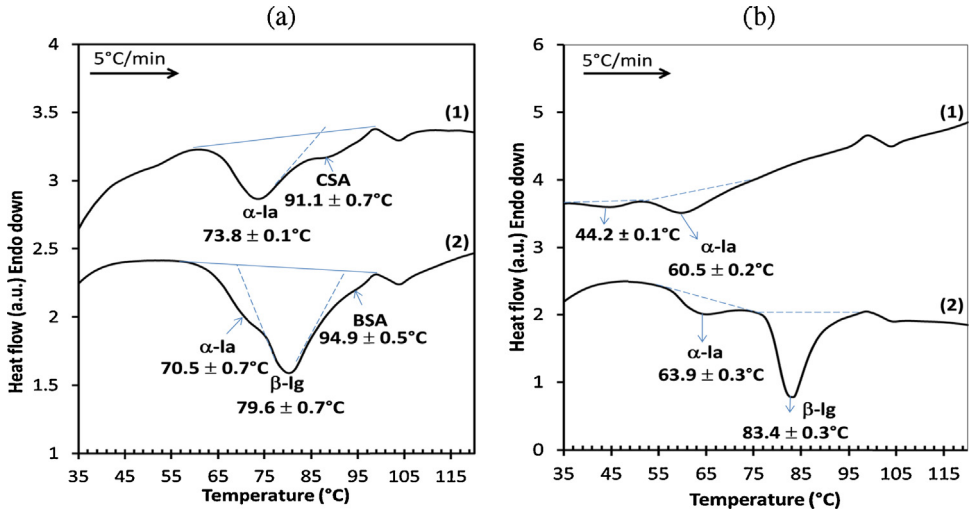

Fig. 5 – Differential scanning calorimetry curves recorded during heating at 5 °C/min of concentrated (a) rennet and (b) acid wheys from camel (1) and cow (2) milks.

stability of purified α-la, β-lg and BSA at pH 6.6 and observed only one peak at temperature lower than 80 °C.

The DSC curves of concentrated camel and cow acid whey proteins (Fig. 5(b)) showed two endothermic peaks. The thermograms give the peak temperatures at 44.2 and 60.5 °C for acid camel whey (Fig. 5(b)(1)). The first peak at 44.2 °C corresponds to the melting of residual fat globules in camel whey. It is proven that the melting point of camel fat is about 41.4 °C (Farah et al., 1989; Karray et al., 2004). The peak at 60.5 °C is interpreted as the denaturation temperature of camel α-la. This result is in accordance with those found by Paulsson et al. (1985). The two endotherms recorded for cow rennet whey at 63.9 and 83.4 °C were interpreted as the thermal denaturation of α-la and β-lg, respectively (Fig. 5(b)(2)). Moreover, the temperature of thermal denaturation of α-la is significantly lower in camel acid whey as compared to cow acid whey (i.e. a delay of 3.4 °C). This could contribute in the explanation of the lower thermal stability of acid wheys from camel as compared to acid wheys from cow.

The DSC curves reveal differences between acid and rennet wheys. The difference of thermal denaturation observed is attributed to the pH on proteins unfolding, equal to 4.6 and 6.7 for cow acid and rennet wheys, respectively. It is admitted that α-la is more heat stable at acidic pH than at neutral pH. This may be due to its structure modification after acidification and heat treatment (Yang et al., 2006). The difference between cow acid (Fig. 5(a)(2)) and rennet whey (Fig. 5(b)(2)) emanates from the difference of their chemical environment. While at neutral pH, β-lg exists as a dimeric form and at 55 °C, it dissociates in a monomeric form, whereas at higher temperatures, β-lg is denatured (Georges et al., 1962). At acidic pH, β-lg is under octameric form considered as more heat stable than at neutral pH. The presence of caseinomacropeptide in cow rennet whey has no effect on the denaturation reaction of whey proteins (Croguennec et al., 2014).

From Fig. 5, camel whey proteins have a temperature of thermal denaturation lower than their bovine counterparts and could then be considered as less heat stable. Actually, the obtained results are in accordance with those of Atri et al. (2010) who showed that, in both holo and apo states, the stability of camel α-la is greater than that of bovine α-la. It is noteworthy, however, that Laleye et al. (2008) have demonstrated the opposite.

4. Discussion and conclusion

Under 100 °C, the thermal treatments in heat exchangers induce fouling of hot surfaces, which emanates essentially from the denaturation of whey proteins. β-lg, representing 50% of the whey proteins in cow milk (Schmitt et al., 2007), is predominant in milk fouling. During heat treatment, β-lg loses its tertiary structure and becomes reactive by exposing a free thiol group. Unfolded β-lg associates with other whey proteins to cause fouling reactions on heat exchangers hot surfaces. It is noteworthy to mention that β-lg denaturation is almost completed at 89.3 °C for 73 s (Petit et al., 2013).

At neutral pH, β-lg thermal unfolding transition is ≈70 °C (de Wit and Swinkles, 1980), whereupon the protein dimer dissociates and molecules begin to unfold. This reveals the free thiol of Cys121, leading to the intermolecular association (Lametti et al., 1997). The free Cys121 of β-lg, with its reactive thiol group, has a pH-dependent activity in its involvement in the denaturation and aggregation behavior during milk heat treatment (Havea et al., 2001). Lapanje and Poklar (1989) found that at acid pH, where the protein is considered to be more stable, a decrease in β-sheet structure was found on heating.

The obtained results have clearly shown that, on the one hand, camel whey is devoid of β-lg (Fig. 4), and on the other hand, after heating camel rennet and acid wheys at 70 °C, the deposit formation is observed (Fig. 2). It was supposed in this study that α-la and CSA are involved in deposition phenomenon. The differences between camel rennet and acid wheys occured in their chemical compositions, mainly in pH (6.51 and 4.3 for camel rennet and acid wheys, respectively) and mineralization (Relkin, 1996). Indeed, pH has an impact on the thermal behavior of whey proteins, mainly α-la, which is the major protein in camel whey.

α-la, which lacks a free thiol, undergoes a thermal unfolding at a lower temperature than does β-lg (Ruegg et al., 1977). The effect of pH on protein stability differs markedly when comparing the denaturation profiles of holo-α-la recorded at neutral pH versus those obtained under acidic conditions. The pH dependent stability of α-la is relatively constant within the pH range of 5.2–8.0, but decreases upon acidification. α-la exists as a partially folded conformer at acid pH. A second partially folded conformer is formed above 60 °C. Hendrix et al. (2000) have shown that at pH values higher than 5, the

heat-denatured state and the unfolded state are indistinguishable. At pH values close to neutrality, heat denaturation occurs at high temperature and yields a totally unfolded polypeptide.

The obtained results have enabled us to highlight the deposit phenomenon during the heat treatment of camel rennet and acid wheys under different conditions. In this study, we identify whey proteins involved in deposition during camel wheys heating. With reference to these findings and previous studies, we present the following hypotheses:

- The deposit phenomenon during camel rennet whey heating could be caused by the denaturation and adherence of α-la and/or CSA.
- During heating camel acid whey, deposit formation might be generated by the denaturation and adherence of α-la and/or CSA and/or caseins traces present in the whey.

Based on this experimental study, future research works will be dedicated to study the deposit composition to validate the provided assumptions, to examine the thermal behavior of camel milk to understand whey proteins behavior in milk and to investigate the thermal behavior of each camel protein separately.

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