

# EFFECT OF RENNIN ENZYME ON CAMEL MILK PROTEINS

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## ABSTRACT

The Enzymatic coagulation time (ECT) was determined. The effect of pH, temperature, and added ammonium chloride on the coagulation time as well as the release of NPN by the action of rennet were also studied. The results show that increasing milk pH value increase the ECT. The ECT of camel milk at pH 6.6 was 5.5 times lower than at pH 7.0. The results indicated that Enzymatic coagulation of camel milk (ECT) responses in a similar manner in which bovine milk coagulation responses to changes in temperature and pH. Preheating of camel milk causes an increase in the ECT at different pH values. The amounts of NPN soluble in 2% trichloro acetic acid (TCA) and 12 % TCA released at 90% of ECT, were 6.8 and 5.4 mg/100 ml respectively. addition of 5ml mol/l ammonium chloride to camel milk leads to higher increase in the Enzymatic coagulation time of camel milk (ECT) in compares with that increase caused by preheating of milk. This means, that the response to changes in pH, temperature and ammonium concentration is the same for camel and bovine milk.

**Key words:** Camel milk. preheating of milk. Ammonium

## 1. INTRODUCTION

There are 17 million camels in the world, of which 12.2 million are in Africa and 4.8 million in Asia (1). The camel is a potentially important source of milk. Indeed, in some countries hosting large camel populations, camel milk is one of the main components of the human diet. The present knowledge about milk production potential is very limited. However, milk production varying between 1,800 and 12,775 kg during a lactation period between 9 and 18 months has been reported (2). Most camel milk is

consumed fresh or when it has just turned sour. Reports on the possibility of obtaining cheese from camel milk are scarce and often contradictory. Some authors report the existence of rennet coagulated cheese made from camel milk while others categorically state that cheese cannot be made from camel milk (3, 4, 5). The present investigation was therefore undertaken to obtain a better insight into the problem of rennet coagulation of camel milk. The study describes the action of rennet on camel milk as well as the effect of pH, temperature, and added ammonium chloride on the coagulum development. The release of non protein nitrogen (NPN) by the action of rennet on camel milk has also been studied.

## 2. AIM OF STUDY

The main objective of this work was to examine the rennet coagulation and some of its properties in camel milk

## 3. MATERIALS AND METHODS

### 3.1 MILK SAMPLES

Milk samples were taken from different area of Camel Farms which are situated in Misurata city which lies 200 km from Tripoli. The milk samples were collected from 15 individual camels. After measuring the pH of the individual samples the milks were mixed to form one batch, skimmed and prepared for coagulation studies in the laboratory.

### 3.2 MILK COAGULATION ACTIVITY DETERMINATION

The rennet was commercial powder from Chr. Hansen with an activity of 1:100,000. Rennet solution of 0.4% was prepared and an appropriate amount of this solution was taken to give a visually observed coagulation time of approximately 5 min in bovine milk. For measurement of the Enzymatic milk coagulation time(ECT) the following two methods were used.

### **3.3 EFFECT OF TEMPERATURE**

The milk samples were adjusted to pH 6.65 by slow addition of 1 M HCl or NaOH placed in a water bath, and the Enzymatic coagulation time (ECT) was measured at temperatures over the range at (25 to 60 °C.)

### **3.4 EFFECT OF pH**

All samples were equilibrated at 30 °C and the Enzymatic coagulation time(ECT) determined at pH between 5.6 and 7.00.

### **3.5 EFFECT OF ADDED AMMONIUM CHLORIDE**

All the samples were adjusted to pH 6.65 and placed in a water bath a 30°C. ammonium sensitivity was evaluated by measuring the Enzymatic coagulation time after addition of an appropriate amount of ammonium chloride

### **3.6 NPN RELEASED BY THE ACTHION OF RENNIN ON MILK**

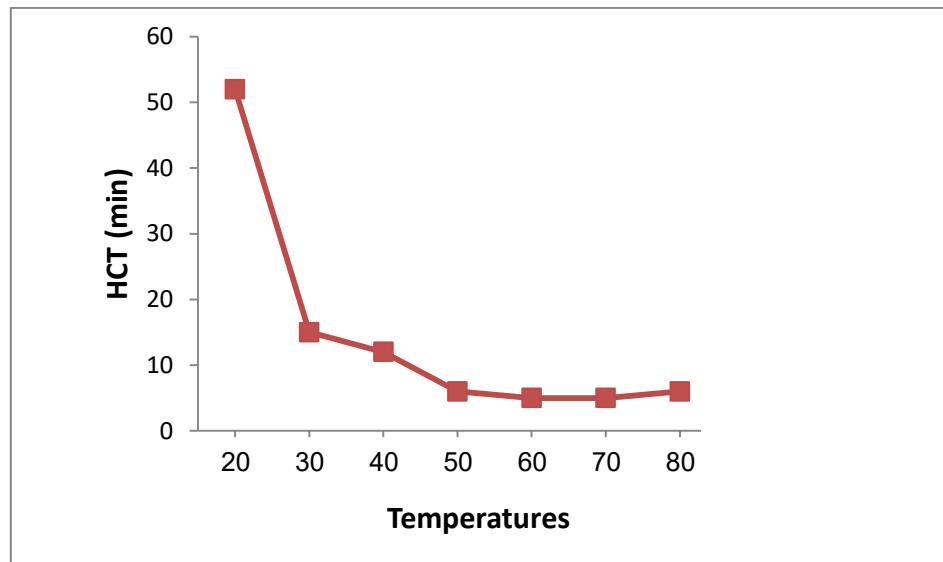
This method is patterned after that described by NITSCHMANN (9). An aliquot rennet solution low enough to give a coagulation time of around 20 min in the milk was added to 25 ml portions of camel milk samples incubated in a water bath at 30°C. At pre-determined intervals trichloro acetic acid (TCA) was added to each portion to obtain a final concentration of 12 % (w/v) in order to stop the reaction and precipitate the milk proteins. The trichloro acetic acid (TCA) soluble NPN, split-off during the action of rennet as well as the total nitrogen (TN) were determined by the Kjeldahl method (10). The rate of liberation of NPN from casein by the action of rennet was measured by monitoring the increase in 12 % trichloro acetic acid (TCA) soluble in N-compounds.

## **4. RESULTS AND DISCUSSION**

### **4.1 EFFECT OF TEMPERATURE ON ECT :**

Temperature between range (25-50C<sup>0</sup>) decreased the Enzymatic coagulation time(ECT) of camel milk at pH 6.6 (Fig. 1). Results indicate that Enzymatic coagulation time(ECT) of camel milk decreased from about 50 minutes to 14 minutes when increased temperature of 25-30C<sup>0</sup> and continue the decreased in the enzymatic coagulation time of camel milk when increased the temperature to higher levels but slowly gradually

until it reaches the lowest value 5.30 minutes at a temperature of 45 and will start to increase simple and did not get Enzyme coagulation time of camel milk when increase temperature to  $55^{\circ}\text{C}$ . And this results agree with what found by (Farah and Bachmann 1987) (13)



**Fig. 1:- Effect temperature on ECT of camel milk at pH 6.6**

#### 4.2 EFFECT OF pH ON THE MILK

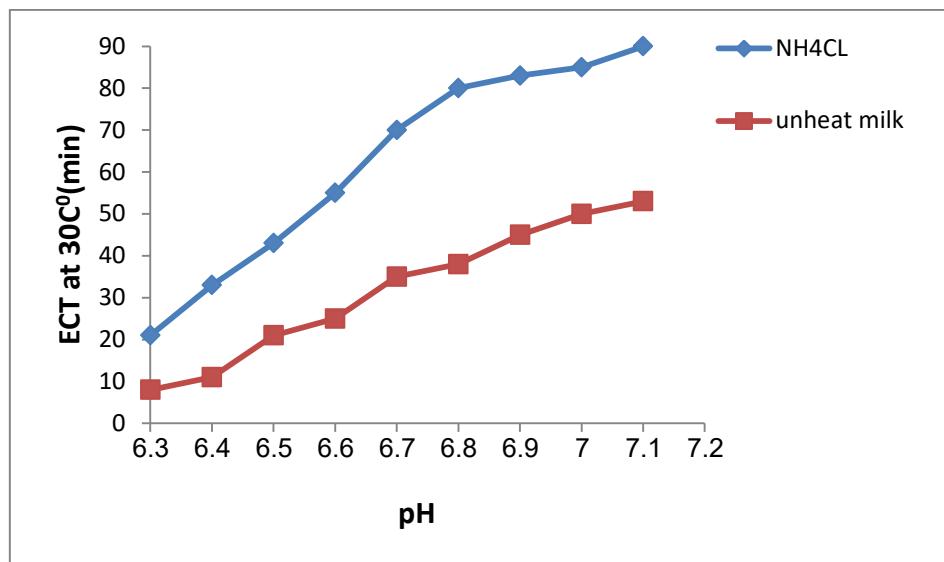
pH values and Enzymatic coagulation times (ECT) of camel and bovine milk are presented in (Table 1). The lowest pH measured in the 15 individual camel milks was at pH 5.7, 5.8 and the highest 7.0. The most frequently observed pH values were 6.5 and 6.6. which agreed with camel milk pH values reported in the literature (11). Enzymatic Coagulation times for each individual camel milk sample as well as those of mixtures were measured and compared with bulk bovine milk adjusted to pH 6.65. The results (Table 1). show that Enzymatic coagulation time of camel milk at pH 6.6 (the border natural), was 12 minutes and increased about 5.5 times to become 67 minutes when pH revalued of milk to 7 While the amount decrease of around 4 times when it declined pH value to 5.6 where, when at least 4 minutes this did not get enzymatic coagulation of milk at pH 7.1 for more than two hours and the results of this study agree with the results of other studies conducted by (17).

**Table 1:** The pH values and ECT of camel milk samples at 30 °c

<b>milk sample No</b>	<b>pH</b>	<b>ECT (min)</b>
1	5.6	4
2	5.7	2
3	5.8	2
4	5.9	4
5	6.0	4
6	6.1	5
7	6.2	5
8	6.3	6
9	6.4	7
10	6.5	8
11	6.6	12
12	6.7	12
13	6.8	13
14	6.9	18
15	7.0	67
Mixture 1-15	6.65 (adjusted)	13
Cow milk	6.65 (adjusted)	5

#### 4.3 EFFECT OF AMMONIUM CONCENTRATION

Results of this study showed that increasing ammonium concentration by addition of 5mmolNH<sub>4</sub>Cl/l to camel milk increased its Enzymatic coagulation times (ECT) more than 3 times multiplier compared with ECT unheated milk at all studied pH values (Fig. 2). The net negative charge on casein micelles of milk plays an important role in controlling its colloidal stability.(18) In addition may be ammonium chloride has inhibitor effect on the action of rennet.

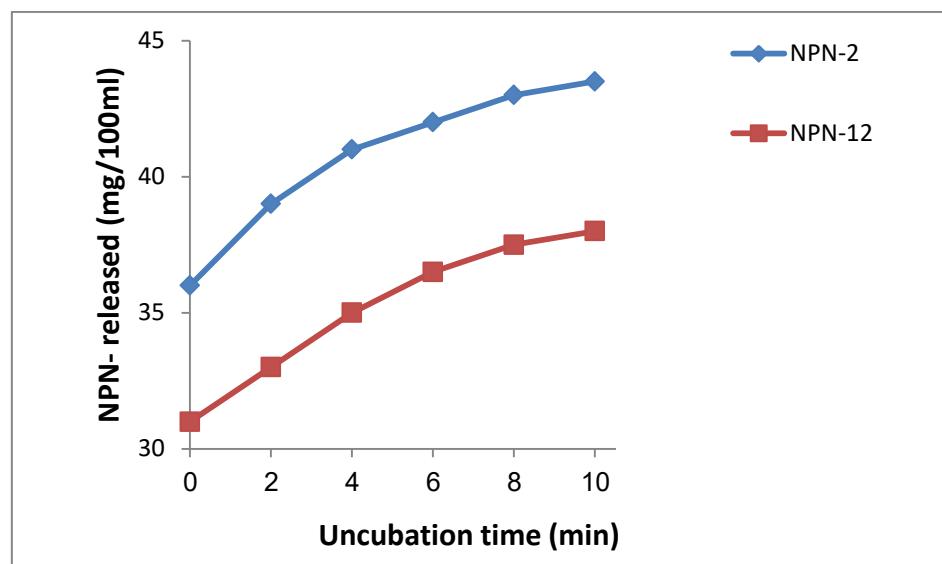


**Fig. 2:-** Effect of added 5mm/l from ammonium chloride on ECT of camel milk at 30°C at different of pH values

In camel milk the Enzymatic coagulation times (ECT) was increased with increasing pH, temperature and added ammonium. This means that the response to changes in pH, temperature and ammonium concentration is the same for camel and bovine milk.

#### 4.4 NPN RELEASED BY THE ACTION OF RENNIN

the amount of NPN released by the action of rennet in camel milk increased at first, reaching a maximum at the coagulation point, and declined then at a slow constant rate (Fig. 3). This is consistent with the findings of MEHAIA (14), who reported the release of NPN soluble in 12% trichloroacetic acid in camel milk. It seems reasonable to assume that a primary reaction of the bovine type occurred between rennet and camel milk caseins. The nature of the released fragments have not been examined. However, MEHAIA (14) reported the existence of glyco- and non-glyco- $\kappa$ -casin in camel casein. Other investigations also reported difficulties in detecting  $\kappa$ -casein in camel milk (15,16).. From the obtained results it can be concluded that camel milk casein is accessible to rennet and the effects of variables like pH, temperature and ammonium chloride on the ECT are similar to that of cow milk but not as pronounced. The action of rennet on camel milk leads to coagulation in the form of flocks with no evidence of gel formation.



**Fig. 3:-** NPN released from camel milk casein by the action of rennet

## 5 CONCLUSION

Our results provide evidence that Enzymatic coagulation time(ECT) of camel milk decreased from about 50 minutes to 14 minutes when increased temperature of 25- (30C<sup>0</sup>). The lowest pH measured in camel milks was at pH 5.7, 5.8 and the highest at 7.0. Addition of 5mmolNH<sub>4</sub>Cl/l to camel milk increased its Enzymatic coagulation times (ECT). The amount of NPN released by the action of rennet increased at first, reaching a maximum at the coagulation point, and declined then at a slow constant rate.

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## تأثير إنزيم الرنين على بروتينات حليب الإبل

### الخلاصة

أجريت هذه الدراسة لمعرفة خصائص تخر حليب الإبل بفعل إنزيم الرنين Rennin وتتأثر درجة الحرارة ودرجة المحموضة (pH) على تلك الخصائص. كما تم دراسة زمن التخثر الأنزيمي (ECT) لحليب الإبل و أيضاً تأثير pH ، والمعاملة الحرارية السابقة وكلوريد الأمونيوم على بعض من جوانب فعل إنزيم الرنين وكذلك الكمية المتحررة من المبيادات النيتروجين غير البروتيني (NPN) بفعل إنزيم الرنين (NPN) بفعل إنزيم الرنين. وأظهرت النتائج أن زيادة قيمة pH للحليب أدى إلى زيادة ECT وان زمن التخثر الأنزيمي لحليب الإبل عند pH 6.6 كان أقل بـ 5.6 مره مما كان عليه عند pH 7.0. أشارت النتائج إلى أن زمن التخثر الأنزيمي (ECT) لحليب الإبل كان مماثل لحليب البقر في الاستجابة للتغيرات في درجة الحرارة وpH. ومعاملة الحرارية السابقة ادت إلى زيادة في زمن التخثر الأنزيمي (ECT) عند قيم pH مختلفة. كمية NPN الدائمة في 2% ، TCA % 12 ، والمتحررة عند 90% من زمن التخثر الأنزيمي كان مقدارها 6.8 ملغم/100ملتر و ملغم/100ملتر على التوالي. إضافة 5مل مول / لتر كلوريد الأمونيوم إلى حليب الإبل ادت إلى زيادة أعلى في زمن التخثر الأنزيمي (ECT) مقارنة مع الزيادة الناتجة عن المعاملة الحرارية السابقة للحليب. وهذا يعني، أن الاستجابة للتغيرات في pH، ودرجة الحرارة وتركيز الأمونيوم هو نفسه بالنسبة لحليب الإبل والأبقار.