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Research Article

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New experimental method for determining the concentration of proteins and lactose in milk and whey

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ABSTRACT

Our objective is to determine the content of protein and of lactose in milk and whey with a new innovative method easy to implement and fast, which is based on the coupling of membrane ultrafiltration operations with electrical conductivity measurements and measures the severity of the Dry Extract (DE). The proteins and lactose determined by the innovative new method was compared with the official method for ten samples. The innovative method gives the results accurate and very close to those obtained by the official method for products of milk and whey. Compared to the official method, the values obtained by this method have a difference about 2% of protein and 1% of lactose.

Keywords: Milk; Whey; Protein; Lactose; Innovative method; Kjeldahl method.

INTRODUCTION

Milk is a white, opaque liquid, twice more viscous than water, slightly sweet taste and the odor not marked [1]. The dairy industry generates large amounts of co-products from the manufacture of cheese and butter. The product we are interested in this work is the nutrient-rich whey primarily the lactose, globular proteins, a non-protein nitrogen fraction, fat and minerals. [2].The composition depends on the origin of the milk and casein coagulation process. The major problem is determining the concentration of protein and lactose. In the literature several methods are used for analysis of protein determination [3,4,5,6] and the lactose [6,7,8].

Generally the dairy industry uses the Kjeldahl method [9] or MilkoScan [6], for the determination of protein content. As regards the content of lactose, it is determined by MilkoScan and brix method. However the official Kjeldhal method has major drawbacks for protein determination [10].

Recovery and recycling of raw materials and the environmental protection materials have long become major economic and political issues. Numerous by-products of the food industry are released into the environment and thus constitute a source of pollution and a considerable loss of certain recyclable materials. Whey, a by-product of the cheese is an example that concerns this work. The quantities of whey are important; about 9.5 liters of whey produced by 1kg of cheese [2]. The value of this by-product has resulted in the implementation of many processes. Yet still about 50% of whey products in the world would not be subject to any treatment [11].

The aim of this study was to determine the content of protein and of lactose in milk and whey with a new innovative method easy to implement and fast which is based on the coupling of membrane ultrafiltration operations with

measurements of electrical conductivity and gravity measurements of Dry Extract (DE). The content of protein and lactose determined by the new innovative method was compared with the official method for ten samples.

EXPERIMENTAL SECTION

Composition of milk

The main constituents of the milk are shown in Table 1.

Table 1: Composition of milk

Minerals salts	9
Lactose ; g/kg	48
Fat; g/kg	37
Protein; g/kg	34
Dry Extract; g/kg	128

Composition of cheese whey

The used whey is a rejection of the plant of the dairy industry of the Meknes city specialized in the production of yoghurt and fresh cheeses. The whey was obtained following an acidic coagulation of pasteurized milk. After milk coagulation, the whey was separated by centrifugation. The table 2 gives the composition of the acid whey.

Table 2: Composition of acid whey

pH	4.3
Conductivity ; µs/cm	7.5
Lactose ; g/kg	43
Fat; g/kg	0.5
Protein; g/kg	5.5
Dry Extract; g/kg	60.26

Experimental and procedure

The experiments were performed in three steps:

Step I: Operation of Ultrafiltration

The pilot plant of Ultrafiltration (UF) is equipped with one module contains one element. The applied pressure over the membrane can be varied from 1 to 10 bars with manual valves. The table 3 gives the characteristic of the membrane used in this operation [12]. The steps of the treatment of whey and milk are shown in figure 1.

Pretreatment:

The whey and the milk undergo mono filtration on a cartridge filter to remove suspended particles and residual lipids.

Operation of Ultrafiltration (UF):

Whey (or milk) pretreated undergoes an ultrafiltration. The **UF** with a volume reduction factor predetermined, will block all protein and fat (**UF**) in the retentate compartment and let the minerals and lactose in the permeate compartment. The Monitoring of the **UF** operation requires the determination of Volume Reduction Factor (**VRF**) given by the following formula:

$$\mathbf{VRF = V_0 / V_R = V_0 / V_0 - V_p}$$

Vo: represents the initial volume of the crude sample (milk or whey), $\mathbf{V_R}$: retentate volume and $\mathbf{V_P}$: Permeate volume.

Table 3: Characteristics of the used membrane

Membrane	P_{max} (bars)	pH	T_{max} (C°)	Material
UF 50 nm	10	3-11	100	Ceramic

Figure 1 shows the diagram of the experimental protocol.

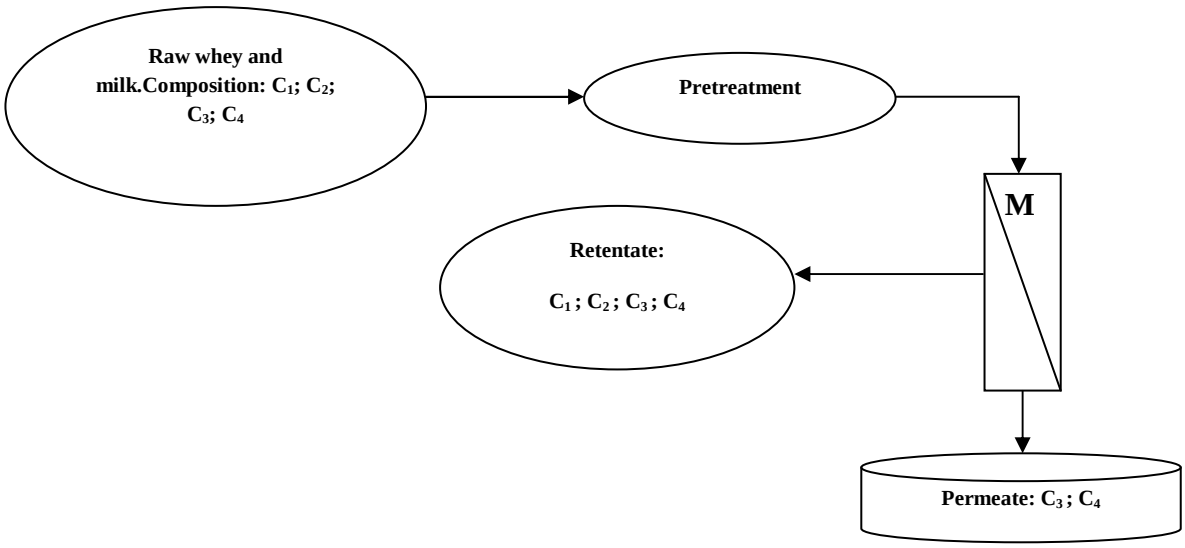

Figure 1: Diagram of ultrafiltration process

C_1: concentration of proteins (g/kg); **C_2**: Concentration of lactose (g/kg), **C_3**: Concentration of fat content (g/kg); **C_4**: Concentration of mineral salts (g/kg); **M**: Module of ultrafiltration

The composition of the whey and milk before and after **UF** is given in Table 4.

Table4: Composition of whey and milk before and after Ultrafiltration

Composition / Parameters	Before UF	After UF	
		Concentrate	Permeate
Fat content (g/kg)	C_3	X C_3	0
Protein (g/kg)	C_1	X C_1	0
Lactose (g/kg)	C_2	C_2	C_2
Minerals (ms/cm)	C_4	C_4	C_4

Step II: Conductivity measurements

The content of minerals is calculated from the measurement of conductivity according to the formula:

$$\textbf{Minerals (g/kg) = EC (ms/cm)* X Factor}$$

EC: Electrical Conductivity

Factor **X** varies depending on the concentration of the product to be treated.

In the range of the milk and whey, the minerals content is derived from the following formula:

$$\textbf{Minerals (g/kg) = EC (ms/cm)* 0,758}$$

Step III: Gravimetric

The determination of the masses of samples retentate and permeate after weighing using a precision balance and after oven-drying is given by the gravimetric measurements.

- Dry Extract (DE) gives the permeate mass of mixture minerals and lactose:

$$\mathbf{M_{Permeate} = M_{Lactose} + M_{Minerals}}$$

- Dry Extract (DE) gives the retentate masse of mixture minerals, lacotse, fat content and protein:

$$\mathbf{M_{Retentate} = M_{Lactose} + M_{Minerals} + M_{Protein} + M_{Fat\ content}}$$

- The lactose content is determined from the mass of permeate sample according to the formula:

$$M_{Lactose} = M_{Permeate} - M_{Minerals}$$

- The protein content is given by the sum of the mass of the retentate sample.

$$M_{Protein} = M_{Retentate} - (M_{Lactose} + M_{minerals} + M_{Fat\ content})$$

- The content of fat is determined by the Gerber method [13].

RESULTS AND DISCUSSION

Results of analysis of protein and lactose content in the milk

We note that for the two curves (protein and lactose) have nearly the same pace for both methods. The results analyses show that the concentrations of protein content and of lactose by the new innovative method are almost constant.

The mean value of milk and whey is respectively about 32; 47 (g/l) and 5.5; 45.5 (g/l). These values are close to what is obtained by other authors [13] so our new innovative method used to determined the protein content and lactose similar to those found by the official method [5,6].

Figures 2 and 3 summarize the results of analysis of protein and of lactose content of milk is determined by two methods. We perform three repetitions for each sample to reduce uncertainty.

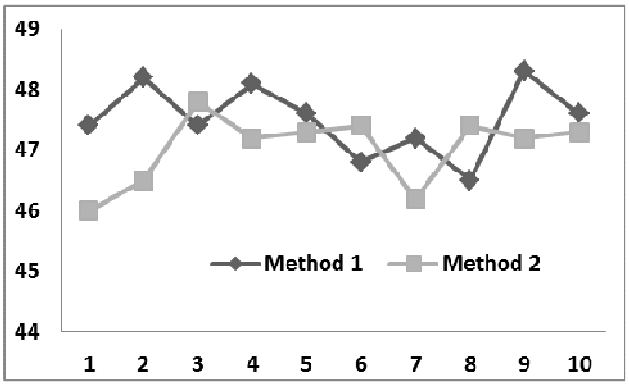

Figure 2: Variation of lactose content for 10 samples of milk by both methods of analysis

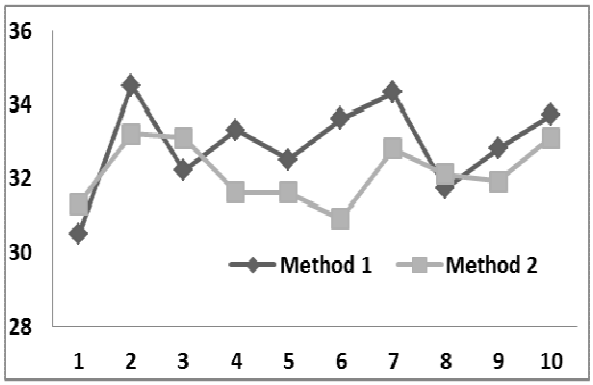

Figure 3: Variation of protein content for 10 samples of milk by both methods of analysis

Method 1: Innovative method; Method 2: Kjeldahl method

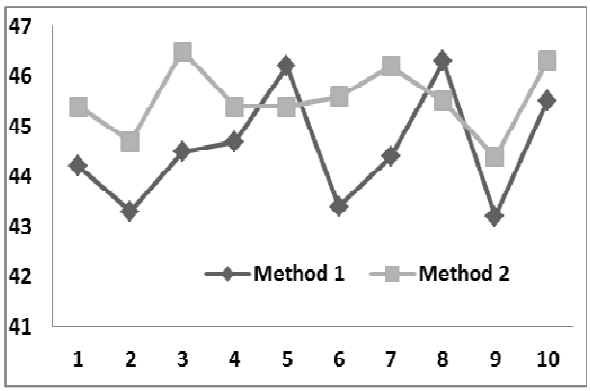

Figure 4: Variation of lactose content for 10 samples of whey by both methods of analysis

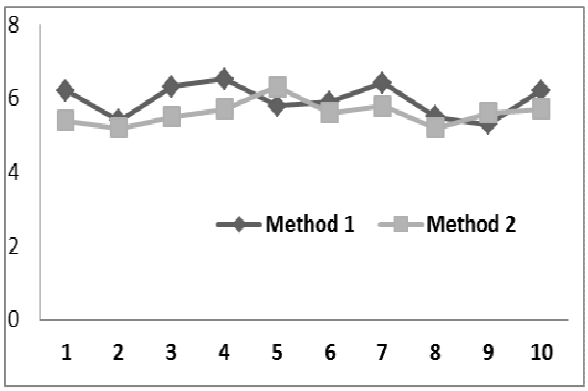

Figure 5: Variation of protein content for 10 samples of whey by both methods of analysis

Method 1*: Innovative method;* ***Method 2****: Kjeldahl method*

Results of analysis of protein and lactose content in the whey

The variation of the lactose content of the 10 samples is the diluted effluent whey by the two methods of analysis varies in a constant manner with a good percentage. For proteins there is a constant variation for both methods.

Figures 2 and 3 summarize the results of analysis of protein and lactose content of whey is determined by two methods. We perform three repetitions for each sample to reduce uncertainty.

DISCUSSION

In Morocco, there has never been reported valuation of by-product of cheese (the whey) that is generally evacuated to the treatment plant without any treatment. This inhibits the dysfunctional step by the presence of organic substances contained in the whey [14].

To address this problem we have tried to develop beginning with the first step to develop the whey from cheese by the recovery of the protein that makes up about 20% of milk protein and has a high nutritional value [15].

In addition to the protein, the whey also contains a significant amount of lactose about 10% [15]. The use of whey to produce value-added products will have a very important advantage economically and environmental [16]. The practice most widespread is the enrichment of cheese milk protein to increase the cheese yield and the production of alcohol from the lactose used as a fermentation substrate. The literature survey shows that the concentration of protein and lactose in the whey is respectively about 6; 45 (g/kg) [17]. To the milk is of the order 35; 49 (g/kg) [18]. Measurements of the concentration of these two components are given by official methods.

For the Protein, Bradford and Lowry [3, 19] have measured the concentration of protein based of the colorimetric and spectroscopic analysis, in this case showed a protein concentration about 6 g/l in the whey.

For the Lactose, official method used for this analysis is that of Allihn-Soxhlet, Spectroscopic [7, 8] shows measurements of the concentration of lactose in the whey of the order of 45 g/kg. Despite the use of these methods for determining the rate of protein/lactose in the both products, the disadvantages present in these methods limits their application.

We recall that the objective of our study is the determination of a new innovative method for the determination of the proteins content and lactose in milk and whey than the official method of analysis has many drawbacks, it is a slow process and requires a lot of time, chemicals and special equipment and can be a risk of leakage of harmful products such as CO_2.To overcome the problems for measuring of protein content and lactose by the official method in our study is adopted a new innovative method that developed in our laboratory, it is an easy and rapid method and does not require a lot of products, which is based on the coupling of three steps, the operations of membrane Ultrafiltration (UF) with measurements of electrical conductivity and gravity measurements of Dry Extract (DE).

For both methods the analysis was verified over a hundred samples of milk and whey, we limit ourselves to presenting the results of ten samples. The figures 2, 3, 4 and 5 confirm the results obtained for both products. The variation curves of protein and lactose in the milk and whey present almost the same curve in the range of 2% of proteins and 1% of lactose determined by the innovative method.

CONCLUSION

The use of the new innovative method has enabled us to respond in part to new data to solve problems of the rate measurements of protein/ lactose and choose the best method.

The new method is based on the coupling of 3 easy steps to implement, fast does not require much time or product and in return gives reliable results. The results of the reliability for this method allow determining the contents of the two components (proteins, lactose) at the same time. This method is applicable for both sample of milk and raw whey and concentrated. Assay results confirm the feasibility and reliability of this new method.

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