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## **Buffalo Milk Characteristics and By-Products**

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**Abstract:** This chapter covers a review of chemical composition and nutrient profile of buffalo milk; several key physical properties (freezing point, surface tension, electric conductivity and thermal stability) are described. Regional manufacturing, utilization and marketing of buffalo milk by-products such as cheese, yogurt, ghee and others are also described and step-by-step flow charts are presented.

**Keywords:** buffalo, dairy, breeds, chemical composition, buffalo milk by-products.

### **1. INTRODUCTION**

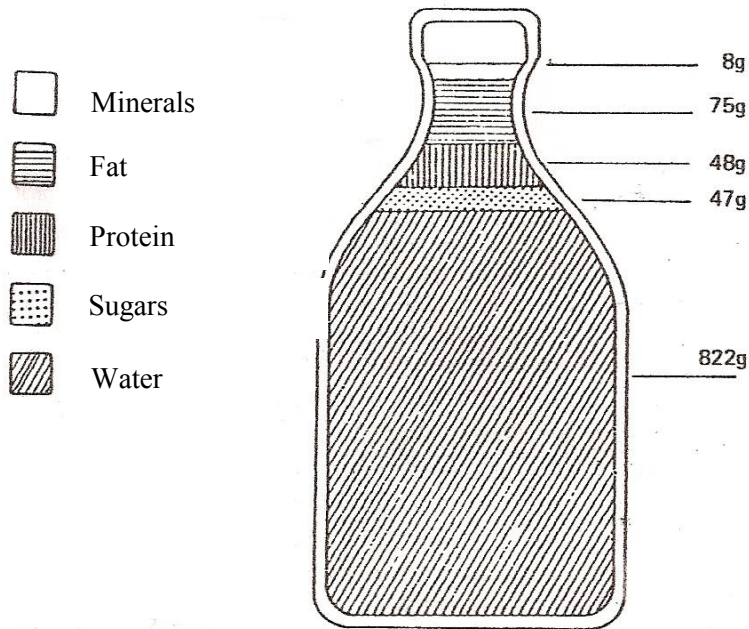
Products derived from fluid bubaline milk, such as butter and ricotta, frescal and mozzarella cheese, have outstanding attributes in terms of product quality and health benefits. Other products, such as fluid milk or ghee, have been adapted by some countries in order to satisfy local market preferences and specific human nutrition demands. In all cases, fluid buffalo milk and its by-products provide excellent nutrient profiles and unique attributes for human nutritional needs; this translates into a wide range of products with high palatability, outstanding cheese yield, and unique health benefits.

### **2. BUFFALO MILK COMPOSITION AND NUTRIENT PROFILE**

Buffalo cow milk quality is considered to be of excellent due to its unique nutrient profile. It is an excellent source of protein, vitamins, minerals and other constituents and it provides calories in countries where the overall supply of essential nutrients is deficient. Bubaline milk is higher in fat, protein, lactose and total solids than milk from cattle (Figure 1). Its fat content is 2.5 to 3 times higher and it provides 40% more calories than its cattle counterpart [1].

Studies conducted in Brazil with Murrah breed showed buffalo milk composition includes conjugated linoleic acids (CLA), considered anti-carcinogen compounds and shown to have beneficial effects on health problems such as obesity, arteriosclerosis and diabetes. This study also demonstrated that supplemental feeding of raw soybean oil further increases CLA content in buffalo milk [2].

## Buffalo Cow Milk Characteristics



**Figure 1.** Average composition of buffalo milk [3].

The main differences between bubaline and cattle milk are:

- Higher butyrose fat (107 % more).
- Lower cholesterol (17 to 24 % less).
- Higher protein (34 % more).
- Higher lactose (19 % more).
- Higher total solid content (48 % more).
- Lower salts (Na, Cl, K).
- Higher overall nutrient values.

Source: [4].

Buffalo cow milk is defined by its nutrient profile; this facilitates its physico-chemical and organoleptic identification. It has higher viscosity, curdling strength and pH [5]. It has characteristic sensory evaluation, slightly sweet flavor and bright white color due to almost complete carotene absence in its fat fraction (3). Buffalo cow milk differences with its cattle counterpart are present also in the colostrum. Environmental factors such as climate conditions, nutrition, breed, management and lactation period influence milk composition. Due to its high casein content, buffalo cow milk is more difficult to digest; in India it is common to add 33% water when the milk is destined for human consumption [6].

The comparative milk composition of cattle and buffalo are shown in Figure 2 [7], whereas changes in composition with lactation stage are shown in Figure 3 [3].

**Figure 2.** Comparative milk composition.

| COMPOSITION      | BUFFALO COW MILK | COW MILK    |
|------------------|------------------|-------------|
| Water            | 84,5             | 87,5        |
| Dry extract      | 16-22            | 11-13       |
| Lean extract     | 7-12             | 6-10        |
| Casein           | 2,8-4,2          | 2,6-3,0     |
| Fat              | 7,0-9,6          | 3,3-4,3     |
| Lactose          | 4,9-5,0          | 4,6-5,2     |
| Total protein    | 4,0-4,6          | 3,0-3,8     |
| Ca (mg/100 g)    | 180-240          | 120         |
| P (mg/100 g)     | 120-140          | 65          |
| Ca/P             | 1.61             | 1.31        |
| Ash              | 0,75-0,85        | 0,80-0,90   |
| Acidity (°SH/50) | 4,2-5,0          | 3,3-3,5     |
| Density (15°C)   | 1.031-1.034      | 1.028-1.035 |
| pH               | 6,6-6,8          | 6,6-6,7     |

**Figure 3.** Changes in bubaline milk composition related to lactation stage.

| Week No.              | Average milk yield | Fat  | Protein | Total solids | Water |
|-----------------------|--------------------|------|---------|--------------|-------|
|                       | <i>Kilograms</i>   |      |         | <i>(%)</i>   |       |
| <b>1. (Colostrum)</b> | -                  | -    | -       | -            | -     |
| <b>2</b>              | 9.06               | 7.77 | 5.58    | 19.06        | 80.94 |
| <b>4</b>              | 10.85              | 6.74 | 4.41    | 17.26        | 82.74 |
| <b>6</b>              | 10.61              | 6.61 | 3.95    | 16.67        | 83.33 |
| <b>8</b>              | 10.31              | 6.30 | 9.82    | 16.23        | 83.77 |
| <b>10</b>             | 9.75               | 6.64 | 9.79    | 16.58        | 83.42 |
| <b>11</b>             | 9.54               | 7.11 | 9.88    | 16.94        | 83.06 |
| <b>13</b>             | 9.25               | 7.15 | 9.93    | 17.17        | 82.83 |
| <b>15</b>             | 8.97               | 7.22 | 4.01    | 17.22        | 82.78 |
| <b>17</b>             | 7.48               | 7.61 | 4.16    | 17.48        | 82.52 |
| <b>19</b>             | 7.95               | 8.01 | 4.20    | 18.07        | 81.93 |
| <b>21</b>             | 7.59               | 8.33 | 4.30    | 18.43        | 81.57 |
| <b>23</b>             | 6.92               | 8.68 | 4.48    | 18.84        | 81.16 |
| <b>25</b>             | 6.55               | 8.80 | 4.50    | 18.94        | 81.06 |
| <b>27</b>             | 6.51               | 8.90 | 4.48    | 19.02        | 80.98 |
| <b>29</b>             | 6.18               | 9.07 | 4.61    | 19.41        | 80.59 |
| <b>31</b>             | 5.65               | 9.10 | 4.66    | 19.63        | 80.37 |
| <b>33</b>             | 5.39               | 9.44 | 4.61    | 19.72        | 80.28 |
| <b>35</b>             | 5.13               | 9.70 | 4.86    | 20.42        | 79.58 |

Fat globules in buffalo cow milk are larger than in cattle counterpart. Therefore, buffalo cow milk fat has higher density and fusion values, a higher saponification and Reickert index; its butiromrefractometer index, Polenske values and Iodine values are lower (Figure 4) [5].

**Figure 4.** Bubaline milk fat analytic constants, compared to cattle milk.

| <b>CONSTANT</b>                   | <b>BUFFALO MILK<br/>FAT</b> | <b>COW MILK<br/>FAT</b> |
|-----------------------------------|-----------------------------|-------------------------|
| <b>Butiro-refractometer index</b> | 42.0                        | 41.2                    |
| <b>Saponification value</b>       | 230.1                       | 227.3                   |
| <b>Reichert value</b>             | 32.3                        | 26.6                    |
| <b>Polenske value</b>             | 1.4                         | 1.7                     |
| <b>Kirchner value</b>             | 28.5                        | 22.1                    |
| <b>Iodine value</b>               | 29.4                        | 33.8                    |
| <b>Fusion point</b>               | 32.0 – 43.5                 | 28.5 – 41.0             |

Analyzing its nutritional value (Figure 5), bubaline milk shows higher resistance to damage due to oxidative conditions, and it is richer in butyric acid and long chain fatty acids such as palmitic and stearic acids. It has higher content of unsaturated fatty acids, and lower content of intermediate-length fatty acids (from C<sub>6</sub> to C<sub>12</sub>). Bubaline milk has higher total and colloidal calcium content, as well as iron and phosphorus. Usually, bubaline milk has more cation than anion concentration, compared to vaccine milk [5].

**Figure 5.** Nutritional values in selected mammals.

| <b>NUTRIENT</b>        | <b>BUFFALO COW</b> | <b>COW</b> |
|------------------------|--------------------|------------|
| <b>Fat (g)</b>         | 6.9                | 3.3        |
| <b>Protein (gm)</b>    | 3.7                | 3.2        |
| <b>Calcium (mg)</b>    | 188                | 113        |
| <b>Iron (mg)</b>       | 0.1                | 0          |
| <b>Phosphorus (mg)</b> | 117                | 97         |

Buffalo cows metabolize all nutritional carotene to vitamin A, which is then transferred to milk, making it readily accessible to the neonate [8]. A study conducted by Ezequiel Patiño, in UNNE (Corrientes University), Argentina, on 960 samples from 40 buffalo cows described in physico-chemical description of bubaline milk for pasture-based systems (Figure 6) [3].

**Figure 6.** Physico-chemical composition of buffalo cow milk in pasture-based systems.

| <b>TRAIT</b>              | <b>MEAN</b> | <b>SE</b> |
|---------------------------|-------------|-----------|
| <b>Density (g/ml)</b>     | 1,0307      | 0,0039    |
| <b>Acidity (° Dornic)</b> | 19,65       | 2,96      |
| <b>pH</b>                 | 6,71        | 0,16      |
| <b>Total solids (%)</b>   | 16,35       | 2,42      |
| <b>Fat (%)</b>            | 7,22        | 1,89      |
| <b>Protein (%)</b>        | 3,85        | 0,92      |
| <b>Lactose (%)</b>        | 4,49        | 0,24      |
| <b>Ash (%)</b>            | 0,83        | 0,08      |

Among all the characteristics studied, titrable acidity (19,65 + - 2,96 ° Dornic) was the trait that exhibited the most variability during lactations. Bubaline milk has a higher value of titrable acidity than cattle milk due to its higher casein content [3]; therefore, the threshold parameters used in the industry for cattle milk in Argentina (13° to 18° Dornic) are not applicable to buffalo cow. The increase in titrable acidity in the last phase of lactation coincides with a drop in milk output and with an increase in total solids due to this volume reduction and consequent protein concentration [3].

The milk used for mozzarella cheese manufacturing, must have a 2:1 fat protein ratio, a minimum fat content of 7.2% and a low value of titrable acidity. Titrable acidity values in Italian Mediterranean breed fluctuate during lactation from 12.0 to 9.0 SH° (Soux-Henkel units/ 100 ml milk) during the first 25 days after calving. Values start at 12.0 at the initiation of lactation, 10.0 by two weeks and drop to 9.0 after 25 days. Conversion used to express acidity levels in ° SH and ° Dornic (°D) can be obtained using the following formulae [3]:

- Milk acidity (°D) = (4.5 x acidity expressed in °SH)/2
- Milk acidity (°SH) = (2 x acidity expressed in °D)/4,5

Normal acidity in Italian Mediterranean breed ranges from 7.0 to 7.8 °SH (Figure 7) [3]. The factors affecting acidity values include a) presence of saprophyte bacteria producing lactic acid through lactose fermentation, b) conservation of milk in dirty refrigerators and slow cooling, c) transport in dirty containers and other contaminants, d) long transportation periods, e) unsanitary milking procedures, f) inadequate foodstuffs (too coarse, moldy or unbalanced mineral mixes) [9].

**Figure 7.** Acidity, pH, density and cryoscopic characteristics of buffalo cow milk.

| LACTATION PERIOD<br>(month) | PHYSICO-CHEMICAL<br>DETERMINATION |      |         |               |
|-----------------------------|-----------------------------------|------|---------|---------------|
|                             | Acidity<br>(°D)                   | pH   | Density | Cryoscopy(°H) |
| 1                           | 20,9                              | 6,86 | 1,0361  | - 0,573       |
| 2                           | 16,2                              | 6,97 | 1,0344  | - 0,579       |
| 3                           | 17,1                              | 6,94 | 1,0344  | - 0,580       |
| 4                           | 18,6                              | 6,91 | 1,0336  | - 0,575       |
| 5                           | 18,9                              | 6,94 | 1,0327  | - 0,580       |
| 6                           | 19,1                              | 6,94 | 1,0337  | - 0,576       |
| 7                           | 18,6                              | 6,90 | 1,0334  | - 0,577       |
| 8                           | 19,2                              | 9,88 | 1,0325  | - 0,574       |
| 9                           | 19,6                              | 6,87 | 1,0326  | - 0,577       |

°D = Dornic degrees; °H = Hortvet degrees

Colostrum is produced by the mammary gland during the early hours post-partum. It has significant higher total solids concentration compared to milk produced during the rest of the lactation (Figure 8). All milk major constituents, together with the minerals (mainly calcium, phosphorus and potassium), and fat-soluble vitamins, are present in bubaline colostrum in higher concentrations compared to cattle. It also contains antibodies that protect young buffalo calf against infections [3]. In order to provide adequate immunity to the calf, it is recommended to ensure colostrum intake of at least 2 liters/calf, within 3-6 hours after birth [10].

**Figure 8.** Mammary gland secretion during the first days after calving.

| <b>TOTAL MILK PERCENTUAL (%)</b> |            |                |                     |                |                  |
|----------------------------------|------------|----------------|---------------------|----------------|------------------|
| <b>Days after calving</b>        | <b>Fat</b> | <b>Protein</b> | <b>Total solids</b> | <b>Lactose</b> | <b>Vitamin A</b> |
| <b>Day 1</b>                     | 9.55       | 9.59           | 26.6                | 7.54           | 1.837            |
| <b>Day 7</b>                     | 7.61       | 5.55           | 18.9                | 4.41           | 0.280            |

Buffalo cow milk has been shown to be a healthy nutrient for humans. It has 58% more calcium, 35% more protein and 20% less cholesterol than cow milk. It is also a richer source of iron, phosphorus, vitamin A and natural antioxidant tocopherol. Peroxidases are part of the mammal's biological defense against pathogens. And peroxidase activity in buffalo cow milk is 2-4 times higher than that in cow milk. Bubaline milk is a perfect option for individuals that are allergic to cattle milk. Larger proportions of other bio protective compounds such as, immunoglobulins, lactoferrin, lysozyme, and lactoperoxidase make buffalo cow milk an excellent choice for a wider range of healthy diets [8].

### **3. CHEMICAL CONSTITUENTS AND PHYSICAL PROPERTIES OF BUFFALO MILK**

#### **3.1. Chemical constituents**

Major constituents of buffalo cow milk are those present in larger amounts: fat, protein and lactose. Minor constituents are present in relatively smaller proportions; those are minerals, vitamins, non-protein nitrogen compounds, pigments, enzymes and others. However, these minor constituents may be so important as the major. Some of them have a fundamental nutritional role, namely calcium, phosphorus and some vitamins (Figure 9) [1].

**Figure 9.** Average values and variation of milk constituents obtained from 24 buffalo herds of Italian Mediterranean breed.

| <b>CONSTITUENT OR PHYSICAL PROPERTY</b> | <b>MEAN VALUE</b> | <b>RANGE</b>  |
|---|-------------------|---------------|
| <b>Specificgravity</b>                  | 1.031             | 1.029 – 1.033 |
| <b>Fat (%)</b>                          | 8.5               | 7.1 – 9.6     |
| <b>Total solids (%)</b>                 | 18.9              | 16.8 – 20.8   |
| <b>Ash (%)</b>                          | 0.84              | 0.79 – 0.90   |
| <b>pH</b>                               | 6.63              | 6.55 – 6.71   |
| <b>Total nitrogen (%)</b>               | 0.710             | 0.571 – 0.809 |
| <b>Nitrogen-derivedcasein (%)</b>       | 0.572             | 0.437 – 0.654 |
| <b>Non-proeicnitrogen (%)</b>           | 0.031             | 0.009 – 0.536 |
| <b>Lactose (%)</b>                      | 4.6               | 4.0 – 5.1     |
| <b>Calcium (%)</b>                      | 0.203             | 0.179 – 0.241 |
| <b>Magnesium (%)</b>                    | 0.020             | 0.0126- 0.027 |
| <b>Potassium (%)</b>                    | 0.139             | 0.085 – 0.187 |
| <b>Sodium (%)</b>                       | 0.075             | 0.047 – 0.102 |
| <b>Phosphorus (%)</b>                   | 0.129             | 0.118 – 0.139 |
| <b>Chlorine (%)</b>                     | 0.065             | 0.056 - .084  |
| <b>Citricacid (%)</b>                   | 0.219             | 0.158 – 0.290 |

(%) = g / 100g milk

Fat globules in buffalo cow milk not only contain a spectrum of triglycerides but also small amounts of liposoluble substances such as cholesterol, vitamin A, D and  $\alpha$ -tocopherol. The high fat content present in buffalo cow milk gives it its unique flavor and makes it a particularly valuable commodity in India and other countries where ghee, a type of clarified butter, is considered an important dairy product. This clarified butter, produced over low heat has excellent conservation properties without the need for refrigeration. In India, diluted buffalo cow milk for human consumption is preferred over cow's milk [1].

Although similar, proteins in bubaline milk are not identical to those in cattle. Buffalo cow milk is higher in casein and serum albumins. Most bubaline casein (98.4%) is present in the form of large micelles (80 a 250 nm vs 70 a 110 nm in cattle). Buffalo cow milk has higher lactoferrin content. This glycoprotein plays a key role in infant protection by inhibiting bacteria such as *Escherichia coli* sp., and also plays an important role in iron absorption [8].

Some of the minor constituents such as non-protein nitrogen (NPN) of the buffalo cow milk have no major incidence in human nutrition. NPNs are compounds related to proteins without being real proteins although they are involved in some protein reactions. Some NPN compounds are present as free amino acids in small and variable quantities. Of these, tyrosine is present at a level of 46 mg/liter and does not vary significantly with lactation period. When milk is stored, this value tends to increase slowly, possibly due to enzymatic hydrolysis of protein (Ferrara et al., 1964). Urea is an important compound: it means the 40-50% of total NPN. Other compounds exist in small quantities: creatine, creatinine, uric acid, ammonia and those hydro soluble vitamins that have nitrogen. Urea nitrogen represents 44.8 and 41.2% of total NPN in the cow and in the buffalo cow, respectively. Creatinine nitrogen is nearly equal in cow (11.7 mg/liter) than in buffalo cow (9.95 mg/liter) milk. Uric acid-derived nitrogen is 5.42 mg/liter in cattle and 1.9-2.4 mg/liter in buffalo cow, and 2.5 a 2.9 mg/liter for cattle and bubaline ammonia-derived nitrogen. Some of these products are mostly considered excreted rather than secreted products. Although urea is an excreted product, it can be utilized by ruminants as a source of nitrogen by microbial conversion and further metabolized via saliva, food and bloodstream.

Enzymes are minor constituents of milk; two of them have commercial significance in fluid milk: peroxidase and alkaline phosphatase. Buffalo cow milk has two to four-fold higher peroxidase activity than cattle's. The optimal pH for enzymatic activity in buffalo cow milk is 7.0 (6.5 for cattle). Only 6'' at 85° C are needed for peroxidase inactivation [11]. Other minor constituents are the minerals, including macro and micro-nutrients. Calcium, very important for human consumption, is a macronutrient needed for adequate bone formation during growth of buffalo calf, and is needed for milk coagulation in the neonate's stomach. It is also industrially important, in the first steps of cheese manufacturing. Under similar conditions, bubaline milk coagulates faster than cattle, even when diluted to double its volume [11]. Average calcium content of buffalo cow milk is 0.18-0.20%, with considerable variations out of these values towards the beginning and end of lactation, and in some animals [1].

Phosphorus, also essential to bone and teeth formation is present in bubaline milk as free phosphate, bound to other elements or in form of esterified casein or hydro soluble esterified phosphate. It may also be present as part of the phosphatide molecule, ionized or in its inorganic form. Although all of these fractions vary, average phosphorus content in bubaline milk is 0.12 – 0.13% (versus 0.10-0.12 in cattle) [1]. Total mineral constituents (total ash) are approximately 0.79% in bubaline milk, versus 0.75% in cattle. Sodium, potassium and chlorine levels are much higher in cattle than bubaline milk. Lactose content is higher in cattle's milk.

Regarding microelements, iron and copper are needed in red blood cell formation as well as for other tissues. These levels of these elements are generally low in bubaline milk and must be provided through supplementation to ensure adequate growth of newborn calves. Other



microelements are present in small quantities in the mammal milk, usually combined or associated with one or more proteins.

Values obtained for buffalo cows in the middle of lactation, expressed in parts per million, are shown in Figure 10 [11]:

**Figure 10.**Microelementprofile of buffalo milk

| <b>ELEMENT</b>    | <b>VALUE (ppm)</b> |
|-------------------|--------------------|
| <b>Iron</b>       | 1.01               |
| <b>Copper</b>     | 0.22               |
| <b>Zinc</b>       | 2.39               |
| <b>Aluminum</b>   | 0.22               |
| <b>Manganese</b>  | 0.17               |
| <b>Lead</b>       | 0.16               |
| <b>Molybdenum</b> | 0.022              |
| <b>Silicone</b>   | 0.21               |

With the exception of aluminum, silicone and lead, all of the above elements are considered essential. All of these elements are high during the first month of lactation, progressively decline towards mid lactation and tend to slightly increase by the end. Other minor constituents present in trace amounts but also considered essential nutrients are the vitamins. Most vitamins (vitamin A, Riboflavin, Vitamin C, etc.), with the exception of tocopherol, carotene, and probably vitamin B12, are found in lower concentration in bubaline milk compared to cattle counterpart. It was determined content of vitamin B complex for buffalo cow milk in mid lactation [11]:

**Figure 11.**Vitaminprofile of buffalo milk

| <b>VITAMIN</b>                    | <b>CONTENT (mg/l)</b> |
|-----------------------------------|-----------------------|
| <b>Thiamine (B<sub>1</sub>)</b>   | 0.81                  |
| <b>Riboflavin (B<sub>2</sub>)</b> | 1.67                  |
| <b>Biotin</b>                     | 2.02                  |
| <b>B<sub>6</sub></b>              | 0.25                  |
| <b>Nicotinic acid</b>             | 0.82                  |
| <b>Pantothenic acid</b>           | 2.02                  |
| <b>B<sub>12</sub></b>             | 4.3 (ug/l)            |

The most apparent visual difference between buffalo cow and cattle milk is the yellowish color of the later due to its higher carotene content. In the bubaline milk the vitamin A level is very superior, giving to it the white color. It is suspected the buffalo cow has an efficient metabolic mechanism that prevents pigments to appear in milk, whatever the carotene content of the diet may be [1]. Vitamin C (ascorbic acid) content is higher in buffalo cow milk (Figure 12) [11].

**Figure 12.** Vitamin content reported for buffalo, domestic cattle and human milk.

| <b>SPECIES</b>      | <b>VITAMIN C VALUE (mg/l)</b> |
|---------------------|-------------------------------|
| <b>Buffalo</b>      | 19.5 – 39.5                   |
| <b>Domestic cow</b> | 7.1 – 7.8                     |
| <b>Human</b>        | 11.3 – 27.0                   |

Another minor component of bubaline milk is citric acid. This is an important constituent because of it is part of non-ionic calcium, which in turn plays a role in curd formation (coagulation). Is present in important quantities, but values reported by different groups of researchers vary; some indicate buffalo cow may have higher content than cattle, while others indicate the opposite. Values

reported for buffalo cow range between 0.180 and 0.224%. Colostrum contains quite more citric acid than the milk produced in the middle of lactation.

### ***3.2. Physical properties***

Buffalo cow milk has a slightly sweet flavor; it is more viscous and whiter than cattle milk due to the absence of carotene in its fat globules. Just as in dairy cow, physical properties exhibit less variability around mid-lactation compared to chemical constituents.

Several studies have reported pH of fresh bubaline milk, with great variation among individuals. In India it was obtained fresh milk from Murrah buffalo cows and reported pH values of  $6.74 \pm 0.08$  (vs. Cebu cattle  $6.63 \pm 0.08$ ). In Italy it was reported an average  $6.78 \pm 0.03$  for fresh milk from Italian Mediterranean buffalo cows, with marked seasonal fluctuations of 6.73 in August (summer) and 6.85 in December (winter). All values were higher in buffalo cow milk. Bubaline and cattle milk tend to lose continuously carbon dioxide and to slightly increase its pH overtime. Subsequently, pH drops due to acid lactic produced by bacterial activity. A mastitis-infected animal will exhibit elevated pH values ( $>7.0$ ) in the infected quarter. The pH of fresh bubaline milk does not appear to be linked to daily output, stage or number of lactation [1, 11].

Specific gravity is another physical constant of fresh bubaline milk. Average herd values are between 1.030 y 1.032, although individuals with wider ranges may be found (versus 1.029 to 1.033 in cows). Low values are generally associated with higher fat percentages [1].

Freezing point of any biological fluid is found below freezing point of pure water. Freezing point depression is still utilized as an indicator of dilution of milk with water. Average freezing point for whole milk is almost the same for buffalo cow and cattle,  $0.544\text{ }^{\circ}\text{C}$  and  $0.545\text{ }^{\circ}\text{C}$ , respectively. Colostrum has a higher depression than milk ( $0.57 - 0.58\text{ }^{\circ}\text{C}$ ). It is generally assumed that any bubaline or cattle milk with a depression of  $0.530\text{ }^{\circ}\text{C}$  or lower should be considered altered [1, 11].

Surface tension of fresh buffalo cow milk is similar to that of cattle, although values may vary due to measurement techniques. On average, bubaline milk is 48.74 dynes/cm and cattle 49.36. Surface tension is inversely affected by temperature [1].

Electric conductivity is affected by chlorhydric acid content; this component tends to increase in mastitis-affected quarters. Average value for “healthy” samples  $6.62\text{ }^{\circ}\text{milimohs}$  for buffalo cow and  $6.69$  for cattle [11].

Fluorescence: When subjected to ultraviolet light, cattle’s milk turns canary yellow color, whereas bubaline’s turns white. When milk is heated to its boiling point, fluorescent color should not be affected. This color does change in the milk that have been adulterated or diluted with water [11].

Curding time: Under similar temperature and concentration conditions, bubaline milk coagulates more rapidly than cattle; this coagulation time may be reduced by adding hydrochloric acid. Noteworthy, due to its high calcium content, buffalo cow milk stability at ambient conditions (Corrientes, Argentina) is lower than cattle, with slower coagulation times [11].

## **4. ELABORATION OF BUBALINE CHEESE AND OTHER BY-PRODUCTS: DESCRIPTION OF MANUFACTURING PROCESS**

### ***4.1. Manufacturing process of white, bubauno and yearling buffalo cheese (Venezuela) [12].***

### **FIRST STAGE (same process for all three cheese types)**

Raw milk (5°C)  
Heating (40°C)  
Partial skimming  
Polyvalent tank (36 -37 °C)  
Addition of rennet (2.5 gr/100 l.)  
Agitation (5 minutes)  
Coagulation (40-45 minutes)  
Curdling  
Setting period (5 minutes)  
Partial removal of whey (Flor de cuajada)

### **SECOND STAGE**

#### ***White cheese***

Addition of salt (1.2% weigh: volume)  
Place in cheese molds  
Pressing  
Storage  
Marketing

#### ***Bubauno cheese***

Heat curd (50°C 10 minutes)  
Place in wicker molds  
Addition of salt (rubbing)  
Maturation at 18°C, skin is rubbed with vegetable oil  
Marketing

#### ***Yearling cheese***

Place in molds  
Pressing  
Salt addition in brine (24%)  
Maturation at 18°C, skin is rubbed with coffee  
Marketing

## ***4.2. Manufacturing process for hand-pulled cheese (mozzarella and provolone) [12]***

### **FIRST STAGE (same process for all cheese types)**

Raw milk  
Heating (38°C)  
Coagulation starter (1%)  
Addition of rennet (2.5 gr/100 l.)  
Coagulation (50-60 min)

Curd cutting (size of a corn kernel)  
Setting period (5 minutes)  
Removal of whey  
Curd kneading point  
Dough kneading (in water at 85°C)

## **SECOND STAGE**

### ***Mozzarella cheese***

Place in molds  
Container with water (18°C)  
Brine or sauce (salsetta)  
Packaging  
Storage and/or marketing

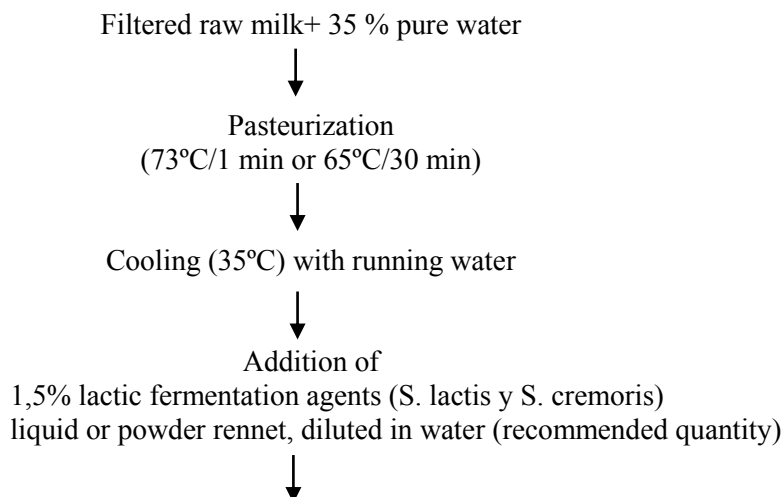
### ***Provolone cheese***

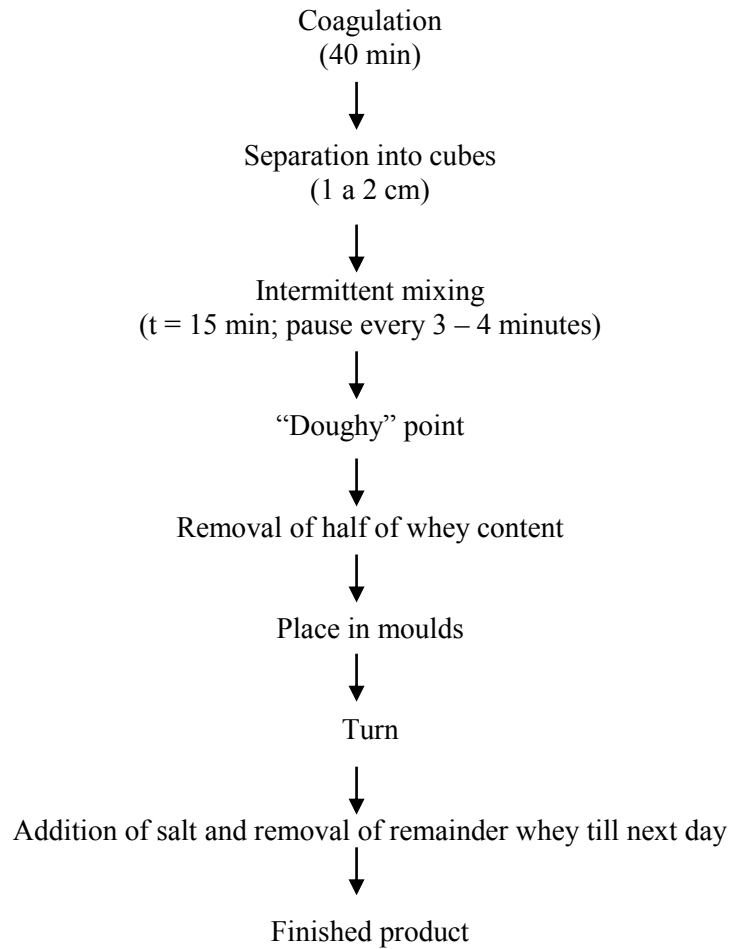
Place in molds  
Brine  
Maturation  
Storage and/or marketing

### ***4.3. Production of criollo (creole) cheese (Brazil)***

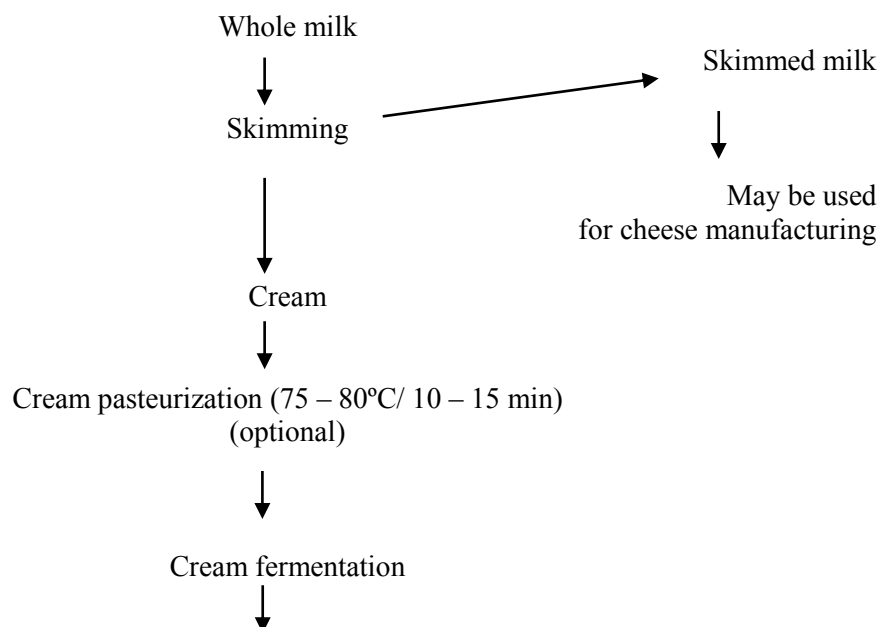
The production of “criollo” or creole cheese (“frescal” for Brazil) starts with milk pasteurization. This can be accomplished by ultra-pasteurization (with the sheet equipment), 72°C for 18”, or traditional pasteurization at 65°C for 30’. Temperature must then be lowered to 35°C, fermentation agents must be added and after 20’ rennet must be added (80 % pepsin and 20 % chymosin). Separation into curd and whey should be noticeable after 25-40’. Curd is then dislodged manually into pieces no bigger than 1 cm<sup>3</sup> (cubes). After this, 50% of whey is withdrawn and the remainder curd plus whey are placed into molds, which are then turned and salted. Product is packed in sealed plastic containers (not vacuum sealed) and kept in cold room for 24 h, after this period the product is ready to be consumed [13].

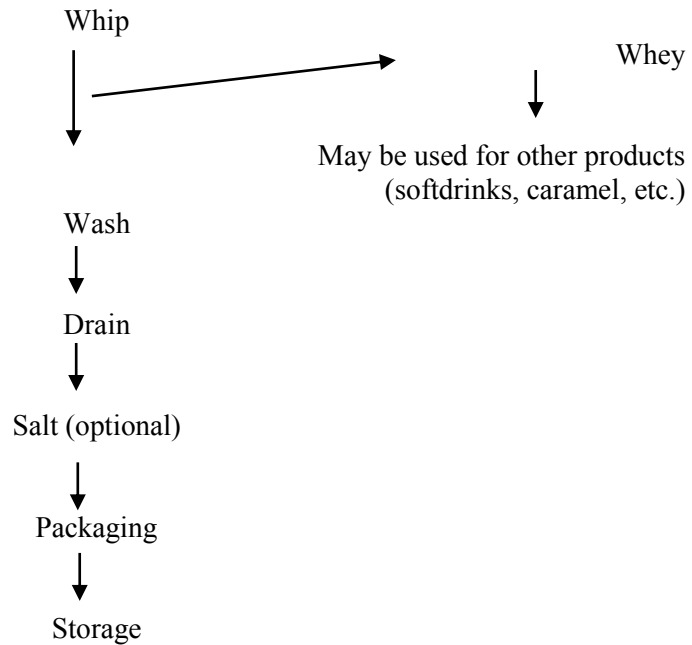
### ***Schematic representation of production process of “criollo” cheese (Brazil: “Queijo Minas Frescal”) [13]***





***Flow chart for butter production (Brazil)***





FLOW DIAGRAM – Butter Elaboration [13]

#### ***4.4. Provola affumicata cheese***

In Italy, buffalo milk is also used to produce "provola affumicata", a smoked version of mozzarella with an increasing demand in the country. It is produced by burning rice or other type of straw to produce smoke under milk containers, while covering the top with a wet cloth in order to trap humidity [13].

#### ***4.5. Mozzarella di Bufala Campana***

##### ***4.5.1. Definition***

The Italian Buffalo Breeders Association (ANASB: Associazione Nazionale Allevatori della Specie Bufalina) defines the "Mozzarella di Bufala Campana" (that means from the "terroir" of Campania Region) in this way: "It is a fresh stretched and kneaded cheese, produced with milk from buffalo cows raised in the region of denomination D.O.P. (Protected Denomination of Origin) and also processed in dairy plants within that same region. The product may have several shapes and presentations that range from 10 to 800 gr. It has ivory color, a thin, smooth crust of 1mm. It is composed of thin multi-layers, slightly elastic within 8-10 h post-threading and becoming softer overtime. It should have no holes and must be produced without conservatives, inhibitors or coloring agents. It should have slight runoff of creamy whey when sliced; scent of lactic fermentation should be noticeable". According to current Italian law, preparation of mozzarella can only be done with buffalo milk.

Milk quality is of utmost importance; animal health, sanitary handling, milking process and storage hygiene are key aspects of the final quality of the product. Under normal conditions, buffalo cow milk contains higher quantity and specific strains of lactobacillus, compared to cattle's. From the metabolic activity of those strains come the particular flavor and aroma, through the production of special substances and composites, which probably have a remarkable influence in the curd acidification during the transformation. Because mozzarella manufacturing is still done using little or no mechanization (handmade), hygiene, sanitary conditions of storage and transport of the milk are essential. Microbial contamination screenings are conducted periodically year-round. Both

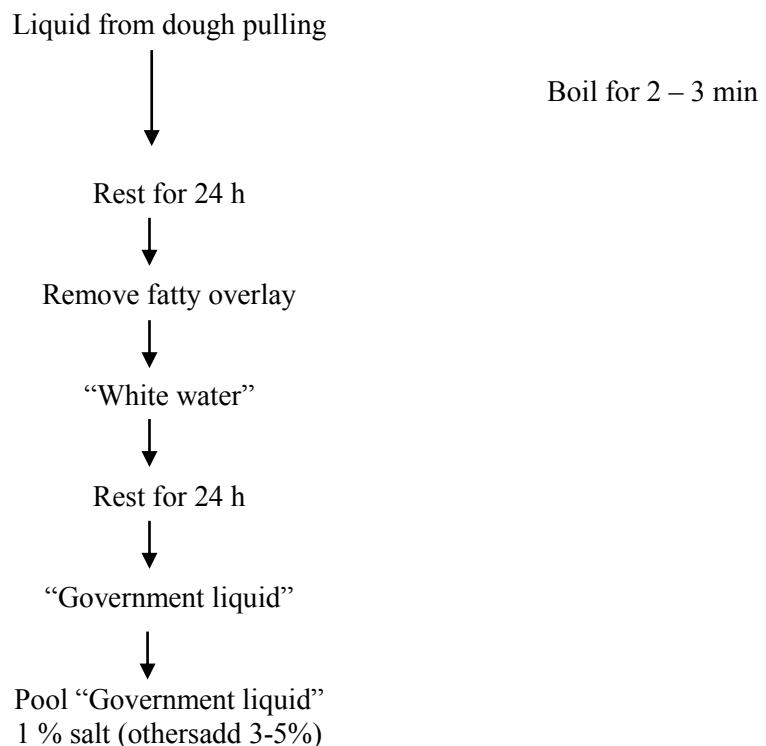
microbiological and hygienic milk qualities are essential in mozzarella elaboration. The manufacturing of mozzarella has been termed a “forward-flow” process; this type of production prevents cross contamination of starting materials [7].

#### **4.5.2. Production examples and aspects of mozzarella cheese production**

“Maina della Torre” Dairy Plant, owned by Prato Verde Factories, is located in Latina Province, Lazio Region, Italy. This plant only processes the milk needed to produce mozzarella within the same day. Their production inputs are: milk, enzymes and whey. After 4-5 hours, once curd and whey are separated, they cut the curd into small cubes and place them in a wooden container with water at 98° C for the handmade stretching and kneading process, where the dough will reach a temperature of 66-67°C. They produce 35-40 kg of cheese per batch. Simultaneously, they obtain the “piccolo latte” or “small milk”, a by-product from the remainder liquid. They have capacity for 5000 liters, although in winter they only process 2500 liters due to reduced demand. Mozzarella cheese yield is 1 kg of cheese of every 4 liters of milk (25% yield). Freshly made mozzarella is placed in salt-water bats (15% salt) for 2 hours; it is then transferred to a 1.5% salt-water solution, called “government liquid”.

La Salamandra SA, in Buenos Aires, Argentina, prepares their “government liquid” by adding 12° g citric acid and 320 g salt to 450 liters of water [14].

#### **4.5.3. Liquids used in mozzarella handling and conservation [13]**



#### **4.5.4. Mozzarella cheese yield**

One hundred kg of buffalo milk will render from 20 (in hot days) to 25 (in cold days), sometimes even 27 kg of mozzarella cheese. In contrast, cattle’s milk yield is 9-12% (100 liters of milk will yield 9-12 kg mozzarella). Ricotta cheese is produced by addition of 3% whole milk to whey. 100 kg of whey renders 1 kg of fresh ricotta. Butter is produced with the fat skimming’s of whole milk; this skimming will reduce the weight of the mozzarella cheese, and therefore is not traditionally done [15].

$\text{Mozzarella (kg)} = \text{milk (kg)} \times [(3,5 \times \% \text{ protein}) + (1,23 \times \% \text{ butirose fat}) - 0,88] / 100$

It must be noted that this formula was obtained from Italian elaboration standards, which accepts considerable humidity content. It is not known the extent of applicability of the formula under other conditions in other countries. Based on the formula, for each 1% increase in milk protein results in a threefold increase in cheese output. Noteworthy, it is not protein content but nitrogen level in milk that is usually routinely measured, since is contained mainly in the proteins with a ratio known to be relatively constant [15].

#### ***4.5.6. Other aspects of the mozzarella cheese production chain***

Buffalo mozzarella is considered to be of higher quality than cattle's. When the product is melted, it results in a continuous thread without clumps. In cattle's mozzarella, there is presence of clumps and strong "cheese" aroma. There are continuous fluctuations in milk protein and fat content throughout the year, particularly due to the pooling of milk from buffalo cows in different stages of lactation. Milk from buffalo cows in the first stage of lactation can make up over 60% of total volume. This results in increased acidity and presents a problem for mozzarella cheese manufacturing; namely, shortened shelf-life and poor organoleptic attributes of the resulting product. Management errors such as energy and protein nutrient deficiencies, excess silage in ration during the first stage of lactation can worsen this problem because the buffalo cow will attempt to balance the nutrient imbalance by mobilizing energy stores with the consequent increase in circulating non-esterified fatty acids.

The tropical origin of the bubaline species implies poor adaptability to low ambient temperatures and wind chill; these factors can lead to an increase in milk acidity particularly in periods of transition from warm to cooler temperatures. The increase in milk acidity is a multivariate phenomenon which can be attributed to one or several factors: a) mesophyll cell count, b) chemical composition of the diet, c) poor milk handling and storage, d) poor milking hygiene leading to higher microbial counts, higher lactose fermentation and higher lactic acid production [16]. It is therefore extremely important to ensure hygiene and product quality throughout the whole cheese production chain. This is particularly true for Italy, which has stringent product quality standards for mozzarella [17]. Buffalo mozzarella consumers are demanding clientele (particularly in Italy); cheese must arrive to markets within the day of productions, while other Italian cities have 2-3 day tolerance standards. Cheese is normally transported in brine, although vacuum-packed cheeses are now considered acceptable.

#### ***4.5.7. Process for reception, manufacture and packaging for mozzarella cheese***

As mentioned previously, mozzarella cheese production is a "forward-flow process", where there is no crossing of starting materials and therefore low risk of cross-contamination [13,18,19].

The complete process for the manufacture of soft cheeses of threaded dough, such as "mozzarella di bufala" is detailed below :

- 1) **Reception of raw milk:** milk arrives in stainless steel milk transport trucks; it has been previously cooled in the farm and shipped within a short period of time to the plant. Transport temperature should not surpass 10° C, with the exception of those farms in which the time lapse between milking and transport is under 2 hours. Milk should be kept at 0 y 4°C.
- 2) **Sanitation:** Milk is sanitized by simple filtration through fine meshes in order to separate coarse contaminant materials; it is then centrifuged at 5000 rpm to separate smaller impurities.
- 3) **Pasteurization:** typically, pasteurization is done at 68-70° C, for a period of 15-20 seconds. In the case of "Mozzarella di Bufala Campana D.O.P.", treatment is conducted in a plate-type thermal exchange system, at 72° C for 15 seconds. In all cases, the thermally-treated milk must be negative to alkaline phosphatase test. Higher temperatures or longer treatment times pose risk of excessive seroprotein precipitation with casein entrapment and



consequent reduction in stretching and kneading properties. The objective of pasteurization is the elimination of contaminating bacteria. However, the process also reduces the content of naturally occurring casein-utilizing microorganisms; it is then necessary to add lactic ferments or commercial yeast to counteract the depleting action of the thermal treatment. In order to maintain good spreading and kneading quality of the dough, handicraft or “artisan” production of mozzarella cheese does not involve pasteurization. This requires that the mesophyll cell count of the buffalo milk used for such process is under 500.000 colony forming units/ml. In addition, originating farms must be Brucellosis and Tuberculosis free. In Brazil, Oliveira recommends no pasteurization for mozzarella cheese production due to its detrimental effects on the protein matrix [13]. It must be noted that most South American farms are pasture based, as opposed to dry low/stable confined; mesophyll counts in these production systems are typically lower [20].

- 4) **Fermentation starters:** milk is heated in stainless steel bats at 36 a 39° C, and lactic fermentation starters from previous batches are added (3-5 liters/100 liters of buffalo milk) (Garofalo – ANASB - 2-3 liters.). The amount of added product should be enough to elevate milk’s pH to 6.3 or 11-12°SH acidity. Most natural fermentation agents contain some thermophile lactobacillus (*Lactobacillus delbrueckii* Subsp. *bulgaricus*), cocci (*Streptococcus thermophiles* and other mesophylls) and large yeast quantities, mostly responsible for the aroma in mozzarella. Ferments are obtained from the fermentation of sweet whey, by heating to 40 - 42° C followed by acidification to 17/18 °SH/50 and 4.3 pH and cooling 8° C.
- 5) **Coagulation:** Rennet is used as the main coagulating agent; it is added at a rate of 1:12.000 -1:15.000 parts as is, diluted in warm water. The amount of rennet added should cause milk coagulation within 15-20 minutes. Rennet is obtained from the inner mucosa of the abomasum of calves; it is rich in chymosin and pepsin. Milk coagulates and forms a curd due to a destabilization process of the calcium micelles, which occurs in response to the low pH and the enzymes present in rennet.
- 6) **Curd separation:** Curd separation is observed when coagulated material starts detaching from the container walls, this happens when whey has reached a 7,5-8,0°SH acidity. The classic tool to dislodge curd is the “herringbone” mixer and sometimes the “lyra-shape” mixing aid. Mixing should be done slowly and in circular movements in order to gently dislodge and break the curd into pieces.
- 7) **Whey fermentation:** After allowing curd to sediment, 2/3 of whey are separated (which can be used for ricotta cheese manufacture), the remaining whey is kept with the curd to promote fermentation.
- 8) **Straining:** Once curd is set (pH close to 5) it is then removed from stainless Steel bats and placed on stainless steel tables in order for remaining whey to drip (3-4 hours).
- 9) **Curd mincing:** Curd is minced in specialized equipment with rotating blades, producing pieces of 2 x 2 cm.
- 10) **Stretching and kneading:** It can be done in a wooden container, with a characteristic wooden stick. It can also be done automatically. In both cases, boiling water (100° C) in added to allow handling of the minced curd. Optimal stretching and kneading temperature range between 68-70° C. The dough should never reach temperatures higher than 39° C and the process should only last 4-5 minutes. Ideal maturation of the dough is normally with pH values between 4.90 and 5.10. It is recommended to start kneading with a pH 5.2 and finish with pH 4.8 (18). Garofalo described subtle variations to this process [19]: “Stretching and kneading is the step in the manufacturing process that most greatly affects the finished product and its yield over liter of milk. Immature or overly matured curds result in loss of consistency and 2-3% reduction in overall cheese yield. Senior cheese masters are able to empirically predict the optimal point for the initiation of stretching and kneading process. Research conducted on the relation between kneading process and curd acidity demonstrated an optimal value of 4.8-4.9, which resulted in minimal weigh loss in brine storage. Currently, there are several mechanical alternatives to accomplish the kneading process; all of them simulate the man-made movements of the dough. Kneading consists of

submerging the curd in hot water (80-85°C) for it to acquire an elastic consistency that allow the operator to mould the dough into different shapes. The dough is kept in the kneading equipment for 15-20 minutes, approximately. In case of handmade kneading, this is done in a wooden container (45 cm height, 70 cm base diameter, 90 cm rim diameter); the dough is gently stirred with a wooden stick and a bowl, while raising it in order to incorporate water rendering a smoother, more shiny and silkier final product. Because of the limited time in small plants, is better not to knead more than 30 kg of dough with 45 litre of water. After the necessary period of time dough is carried to the shape phase.

- 11) **Cutting or “mozzatura”**: This step can be done manually or mechanically, with equally satisfactory results. Cutting of mozzarella by hand is done with two operators; one of them holds large pieces of dough (3-4 kg) while another, using index and thumb fingers separates and pinches (“mozza”) 20-800 g pieces. Mechanization of his process results in more uniform pieces.
- 12) **Cooling**: Cooling is accomplished by submerging in cold water (10-12° C) for 30 minutes. Cold shock creates a protective crust, trapping humidity within the product [13].
- 13) **Salting**: Done in “salsetta”: (2-5 % of salt) or brine (“salmoia”: 10 % of salt), salt concentration varies with weigh and size of pieces. Although some manufacturers combine the salting with the kneading process, “mozzarella di bufala” does not render itself well to this because it requires some time exposed to brine in order to form the crust (de 5 a 15 hours). The salting process for the “Mozzarella di Bufala Campana DOP” [19] is done by sequential immersions in saline solutions at different concentrations (8 to 12 %, acidity 9 to 11°SH/50). Salt penetrates the cheese by diffusion, speed of penetration depends on salt concentration of the brine, temperature and shape of the mozzarella piece.
- 14) **Packaging and distribution**: Traditionally it is packaged with brine, although vacuum sealing is now available and guarantees acceptable conditions at 20°C for a maximum of 10 days. The ANASB [19] recommends using the same kneading liquids, with added salt and lactic acid. Another alternative to extend shelf life during summer months is to use salted, pasteurized water with lactic acid. Noteworthy, shelf life is primarily connected to the quality of the starting materials (raw or pasteurized milk) and the sanitary practices implemented throughout the manufacturing process [18,19].

#### ***4.5.8. A slightly different production for mozzarella is followed in Alagoas, Brazil [21]:***

1. Add water (90 % milk + 10 % water).
2. Pasteurize.
3. Add 40 ml calcium chloride for each 100 liters of milk.
4. Add 3 % fermented whey at 40°D (Dornic), or 2 % at 60°D, or 1 % at 120°D.
5. Allow fermentation for 20-30 minutes; ideally, acidity should be 21°D at the addition of rennet.
6. Temperature should be 38°C at the addition of rennet.
7. Dissolve rennet in water (2 liters).
8. Add rennet slowly, while stirring; this process should last 1-2 minutes. Let milk stand.
9. Rennet should cause the formation of curd within 30-45 minutes. If this occurs in less than 30 minutes, the product will turn sour. If it occurs in over 45 minutes, cheese yield will be reduced.
10. Once curd is set, it is separated with a lira type tool; in sidewise and up and down motions.
11. In order to obtain a soft product, curd pieces should be 1.5 to 2 cm.
12. Curd must be allowed to set for 3 minutes, in order to allow crust formation.
13. Initial mixing: Stir slowly, increasing agitation gradually for 20 minutes.
14. Allow product to sediment to bottom of container.
15. Second mixing can be done in two ways:
  - a. *By heating up the container walls*: heat slowly, stirring gradually and increasing speed while preventing clump formation. Dough should have an ideal consistency at 38°C.
  - b. *By adding hot water*: This technique is frequently termed “lactose removal”, because it will eliminate at least part of the lactose content. Part of the whey is removed. Water at 100°C is added

with a narrow spout container. Direct contact of the dough with hot water must be avoided at all times, in order to avoid rubbery consistency. Dough is stirred slowly; speed is then increased as the temperature of the dough starts to increase. Dough should reach ideal consistency by 38°C.

16. Let dough stand with whey until it reaches kneading point (around 4-5 hours). The longer the time dough sits in whey, the longer kneading time it will require.

17. Test kneading point: dough should stretch for about a meter.

18. Kneading: knead with water (85-90°C), the amount of water should be 1.5-2 times the weight of dough to be kneaded.

19. Salting:

a. *Mozzarella logs for slicing*: Use 2 – 2.5% brine. Work dough, close balls and give shape. Place cheese in ice water, inside a mould for about 1 hour. Withdraw from mould and let water drain, keeping in cold room.

b. *Mozzarella in round pieces*: Withdraw from ice water. Place in brine (20%) for 5-10 minutes, depending on the size of the ball. Place balls in brine for final storage. They should have the same acidity as the dough, to prevent brine from becoming milky.

20. Packaging: In case of mozzarella logs, packaging is vacuum sealed. In the case of mozzarella balls, they are packaged with adequate volume of brine for balls to remain fully immersed.

## 5. PRODUCTION AND UTILIZATION OF MILK BY-PRODUCTS IN DIFFERENT COUNTRIES

All derivatives from bubaline milk are considered functional foods: those that provide benefits for human health. The main products obtained from buffalo cow milk are fresh, “frescal” or “criollo” (creole) cheese (Argentina, Venezuela, Brazil), butter, yoghurt (Bulgaria), mozzarella, provola (Italy) and other cheeses in India, such as cheddar, karnal, brick, surati and melted.

According to their water content [11], cheese can be classified as shown in Figure 13:

**Figure 13:** cheese classification according to water content, denomination and country of origin.

| CLASSIFICATION    | WATER CONTENT (%) | DENOMINATION AND COUNTRY OF ORIGIN  |
|-------------------|-------------------|---|
| Soft cheeses      | >45               | Karish, Mish and Domiati ( <i>Egypt</i> )<br>Madhfor ( <i>Irak</i> )<br>Mozzarella ( <i>Italy</i> )<br>Alghab ( <i>Syria</i> )<br>Vladeasa ( <i>Rumania</i> ) |
| Semi-soft cheeses | 40-45             | Beyazpeineri ( <i>Turkey</i> )  |
| Hard cheeses      | < 40              | Braila ( <i>Rumania</i> )<br>Rahss ( <i>Egypt</i> )<br>White ( <i>Bulgaria</i> )<br>Akkawi ( <i>Syria</i> )   |

Another common classification of cheeses is according to their acidification type: enzymatic coagulation (curd) and acid coagulation (natural or bacterial). Many cheeses are subjected to mixed coagulations, most with higher prevalence of acid coagulation in the process [22]. Due to different characteristics of the casein micelle and fat of buffalo cow milk, the main problems associated in cheddar cheese manufacturing have been acidity, coagulation times, humidity retention and delayed proteolysis and lipolysis. Hard cheeses made with buffalo milk are more difficult to dehydrate due to the higher protein content, which in turn results in more water retention. Pasteurization is only

done when the microbial count is high (this seldom occurs).

Fermented dairy products have been manufactured from buffalo cow milk with good results. Yoghurt derived from bubaline milk is considered a delicacy [23], its rich, creamy texture due to its higher calcium and casein content. Protein and vitamin E are also higher in this product, with low cholesterol. Recently, probiotics have been added to this product giving to it an unique flavor and biological properties. In Bulgaria, Rumania and Albania, buffalo milk is almost exclusively destined to yoghurt manufacture due to its high demand by local markets.

“Dahi” yoghurt, of superior consistency and texture, is preferably manufactured with buffalo cow milk due to its higher protein, fat and solids content and also due to the bigger size of fat globules, casein micelle and calcium in colloidal state [22]. Some typical Indian products are elaborated with buffalo cow milk, due to its quality and to the consumer preference. The most important is "Khoa", a coagulated dairy product obtained by milk dehydration (water content 19-25%). "Khoa" prepared from buffalo cow milk has a unique quality, consistence and texture, that makes it preferred over the same product prepared from cattle's milk. "Khoa" is used as a base ingredient for the preparation of widely popular sweets and candy. "Paneer" is another typical Indian buffalo cow dairy product. It contains an acid coagulated solid extract that is used as ingredient to cook vegetables in the north of the country. This product is obtained by acidification of milk with acid curd or lemon juice; it is then filtered and pressed without addition of salt. It must be consumed within 3 days, usually with spicy sauces and vegetables and with a special round, dry and hollow type of bread, the “ciabatti”. The "Cottage" cheese is prepared by acid coagulation in Europe and North America is a similar product [24].

### ***India, Turkey and Iran***

In India, fluid milk diluted with 33% water for human consumption is the principal destination of the production. The second product in importance is the ghee, a clarified butter with 99% fat that can be stored under ambient conditions. Buffalo cow ghee is almost translucent (as opposed to the yellowing color in cattle-derived ghee) due to absence of carotenoids. Ghee has been used since ancestral times in these countries, both as a food ingredient and as a part of the religious Hindi rituals. The product is perfectly fit to be used under tropical conditions and is the only animal fat source in the Indian predominantly vegetarian diet. The product is also steadily consumed by Muslim countries of Central Asia and Middle East. Over 40% of the milk produced in India is destined to ghee manufacture, with an annual average of 480.000.000 kg. Average human intake is 1.2 kg/head/year. Of this, 80 % is destined to food preparation, 18 % for bakery and 2 % for religious ceremonies. Ghee contains 99-99,5 % fat and no more than 0.5 % humidity. Ghee is either prepared from butter or from boiled milk skin ceam, following one of two methods: butter mechanical separation of skin cream hand-beaten (“Desi”), the latter being the method of choice for most ghee produced in India. “Desi” means common, or in other words, popular. In this country, buffalo milk as dry powder is also used for infant formulas. Specialists at Karnal Institute have introduced modifications in buffalo cow milk in order to make it more adequate for human consumption as fluid milk, resembling the profile of maternal milk [24]. Indian dairy cooperatives generally reduce the fat content of the product by dilution with 33% water, reaching a composition (water, fat, protein, solids) similar to cattle milk.

In Turkey, buffalo cow milk is specially sought after for its contribution to the famous turkish breakfasts. Whipped cream is mixed with warm milk, then slowly cooled in shallow pans, reheated and finally cut in the next morning. The result is the famous Qeymac, Keimak or Kaimak: a type of ghee, a semi-liquid clarified butter highly demanded by the population [25].

In Iran, farmers boil buffalo cow milk overnight, place it in shallow pans and collect the cream the following morning which is then sold in open markets. This ghee-type product, called Qeymac, Keimak, Kaimak in turkish, or Sarshir in persian, is very popular in restaurants and patisseries. Fat extracted from buffalo cow milk is partially sold to dairy product dealers, and is also used for the manufacture of salty yoghurt, a traditional product called Mast in Persia. Most of the elaboration is done by women.

### ***Brazil, Argentina, Venezuela and Bolivia***

In Brazil, in order to make better use of the advantageous properties of buffalo cow milk in the manufacture of other dairy products; a significant expansion of industrial units was undertaken in the early 90'. This expansion allowed producers to obtain a return on their product that almost doubled the return from cattle's milk. In addition, the price levels of buffalo cow milk products remained steady throughout the year. This in turn stimulated the establishment of important so-called 'cuencas' or 'hotspots' for buffalo cow milk production, particularly in the southwest of the country [26]. Noteworthy, while cattle milk demand has been dropping due to economic causes, buffalo cow milk plants have experienced an annual growth of 32.3% in processed milk between the years 2001 and 2005.

In addition to the traditional demand for mozzarella cheese, other less traditional buffalo cow products are experiencing an increase in interest and demand in Brazil, namely frescal, ricotta, provolone cheese, caramel, curd, yogurt and others. With the objective to promote buffalo cow milk, the Brazilian Buffalo Breeders Association (ABCB) has implemented a monitoring program "Selo de Pureza" (Purity Seal) since 2001. All products from participating farms are certified by protein electrophoresis, with guarantee of no contamination with other milk sources. As of 2007, 111 farms producing around 3.2 million liters per year were participating of the program. It is estimated that Brazil currently produces 90 million liters of bubaline milk (approximately 40.000 lactating buffalo cows), with over 150 dairy plants manufacturing some kind of bubaline dairy product. Brazilian domestic demand of buffalo cow dairy products is relatively steady throughout the year. Therefore, farmers are using reproductive biotechnologies in order to de-seasonalize calvings in order to be able to provide a more steady supply of product [26]. According to R. Amaral [27] ten years ago 80 % of the producers were oriented towards meat production; today more than half of those have switched their focus towards dairy.

Santa Eliza Farm, in Dourado, Sao Paulo State, Brazil, has its own dairy plant (Dourado Buffalo Dairy Products Factory) processing 4.000 liters/day from their own and nearby milking farms. They apply standardized good manufacturing practices, taking special care of the environment; their de-seasonalization strategy allows them to maintain steady production year-round by milking 200 animals out of the breeding season. Their registered trademark, mozzarella Dourado, is one of the most prestigious products in the Brazilian market. They supply 85% of production to Sao Paulo (280 km of distance), 10% to Rio do Janeiro and the remaining production to the rest of the country. Laguna Farm [15] recently received an award by the State of Ceará. It is a family integrated production system, from farm to market, mainly supplying their products to Fortaleza, a city of 2.3 million inhabitants. Their products include "frescal" (regular and oregano) and "criollo" (creole) cheese (less humid than the "frescal"), ricotta and whole milk. Their production does not include mozzarella, due to its inherent lower cheese yield, compared to the other cheeses. According to their economic analysis, bubaline cheese production is more profitable than cattle's due to its lower input requirements on milk volume, forage, equipment and also because its market is about 15% higher. A growing number of small-scale farmers are switching to bubaline milk in those regions of Brazil where dairy plants specialized in buffalo milk have settled. Even with the typical low production of poorly improved buffalo herds, their returns on investment are still higher than that obtained with cattle [26].

In Argentina, a dairy farm owned by González Fraga, in Torres, Luján, Buenos Aires Province, is the buffalo milk supplier for La Salamandra S.A. The group covers all aspects of the production chain from farm to market outlets, with a registered trademark. The industrial plant produces 22 kg mozzarella/100 liters of milk. Several niche opportunities exist today in their local market due to the significant influence of the Italian culture, especially in urban populations. This demand still remains to be satisfied. La Salamandra S.A. markets their products using top restaurants and exports 20% of their cheese production to Chile. Other dairy plants market criollo (creole) type cheeses

(Formosa, Corrientes and Misiones Provinces) and flavored, semi-hardened paste cheeses (Santa Úrsula, owned by Sons of Raúl Maglietti S.R.L.) in Formosa.

Pangolares Ranch, owned by the Argentinian Alberto Duhau and brothers, is located near Maturín city, Monagas State, Venezuela, and produces “Feta” cheese, a Greek, salty type cheese similar to the Venezuelan criollo (creole), that yields 22-23%. They also produce “white cheese” (of soft consistence), Gouda cheese (semi-hard), mozzarella and ricotta. In its institutional presentation, mozzarella is packaged in a container with 2.5 liters of salt water and mozzarella 16 balls of 160 g/each, their products are sold to the towns of Maturín, Puerto Ordaz, Puerto La Cruz, Caracas and Valencia (in Venezuela); in its home presentation is done with two balls of 160 g/each, or with 16 of 20 g [15].

El Carmen Ranch, in Santa Cruz, Bolivia, owned by Robert Haab, produces several types of cheeses such as melted cheese, ricotta and others. Their trademark, “Caserti”, has a strong presence in the local market [15].

### *Italy*

Italian cheese market is complex due to market restrictions for fresh cheeses and constrains on shelf life. They key aspects of “Mozzarella di Bufala Campana” (MBC) are: Protected Designation of Origin (DOP) and the merge of cheese and breed, the production of mozzarella cheese from milk exclusively from buffalo cows of Italian Mediterranean breed. Another important aspect is the inclusion of mozzarella cheese in traditional Italian recipes such as pizza, caprese salad and others. Storage in salty water results in a progressive softening of the mozzarella, the degree of softening depends on the calcium content. The higher the calcium content, the firmer the cheese. Calcium content is also important in pizza preparation, because the Calcium micelle aids in reducing the acidity of the tomato sauce. When baked, care should be taken not to cause “browning” of the cheese, through turning residual sugars (glucose and galactose) into lactic acid, using cultures of lactic acid bacterial rests [23]. Destination of Italian exports of mozzarella for the year 2005 are shown in Figure 14.

**Figure 14.** Country of destination of mozzarella Italian exports for 2005

| <b>COUNTRY OF DESTINATION</b> | <b>% OF ITALIAN EXPORTS (MOZZARELLA) (2005)</b> | <b>VARIATION (%) FROM PREVIOUS YEAR (2004)</b> |
|-------------------------------|---|--|
| <b>France</b>                 | 21.4  | +1   |
| <b>Germany</b>                | 18.1  | +0.7   |
| <b>UnitedStates</b>           | 5.2   | -0.8 (“Euro effect”)                           |
| <b>UnitedKingdom</b>          | 12.1  | -0.6   |
| <b>Japan</b>                  | 8   | +2.8   |
| <b>Switzerland</b>            | 6.8   | -0.8   |
| <b>Spain</b>                  | 6.2   | +2.5   |
| <b>Belgium</b>                | 3.4   | +0.7 %   |
| <b>Canada</b>                 | 2.3   | +0.4   |

Exports to Japan have been undergoing a growing trend (2001-2005); it has grown from 2 % in 2001 to 8.7 % in 2005, with an increase of 2.8 % respect 2004. Fifty-one percent of exports are within countries of the European Community [7].

The limitations of the Italian market are associated to the cheese industry organization and distribution problems. For this reason, many breeders (producers) and cooperatives prefer to market their products themselves in order to gain added value and improve their income. The wide diversity within typical mozzarella is a considerable marketing problem. Mozzarella produced by

small plants is typically produced using unique yeasts and natural microorganisms; its shelf life is normally three to five days and may not be extended by refrigeration. On the other hand, mozzarella manufactured by larger plants is produced without yeasts and microorganisms for long-term (30 day) conservation by refrigeration. This latter product, which is destined to the large scale grocery store and export market, is not as soft and moist. Both products have the same denomination and DOP, but are quite different. Although both excellent products, the striking difference between the two may confuse the general public. Mozzarella cheese is usually presented as balls or “polpettas” of variable size (20-800 gr), as small bites or “bocconcino” and braided or “treccia” [22].

In Italy the price of buffalo milk is four times the price of cattle’s cheese. Italian mozzarella producers started to pasteurize their milk in the early 90’s. Even though pasteurization reduces milk yield significantly, it is still required by the markets due to sanitary issues and shelf life. Italian consumers have progressively learned to value the certificate of origin of their product; hence, the status of DOP (Denominazione di Origine Protetta) and DOC (Denominazione di Origine Controllata) are important accomplishments. The internal demand has also increased steadily in latest years; 63 % of mozzarella is consumed by the south of Italy, with an average family intake of 1,7 kg/year in southern Italy, meanwhile the national average is of 1 kg [19].

“La Garofalo” Dairy Factory, in Caserta city, processing 60.000 liters/day, is the largest in Italy. The plant is completely automatized, with 20 hour-day work shifts. Forty- percent of their production goes to the export market (Germany, France, Switzerland, Belgium, Canada, USA and Russia). The price is the same for the local market whole-salor and for the international trader: 8 E/kg. With the exception of the manual kneading, the rest of the manufacturing process is completely automated, including packaging. Each holding tank has a capacity of 1000 liters; pasteurization is done at 72° C for 15”, and cooling at 4° C in a water-milk laminar counter flow. Riva Bianca Ranch, in Paestum, Salerno Province, processes 5000 liters/day. They consider mozzarella an excellent business: it contains 66% of water. Riva Bianca produces ricotta, mozzarella and scamorza. The last one (scamorza) has the highest shelf-life among their products; this product has an 18 % yield and the production cycle lasts at least 45 days. It is a high quality cheese that allows producers the flexibility to utilize all surplus milk when the demand for mozzarella temporarily drops.

In Italy, a Technical Committee of the Scientific Consortium for the Protection of “Mozzarella di Bufala Campana” was established in 1999, and standardized parameters for health management, nutrition and body condition score of bubaline herds were outlined [28]. Finally, because mozzarella made from cattle’s milk is 20% cheaper than its buffalo cow counterpart, it is important to be able to distinguish between the two products. 1) Color: buffalo cow mozzarella is whiter, pearlier and glossier whereas cattle’s mozzarella has a yellowish color. 2) Fat content: buffalo cow mozzarella has a higher (20-30%) fat content than cattle’s. 3) Texture: the surface of buffalo cow mozzarella should appear smooth, whereas cattle’s is bit rougher. Its outer layer should be thin and when open, interior should be moist and creamy whey runoff should be noticeable. Fresh buffalo cow mozzarella is more elastic than cattle’s. 4) Flavor: buffalo mozzarella cow is sweeter than cattle’s, with a slightly acidic finish. Buffalo milk also can be differentiated from cattle’s milk by protein electrophoresis. In Argentina, this test is conducted by the Industrial Technology National Institute – Dairy (INTI – LÁCTEO). In Brazil, the “selo de pureza” is awarded by the Breeders Association. Purity control conducted by protein electrophoresis has an accuracy of 90 %. There are also molecular probes available to conduct such evaluation, with even higher accuracy [29].

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