



## Production of Laban

Catherine Béal, Gisèle Chammas

► **To cite this version:**

Catherine Béal, Gisèle Chammas. Production of Laban. Handbook of Plant-Based Fermented Food and Beverage Technology, CRC Press, pp.181 - 212, 2012, 10.1201/b12084-14 . hal-01511556

**HAL Id: hal-01511556**

**<https://hal-agroparistech.archives-ouvertes.fr/hal-01511556>**

Submitted on 18 Nov 2019

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

# Production of laban

**Catherine Béal**

**Gisèle Chammas**

**In: Hui YH, Özgül Evranuz E, editors. Handbook of animal-based fermented food and beverage technology. 2nd Edition ed. Boca Raton, Florida, USA: CRC Press. p 181-211.**

1.	Introduction.....	3
2.	Laban market in Lebanon and other countries.....	4
3.	Lactic acid fermentation of laban.....	5
3.1.	Microbiology of laban fermentation.....	5
3.1.1.	Microorganisms.....	5
3.1.2.	General characteristics of lactic acid bacteria used for laban fermentation.....	7
3.1.3.	Associative growth.....	7
3.2.	Environmental factors that affect lactic acid bacteria metabolism.....	8
3.2.1.	Physico-chemical factors.....	8
3.2.2.	Microbiological factors.....	9
3.3.	Biochemistry of laban fermentation.....	10
3.3.1.	Lactose catabolism.....	10
3.3.2.	Nitrogen catabolism.....	11
3.3.3.	Lipid metabolism.....	12
3.3.4.	Other metabolisms.....	12
4.	Influence of lactic acid fermentation of laban properties.....	12
4.1.	Impact of lactic acid fermentation on physical properties of laban.....	12
4.1.1.	Microstructure of laban.....	12
4.1.2.	Physical properties of laban.....	14
4.2.	Impact of lactic acid fermentation on laban flavor.....	15
4.2.1.	Taste of laban.....	15
4.2.2.	Aroma compounds of laban.....	16

4.3.	Impact of lactic acid fermentation on shelf life of laban .....	17
4.4.	Modification of nutritional value of milk .....	18
4.4.1.	Decreasing lactose malabsorption .....	18
4.4.2.	Increasing protein digestibility .....	19
4.4.3.	Effect on vitamins content .....	19
4.4.4.	Effect on calcium assimilation .....	20
4.4.5.	Health effect from probiotic bacteria .....	20
5.	General process of laban manufacture .....	22
5.1.	Milk reception and analysis.....	22
5.2.	Standardization of milk fat .....	22
5.3.	Standardization of protein content.....	24
5.4.	Homogenization .....	25
5.5.	Heat treatment and cooling .....	25
5.6.	Inoculation and fermentation .....	26
5.6.1.	Inoculation modes .....	26
5.6.2.	Incubation of laban.....	27
5.6.3.	Kinetic of laban fermentation .....	27
5.6.4.	Controls during laban fermentation.....	28
5.7.	Cooling.....	28
6.	Packaging and storage of laban.....	29
6.1.	Packaging of laban.....	29
6.1.1.	Packaging materials.....	29
6.1.2.	Packaging systems .....	30
6.1.3.	Storage of the products.....	31
6.2.	Shelf life of laban .....	31
7.	Evaluation of Laban quality .....	32
7.1.	Microbiological characteristics.....	32
7.2.	Physico-chemical characteristics.....	33
7.3.	Sensory evaluation .....	34
8.	Conclusion .....	36
9.	References.....	36

## 1. Introduction

Laban is a fermented milk produced in Lebanon and some Arab countries. It is obtained from lactic acid fermentation, thus leading to acidification and coagulation of milk. Historically, these fermented dairy products appeared as a means to preserve milk, but they now gain a high interest, due to their pleasant sensory properties (freshness, acidity, mouth coating). They represent an interesting alternative to milk and cheese consumption. If the historical and geographical origin of laban has never been precisely established, the first known products appeared in the Middle-East, 10 or 15 000 years ago. They are now commercialized within a large number of countries, as they are essential in the Arab diet. Lebanon and Arab countries are the greatest consumers, together with countries of North Africa.

Laban is obtained through the lactic acid fermentation of heat-treated cow milk by thermophilic starters such as *Streptococcus thermophilus*, *Lactobacillus acidophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*, at 40-45°C (Baroudi & Collins, 1976; Chammas, Saliba, Corrieu, & Béal, 2006). Depending on the product, the milk used may be full-fat, partially skimmed or fully skimmed, thus leading to the commercialization of whole-fat, low-fat and fat-free laban (Libnor, 1999).

In Lebanon, laban is manufactured by both industrial and small traditional producers, each of them using different manufacturing practices (Surono & Hosono, 2003). Traditional producers mainly use artisanal starters, consisting of a mixed culture of unknown number of undefined and indigenous thermophilic strains. This procedure generates diversified and typically flavored products that are generally preferred by consumers (Wemekamp-Kamphuis, Karatzas, Wouters, & Abee, 2002). The industrial production of laban is carried out by associating selected starters showing well-characterized properties and by using a standardized process. This practice allows achieving the desired metabolic activity and technological properties of the strains during their growth in milk. It also leads to a constant quality of the products and a high level of reproducibility of the processes, but to the detriment of the diversity of flavors, which is lower comparing to traditional products.

The composition and characteristics of laban differ according to the production scale. As a general rule, laban has a titratable acidity of about 1% and a pH of 4.0 (Baroudi & Collins, 1976). Table 1 shows the gross composition of laban, which has been determined recently by (Guizani, Kasapis, & Al-Ruzeiki, 2001; Musaiger, Al-Saad, Al-Hooti, & Khunji, 1998). The fat content depends on the kind of laban: a whole milk laban contains 3.1 % of fat, whereas a low fat product comprised only 1.3 % of fat (Musaiger et al., 1998).

**Table 1. Physico-chemical composition of Laban**

Kind of product Parameter	Experimental laban	Whole fat laban	Low fat laban	Home-made laban	Commercial laban	Artisanal laban	Tunisian Leben
pH	4.25	4.2-4.9	4.3-4.6	3.98 ± 0.13	4.52 ± 0.03	Nd	4.29-4.45
Titratable acidity (%)	0.9-1.2	0.6-1.1	0.8	1.12 ± 0.12	0.77 ± 0.04	0.9-1.75	Nd
Total solids (%)	Nd	11.1-13.1	9.7-11.5	6.29 ± 0.21	10.87 ± 0.32	Nd	7.05-7.4
Protein (%)	Nd	2.5-4.6	2.9-3.5	2.11 ± 0.17	3.15 ± 0.15	Nd	1.86-2.56
Fat (%)	Nd	2.5-4.3	1.2-1.4	1.12 ± 0.35	3.50 ± 0.17	3.3-4.2	1.48-3.50
Lactose content (%)	Nd	3.3-4.2	3.7	Nd	Nd	2.65-3.99	1.90-2.59
Reference	Baroudi and Collins 1975	Musaiger et al. 1998	Musaiger et al. 1998	Guizani et al. 2001	Guizani et al. 2001	Chammas et al. 2006	Samet-Bali et al. 2009

## 2. Laban market in Lebanon and other countries

In the last decades, the consumption of dairy products increased at extremely high rates in the Middle East due to their nutritional value: they are considered as cheap sources of animal protein and well-known source of calcium in the human diet. Currently, the per capita consumption of dairy products is 152 kg/y (equivalent to milk quantity) in Lebanon, 117 kg/y in Syria, 78 kg/y in Jordan and 54 kg/y in Saudi Arabia (Al Ammouri, 2006; Alqaisi, Ndambi, & Hemme, 2009; BMI Business Monitor International, 2010; Lebanese Ministry of Agriculture, 2007). In Lebanon, laban is consumed as such or used in the preparation of a wide variety of dishes. Therefore, the annual consumption of this fermented milk is the highest among all the countries of the region.

Laban is the main dairy product manufactured in the Middle East and the most consumed dairy product in Lebanon (Nasreddine, Hwalla, Sibai, Hamzé, & Parent-Massin, 2006). The laban consumption reaches 25 Kg per year per person, followed by cheese (12.8 Kg per year per person) and then by labneh or strained laban (10 Kg per year per person). According to (Lemoine, 2002), 40 tons of laban are produced annually by 135 factories in Lebanon. Other 165 small-scale dairy processing units are scattered all over the country and deliver their production to neighborhood market. The biggest dairy companies in Lebanon are Karoun Dairy and Liban Lait. Almarai is a well-known Saudi Arabian dairy company. Today, it becomes the largest producer and exporter of milk and dairy products in the Middle East. The company production capacity is 1.8 million liters of milk per day. According to (Sadi & Henderson, 2007), about 60 % of Almarai raw milk is fermented to make laban. Probiotic culture laban is also manufactured.

The level of export of dairy products is still very low in the Arab countries compared to Saudi Arabia where dairy exports are increasing at a high rate. Although Syria is the biggest producer among these countries, small quantities are processed by the dairy factories with restrained infrastructure and technology, as compared to on-farm and artisanal production. On the other hand, in Saudi Arabia, the processing units are more developed, and sophisticated technology is used with higher percentage of milk delivered to dairy (Alqaisi, Ndambi, Uddin, & Hemme, 2010). Laban is one of the most dairy products exported, followed by unsweetened milk. The main destinations of these exports are the neighboring

gulf countries, Iraq, Jordan, Lebanon, Yemen and selected African markets (Alqaisi et al., 2010).

Due to the long tradition of milk and fermented milk products consumption in the Middle East, the potential for further development in the dairy sector of this region is greatly possible. Dairy cow population is estimated at 77,000, 773,000, 50,000, and 111,600 cows for Lebanon, Syria, Jordan, and Saudi Arabia, respectively (Alqaisi et al., 2009; Alqaisi et al., 2010; Anonymous, 2008; Lebanese Ministry of Agriculture, 2007). Total milk produced in 2007 was estimated at 0.24, 2.63, 0.28, and 1.34 million tons in Lebanon, Syria, Jordan, and Saudi Arabia, respectively (Alqaisi et al., 2010; Lebanese Ministry of Agriculture, 2007).

In all Arab countries, increasing feed prices are a major risk factor in the dairy industry, mainly because these countries rely basically on imported feedstuff in feeding animals. Additionally, the availability of water resources, particularly in Saudi Arabia and Jordan, is another limiting factor in fodder production.

### **3. Lactic acid fermentation of laban**

As a fermented dairy product, laban is obtained from lactic acid fermentation that corresponds to the transformation of carbohydrates into lactic acid as a major metabolic end-product, with the aid of specific microorganisms called lactic acid bacteria. This bioreaction leads to important biochemical, physico-chemical and sensory changes of milk.

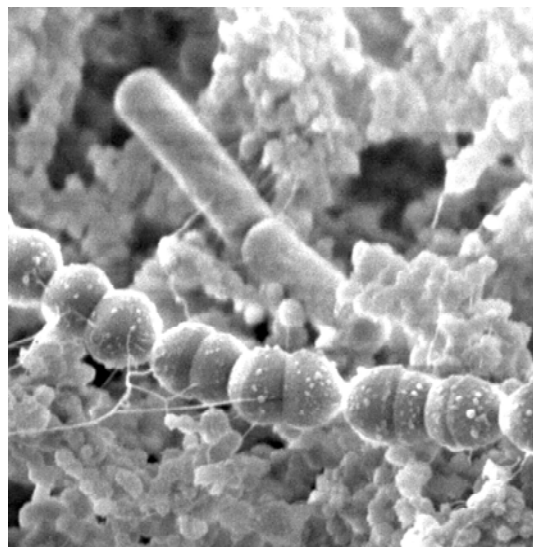
The first outcome of the lactic acid fermentation is to increase the shelf-life of the products, by inhibiting the growth of microbial spoilage and the occurrence of enzymatic reactions. It allows obtaining safe products, free of pathogenic microorganisms, as a result of product acidity. It also confers the products some specific nutritional and sensory properties, such as texture and flavor.

#### **3.1. Microbiology of laban fermentation**

##### **3.1.1. Microorganisms**

According to the authors and to the countries, the microbial composition of Laban differs. (Baroudi & Collins, 1976) identified five microorganisms in artisanal products, corresponding to three bacterial species, *Streptococcus thermophilus*, *Lactobacillus acidophilus* and *Leuconostoc lactis* and two kinds of yeasts, *Kluyveromyces fragilis* and *Saccharomyces cerevisiae*. (Guizani et al., 2001) characterized some artisanal and commercial Laban from Sultanate of Oman. They indicated that *Lactococcus lactis* subsp. *lactis*, *Lactococcus lactis* subsp. *lactis* biovar *diacetylactis*, *Lactococcus lactis* subsp. *cremoris* and *Lactobacillus plantarum* were responsible for the fermentation of traditional laban, whereas *Lactobacillus acidophilus*, bifidobacteria and *Streptococcus thermophilus* were responsible for the production of commercial laban. (Chammas, Saliba, Corrieu et al., 2006) identified 96 strains isolated from 18 Laban products that were collected in small-scale farms located in 15 areas distributed throughout the Lebanese territory and one commercial

product manufactured by using a mixture of European commercial starters. The herds corresponded to Friesland cattle and to Baladi, a draft breed on Lebanon. In all of these products, two bacterial species were identified, belonging to *Streptococcus thermophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*. As an illustration, Figure 1 allows visualizing some cells of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* enclosed in a milk coagulum. More recently, (Samet-Bali, Bellila, Ayadi, Marzouk, & Attia, 2009) demonstrated that the production of industrial Leben in Tunisia was triggered by the action of *Lactococcus lactis* subsp. *lactis*, *Lactococcus lactis* subsp. *diacetylactis* and *Lactococcus lactis* subsp. *cremoris*. From this information, different bacterial species are mentioned as triggering lactic acid fermentation in Laban. The differences may be ascribed to the different countries in which the samples have been collected, and to the level of industrialization of the products.



**Figure 1. Photograph of laban comprising *S. thermophilus* and *L. bulgaricus* subsp. *bulgaricus* cells (Photograph from J.P. Tissier, INRA)**

Whatever the considered study, the microorganisms are always used in mixed cultures, combining at least two species, and often many strains of each species. In most of the studies, the strains were not selected by the authors as they were isolated from artisanal products. On the contrary, in industrial plants, only the two species *Streptococcus thermophilus* and *Lactobacillus bulgaricus* are encountered, thus indicating that industrial laban is close to yogurt. By analogy with yogurt, the criterion of choice of the strains relies on technological considerations (acidification activity and bacteriophage resistance) and sensory properties (exopolysaccharide production, aroma compounds synthesis, post-acidification).

### 3.1.2. General characteristics of lactic acid bacteria used for laban fermentation

Lactic acid bacteria are Gram-positive bacteria, non-sporulating and non-respiring rods or cocci. Their low guanine + cytosine (G+C) content is comprised between 33% and 54%. They are known to synthesize large amounts of lactic acid from lactose, either as the main end-product (homofermentative) or in combination with carbon dioxide and ethanol (heterofermentative). Both L(+), D(-) or racemic lactic acid are synthesized, according to the species. For example, *Streptococcus thermophilus* produces only L(+) lactic acid, whereas *Lactobacillus delbrueckii* subsp. *bulgaricus* synthesizes D(-)lactic acid, both of these species being homofermentative. According to the genus, they grow at temperatures of 25-30°C (mesophilic bacteria) or 37-45°C (thermophilic bacteria), but not at 15°C. These characteristics have to be taken into account when associating two species in a same product.

### 3.1.3. Associative growth

Simultaneous growth of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* leads to a positive indirect interaction between these two species that is used to be termed as symbiosis (Rajagopal & Sandine, 1990). This interaction results in an increase of acidification rate and bacterial concentrations in mixed culture, as compared to pure cultures (Béal & Corrieu, 1998). The production of aroma compounds (mainly acetaldehyde) and physical stability of the product (syneresis) are also improved in mixed cultures. The stimulation of *S. thermophilus* by *L. delbrueckii* subsp. *bulgaricus* operates through the proteolytic activity of the lactobacilli, which liberates peptides and amino-acids that stimulate the growth of the streptococci (Courtin, Monnet, & Rul, 2002). In return, *S. thermophilus* synthesizes formic acid and CO<sub>2</sub> that promote the growth of *L. delbrueckii* subsp. *bulgaricus* (Ascon-Reyes, Ascon-Cabrera, Cochet, & Lebeault, 1995; Perez, De Antoni, & Anon, 1991).

When other bacteria are associated with *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, other kinds of interaction take place. *L. delbrueckii* subsp. *bulgaricus* limits the development of *Lactobacillus acidophilus* (competition and inhibition phenomena), whereas associative growth is observed between *S. thermophilus* and *L. acidophilus* (Vinderola, Mocchiutti, & Reinheimer, 2002).

Finally, specific inhibition phenomena may occur, as a result of bacteriocin production. Bacteriocins are small and thermostable proteins produced by some lactic acid bacteria to inhibit the growth of closely related strains. By considering *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, less than 10 bacteriocins have been identified (A. Y. Tamime & R. K. Robinson, 1999). Nevertheless, this phenomenon makes it necessary to verify the biological compatibility of strains before associating them for laban production.



## **3.2. Environmental factors that affect lactic acid bacteria metabolism**

Growth and acidification of lactic acid bacteria are strongly influenced by physical, chemical and microbiological environmental factors.

### **3.2.1. Physico-chemical factors**

Temperature is the first environmental factor to be taken into account during laban manufacture. It acts directly on chemical and biochemical reaction rates. As at least two species of microorganisms are associated during laban fermentation, they first have to be compatible by considering their optimal temperature for growth. By considering the species involved in laban fermentation, the optimal temperature is comprised between 37°C and 42°C (Adamberg, Kask, Laht, & Paalme, 2003; Béal, Louvet, & Corrieu, 1989; Chammas, Saliba, Corrieu et al., 2006). Nevertheless, in artisanal productions, the temperature is generally lower, thus favoring the growth of other microorganisms (Baroudi & Collins, 1976). The fermentation temperature also affects the physical properties of the gels, a lower temperature leading to a stronger texture (Béal, Skokanova, Latrille, Martin, & Corrieu, 1999).

Water activity ( $a_w$ ) in milk products is mainly related to NaCl or sugar concentration (Labuza, 1980). By reducing  $a_w$ , the fraction of free water decreases and the availability of the nutrients is affected (Fajardo-Lira, Garcia-Garibay, Wachter-Rodarte, Farrés, & Marshall, 1997). In fermented dairy products, NaCl has been shown to limit post-acidification together with an increase of mortality (Lacroix & Lachance, 1988). Nevertheless, as in laban, no saccharose nor NaCl are introduced in milk before or during fermentation, water activity is generally not affected.

Milk composition has a strong influence on the growth and acidification by lactic acid bacteria. If lactose and minerals concentrations are high enough in milk (Desmazeaud, 1990), the nitrogenous fraction (amino acids and oligopeptides) is insufficient and starvation may occur at the end of fermentation. Moreover, when present in milk, some specific components have a negative impact on bacterial growth (Reiter, 1978): lactenins, lactoperoxidase/thiocyanate/hydrogen peroxide,  $H_2O_2$ , antibiotic residues, detergent and disinfectant residues, insecticide residues or somatic cells.

Thermal treatment of milk before fermentation acts positively on bacterial metabolism, thus reducing the fermentation time. This stimulatory effect is explained by several factors: elimination of undesirable and pathogen microorganisms thus reducing competition phenomena during growth, restriction of some antibacterial substances levels that are naturally present in milk (agglutinins, lactoperoxidase, toxic sulfides), release of cysteine and glutathione that act as antioxidant, and production of small quantities of formic acid from lactose (A.Y. Tamime & Robinson, 2007b). Finally, it contributes to slightly increase the amounts of amino acids and small peptides in milk.

As a last environmental factor, pH is very important for growth of lactic acid bacteria (Béal & Corrieu, 1991). It affects the nutrients availability, the cellular membrane

permeability and the enzymatic reaction rates (Rault, Bouix, & Béal, 2009). During laban production, pH is not controlled and thus, decreases during fermentation. This acidification is a major factor that slows down bacterial metabolism and confers the product some important technological characteristics. Moreover, undissociated lactic acid concentration increases in milk as a result of fermentation and pH decrease. As bacterial membrane is permeable to non-dissociated lactic acid, the inhibitory effect of acidification is strong (Amrane & Prigent, 1999), thus leading to low bacterial concentration at the end of the laban fermentation.

### 3.2.2. Microbiological factors

The choice of bacterial strains depends on several factors, referring on technological properties and performances. The main technological properties of lactic acid bacteria concern their acidification activity (Corrieu, Spinnler, Jomier, & Picque, 1988), their capacity to metabolize carbohydrates, their proteolytic and lipolytic activities and their ability to produce aroma compounds, exopolysaccharides or bacteriocins (Béal, Marin, Fontaine, Fonseca, & Obert, 2008). The selection of a lactic acid starter also takes into account the bacterial growth rate and metabolic rates, the interactions of the strain with other species and its sensitivity to bacteriophages. The ability of the bacteria to be frozen or freeze-dried is also important, in order to permit their stabilization before use (Béal et al., 2008).

The inoculum level affects the fermentation rate, which is more rapid with a high inoculum rate. As a general rule, inoculation is done at a concentration of  $10^6$  CFU/mL, in order to simultaneously shorten the fermentation and limit the costs. When using a direct inoculation, the inoculum rate in milk varies between 2.5 g and 70 g / 100 L, whereas a 1 L / 100 L is generally necessary for an indirect inoculation.

The balance between the bacterial species influences the fermentation kinetic. When streptococci are involved in the fermentation, they always predominate at the end of acidification, even if the initial ratio is generally well balanced (1:1) between the species *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* (Béal & Corrieu, 1991). The existence of a positive mutual interaction between these two species partly explains this phenomenon (Rajagopal & Sandine, 1990).

Finally, when bacteriophages arise in a production plant, they strongly affect the fermentation, thus leading to serious economic losses in laban industry. By considering *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, two kinds of bacteriophages may be encountered, both of them belonging to the *Siphoviridae* family (Krusch, Neve, Luschei, & Teuber, 1987). The virulent or lytic bacteriophages lyse the bacterial host during infection, whereas the temperate, prophage or lysogenic bacteriophages, which insert their genome in the host chromosome, do not lyse the host cells (Neve, 1996; Quiberoni, Guglielmotti, Binetti, & Reinheimer, 2004). In order to avoid phage attack, the combined use of aseptic techniques for the propagation of starter cultures, proper heat treatment of the milk and daily rotation of bacteriophage unrelated strains or phage-resistant strains is strongly recommended.

### 3.3. Biochemistry of laban fermentation

Lactic acid bacteria commonly use milk nutrients to permit their growth as well as metabolite production. Most of these reactions are answerable to lactose catabolism that corresponds to the main functionality of lactic acid bacteria and is essential to obtain a high grade fermented product (flavor and stability). Some anabolism reactions are also important as they act on the synthesis of exopolysaccharides, aroma compounds or preservative compounds.

#### 3.3.1. Lactose catabolism

Lactic acid fermentation is defined by the following simplified biochemical reaction:

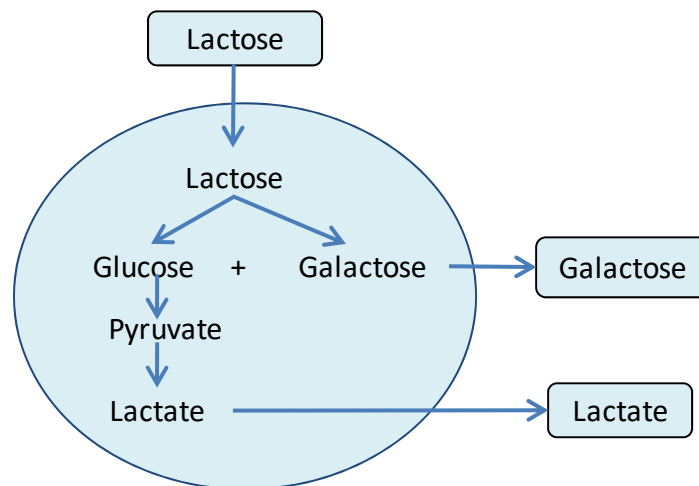
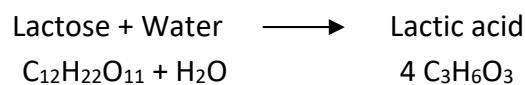
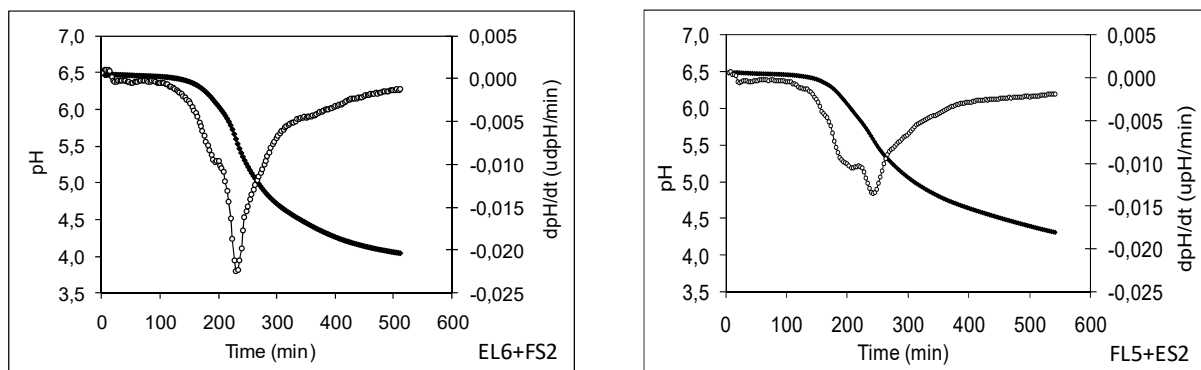


Figure 2. Main oxydo-reduction reactions involved in lactose catabolism

The catabolism of lactose into lactic acid occurs according to four steps that are summarized on Figure 2. Since the lactose catabolism takes place inside the cells, the entry of lactose represents the first step of this pathway. In *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, it involves cytoplasmic proteins such as ATP dependant permease that translocates lactose without chemical modification. This LacS permease acts according two ways: a symport lactose/H<sup>+</sup> and an antiport lactose/galactose, this last one being preferred when galactose is present (Poolman et al., 1996). Lactose is then hydrolyzed into glucose and galactose by  $\beta$ -galactosidase (Thompson & Gentry-Weeks, 1994). Glucose is catabolized to pyruvate according to Embden Meyerhof pathway, whereas galactose is excreted from the cells in *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*. In mesophilic lactic acid bacteria, galactose is catabolized into pyruvate through Leloir pathway. These reactions drive the synthesis of energy (2 ATP/glucose). The third step corresponds to the reduction of pyruvate into lactate, using lactate-dehydrogenase as catalyst (Thompson & Gentry-Weeks, 1994). This reaction allows the cofactor NAD<sup>+</sup> to be oxidized. According to the species, L(+),

D(-) or racemic lactic acid is synthesized. Finally, lactate is secreted from the cells, due to a symport with  $H^+$ . The increase of lactate concentration in milk undergoes acidification, together with an inhibition of bacterial growth that stops before lactose starvation. This inhibition combines the effects of lactic acid accumulation with resulting pH decrease.

Figure 3 shows the typical acidification kinetics obtained during manufacture of two laban samples (Chammas, 2006). Acidification kinetics were different among the two samples, the first one being highly acidifying as pH 4.5 was obtained after 334 min, whereas the second one needed 485 min to reach pH 4.5, thus indicating that the strains used were less active.



**Figure 3. Acidification kinetic of two laban samples (from Chammas 2006)**

Strains *S. thermophilus* FS2 and *L. bulgaricus* subsp. *bulgaricus* EL6

Strains *S. thermophilus* ES2 and *L. bulgaricus* subsp. *bulgaricus* FL5

### 3.3.2. Nitrogen catabolism

Because they are unable to catabolize mineral nitrogen, lactic acid bacteria use the milk proteins as a nitrogen source during laban fermentation. Milk contains about 32 g/L nitrogen compounds that are shared into a soluble fraction (free amino acids, small peptides, whey proteins, nitrogenous bases, urea and group B vitamins) and an insoluble fraction composed of the caseins (Desmazeaud, 1990). Lactic acid bacteria may use the free amino acids, the small peptides and the whey proteins contained in milk as a consequence of their proteolytic activity, although it is considered as weak and variable according to the species (Fernandez-Espla, Garault, Monnet, & Rul, 2000; Stefanitsi & Garel, 1997). Nevertheless, as they cannot synthesize all amino acids, an exogenous supply of at least leucine and valine is necessary to fulfill the bacterial needs (Letort & Juillard, 2001). These components take part into many cellular functions such as protein and enzyme synthesis that are used for the biosynthesis of cell constituents. The liberation of some amino acids into milk is important as they stimulate the growth of some species such as *Streptococcus thermophilus* (Courtin et al., 2002). Some peptides that are liberated may also act as precursors for the synthesis of flavor compounds. Among them, threonine is converted, thanks to a threonine aldolase, into acetaldehyde that is a major compound of laban aroma (Ott, Germond, & Chaintreau, 2000).

### **3.3.3. Lipid metabolism**

Milk lipids represent between 33 to 47 g/L of solid fraction of cow milk, mainly on the form of triacyl glycerols (96-98%). They act as a source of essential fatty acids, which cannot be synthesized by animals, and of fat-soluble vitamins (A, D, E, K). Lactic acid bacteria display limited lipolysis, as a consequence of lipase and esterase activities, which are species- and strain-dependent (Kilcawley, Wilkinson, & Fox, 1998). The lipase activity may also contribute towards the flavor and rheological properties of the dairy products.

### **3.3.4. Other metabolisms**

Bacterial metabolism also includes less important compounds, such as nitrogenous bases, vitamins and minerals. By considering the vitamins, lactic acid bacteria used for laban fermentation need biotin, niacin, riboflavin, vitamin B<sub>12</sub> and pantothenic acid, whose concentrations decrease in milk as a result of microbial catabolism (Béal et al., 2008). Conversely, depending on the strain, niacin, folic acid, and in to a lesser extend riboflavin, thiamin, vitamin B<sub>6</sub> and vitamin B<sub>12</sub> are actively synthesized by these lactic acid bacteria during milk fermentation (Rao, Reddy, OPulusani, & Cornwell, 1984).

Some nitrogenous bases that are essential for nucleic acids synthesis are brought within milk (Zink, Elli, Reniero, & Morelli, 2000). Nevertheless, a high variability among the needs exists within species and strains, thus explaining some differences observed for bacterial growth in milk.

Finally, some cationic compounds are involved in metabolic pathways. Among them, manganese, magnesium and potassium are often concerned (Boyaval, 1989).

## **4. Influence of lactic acid fermentation of laban properties**

During fermentation, lactic acid bacteria induce strong changes on milk composition and properties. These changes concern the physical properties of the gel, the flavor of the product, the shelf life of the laban and its nutritional characteristics.

### **4.1. Impact of lactic acid fermentation on physical properties of laban**

#### **4.1.1. Microstructure of laban**

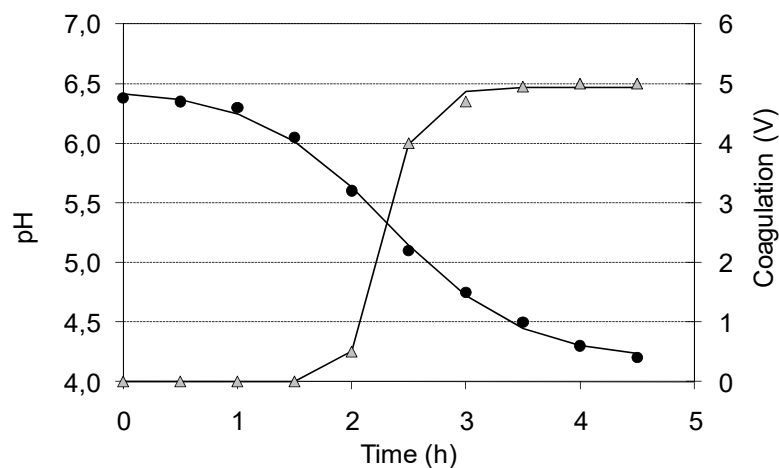
The microstructure of laban consists of a protein matrix composed of aggregated casein micelle chains and clusters, surrounding fat globules. Among these proteins, caseins  $\alpha$ S1,  $\alpha$ S2,  $\beta$  and  $\kappa$  (relative proportion: 4/1/3.7/1.4) account for around 80% of total nitrogen fraction of cow milk.

During laban fermentation, a destabilization of the casein complex occurs as a result of acidification, thus leading to aggregation and gel formation. The lactic acid that is excreted by lactic acid bacteria progressively converts the colloidal calcium/phosphate complex in the micelle to a soluble calcium phosphate fraction. The casein micelles are then gradually

depleted of calcium, which leads to coagulation of the casein at pH 4.6–4.8. The reactions of dissociation and aggregation of casein micelles can be summarized as follows:

- Between 6.7 and 5.8, calcium phosphate is partly solubilized thus inducing a dissociation of the casein micelles;
- Between pH 5.8 and 5.3, solubilization of calcium phosphate is entire. Protein interactions decrease inside the micelles and gelification starts, thanks to appearance of hydrophobic links between serum proteins;
- Between pH 5.3 and 4.8, dissociated caseins are integrated into acido-micelles whose hydration is reduced as compared to native micelles;
- Below pH 4.8, no more electrostatic repulsion occurs between acido-micelles because of their isoelectric pH (pH 4.6). As a consequence, hydrophobic interactions increase, thus enhancing aggregation of casein and strengthening the protein network.

This phenomenon is displayed on Figure 4 that shows the time course of coagulation as a function of acidification.



**Figure 4. Time course of acidification and coagulation during laban manufacture**  
 ● pH, △ coagulation

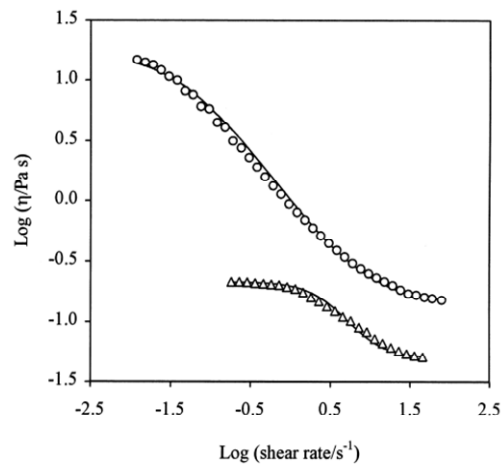
Depending on the acidification rate and the strains used, the coagulation is accompanied by syneresis. This phenomenon that is commonly observed in laban corresponds to the shrinkage of the gel that induces whey formation on the surface of the product. In laban, it can be reduced either by increasing the non-fat solid content of milk prior to fermentation or by heating the milk, for example, at 90°C for 10 min or at 120°C for 2 min. Heating causes denaturation of whey proteins, especially  $\beta$ -lactoglobulin, and weakens their interaction with the casein micelles. The whey protein-coated micelles form a finer gel than that formed from unheated or HTST pasteurized milk, with less tendency to syneresis.

### 4.1.2. Physical properties of laban

The physical structure of laban has been extensively studied by (Chammas, Saliba, & Béal, 2006). It is mainly the consequence of milk coagulation, as exopolysaccharides production has never been demonstrated in this fermented product (Duboc & Mollet, 2001). Six main descriptors of textural properties are used to characterize laban (Chammas, Saliba, Corrieu et al., 2006).

Quantification of laban texture is first obtained by measuring the firmness, the cohesiveness and the fracturability of the product. These characteristics can be determined by means of a texture analyzer, which allows measuring the force of penetration of a probe in the laban sample at a well-defined speed and at a given temperature, thus leading to the determination of the texture profile of the sample (Chammas, Saliba, Corrieu et al., 2006). For each force–time curve, obtained by two successive compressions of each sample, the force (in N) required to disrupt the sample is used as a measure of fracturability, the maximum force (F1, in N) recorded during the first compression as a measure of firmness and the ratio (dimensionless) of the area under the force curve measured during the second compression (A2) to the one measured during the first compression (A1) as a measure of cohesiveness. By measuring these parameters for 96 samples of milk fermented with *S. thermophilus* and *L. bulgaricus* isolates taken from laban samples, (Chammas, Saliba, Corrieu et al., 2006) obtained high firmness values comprised between 0.32 and 0.52 N. Fracturability of these samples ranged between 0.21 and 0.4 N and cohesiveness varied from 0.5 and 1.1, thus showing a high diversity among strains.

Laban also exhibits thixotropic rheological properties, i.e. the viscosity of the product decreases as the rate of shear increases. A typical relationship between viscosity and shear rate is shown on Figure 5 (Guizani et al., 2001). These properties are quantified by the apparent viscosity (Pa.s), which represents the flow behavior (where sample structure is destroyed) and the complex viscosity (Pa.s) that represents the viscoelastic behavior (where sample structure is not affected) of the product. These rheological measurements are generally performed by means of a rheometer, equipped with a cone-and-plate system (Chammas, Saliba, Corrieu et al., 2006). These authors measured the rheological properties of 96 fermented milks made with strains isolated from laban samples. The apparent viscosity slightly varied between 0.11 and 0.32 N, whereas the complex viscosity strongly differed, from 0.24 to 12.63 N. These values were lower than those measured with yogurt samples, thus indicating that strains used for laban manufacture can be considered as non-ropy strains, which is in agreement with the low cohesiveness and the high firmness of the products obtained with these bacteria (Chammas, Saliba, Corrieu et al., 2006) and with the results obtained by (Duboc & Mollet, 2001).



**Figure 5. Viscosity variation as a function of shear rate for commercial (○) and home-made (Δ) laban (From Guisani et al, 2001).**

Finally, as syneresis may occur, it has been determined by (Chammas, Saliba, Corrieu et al., 2006) for different laban products. Syneresis can be measured by gently collecting the whey on the surface of the samples at 4°C. The amount of collected whey is expressed in percentage (v/v) of fermented milk sample. It varied between 1.9% and 13.7% by considering the previous 96 different fermented milks (Chammas, Saliba, Corrieu et al., 2006).

These rheological and textural properties are major parameters of quality of laban. They can be controlled by varying the strains used, but also the total solids content of the milk, the heat treatment and homogenization of the milk.

Besides laban production, manufacture process of Labneh and other Middle Eastern fermented milks differs as these fermented products are concentrated by removing part of the serum (whey). This is done traditionally by stirring and then straining the laban in cloth or animal-skin bags. More recent technologies are now available, using membrane techniques (ultrafiltration and reverse osmosis) or centrifugation (Nsabimana, Jiang, & Kossah, 2005). The main differences with laban concern the rheological properties of labneh, which are the consequence of its higher total solids level (230-250 g/kg) (Ozer & Robinson, 1999).

## **4.2. Impact of lactic acid fermentation on laban flavor**

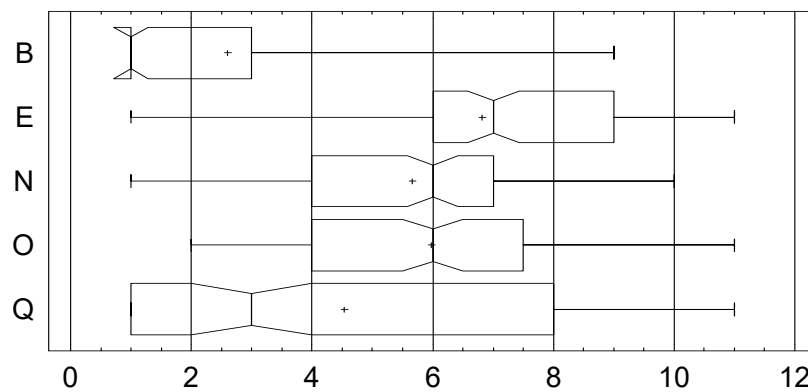
Lactic acid bacteria strongly determine the flavor of laban, for two main reasons: acidification and aroma compounds synthesis.

### **4.2.1. Taste of laban**

Lactic acid, which is the main fermentation metabolite, confers the laban a typical sharp and acidic taste. Depending on the product, the acid level may differ, artisanal products being more variable than industrial products. From sensory analysis or sourness, (Chammas, Saliba, & Béal, 2006) demonstrated that the acidity of 5 labans differs, as the ratings are



comprised between 1 and 11, on a 0-12 scale (Figure 6). This sourness was related to titratable acidity that varies between 9.2 and 17.5 g/kg. Among the tested samples, traditional laban samples shows lower acidity (9.2 to 17.4 g/kg) than laban elaborated with commercial strains (17.5 g/kg). These values are higher than those observed for yogurts, where lactic acid content varies between 9 and 10 g/kg (A. Y. Tamime & R. K. Robinson, 1999).



**Figure 6. Boxplot showing the distribution of ratings by panelists for five samples of laban for sourness (from Chammas 2006)**

(+) = represents the mean value of ratings given by the panelists for each sample

#### 4.2.2. Aroma compounds of laban

Lactic acid bacteria are responsible for the synthesis of aroma compounds, which contribute to the aroma of laban. These compounds include non-volatile acids (lactic and pyruvic acids), volatile acids (formic, acetic and butyric acid), carbonyl compounds (acetaldehyde, acetone, acetoin or diacetyl) and some miscellaneous compounds (certain amino acids, components formed by thermal degradation of protein, fat or lactose).

The identification of volatile compounds of fermented milks is generally done by gas chromatography. From (Chammas, Saliba, & Béal, 2006), aroma molecules have to be first extracted by using solid-phase microextraction (SPME) technique before being analyzed by gas chromatography coupled to mass spectrometry (GC-MS).

These authors demonstrated that twelve main aroma compounds were found in laban samples fermented by *S. thermophilus* and *L. bulgaricus*. Two compounds, furancarboxaldehyde and furanmethanol, were brought into the cultures by the heat-treated milk, where their relative content was equal to 18% and 28%, respectively. Four compounds were synthesized by both *S. thermophilus* and *L. bulgaricus* but in different amounts. They represented around 50% of the total flavor profile of *L. bulgaricus* cultures, but only 5% in *S. thermophilus* cultures. Milks fermented with *S. thermophilus* allows obtaining fermented milks with high contents of 2,3-butanedione, acetoin and 2,3-pentanedione (87%) whereas flavor profile of *L. delbrueckii* subsp. *bulgaricus* fermented milks included acetic, butanoic and hexanoic acids (48%) and acetaldehyde and acetone

(16%). Besides these species related differences, some diversity exists within a bacterial species (Chammas, Saliba, & Béal, 2006).

These sensory properties are major parameters of flavor of laban. They can be controlled by varying the kind and the number of strains used, as well as the balance between strains, in order to achieve fermented products with various flavor characteristics. From Table 2, strong differences exist between pure and mixed cultures, after milk fermentation, whereas differences between mixed cultures are more subtle. All labans obtained from mixed cultures display a complex aroma composition, including all kind of aroma compounds produced by the two species *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*.

**Table 2. Main aroma compounds identified in laban produced with two species of *S. thermophilus* (ES2 and FS2) and two species of *L. delbrueckii* subsp. *bulgaricus* (EL6 and FL5) (from Chammas, 2006)**

Strains	Acetaldehyde	Acetone	2-3, butanedione	2-3, pentanedione	Acetoin	Acetic acid	Butanoic acid	Hexanoic ac
ES2	0	2,53	59,84	4,83	24,13	1,16	2,76	1,70
FS2	0	4,67	54,24	5,41	24,97	1,03	3,03	1,81
EL6	5,00	8,40	0,00	0,00	0,00	18,05	16,47	10,47
FL5	6,43	12,93	0,00	0,00	0,00	24,55	15,92	11,01
EL6+ES2	1,44 <sup>b</sup>	3,53 <sup>b</sup>	25,18 <sup>a</sup>	23,61 <sup>c</sup>	24,91 <sup>f</sup>	5,88 <sup>b</sup>	4,97 <sup>b</sup>	3,73 <sup>b</sup>
EL6+FS2	1,14 <sup>ab</sup>	1,94 <sup>a</sup>	47,29 <sup>cd</sup>	22,18 <sup>c</sup>	13,80 <sup>bc</sup>	5,01 <sup>ab</sup>	3,46 <sup>ab</sup>	2,46 <sup>ab</sup>
EL6+ES2+FS2	1,11 <sup>ab</sup>	2,10 <sup>a</sup>	37,11 <sup>b</sup>	29,21 <sup>d</sup>	16,80 <sup>de</sup>	4,20 <sup>ab</sup>	3,47 <sup>ab</sup>	2,46 <sup>ab</sup>
FL5+ES2	0,85 <sup>a</sup>	1,62 <sup>a</sup>	52,83 <sup>de</sup>	18,34 <sup>b</sup>	15,36 <sup>cd</sup>	3,37 <sup>ab</sup>	3,09 <sup>ab</sup>	2,12 <sup>a</sup>
FL5+FS2	0,85 <sup>a</sup>	1,47 <sup>a</sup>	58,94 <sup>ef</sup>	16,52 <sup>b</sup>	10,61 <sup>a</sup>	2,83 <sup>a</sup>	3,33 <sup>ab</sup>	2,38 <sup>ab</sup>
FL5+ES2+FS2	0,60 <sup>a</sup>	1,24 <sup>a</sup>	55,01 <sup>f</sup>	17,86 <sup>b</sup>	14,52 <sup>c</sup>	2,60 <sup>a</sup>	2,77 <sup>a</sup>	1,90 <sup>a</sup>
EL6+FL5+ES2+FS2	0,94 <sup>ab</sup>	1,67 <sup>a</sup>	45,15 <sup>c</sup>	24,39 <sup>c</sup>	15,83 <sup>cd</sup>	3,00 <sup>a</sup>	3,64 <sup>ab</sup>	2,37 <sup>ab</sup>
EL6+FL5+ES2	0,69 <sup>a</sup>	1,29 <sup>a</sup>	36,60 <sup>b</sup>	31,34 <sup>d</sup>	18,42 <sup>e</sup>	4,24 <sup>ab</sup>	2,85 <sup>a</sup>	1,84 <sup>a</sup>
EL6+FL5+FS2	0,86 <sup>ab</sup>	1,27 <sup>a</sup>	62,71 <sup>e</sup>	11,66 <sup>a</sup>	12,39 <sup>ab</sup>	3,15 <sup>ab</sup>	2,60 <sup>a</sup>	2,20 <sup>a</sup>

Different letters account for statistically different values, according to multiple comparison test of Newman Keuls at 99.9 % for acetaldehyde, acetone and acetic, hexanoic and butanoic acids, 99 % for 2.3-pentanedione and 95 % for 2.3-butanedione and acetoin.

### 4.3. Impact of lactic acid fermentation on shelf life of laban

The shelf life of laban is defined as the time during which the product can be stored and consumed, without damaging the quality of the product. It is defined according to hygienic and sensory features. By considering hygienic properties, the low pH of the laban allows increasing the shelf life of the product as compared to that of pasteurized milk. The growth of bacterial pathogens and psychrotrophic bacteria is definitively restrained, whereas yeasts and molds that are acid-tolerant can spoil retail products. This resistance limits the shelf life of the laban to 30 days.

By considering sensory characteristics, post-acidification may occur, depending on the strains used (Kneifel, Jaros, & Erhard, 1993; Sofu & Ekinci, 2007). As a general rule, strains exhibiting high acidification activity are not recommended for mild products manufacture, since they may favor post-acidification during storage at 4°C. *L. delbrueckii* subsp. *bulgaricus* is generally involved in post-acidification as mainly D(-) lactic acid increases in the product during storage (Imhof & Bosset, 1994). This species is acid-tolerant, and has the ability to

produce lactic acid to levels of 1.7 g/100 mL, or even above depending on the strain (Robinson, Lucey, & Tamime, 2006).

The post-acidification is often associated with an increase of viscosity of the product (Martin, Skokanova, Latrille, Béal, & Corrieu, 1999), due to the increase in lactic acid and to exopolysaccharide production (Saint-Eve, Lévy, Le Moigne, Ducruet, & I., 2008), and with other defects such as syneresis (Kneifel et al., 1993). Thus, after fermentation, cooling must be done as rapidly as possible, so that post-acidification in product remains less than 0.3 pH units (Spreer, 1998). Besides this, starter cultures with reduced post-acidification behavior have been developed and are offered by many culture manufacturers (Kneifel et al., 1993).

From these considerations, in Lebanon, the usual shelf life of laban is comprised between 14 and 30 days.

#### **4.4. Modification of nutritional value of milk**

Fermented milks have been recognized by nutritionists as being beneficial to human health. By comparing with liquid milk, consumption of these products improves lactose digestion in lactose-intolerant individuals (Parra, Martinez de Morentin, Cobo, Lenoir-Wijnkoop, & Martinez, 2007) and protein digestibility (Beshkova, Simova, Frengova, Simov, & Adilov, 1998; Serra, Trujillo, Guamis, & Ferragut, 2009), increases the level of B-complex vitamins (Crittenden, Martinez, & Playne, 2003; Fabian, Majchrzak, Dieminger, Meyer, & Elmadfa, 2008), enhances calcium assimilation (Parra et al., 2007), and may assure health promoting effects related to probiotic (Uyeno, Sekiguchi, & Kamagata, 2008).

##### **4.4.1. Decreasing lactose malabsorption**

Lactose intolerance is the inability to digest significant amounts of lactose, the predominant sugar of milk. This inability results from a shortage of the enzyme lactase, which is normally produced by the cells that line the distal ileum (Mc Bean & Miller, 1998). Lactase breaks down milk sugar into simpler forms (glucose and galactose) that can be absorbed into the bloodstream. Lactose maldigestion is a relevant factor influencing milk and dairy product consumption, since lactase-deficiency often produces gastrointestinal symptoms after lactose intake (Parra et al., 2007). As undigested lactose reaches the colon, it is fermented by gas-producing bacteria and may cause bloating, flatulence, abdominal pain and diarrhea (Jarvis & Miller, 2002).

Laban and other fermented milks are better tolerated than milk by lactose-intolerant individuals (Varela-Moreiras, Antoine, Ruiz-Roso, & Varela, 1992). During the fermentation of milk, lactic acid bacteria hydrolyze the lactose and produce lactic acid as their main product, thus leading the lactose content of milk to drop from around 5% to 3%. In addition, after ingestion, some viable starter cultures reach the duodenum and participate in lactose digestion as they contain  $\beta$ -galactosidase activity (Pochart, Dewit, Desjeux, & Bourlioux, 1989), which could. These effects are observed only when living bacteria reach the intestine,

as a heat treatment of the fermented milk diminishes the effect on lactose digestibility due to enzymatic inactivation (Parra et al., 2007).

#### **4.4.2. Increasing protein digestibility**

Biochemical changes of milk during laban fermentation include not only carbohydrate metabolism and production of flavor components, but also a slight but significant degree of proteolysis. Many authors studied the properties of the proteolytic systems of lactic acid bacteria, revealing that proteolytic activity presents important variations, not only among the species but also among the strains belonging to the same species (Law & Haandrikman, 1997; Shihata & Shah, 2000). By considering casein hydrolysis, these variations are related to cell wall proteases, to transport systems of peptides from extracellular to intracellular media, and to cytoplasmic peptidases that produce amino acids (Savijoki, Ingmer, & Varmanen, 2006; Tzvetkova et al., 2007). Conversely, the cleavage of whey proteins during the fermentation by lactic acid bacteria involved in laban production remains undetectable (Bertrand-Harb, Ivanova, Dalgalarondo, & Haertle, 2003)

Many studies show the effect of lactic acid fermentation on protein digestibility and human health. According to (Béal & Sodini, 2003), the digestibility of the protein fraction of milk increases after yogurt fermentation, due to a modification of protein structure that facilitates the action of the proteolytic enzymes during the intestinal transit. In addition, fresh yoghurt intake results in higher acute leucine assimilation than during intake of pasteurized product (Parra & Martínez, 2007). By considering some specific strains, proteolysis lead to the formation of bioactive peptides, which are encrypted within the primary structure of milk proteins (Hayes, Stanton, Fitzgerald, & Ross, 2007) and are of special interest for their potential biological activities (Serra et al., 2009). These bioactive health-beneficial peptides are called  $\beta$ -casomorphins (Schieber & Brückner, 2000). They act as histamine releaser and are believed to be produced from milk proteins by *Lactobacillus* strains as reported by (Savijoki et al., 2006).

Casein hydrolysis continues during laban storage, thus leading to an increase in the total concentration of free amino acids that becomes four times higher than in milk after two days of storage at 4°C (Beshkova et al., 1998). An increase in hydrophobic bioactive peptides also occurs (Serra et al., 2009). Taking into account these considerations, the maintenance of living microorganisms in these products would be crucial for the health benefits claimed for fermented milks.

#### **4.4.3. Effect on vitamins content**

During milk fermentation, some vitamins are utilized by lactic acid bacteria while others are actively synthesized. The extent of synthesis and metabolism of these vitamins by bacterial cultures depends on the strain of bacteria used, the size of the inoculum and the conditions of fermentation (Fabian et al., 2008; Kneifel, Holub, & Wirthmann, 1989). Among the B-complex vitamins, an increase of the vitamin content of yogurt was reported for folic

acid, pyridoxine and biotin (Béal & Sodini, 2003; Crittenden et al., 2003). However, folic acid level remains relatively low in terms of recommended daily allowance, even though inoculum species were judiciously selected (Crittenden et al., 2003). The results of the study conducted by (Fabian et al., 2008) indicate that daily consumption of 200 g of yogurt for 2 weeks can contribute to the total intake of thiamin (vitamin B1) and riboflavin (vitamin B2).

Based on its ability to synthesize vitamins, it is supposed that lactic acid bacteria ingested with laban, which are the same than in yogurt, could be a supplementary source of vitamins in human nutrition.

#### **4.4.4. Effect on calcium assimilation**

Dairy products, and specifically laban, are important sources of calcium (Gambelli, Manzi, Panfili, Vivanti, & Pizzoferrato, 1999). However, calcium absorbability, or the availability of calcium for absorption by the intestines, is the first step towards bioavailability of calcium from these sources. According to (Takano & Yamamoto, 2003), calcium assimilation after consumption of both milk and fermented milk is comparable, whereas (Béal & Sodini, 2003) reported higher calcium uptake from fermented milk comparing to milk. In fact, by containing live starters, laban is considered as an important calcium source, since it can involve optimized calcium assimilation (Parra et al., 2007).

Lower acute calcium assimilation is obtained from pasteurized as compared to fresh fermented milk products. The negative effect of pasteurization is due, from one hand, to a decrease in lactose absorption and consequently to a decrease in calcium assimilation and from the other hand, to changes in the structure of the yogurt related to the pasteurization process that could also modify the calcium disposal (Parra et al., 2007). In fact, some proteins are denatured and aggregated during the heating process, and a non-specifically binding of proteins to calcium has been described, decreasing the availability of calcium to be absorbed by the human body (Halpern, 1993).

#### **4.4.5. Health effect from probiotic bacteria**

Fermented milks are considered as good probiotic-carrier food with health-promoting effects. Probiotics are defined as “live microorganisms which, when administered in adequate amounts, confer a health benefit on the host” (Sanders, 2003). The application of the term “probiotic” to the starter bacteria *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* is still debated. Moreover, no specific study has been conducted by considering the bacteria involved in laban production.

Survival during passage through the gastrointestinal tract is generally considered a key feature for probiotics to preserve their expected health-promoting effects (Holzapfel, Haberger, Geisen, Bjorkroth, & Schillinger, 2001). There have been conflicting studies concerning the recovery of *L. delbrueckii* subsp. *bulgaricus* and *S. thermophilus* from faecal samples after daily ingestion of fermented milk. Some authors confirmed that yogurt bacteria can be retrieved from feces (Elli et al., 2006; Guarner & Malagelada, 2003; Mater et

al., 2005), while others obtained negative results (Del Campo et al., 2005; Pedrosa et al., 1995). As an illustration, the results obtained by (Marteau, Minekus, Havenaar, & Huis In't Veld, 1997) show that survival of *L. delbrueckii* subsp. *bulgaricus* and *S. thermophilus* is two times lower in a gastric juice, than that of *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (figure 7). By sampling intestinal liquid by intubation, (Pochart et al., 1989) observed that after ingestion of  $10^{11}$  CFU/mL, only 1% survived in the duodenum ( $10^9$  CFU/mL). These bacteria were characterized by a high lactase activity. These probiotic traits are strictly strain specific and therefore, the diversity of these results is explained by the use of different strains in the different studies.

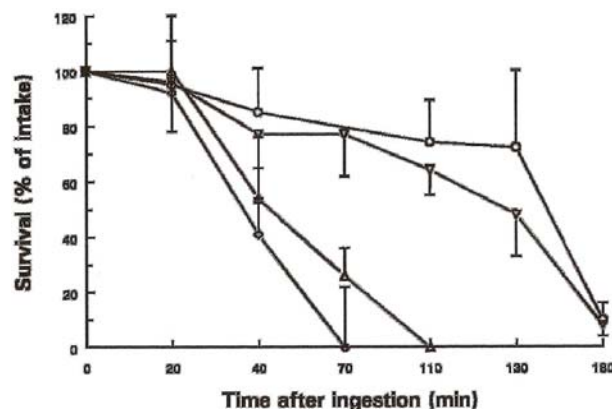


Figure 7. Comparative survival of ingested *Bifidobacterium bifidum* (○), *L. acidophilus* (▽), *L. bulgaricus* (△) and *S. thermophilus* (◇) in a gastric juice (From Marteau et al. 1997)

Values are expressed as mean percentages ( $\pm$ SE) of live bacteria relative to the ingested numbers ( $n = 6$  for each strain)

The consumption of probiotic products is helpful in maintaining good health, as demonstrated by the following beneficial effects of probiotic bacteria in humans:

- Control of intestinal infections by producing inhibitory/antimicrobial substances such as organic acids and bacteriocins and by stimulating the immune system (Ayar, Elgün, & Yazici, 2005; Fioramonti, Theodorou, & Bueno, 2003; Lourens-Hattingh & Viljoen, 2001; Uyeno et al., 2008);
- Reducing lactose intolerance by producing  $\beta$ -galactosidase (Lourens-Hattingh & Viljoen, 2001);
- Control of hypercholesterolemia (Ayar et al., 2005; Lourens-Hattingh & Viljoen, 2001);
- Anticarcinogenic activity (Lourens-Hattingh & Viljoen, 2001).

From this information, interest for the consumption of products containing probiotic microorganisms is increasing and the market is continuously expanding, mainly in European countries (Uyeno et al., 2008). In Lebanon and other Arab countries, the market of these fermented products remains very small but is going to increase.

## **5. General process of laban manufacture**

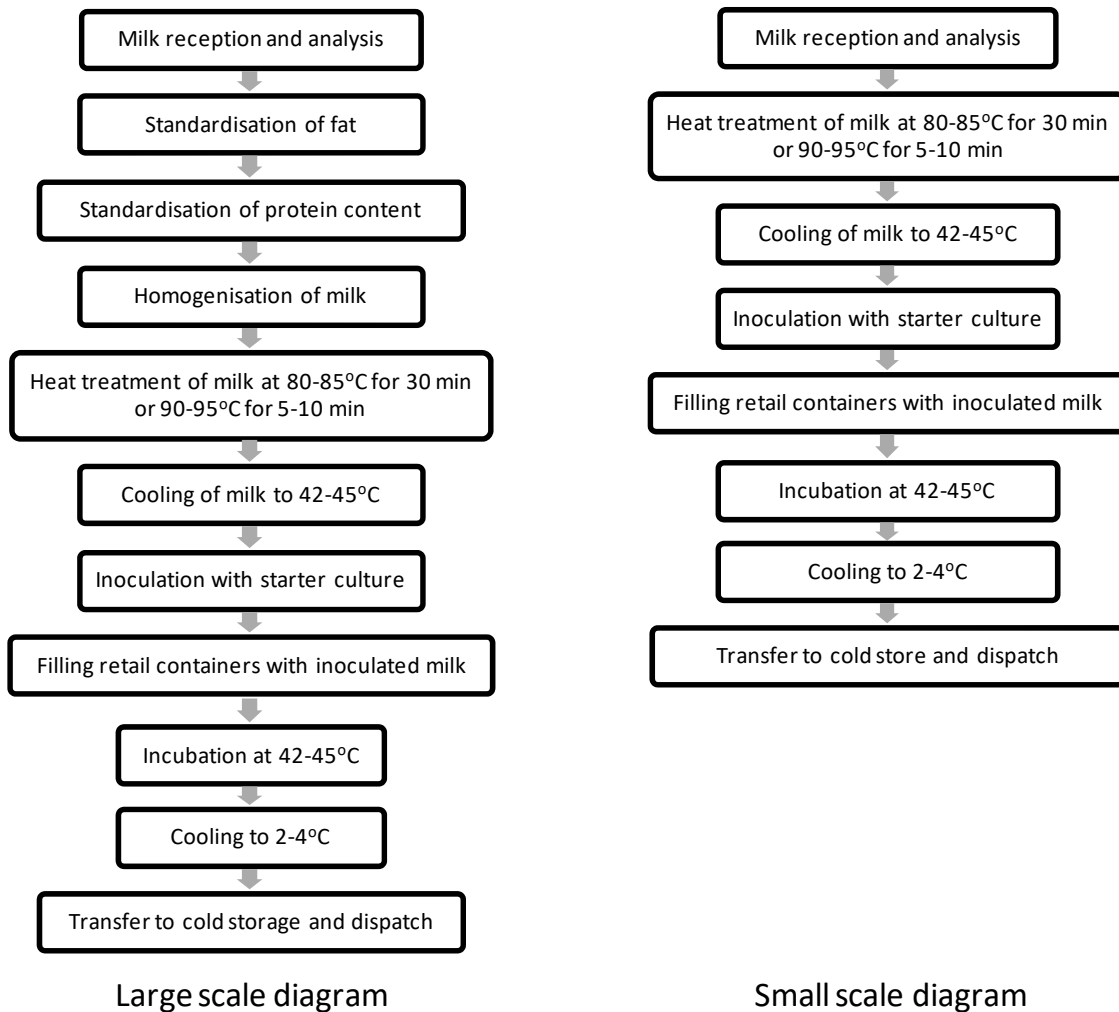
The general flow diagrams of laban manufacture are shown on Figure 8, for small scale and large scale production plants. They differ significantly according to artisanal and industrial units, the last one being more complicated and controlled than the first one.

### **5.1. Milk reception and analysis**

After being collected, fresh milk is stored in refrigerated tanks and transported into refrigerated road tankers to the production plant. At arrival, milk is first controlled, then pumped and filtrated to discard any solid residues (straw, leaves) and stored at least at 5°C in silos. These tanks, which are made of stainless steel, may reach 100 000 liters at industrial scale. Temperature control is achieved by using a jacket in which cold water is injected and circulates.

### **5.2. Standardization of milk fat**

Cow milk is constituted of water, lactose, fat, proteinaceous compounds (caseins and native serum proteins), other nitrogen compounds and minerals. The average composition of milk varies according to the breed, the lactation stage, the cattle feeding of the animals and the season. For a better regularity of the quality of the industrial products, it is standardized by considering both fat and protein contents.



**Figure 8. General flow diagrams of laban manufacture for large scale and small scale production plants**

Fat content of cow milk fluctuates between 3.8 and 4.2%. Standardization allows this fat content to achieve the desired value in the product, which varies between 0.5% and more than 3%, depending on the laban product (fat-free, low-fat or whole fat laban). This is done by first skimming the fat off the milk, and then by mixing the skimmed milk with cream in convenient proportions.

As an example, one shall use the following calculation to prepare a mix of 10 000 kg of milk at 1.6 % of fat. If  $m$  is the quantity of skimmed milk containing 0.1 % of fat and  $c$  the quantity of cream composed of 50 % of fat, the following number system has to be used:

$$m + c = 10\ 000$$

$$0.1 \cdot m + 50 \cdot c = 1.6 \cdot 10000$$

Then:

$$x = 9\ 700 \text{ and } y = 300$$

Finally, one shall mix 300 kg of cream with 9 700 kg of skimmed milk to obtain 10 000 kg of milk with 1.6 % of fat.

Fat separation is done by using centrifugal separators, at a temperature of 70°C. The skimming efficiency of these separators allows obtaining a residual fat in skim milk lower



than 0.07 %. The final fat content in milk is achieved by proportional mixing of skim milk and cream. At industrial scale, this operation is carried out automatically by using flow-meters, densimeters and pressure gauges to allow on-line calculation and monitoring of the fat content in the final product. Flow rates range between 7 000 and 45 000 L/h and accuracy of the final fat content is higher than 0.03 %.

### 5.3. Standardization of protein content

The total nitrogen content of cow milk varies between 2.9% and 3.7% all along the year. In order to reduce this variability and to allow the protein content to reach a value comprised between 3.2% and 5% in laban, milk is supplemented with dairy proteins. This standardization allows the laban to become more firm and to reduce syneresis in the product. It also increases the viscosity of the fermented milks.

Various practices are used to complete protein fortification. In traditional small production plants, milk is heated before fermentation to favor partial evaporation and to reduce the volume to achieve a protein content of around 5%. In larger production plants, milk supplementation is done by adding skimmed milk powder, concentrated milk, buttermilk powder, whey powder, whey protein concentrates or casein hydrolysates (A.Y. Tamime & Robinson, 2007a). Concentration by vacuum evaporation is also employed, by incorporating a single effect plate evaporator into a processing line.

As an example, one shall use the following calculation to prepare a mix of 10 000 kg of milk at 4.5 % of protein. If  $m$  is the quantity of skimmed milk containing 3.3 % of protein and  $p$  the quantity of skimmed milk composed of 34 % of protein, the following number system has to be used:

$$\begin{aligned}m + p &= 10\,000 \\3.3 \cdot m + 34 \cdot p &= 4.5 \cdot 10\,000\end{aligned}$$

Then:

$$m = 9\,610 \text{ and } p = 390$$

Finally, one shall mix 390 kg of skimmed milk powder with 9 610 kg of skimmed milk to obtain 10 000 kg of milk with 4.5 % of protein content.

During incorporation of dry ingredients such as milk powder or derived milk products into the aqueous phase (milk standardized in fat content), a complete dispersion and hydration is required, in order to avoid formation of lumps. It is also important to circumvent the incorporation of air into the product, in order to limit foam formation during mixing. Different kinds of equipments exist, either batch or continuous, all of them allowing high-quality cleaning in place. The mixing funnel involving a Venturi valve is the simplest system. As a general procedure, a tank is firstly filled with the fat standardized milk at 40-45 °C that is pumped at a flow rate of about 25 m<sup>3</sup>/h. The dry ingredients are then added into the funnel where they are progressively incorporated into the milk, thanks to the Venturi effect and the circulation of the milk. As alternative, on-line systems also exist that allow continuous incorporation of the milk powder. Mixing rates as high as of 45 kg/min are obtained with these systems.

## **5.4. Homogenization**

As it contains fat, milk is homogenized to provide a stable fat-in-water emulsion, in order to prevent creaming during fermentation at industrial scale. By reducing the size of fat particles, homogenization also helps to increase the viscosity of the product, to reduce syneresis during storage of the product and to enhance the whiteness of the milk. It is generally conducted before heat treatment or during heating of milk. Nevertheless, the study of (McKenna, 1987) demonstrates that downstream homogenization (post-pasteurization) allows obtaining a more stable laban by considering viscosity and syneresis.

Homogenization is a mechanical treatment that decreases the average diameter and increases the number and surface area of the fat globules in milk. Homogenizers used in laban production are composed of one or two stages. With high fat content products, two stages are needed. Homogenization is done by passing standardized milk under high pressure through a tiny orifice (0.1 mm diameter). The velocity is generally comprised between 100 and 400 m/s between the valve and the valve seat. The residence time ranges between 10 and 15  $\mu$ s and the flow rate is comprised between 4 000 and 20 000 L/h. During homogenization, pressure and temperature are controlled, at 10-20 MPa and 65-70°C, respectively. After homogenization, the mean diameter of fat globules is reduced from 3  $\mu$ m (range 1-10  $\mu$ m) to 0.5  $\mu$ m (range 0.2-2  $\mu$ m).

## **5.5. Heat treatment and cooling**

After homogenization, the mix is heat-treated in order to reduce the microbial contaminations and to improve the physical properties of the fermented milk (viscosity, water holding). It aims to degrade 80% of the two main serum proteins,  $\alpha$ -lactalbumin and  $\beta$ -lactoglobulin. When being denaturated, these two proteins attach themselves to casein surface through disulfur bonds. This phenomenon allows avoiding the casein micelles to link in large aggregates during further acidification, thus reducing syneresis and permitting a good consistency of the products.

Manufacturers provide equipments for batch or continuous heat treatment. Batch processes are conducted in a jacketed vat, by injecting hot water or steam in the double jacket or by means of heating coils surrounding the inner jacket. During heating and holding periods, the mix shall be agitated. Barem used for batch heat treatment is generally 85-90°C for 15-30 min.

Continuous processes are generally preferred for large scale production, as bulk milk is processed in a short span of time. It also allows reducing the vat volume, thus increasing productivity and saving costs. For continuous heat treatment, either scraped surface heat exchangers or plate heat exchangers are used, the last one being more popular. The main advantage of plate heat exchangers is that they offer a large transfer surface that is readily accessible for cleaning. Overall heat transfer coefficients are in the range of 2 400-6 000 J/m<sup>2</sup>.s.°C. Barems for heat treatment vary according to the industrial plant: 30 min at 85°C, 5

min at 90-95°C or 3 s at 115°C. The most used conditions are 92°C for 5-7 min, with a flow rate comprised between 4 000 and 20 000 L/h.

After heat treatment, milk is cooled down to fermentation temperature. Sometimes, cooling the milk at 4°C is necessary before or after inoculation, in order to delay incubation. It is then stored in vats under overpressure, for few hours.

## **5.6. Inoculation and fermentation**

Fermentation of heat treated milk starts by inoculating the selected bacteria (mainly lactobacilli and streptococci) into the milk. After inoculation, the liquid product is packed and the fermentation takes place in the retail containers. The process involves seven stages:

- Stabilization of milk temperature at fermentation temperature (generally 42°C during laban production)
- Addition of the starter culture to the milk
- Mixing in order to obtain a well homogenized mix
- Aseptic dispatching of the product into the retail containers
- Sealing of the retail containers
- Incubation at 42°C for a few hours controlled temperature cabinets or tunnels
- Cooling of the product in order to stop the fermentation

### **5.6.1. Inoculation modes**

Inoculation mode depends on the manufacturing scale. During traditional laban production, inoculation is done by using an artisanal starter consisting of an unknown number of undefined strains. These starter cultures are composed of fermented samples taken from a previous laban production. This procedure implies that artisanal starter composition is unknown and strongly variable, thus leading to a variable quality of the product (Chammas, Saliba, Corrieu et al., 2006).

Industrial starters are characterized by well-defined strains, which are combined in well defined balance. This procedure allows providing products with the desired characteristics that result from the metabolic activity and the technological properties of the strains during their growth in milk. This leads to a standard quality of the products and a high level of reproducibility of the processes, but lower sensory properties (Chammas, Saliba, & Béal, 2006). By considering large scale processes, direct vat inoculation is commonly employed. The starters are bought from industrial starter manufacturers which sell them either on frozen form (storage below -40°C) or on freeze-dried form (storage at or below 4°C). The inoculation rate is generally comprised between  $5 \times 10^5$  and  $5 \times 10^6$  CFU/mL. Semi-direct inoculation is also employed, thus involving a preculture step before laban inoculation. The cost of the starters is then reduced.

The balance between the bacterial species strongly influences the fermentation kinetic and the quality of the final product. During laban production, a ratio 1:1 between *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, is generally employed. The choice of the

strain also affects the acidification rates and the sensory characteristics of the products, as shown by (Chammas, Saliba, Corrieu et al., 2006).

### 5.6.2. Incubation of laban

Incubation of inoculated milk takes place directly in the cups. Depending on the size of the production plant, it is carried out either in warm rooms or tunnels at a temperature of 42°C. By considering small scale manufacturers, fermentation generally takes place in insulated chambers, inside which forced hot air is circulated. When incubation is finished, it is replaced by chilled air to cool down the product to 4°C.

In larger production plants, tunnel systems are preferred as they allow a continuous operation and are energy saving as compared to warm rooms. Two different tunnels are generally combined (one for incubation, one for cooling), but single tunnels with an incubation section and a cooling section are also available. The pallets with the laban cups move inside the tunnel with the help of conveyors. The length of the tunnel and the moving speed are calculated as a function of fermentation duration.

### 5.6.3. Kinetic of laban fermentation

During laban production, fermentation occurs according to Figure 9, which shows the time course of bacterial growth, substrate and lactic acid concentration as well as pH of milk.

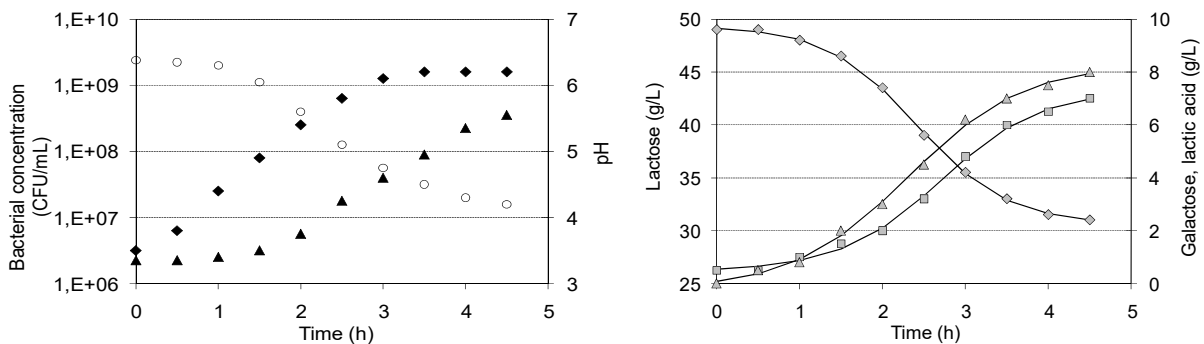


Figure 9. Time course of bacterial growth, substrate consumption, metabolite production and pH decrease during laban manufacture

◆ *S. thermophilus*, ■ *L. delbrueckii* subsp. *bulgaricus*, ○ pH, ◇ lactose, galactose, △ lactic acid

This kinetic was obtained by using a mixed culture of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*. From this figure, the growth of *S. thermophilus* starts earlier than that of *L. delbrueckii* subsp. *bulgaricus*. This is explained by the requirements of the lactobacilli in growth factors, formic acid and CO<sub>2</sub> that are synthesized by the streptococci (Ascon-Reyes et al., 1995). The development of *S. thermophilus* stops after 3.5 h at pH 4.5, because of the sensitivity of this species to low pH. Figure 9 also shows that *L. delbrueckii* subsp. *bulgaricus* starts to grow after 2h of fermentation. As lactobacilli

are more resistant to acidity, their growth continues until 6 h. The final ratio between these two species is generally unbalanced, in favor to the streptococci.

The lactose concentration, which is the main carbon substrate for bacterial growth, decreases from 49 g/L to 31 g/L. A significant lactose concentration remains in the final product. In the same time, the galactose and lactic acid concentrations increase in the fermented milk, thus reaching 7 g/L and 8 g/L, respectively in laban. The molar yield of lactic acid (calculated as the ratio between formed lactic acid and consumed lactose) is equal to 90%, thus corresponding to a homofermentative metabolism.

The pH of the milk decreases from pH 6.4 down to pH 4.2. This acidification is inversely correlated to the increase of the lactic acid concentration. As explained previously, this pH decrease induces the gel formation. This is confirmed by the measurement of the electrical signal obtained from a coagulation sensor, which varies as a function of pH as shown on Figure 4.

Finally, the laban composition is characterized by a bacterial concentration that is higher than  $10^9$  CFU/g, a level of acidity that depends on the time at which the fermentation is stopped, a lactose concentration that is lower than that found in milk, and the presence of specific compounds that may influence the sensory properties of the final product.

#### **5.6.4. Controls during laban fermentation**

The controls during laban production are quite basic. They mainly concern the temperature, and sometimes the pH or the titratable acidity that are checked in taken samples.

During incubation, the first operating condition to be controlled is the temperature. For laban fermentation, it is maintained between 40 and 45°C, the precise value depending on the strains used. In some artisanal production units, the temperature may be lower (30-37°C), thus leading to a longer fermentation time.

The length of the incubation is the second important parameter to be controlled. It depends on the final pH and acidity that are required. By considering laban, the length is comprised between 3 h (competitive industrial plants) and 12 h (artisanal production units). These values correspond to a final pH of 4.8 to 4.2 in the product, and a final titratable acidity level of 9.2 to 17.5 g/kg.

#### **5.7. Cooling**

A rapid cooling of the product is necessary to stop the fermentation, thus allowing the metabolic and enzymatic activities to cease. A quick cooling is also useful to achieve a more uniform quality of the product, and to prevent post-acidification. Cooling starts when the acidity of the product reaches the required value and lasts between 30 min and 1 h. For laban production, the cooling begins when the lactic acid level reaches 0.8 to 1% (pH 4.8 to 4.7), in order to obtain a final acidity in the product comprised between 1.2 to 1.4%.

The cooling is done by directly decreasing the temperature from incubation temperature to 5°C, either in a cold chamber, by circulating cooled air, or in a chill tunnel. This last practice allows a more rapid cooling, and then a lower post-acidification than the first one. Nevertheless, very rapid cooling may lead to whey separation, and the investment is much more expensive.

After cooling, laban cups are maintained at low temperature, i.e. between 4 and 8°C during storage, transport and distribution.

## **6. Packaging and storage of laban**

Packaging is crucial for safe delivery of laban to consumers. It constitutes also the most effective means of communication between dairy products manufacturer and eventual consumers.

Packaging is defined by (Brody, 2008) as “the totality of all elements required to confine the product within an envelope that functions as a barrier between the product and the environment, which is invariably hostile to the contained product unless the protection afforded by packaging is present”. It should meet specific requirements such as providing protection, easy handling, and offering a vehicle for a message. Environmental factors including temperature, moisture, oxygen, shock, compression and human disturbances have to be taken into account.

### **6.1. Packaging of laban**

#### **6.1.1. Packaging materials**

Among the primary packages used for fermented milks, plastic tubs (capacity 500 g, 1 Kg, 2 Kg and 5 Kg) and cups (capacity 125 g and 500 g) are the most common. The term “plastic” describes a family of materials derived from petrochemical sources, capable of being shaped or molded. Plastic package materials are characterized by their light weight, relative ease of fabrication, low cost and their malleability. The most commonly used plastic packaging materials for storage of fermented milks such as laban are high density polyethylene (HDPE), polypropylene (PP) and polystyrene (PS) (Cooper, 2007; Cutter, 2002). HDPE is used for the manufacture of tubs and their lids, while PP and PS are used for the manufacture of cups.

- HDPE is a semi-rigid translucent plastic. This polymer has good moisture and water resistance, but very poor barrier properties to gases. It has good heat resistance (up to about 100-120°C), making it suitable for “hot fill” and pasteurization. Usually, HDPE is used to form bottles for milk, drinkable yogurt and laban.
- PP is stiffer than HDPE and has good clarity, high moisture resistance but low gas barrier properties. Polypropylene is converted with the addition of thermoforming to make injection molded cups for yogurt and laban.

- PS is a hard polymer, with excellent transparency and good structural properties, but with poor oxygen and water vapor barrier properties. High-impact polystyrene (HIPS), made with incorporation of a blowing agent, is a material used commonly for packaging of yogurt and related products world-wide. However, oxygen diffuses into the product through the HIPS packaging during storage (Talwalkar, Miller, Kailasapathy, & Nguyen, 2004). Packaging alternatives to HIPS have been developed: polystyrene-based gas barrier which is effective in preventing diffusion of oxygen into the product during storage (Talwalkar et al., 2004) and polystyrene-based gas barrier with an active packaging film that can actively scavenge oxygen from the product (Cutter, 2002; Talwalkar et al., 2004).

Among these materials, polystyrene packaging is preferred for limiting aroma compound losses from the product and for avoiding the development of odor and aroma defects (Saint-Eve et al., 2008).

### **6.1.2. Packaging systems**

As laban is a set-style plain fermented milk, fermentation takes place in the final cups. In Lebanon and other Arab countries, traditional laban is produced by an old-age practice that entails a manual filling (for tubs capacity 1 Kg, 2 Kg and 5 Kg) and container uncovering during incubation. This process is slow, cumbersome, labor-intensive and unhygienic. The use of modern processing lines, that minimize manual handling during production and contamination during incubation, potentially yields a product with superior microbiological quality as compared to the traditional method. After inoculation, milk is pumped to the filling machine. Filled and sealed cups and tubs are placed on pallets and transferred to the incubation room. After fermentation, they are transported into cooling room.

Filling machines are designed to fill laban in two types of packaging systems (Robinson et al., 2006):

- Preformed containers are filled with laban then covered with an aluminum foil lid that is heat-sealed to the container (cups capacity 125 g) or covered with snap-on lid for tubs capacity 500 g, 1 Kg and 2Kg.
- Form-fill-seal containers are produced during the filling operation. Filling machine is fed by a roll of film, thermoforms cups, fills them and seals them with a foil lid. This system is applicable to 125 g-capacity cups and is suitable for large-scale operations.

The choice of the packaging machine and the type of container are influenced, among other considerations, by the marketing concepts and consumer acceptability (A. Y. Tamime, Robinson, & Latrille, 2001).

### 6.1.3. Storage of the products

Laban must be stored and transported in such a way that the product is not negatively influenced by the environment. For this purpose, to safeguard the quality of the product, temperatures below 4°C are chosen by most of manufacturers.

Laban containers are stored in cold refrigerated rooms (temperature between 2 and 4°C) for delivery to grocery stores or warehouses for distribution. The vehicles used for the distribution are mechanically refrigerated at the same temperatures as in the storage facilities in the factory (between 2 and 4°C).

## 6.2. Shelf life of laban

Shelf life of a product is the recommended time, during which the defined quality remains acceptable under specified conditions of distribution, storage and display. The shelf-life of foods is determined according to the proportion of product failure tolerated by the health risks that might result from consumption of failed/expired products (Labuza, 2000). For products that, practically, do not pose health hazards to consumers, shelf-life is determined at probability of failure of 50 % (L. Hough, Puglieso, Sanchez, & Da Silva, 1999).

Due to its inherent inability to support growth of pathogens, presumably due to its low pH (Muir and Banks, 2000), laban has an excellent safety record. *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* prevent the survival of *Escherichia coli* (Kasimoğlu & Akgün, 2004), *Listeria monocytogenes* (Gohil, Ahmed, Davies, & Robinson, 1995) and *Enterobacter* (Shaker, Osaili, & Ayyash, 2008) during the processing and post-processing stages.

However, as laban is sold with live bacteria, sensory properties of the product will change until they reach a limit beyond which the consumer will reject the product. According to (L. Hough et al., 1999), the shelf life of food products depends on the interaction of the food with the consumer: hence, a laban with long storage period may be accepted by a consumer who likes high-acid flavor but rejected by another consumer who does not like high-acid products (Curia, Aguerrido, Langohr, & Hough, 2005; Salvador, Fiszman, Curia, & Hough, 2005). Practically, changes in physical, chemical and microbiological structure of laban determine the storage and shelf life of the product.

A study conducted by (Saint-Eve et al., 2008) shows that a rapid evolution of low-fat yogurts stored at 4°C occurred during the first 14 days of storage, at the sensory and physicochemical levels. However, defects of the sensorial quality of whole-fat yogurts take longer time to appear than fat-free yogurts (Curia et al., 2005; Saint-Eve et al., 2008; Salvador et al., 2005).

Although low pH of laban inhibits the growth of many bacterial pathogens, psychrotrophic bacteria, yeasts and molds are acid-tolerant and can spoil retail products within anticipated shelf life of 21 days (A. Y. Tamime et al., 2001). Thus, in addition to deterioration of physicochemical properties, microbiological counts are used as indices for the end of shelf life (G. Hough, Langohr, Gómez, & Curia, 2003; Muir, 1996). Psychrotrophic bacteria have been reported as being the major determinant of shelf-life of yogurt and



related products (Lewis & Dale, 2000). The detrimental level to flavor quality of fermented milks is higher than  $10^7$  cfu/g (Bishop & White, 1986). Flavor defects have been reported when counts of yeasts and molds reach levels of  $10^5$  cfu/g (Kadamany, Khattar, Haddad, & Toufeili, 2003; A.Y. Tamime & Robinson, 2007b). (Al-Tahiri, 2005) reported a significant production of gas and the presence of unpleasant flavors in highly contaminated samples by yeasts and molds. According to (Sofu & Ekinici, 2007), there was an increase in the percentage of the total area having color of pale-greenish-yellow, grayish-yellow, light grayish-green and yellowish-green, 14 days after the storage of yogurt samples. The presence of these colors is associated with microbial spoilage of the product.

Consequently, shelf life of laban depends on the level of contamination and the production procedure (artisanal or industrial scale). Yogurt may be sold up to 21 days (Robinson & Itsaranuwat, 2006), 28 days (Salvador et al., 2005) or 35 days (Curia et al., 2005; Robinson & Itsaranuwat, 2006) after manufacture. In Lebanon, shelf life of laban varies between 14 and 30 days.

## 7. Evaluation of Laban quality

Before commercialization, laban is evaluated in terms of microbiological, physico-chemical and sensory aspects. These controls are done systematically for industrial products, whereas they remain insufficient for artisanal products.

### 7.1. Microbiological characteristics

The microbiological examination of the finished product includes checks for the survival of the starter organisms, as well as for the presence of undesirable spoilage species. According to the Lebanese legislation (Libnor, 1999), *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* dominate in the product, while other species of lactic acid bacteria may be also present in artisanal products. According to (Libnor, 1999), live starter bacteria must be abundant in laban until the end of the shelf life of the product, but the required counts are not specified. As a comparison, in yogurt, total bacterial population of starter origin should be at least  $10^6$  cfu/mL from French legislation (Loones, 1994), but it generally exceed  $2 \times 10^9$  cfu/mL (A. Y. Tamime et al., 2001).

Laban has a pH below 4.3 and therefore should be considered as a safe product. However, (Robinson & Itsaranuwat, 2006) reported that occasional checks for specific pathogens such as *Salmonella* spp. and *Listeria monocytogenes* are necessary. As fixed by the Lebanese standards, the total count of *Staphylococcus aureus* must be lower than 10 cfu/g and *Salmonella* should be absent in 25 g of sample (Libnor, 2007).

Microbiological check covers also yeast and molds, coliform bacteria and eventually foreign flora. Acid-tolerant yeasts that may grow in laban and affect its quality involve some species of *Saccharomyces*, *Kluyveromyces marxianus* var. *lactis* and *Kluyveromyces marxianus* var. *marxianus* (Robinson & Itsaranuwat, 2006; Robinson et al., 2006). Molds such as *Mucor* spp., *Rhizopus* spp., *Penicillium* spp. and *Aspergillus* spp. can grow at the product-

air interface (Robinson et al., 2006). In yogurt, total colony counts for non-starter bacteria should be lower than  $10^3$  cfu/g and yeast and mold counts should be inferior to 10 cfu/mL (A. Y. Tamime et al., 2001). However, the Lebanese standards tolerate higher values. Total and faecal coliform counts must be lower than  $5 \times 10^2$  cfu/g and yeast and mold counts should remain lower than  $10^3$  cfu/g (Libnor, 2007).

## 7.2. Physico-chemical characteristics

The physico-chemical tests cover fat content, solids-non-fat content, pH value, titratable acidity, foreign water content and gel firmness.

Retail products are designed as fat-free, low-fat and whole-fat laban with a fat content set by the legislation as follows (Libnor, 1999): less than 0.5 % for fat-free products, between 0.5 and 3 % for low-fat products, and not less than 3 % for whole-fat products. In Lebanon, the mean value of fat content in commercial whole-fat laban is 3.5 %, while in traditional laban the fat content varies between 3.3% and 4.2 % (Chammas, Saliba, & Béal, 2006). According to (Musaiger et al., 1998), the fat content of low fat laban is comprised between 1.2 and 1.4%. The production of diversified laban according to its fat content is carried out to satisfy diet-conscious consumers, but it has an effect on the acceptability of the products. Fat content affects the creamy smell, the flavor and the thickness of laban (Chammas, Saliba, & Béal, 2006).

Solids-non-fat (SNF) content indicates protein, lactose and mineral contents of the product. It depends on the degree of fortification of milk solids. The minimum SNF required in laban is 8.5 % follows (Libnor, 1999). This level is essential to obtain a firm coagulum, as stabilizers are not permitted under local regulations. In the products analyzed by (Musaiger et al., 1998), the SNF content was comprised between 8.8% and 9.3%, thus satisfying these rules. The mineral content was mainly characterized by potassium (132-146 mg/100g), calcium (120-128 mg/100g), phosphorus (101-104 mg/100g), sodium (56-81 mg/100g) and magnesium (11-12 mg/100g) (Musaiger et al., 1998).

Acidity of laban is important with respect to public safety and to product quality. Mild products with pH values higher than 4.5 can allow the survival of *Salmonella* for up to 10 days (Al-Haddad & Robinson, 2003) or *Escherichia coli* for up to 7 days (Massa, Altieri, & Quarante de Pace, 1977). The pH of laban has been determined by many authors. According to (Musaiger et al., 1998) it is comprised between 4.4 and 4.5, from (Baroudi & Collins, 1976) it is lower (pH 4.25) and from (Guizani et al., 2001), it depends on the manufacture scale (pH 4 in artisanal products and pH 4.5 in commercial products).

The acidity of final product is monitored according to consumer preference. Lactic acid content should not exceed 1.5 % in laban (Libnor, 1999). However, products available on the market show variable acidity values, ranging between 0.8 and 1.75 % (Chammas, Saliba, & Béal, 2006; Musaiger et al., 1998). As a consequence, laban is more acidic than other fermented milk products, where the lactic acid content is 0.9-1 % in yogurt (Musaiger et al., 1998; A.Y. Tamime & R.K. Robinson, 1999), 0.9-1.2 % in zabady produced in Egypt (Abd El-Salam, 2003), and 0.77 % in laban from Sultanate Oman (Guizani et al., 2001) respectively.

Although firmness and viscosity of laban are requested by the consumers, there is no legislation set to evaluate these two parameters. The study carried out by (Chammas, Saliba, & Béal, 2006) shows that there are big differences between laban samples in terms of gel firmness and apparent viscosity, due to variations in starter culture strains. From (Chammas, Saliba, & Béal, 2006), apparent viscosity of laban is comprised between 0.18 and 0.28 Pa.s, which is consistent with that of yogurt prepared with 3% fat in milk (apparent viscosity of 0.23 Pa.s, (Shaker, Jumah, & Abu-Jdayil, 2000)). Finally, even though no specific study concern laban, processing parameters may also affect the physical characteristics of this product, as demonstrated with yogurt (Béal et al., 1999; Hassan, Frank, Schmidt, & Shalabi, 1996; Lucey & Singh, 1998).

### **7.3. Sensory evaluation**

Consumers' perception of fermented milk products are mainly related to health, nutrition, sensory characteristics and pleasure (Ares, Giménez, & Gámbaro, 2008). Manufacturers should therefore ensure that their product is meeting the expectations of the consumers concerning natural laban. In Lebanon, labans with different acidity, texture and aroma profiles are present on the market to comply the consumers' preference.

From a sensory point of view, laban is characterized by its acidity, texture and aroma. As compared to other fermented milks, laban is more acidic (Chammas, Saliba, & Béal, 2006; Musaiger et al., 1998). Increase in fat and total solids content decreases the perception of acidity in yogurt (Robinson & Itsaranuwat, 2006) and leads to the appearance of granulated texture (Trachoo & Mistry, 1998).

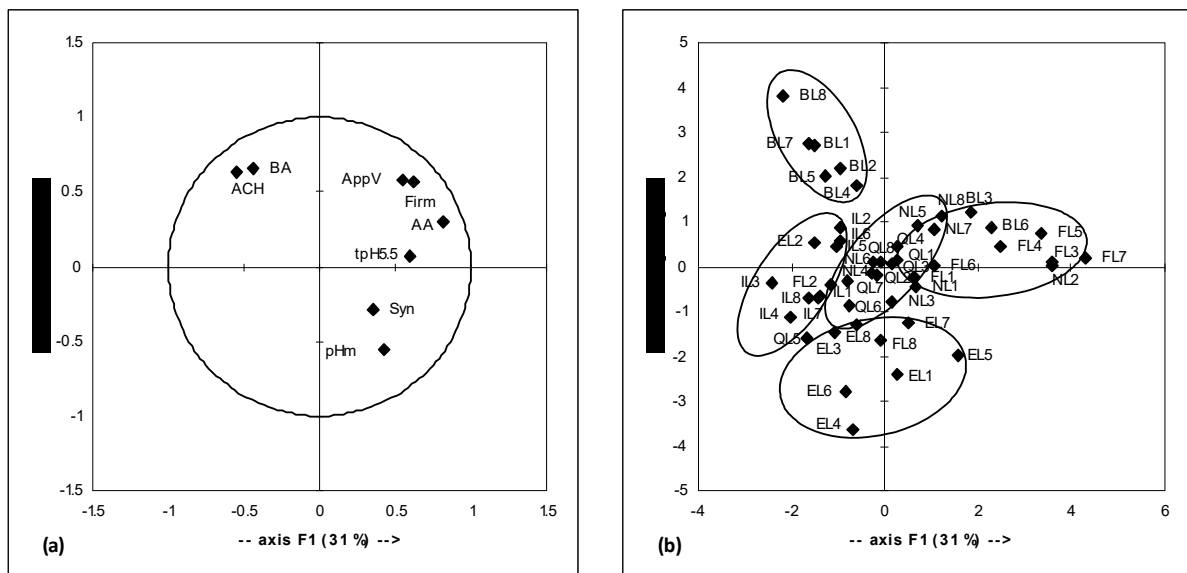
Laban is composed of a firm gel, as a result of the increase in lactic acid level (Saint-Eve et al., 2008). The composition of the starter strongly influences the gel firmness (Chammas, Saliba, & Béal, 2006). Laban is also characterized by its aroma, which intensity reflects the presence of aromatic compounds such as acetaldehyde, organic acids, 2,3-butanedione and acetoin (Chammas, Saliba, & Béal, 2006; Ott, Hugi, Baumgartner, & Chaintreau, 2000).

Sensory analysis is considered as an important technique to determine product quality. (Murray, Delahunty, & Baxter, 2001) reported that descriptive sensory tests profiled products on all their perceived sensory characteristics by training a group of panelists to reliably identify and score product attributes. Pre-established lists of sensory attributes related to fermented milk products can be found in the literature (Bodyfelt, Tobias, & Trout, 1988; Civille & Lyon, 1996; Martin et al., 1999). (Chammas, Saliba, & Béal, 2006) used a set of 23 attributes to differentiate samples of laban in terms of appearance, color, texture, odor, taste, and after-taste. Eight sensory attributes were related to appearance, color, and texture. Five attributes were used to describe odor (yogurt, creamy, butter, burnt, and yeast), five characteristics to represent flavor (sourness, bitterness, astringency, sweetness, and saltiness), and five descriptors to characterize aftertaste (sourness, bitterness, astringency, sweetness, and saltiness). These attributes, as well as their definitions, are given in Table 3.

**Table 3: Attributes used in the sensory evaluation of laban (from Chammas et al. 2006)**

Perception category	Attributes	Definition
Appearance	Gel firmness	Absence of syneresis
	Color	Scale yields from white to yellow
Texture assessed with the spoon	Gel-like	Product's ability to wiggle similar to a gelatin dessert
	Smooth	Presence of grain-size particles in the gel quantified by visual inspection of the spoon's back
	Thick	Product's ability to flow from the spoon
	Slimy	Product's ability to flow in a continuous way from the spoon
Texture assessed in the mouth	Thick	Product's flowing resistance assessed by pressing one spoonful of the product between the tongue and the palate
	Mouth coating	Product's ability to form a film lining the mouth

From the study of (Chammas, Saliba, & Béal, 2006), the physical properties of laban strongly vary according to the product under consideration. They demonstrate a variety of scores (comprised between 1 and 5.4 on a maximal scale of 0-6) by considering 19 laban samples. Figure 10 shows of the principal component analysis of the sensory ratings obtained from various labans samples, collected all around Lebanese Territory. This figure confirms that laban is a product with various sensory characteristics, thus encountering the diverse expectations of a wide range of consumers.



**Figure 10. Graphical representation of the principal component analysis of the sensory ratings for labans showing PC1 versus PC2: (a) Factor loadings; (b) Factor scores (From Chammas et al. 2006a)**  
 Abbreviations: gel firmness (Firm), color (Col), gel-like (Gel), smooth (Smoo), thickness assessed by the spoon (This), slimy (Slim), thickness assessed by the mouth (Thim), mouth coating (Mcoat), odor of yogurt (Yog), odor of butter (Butt), sourness (Sour) and sweetness (Sweet).

## 8. Conclusion

Laban is a major dairy product in the diet of many Middle Eastern countries. Production of laban is the result of the use of lactic acid bacteria, which lead to milk acidification, aroma compounds synthesis and texture development. Strong differences exist between artisanal and commercial products, owing to the strains used but also the manufacture process. These differences generate a great diversity in sensory properties, artisanal products being more typically flavored than commercial labans. On the other hand, microbiological traits are less controlled in artisanal products, thus inducing flavor defects, leading to insufficient process regularity and decreasing hygienic safety.

In the future, some progress have to be done in order to develop specific strains for laban, thus reaching consumers' preferences together with guaranteeing high safety and regularity of the products. This progress may also concern the definition of well defined starters, by combining these specific strains at well defined balances. Moreover, based on their health benefits for consumers, the development of probiotic strains for laban manufacture will permit an increase in laban consumption.

## 9. References

- Abd El-Salam, M. H. (2003). Fermented milks: Middle East. In H. Roginski, J. W. Fuquay & P. F. Fox (Eds.), *Encyclopedia of dairy sciences* (pp. 1041-1045). London: Academic Press.
- Adamberg, K., Kask, S., Laht, T. M., & Paalme, T. (2003). The effect of temperature and pH on the growth of lactic acid bacteria: a pH-auxostat study. *International Journal of Food Microbiology*, *85*(1-2), 171-183.
- Al-Haddad, K. S. H., & Robinson, R. K. (2003). Survival of salmonellae in bio-yoghurts. *Dairy Industries International*, *69*(7), 16-18.
- Al-Tahiri, R. (2005). A comparison on microbial conditions between traditional dairy products sold in Karak and same products produced by modern dairies. *Pakistan Journal of Nutrition*, *4*(5), 345-348.
- Al Ammouri, N. (2006). Comparative advantage of cow milk in Syria. In (Vol. 25, pp. 20). Syria: National Agricultural Policy Center (NAPC).
- Alqaisi, O., Ndambi, O. A., & Hemme, T. (2009). Development of milk production and the dairy industry in Jordan. *Livestock Research for Rural Development*, *21*(7), on-line edition.
- Alqaisi, O., Ndambi, O. A., Uddin, M. M., & Hemme, T. (2010). Current situation and the development of the dairy industry in Jordan, Saudi Arabia, and Syria. *Tropical Animal Health Production*, *42*, 1063-1071.
- Amrane, A., & Prigent, Y. (1999). Differentiation of pH and free lactic acid effects on the various growth and production phases of *Lactobacillus helveticus*. *Journal of Chemical Technology and Biotechnology*, *74*, 33-40.
- Anonymous. (2008). Syrian agricultural database: NAPC National Agricultural Policy Center: <http://www.napcsyr.org/sadb.htm>.

- Ares, G., Giménez, A., & Gámbaro, A. (2008). Understanding consumers' perception of conventional and functional yogurts using word association and hard laddering. *Food Quality and Preference*, *19*(7), 636-653.
- Ascon-Reyes, D. B., Ascon-Cabrera, M. A., Cochet, N., & Lebeault, J. M. (1995). Indirect conductance for measurements of carbon dioxide produced by *Streptococcus salivarius* spp. *thermophilus* TJ 160 in pure and mixed cultures. *Journal of Dairy Science*, *78*(1), 8-16.
- Ayar, A., Elgün, A., & Yazici, F. (2005). Production of a high nutritional value, aromatised yogurt with the addition of non-fat wheat germ. *Australian Journal of Dairy Technology*, *60*(1), 14-18.
- Baroudi, A. A. G., & Collins, E. B. (1976). Microorganisms and characteristics of Laban. *Journal of Dairy Science*, *59*(2), 200-202.
- Béal, C., & Corrieu, G. (1991). Influence of pH, temperature and inoculum composition on mixed cultures of *Streptococcus thermophilus* 404 and *Lactobacillus bulgaricus* 398. *Biotechnology and Bioengineering*, *38*(1), 90-98.
- Béal, C., & Corrieu, G. (1998). Production of thermophilic lactic acid starters in mixed cultures. *Lait*, *78*, 99-105.
- Béal, C., Louvet, P., & Corrieu, G. (1989, Novembre). *Optimal controlled pH and temperature for growth and acidification of Streptococcus thermophilus 404 and Lactobacillus bulgaricus 398*. Paper presented at the EEC Sectorial Meeting, Cultures Collections, Gènes, Italie.
- Béal, C., Marin, M., Fontaine, E., Fonseca, F., & Obert, J. P. (2008). Production et conservation des ferments lactiques et probiotiques. In G. Corrieu & F. M. Luquet (Eds.), *Bactéries lactiques : De la génétique aux ferments* (pp. 661-785). Paris: Tec&Doc Lavoisier.
- Béal, C., Skokanova, J., Latrille, E., Martin, N., & Corrieu, G. (1999). Combined effects of culture conditions and storage time on acidification and viscosity of stirred yoghurt. *Journal of Dairy Science*, *82*, 673-681.
- Béal, C., & Sodini, I. (2003). Fabrication des yaourts et des laits fermentés. In *Techniques de l'Ingénieur* (Vol. n° F6315, pp. 1-16). Paris.
- Bertrand-Harb, C., Ivanova, I. V., Dalgalarrodo, M., & Haertle, T. (2003). Evolution of  $\beta$ -lactoglobulin and  $\alpha$ -lactalbumin content during yoghurt fermentation. *International Dairy Journal*, *13*(1), 39-45.
- Beshkova, D. M., Simova, E. D., Frengova, G. I., Simov, Z. I., & Adilov, E. F. (1998). Production of amino acids by yogurt bacteria. *Biotechnology Progress*, *14*, 963-965.
- Bishop, J. R., & White, C. H. (1986). Assessment of dairy product quality and potential shelf life. *Journal of Food Protection*, *49*, 739-753.
- BMI Business Monitor International. (2010). Saudi Arabia Agribusiness Report Q3: <http://www.fastmr.com>. 52.
- Bodyfelt, F. W., Tobias, J., & Trout, G. M. (1988). *The sensory evaluation of dairy products*. New York: Van Nostrand Reinhold.

- Boyaval, P. (1989). Lactic acid bacteria and metal ions. *Lait*, 69(2), 87-113.
- Brody, A. L. (2008). Packaging milk and milk products. In R. C. Chandan, A. Kilara & N. P. Shah (Eds.), *Dairy processing and quality control* (pp. 443-464). Iowa, USA: Wiley-Blackwell.
- Chammas, G. I. (2006). *Caractérisation et utilisation de souches de bactéries lactiques thermophiles locales dans le développement de produits laitiers fermentés au Liban*. Unpublished Thèse de doctorat, Institut National Agronomique Paris-Grignon, Paris.
- Chammas, G. I., Saliba, R., & Béal, C. (2006). Characterization of the fermented milk "Laban" with sensory analysis and instrumental measurements. *Journal of Food Science*, 71(2), S156-162.
- Chammas, G. I., Saliba, R., Corrieu, G., & Béal, C. (2006). Characterization of lactic acid bacteria isolated from fermented milk " laban ". *International Journal of Food Microbiology*, 110(1), 52-61.
- Civille, G. V., & Lyon, B. G. (1996). *Aroma and flavor lexicon for sensory evaluation: terms, definitions, references, and examples*. Pennsylvania: American Society for Testing and Materials (ASTM).
- Cooper, I. (2007). Plastics and chemical migration into food. In K. A. Barnes, R. Sinclair & D. H. Watson (Eds.), *Chemical migration and food contact materials* (pp. 228-250). Cambridge, England: Woodhead Publishing Ltd.
- Corrieu, G., Spinnler, H. E., Jomier, Y., & Picque, D. (1988). Automated system to follow up and control the acidification activity of lactic acid starters. French: INRA.
- Courtin, P., Monnet, V., & Rul, F. (2002). Cell-wall proteinases PrtS and PrtB have a different role in *Streptococcus thermophilus* / *Lactobacillus bulgaricus* mixed cultures in milk. *Microbiology*, 148, 3413-3421.
- Crittenden, R. G., Martinez, N. R., & Playne, M. J. (2003). Synthesis and utilisation of folate by yoghurt starter cultures and probiotic bacteria. *International Journal of Food Microbiology*, 80, 217-222.
- Curia, A., Aguerri, M., Langohr, K., & Hough, G. (2005). Survival analysis applied to sensory shelf life of Yogurts - I: Argentine formulations. *Sensory and Nutritive Qualities of Food*, 70(7), S442-S445.
- Cutter, C. N. (2002). Microbial control by packaging: a review. *Critical Reviews in Food Science and Nutrition*, 42(2), 151-161.
- Del Campo, R., Bravo, D., Cantón, R., Ruiz-Garbajosa, P., García-Albiach, R., Montesi-Libois, A., et al. (2005). Scarce evidence of yogurt lactic acid bacteria in human feces after daily yogurt consumption by healthy volunteers. *Applied and Environmental Microbiology*, 71(1), 547-549.
- Desmazeaud, M. (1990). Le lait milieu de culture. *Microbiologie Aliments Nutrition*, 8, 313-325.
- Duboc, P., & Mollet, B. (2001). Applications of exopolysaccharides in the dairy industry. *International Dairy Journal*, 11(9), 759-768.

- Elli, M., Callegari, M. L., Ferrari, S., Bessi, E., Cattivelli, D., Soldi, S., et al. (2006). Survival of yogurt bacteria in the human gut. *Applied and Environmental Microbiology*, 72(7), 5113-5117.
- Fabian, E., Majchrzak, D., Dieminger, B., Meyer, E., & Elmadfa, I. (2008). Influence of probiotic and conventional yoghurt on the status of vitamins B1, B2 and B6 in young healthy women. *Annals of Nutrition and Metabolism*, 52, 29-36.
- Fajardo-Lira, C., Garcia-Garibay, M., Wachter-Rodarte, C., Farrés, A., & Marshall, V. M. (1997). Influence of water activity on the fermentation of yogurt made with extracellular polysaccharide-producing or non-producing starters. *International Dairy Journal*, 7(4), 279-281.
- Fernandez-Espla, M. D., Garault, P., Monnet, V., & Rul, F. (2000). *Streptococcus thermophilus* cell wall-anchored proteinase: Release, purification, and biochemical and genetic characterization. *Applied and Environmental Microbiology*, 66(11), 4772+.
- Fioramonti, J., Theodorou, V., & Bueno, L. (2003). Probiotics: what are they? What are their effects on gut physiology? *Best Practice & Research Clinical Gastroenterology*, 17(5), 711-724.
- Gambelli, L., Manzi, P., Panfili, G., Vivanti, V., & Pizzoferrato, L. (1999). Constituents of nutritional relevance in fermented milk products commercialised in Italy. *Food Chemistry*, 66, 353-358.
- Gohil, V. S., Ahmed, M. A., Davies, R., & Robinson, R. K. (1995). The incidence of *Listeria* in foods in the United Arab Emirates. *Journal of Food Protection*, 58, 102-104.
- Guarner, F., & Malagelada, J. R. (2003). Gut flora in the health and disease. *Lancet*, 360, 512-519.
- Guizani, N., Kasapis, S., & Al-Ruzeiki, M. (2001). Microbial, chemical and rheological properties of laban (cultured milk). *International Journal of Food Science and Technology*, 36(2), 199-205.
- Halpern, G. M. (1993). Benefits of yogurt. *International Journal of Immunotherapy*, 9, 65-68.
- Hassan, A. N., Frank, J. F., Schmidt, K. A., & Shalabi, S. I. (1996). Textural properties of yogurt made with encapsulated nonropy lactic cultures. *Journal of Dairy Science*, 79(12), 2098-2103.
- Hayes, M., Stanton, C., Fitzgerald, G., & Ross, R. P. (2007). Putting microbes to work: Dairy fermentation, cell factories and bioactive peptides. Part II: Bioactive peptides functions. *Biotechnology Journal*, 2, 435-449.
- Holzapfel, W. H., Haberer, P., Geisen, R., Bjorkroth, J., & Schillinger, U. (2001). Taxonomy and important features of probiotic microorganisms in food and nutrition. *American Journal of Clinical Nutrition*, 73(2), 365S-373S.
- Hough, G., Langohr, K., Gómez, G., & Curia, A. (2003). Survival analysis applied to sensory shelf life of foods. *Journal of Food Science*, 68, 359-362.
- Hough, L., Puglieso, M. L., Sanchez, R., & Da Silva, O. M. (1999). Sensory and microbiological shelf -life of commercial ricotta cheese. *Journal of Dairy Science*, 82, 454-459.



- Imhof, R., & Bosset, J. O. (1994). Relationships between microorganisms and formation of aroma compounds in fermented dairy products. *Zeitschrift für Lebensmittel Untersuchung und Forschung*, 198, 267-276.
- Jarvis, J. K., & Miller, G. D. (2002). Overcoming the barrier of lactose maldigestion to reduce health disparities. *Journal of the National Medical Association*, 94(2), 55-66.
- Kadamany, E. A., Khattar, M., Haddad, T., & Toufeili, I. (2003). Estimation of shelf-life of concentrated yogurt by monitoring selected microbiological and physicochemical changes during storage. *Lebensmittel-Wissenschaft und-Technologie*, 36, 407-414.
- Kasimoğlu, A., & Akgün, S. (2004). Survival of *Escherichia coli* O157:H7 in the processing and post-processing stages of acidophilus yogurt. *International Journal of Food Science and Technology*, 39, 563-568.
- Kilcawley, K. N., Wilkinson, M. G., & Fox, P. F. (1998). Review: enzyme modified cheese. *International Dairy Journal*, 8, 1-10.
- Kneifel, W., Holub, S., & Wirthmann, M. (1989). Monitoring of B-complex vitamins in yogurt during fermentation. *Journal of Dairy Research*, 56, 651-656.
- Kneifel, W., Jaros, D., & Erhard, F. (1993). Microflora and acidification properties of yogurt and yogurt-related products fermented with commercially available starter cultures. *International Journal of Food Microbiology*(18), 179-189.
- Krusch, U., Neve, H., Luschei, B., & Teuber, M. (1987). Characterization of virulent bacteriophages of *Streptococcus salivarius* subsp. *thermophilus* by host specificity and electron microscopy. *Kieler Milchwirtschaftliche Forschungsberichte*, 39(3), 155-167.
- Labuza, T. P. (1980). Influence of water activity on food product stability. *Food Technology*, 34(4), 36-41.
- Labuza, T. P. (2000). The search for shelf life. *Food Testing and Analysis*, 6, 26-36.
- Lacroix, C., & Lachance, O. (1988). Effect de l'Aw sur la survie de *Lactobacillus bulgaricus* et *Streptococcus thermophilus* et le développement d'acidité dans le yogourt conservé au froid. *Canadian Institute of Food Science and Technology Journal*, 21(5), 501-510.
- Law, J., & Haandrikman, A. (1997). Review article: Proteolytic enzymes of lactic acid bacteria. *International Dairy Journal*, 7, 1-11.
- Lebanese Ministry of Agriculture. (2007). *State of agriculture in Lebanon 2006-2007*. Lebanon.
- Lemoine, R. (2002). La filière laitière libanaise : Un potentiel d'investissements dans l'élevage et l'industrie. *Revue Laitière Française*, 619, 12-15.
- Letort, C., & Juillard, V. (2001). Development of a minimal chemically-defined medium for the exponential growth of *Streptococcus thermophilus*. *Journal of Applied Microbiology*, 91(6), 1023-1029.
- Lewis, M., & Dale, R. H. (2000). Chilled yogurt and other dairy desserts. In D. Man & A. Jones (Eds.), *Shelf life evaluation of foods* (pp. 89-109). Maryland: Aspen Publishers.
- Libnor. (1999). Fermented milks, Standard n° 33. In *Lebanese Standards Institution, Ministry of Industry*. Beirut, Lebanon.

- Libnor. (2007). Lebanese standards of dairy products: microbiological limits, standard n° 510. In *Lebanese Standards Institution, Ministry of Industry*. Beirut, Lebanon.
- Loones, A. (1994). Laits fermentés par les bactéries lactiques. In H. de Roissart & F. M. Luquet (Eds.), *Bactéries lactiques* (pp. 135-154). Uriage, France.
- Lourens-Hattingh, A., & Viljoen, B. C. (2001). Yogurt as probiotic carrier food. *International Dairy Journal*, 11(1-2), 1-17.
- Lucey, J. A., & Singh, H. (1998). Formation and physical properties of acid milk gels: A review. *Food Research International*, 30(7), 529-542.
- Marteau, P., Minekus, M., Havenaar, R., & Huis In't Veld, J. H. J. (1997). Survival of lactic acid bacteria in a dynamic model of the stomach and small intestine: validation and the effects of bile. *Journal of Dairy Science*, 80(6), 1031-1037.
- Martin, N., Skokanova, J., Latrille, E., Béal, C., & Corrieu, G. (1999). Influence of fermentation and storage conditions on the organoleptic properties of plain low fat stirred yoghurts. *Journal of Sensory Studies*, 14(2), 139-160.
- Massa, S., Altieri, V., & Quarante de Pace, R. (1977). Survival of *Escherichia coli* 0157 :H7 in yoghurt during preparation and storage at 4°C. *Letters in Applied Microbiology*, 24, 347-350.
- Mater, D. D. G., Bretigny, L., Firmesse, O., Flores, M.-J., Mogenet, A., Bresson, J.-L., et al. (2005). *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* survive gastrointestinal transit of healthy volunteers consuming yoghurt. *FEMS Microbiology Letters*, 250, 185–187.
- Mc Bean, L. D., & Miller, G. D. (1998). Allaying fears and fallacies about lactose maldigestion. *Journal of the American Dietetic Association*, 98(6), 671-676.
- McKenna, A. B. (1987). Effects of homogenization pressure and stabilizer concentration on the physical stability of longlife Laban. *New Zealand Journal of Dairy Science and Technology*, 22(2), 167-174.
- Muir, D. D. (1996). The shelf life of dairy products: 2 Raw milk and fresh products. *Journal of the Society of Dairy Technology*, 49, 44-48.
- Murray, J. M., Delahunty, C. M., & Baxter, I. A. (2001). Descriptive sensory analysis: past, present and future. *Food Research International* 34, 461-471.
- Musaiger, A. O., Al-Saad, J. A., Al-Hooti, D. S., & Khunji, Z. A. (1998). Chemical composition of fermented dairy products consumed in Bahrain. *Food Chemistry*, 61(1-2), 49-52.
- Nasreddine, L., Hwalla, N., Sibai, A., Hamzé, M., & Parent-Massin, D. (2006). Food consumption patterns in an adult urban population in Beirut, Lebanon. *Public Health Nutrition*, 9(2), 194-203.
- Neve, H. (1996). Bacteriophage. In T. M. Cogan & J.-P. Accolas (Eds.), *Dairy Starter Cultures* (pp. 157–189). New York: VCH Publishers.
- Nsabimana, C., Jiang, B., & Kossah, R. (2005). Manufacturing, properties and shelf life of labneh: a review. *International Journal of Dairy Technology*, 58(3), 129-137.

- Ott, A., Germond, J. E., & Chaintreau, A. (2000). Origin of acetaldehyde during milk fermentation using C-13- labeled precursors. *Journal of Agricultural and Food Chemistry*, 48(5), 1512-1517.
- Ott, A., Hugi, A., Baumgartner, M., & Chaintreau, A. (2000). Sensory investigation of yoghurt flavour perception: mutual influence of volatiles and acidity. *Journal of Agricultural and Food Chemistry*, 48, 441-450.
- Ozer, B. H., & Robinson, R. K. (1999). The behaviour of starter cultures in concentrated yoghurt (Labneh) produced by different techniques. *Lebensmittel-Wissenschaft und-Technologie*, 32(7), 391-395.
- Parra, D. M., Martinez de Morentin, B. E., Cobo, J. M., Lenoir-Wijnkoop, I., & Martinez, J. A. (2007). Acute calcium assimilation from fresh or pasteurized yogurt depending on the lactose digestibility status. *Journal of the American College of Nutrition*, 26(3), 288-294.
- Parra, D. M., & Martínez, J. A. (2007). Amino acid uptake in lactose intolerance from a probiotic milk. *British Journal of Nutrition*, 98(Suppl 1), S101-S104.
- Pedrosa, M. C., Golner, B. B., Goldin, B. R., Barakat, S., Dallal, G. E., & Russell, R. M. (1995). Survival of yogurt-containing organisms and *Lactobacillus gasseri* (ADH) and their effect on bacterial enzyme activity in the gastrointestinal tract of healthy and hypochlorhydric elderly subjects. *American Journal of Clinical Nutrition*, 61, 353-359.
- Perez, P. F., De Antoni, G. L., & Anon, C. (1991). Formate production by *Streptococcus thermophilus* cultures. *Journal of Dairy Science*, 74(9), 2850-2854.
- Pochart, P., Dewit, O., Desjeux, J.-F., & Bourlioux, P. (1989). Viable starter culture, b-galactosidase activity, and lactose in duodenum after yogurt ingestion in lactase-deficient humans. *American Journal of Clinical Nutrition*, 49, 828-831.
- Poolman, B., Knol, J., Henderson, P. J., Liang, W. J., Le Blanc, G., Pourcher, T., et al. (1996). Cation and sugar selectivity determinants in a novel family of transport proteins. *Molecular Microbiology*, 19, 911-922.
- Quiberoni, A., Guglielmotti, D., Binetti, A., & Reinheimer, J. (2004). Characterization of three *Lactobacillus delbrueckii* subsp. *bulgaricus* phages and the physicochemical analysis of phage adsorption. *Journal of Applied Microbiology*, 96(2), 340-351.
- Rajagopal, S. N., & Sandine, W. E. (1990). Associative growth and proteolysis of *Streptococcus thermophilus* and *Lactobacillus bulgaricus* in skim milk. *Journal of Dairy Science*, 73(4), 894-899.
- Rao, D. R., Reddy, A. V., OPulusani, S. R., & Cornwell, P. E. (1984). Biosynthesis and utilization of folic acid and vitamine B12 by lactic acid cultures in skim milk. *Journal of Dairy Science*, 67, 1169-1174.
- Rault, A., Bouix, M., & Béal, C. (2009). Fermentation pH influences the physiological-state dynamics of *Lactobacillus bulgaricus* CFL1 pH-controlled culture. *Applied and Environmental Microbiology*, 75(13), 4374-4381.
- Reiter, B. (1978). Antimicrobial systems in milk. *Journal of Dairy Research*, 45(1), 131-147.
- Robinson, R. K., & Itsaranuwat, P. (2006). Properties of yoghurt and their appraisal. In T. A.Y. (Ed.), *Fermented milks* (pp. 76-94). Oxford, UK: Blackwell Science Ltd.

- Robinson, R. K., Lucey, J. A., & Tamime, A. Y. (2006). Manufacture of yoghurt. In A. Y. Tamime (Ed.), *Fermented milks* (pp. 53-75). Oxford, England: Blackwell Science Ltd.
- Sadi, M. A., & Henderson, J. C. (2007). In search of greener pastures, Al-Marai and dairy food business in Saudi Arabia. *British Food Journal*, *109*(8), 637-647.
- Saint-Eve, A., Lévy, C., Le Moigne, M., Ducruet, V., & I., S. (2008). Quality changes in yogurt during storage in different packaging materials. *Food Chemistry*, *110*, 285-293.
- Salvador, A., Fiszman, S. M., Curia, A., & Hough, G. (2005). Survival analysis applied to sensory shelf life of yogurts - II: Spanish formulations. *Sensory and Nutritive Qualities of Food*, *70*(7), S446-S449.
- Samet-Bali, O., Bellila, A., Ayadi, M. A., Marzouk, B., & Attia, H. (2009). A comparison of the physicochemical, microbiological and aromatic composition of traditional and industrial Leben in Tunisia. *International Journal of Dairy Technology*, *63*(1), 98-104.
- Sanders, M. E. (2003). Probiotics: considerations for human health. *Nutrition Reviews*, *61*, 91-99.
- Savijoki, K., Ingmer, H., & Varmanen, P. (2006). Proteolytic systems of lactic acid bacteria. *Applied Microbiology and Biotechnology*, *71*(4), 394-406.
- Schieber, A., & Brückner, H. (2000). Characterization of oligo- and polypeptides isolated from yoghurt. *European Food Research and Technology* *210*, 310-313.
- Serra, M., Trujillo, A. J., Guamis, B., & Ferragut, V. (2009). Proteolysis of yogurts made from ultra-high-pressure homogenized milk during cold storage. *Journal of Dairy Science*, *92*, 71-78.
- Shaker, R. R., Jumah, R. Y., & Abu-Jdayil, B. (2000). Rheological properties of plain yogurt during coagulation process: impact of fat content and preheat treatment of milk. *Journal of Food Engineering*, *44*(175-180).
- Shaker, R. R., Osaili, T. M., & Ayyash, M. (2008). Effect of thermophilic lactic acid bacteria on the fate of *Enterobacter zakazakii* during processing and storage of plain yogurt. *Journal of Food Safety*(28), 170-182.
- Shihata, A., & Shah, N. P. (2000). Proteolytic profiles of yogurt and probiotic bacteria. *International Dairy Journal*, *10*(5-6), 401-408.
- Sofu, A., & Ekinici, F. Y. (2007). Estimation of storage time of yogurt with artificial neural network modeling. *Journal of Dairy Science*, *90*(7), 3118-3125.
- Spreer, E. (1998). *Milk and dairy product technology*. New York: Marcel Dekker Inc.
- Stefanitsi, D., & Gareil, J. (1997). A zinc dependent proteinase from the cell wall of *Lactobacillus delbrueckii* subsp. *bulgaricus*. *Letters in Applied Microbiology*, *24*, 180-184.
- Surono, I. S., & Hosono, A. (2003). Fermented milks: types and standards of identity. In H. Roginski, J. W. Fuquay & P. F. Fox (Eds.), *Encyclopedia of Dairy Sciences* (pp. 1018-1023). London: Academic Press.
- Takano, T., & Yamamoto, N. (2003). Fermented milks: Health effects of fermented milks. In H. Roginski, J. W. Fuquay & P. F. Fox (Eds.), *Encyclopedia of dairy sciences* (Vol. 2, pp. 1063-1069). London: Academic Press.

- Talwalkar, A., Miller, C. W., Kailasapathy, K., & Nguyen, M. H. (2004). Effect of materials and dissolved oxygen on the survival of probiotic bacteria in yoghurt. *International Journal of Food Science and Technology*, *39*, 605-611.
- Tamime, A. Y., & Robinson, R. K. (1999). Traditional and recent developments in yoghurt production and related products. In C. press (Ed.), *Yoghurt - Science and technology* (Vol. Chap. 5, pp. 306-388). Cambridge (England): Woodhead Publishing Limited.
- Tamime, A. Y., & Robinson, R. K. (1999). *Yoghurt: science and technology*. Cambridge, England: Woodhead publishing limited.
- Tamime, A. Y., & Robinson, R. K. (2007a). Background to manufacturing practice. In A. Y. Tamime & R. K. Robinson (Eds.), *Yoghurt: science and technology* (3rd ed., pp. 11-128). Boca Raton, Florida: CRC Press.
- Tamime, A. Y., & Robinson, R. K. (2007b). *Yoghurt: science and technology* (3rd ed.). Boca Raton, Florida: CRC Press.
- Tamime, A. Y., Robinson, R. K., & Latrille, E. (2001). Yogurt and other fermented milks. In A. Y. Tamime & B. A. Law (Eds.), *Mechanisation and automation in dairy technology* (pp. 152-203). Sheffield, England: Sheffield Academic Press.
- Thompson, J., & Gentry-Weeks, C. R. (1994). Métabolisme des sucres par les bactéries lactiques. In H. De Roissard & F. M. Luquet (Eds.), *Bactéries lactiques* (Vol. 1, pp. 239-290). Uriage: Lorica.
- Trachoo, N., & Mistry, V. V. (1998). Application of ultrafiltered sweet buttermilk and buttermilk powder in the manufacture of nonfat and low fat yogurts. *Journal of Dairy Science*, *81*, 133-138.
- Tzvetkova, I., Dalgalarondo, M., Danova, S., Iliev, I., Ivanova, I., Chobert, J. M., et al. (2007). Hydrolysis of major dairy proteins by lactic acid bacteria from Bulgarian yogurts. *Journal of Food Biochemistry*, *31*(5), 680-702.
- Uyeno, Y., Sekiguchi, Y., & Kamagata, Y. (2008). Impact of consumption of probiotic lactobacilli-containing yogurt on microbial composition in human feces. *International Journal of Food Microbiology*, *122*, 16-22.
- Varela-Moreiras, G., Antoine, J. M., Ruiz-Roso, B., & Varela, G. (1992). Effects of yogurt and fermented-then-pasteurized milk on lactose absorption in an institutionalized elderly group. *Journal of the American College of Nutrition* *11*(2), 168-171.
- Vinderola, C. G., Mocchiutti, P., & Reinheimer, J. A. (2002). Interactions between lactic acid starter and probiotic bacteria used for fermented milk products. *Journal of Dairy Science*, *85*(4), 721-729.
- Wemekamp-Kamphuis, H. H., Karatzas, A. K., Wouters, J. A., & Abee, T. (2002). Enhanced levels of cold shock proteins in *Listeria monocytogenes* LO28 upon exposure to low temperature and high hydrostatic pressure. *Applied and Environmental Microbiology*, *68*(2), 456-463.
- Zink, R., Elli, M., Reniero, R., & Morelli, L. (2000). Growth medium for lactobacilli containing amino acids, nucleosides and iron. USA: Nestec S.A.

