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Full Length Research Paper

Enumeration and identification of lactic microflora in Algerian goats' milk

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A total of 153 strains of lactic acid bacteria were isolated from Algerian goats' milk. The strains were identified according to morphological, biochemical and physiological criteria, as well as the use of the API system and SDS-PAGE technique. Identification of the isolates revealed the presence of six genera: *Enterococcus* (41.82%), *Lactobacillus* (29.40), *Lactococcus* (19.60%), *Leuconostoc* (4.57%), *Streptococcus thermophilus* (3.26%) and *Pediococcus* (1.30%). The predominant strains belong to *Enterococcus faecium* (24 isolates), *Enterococcus durans* (22 isolates), *Lactococcus lactis* subsp. *lactis* (25 isolates), *Lactobacillus rhamnosus* (9 isolates) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (7 isolates).

Key words: Lactic acid bacteria, goat's milk, identification, enumeration.

INTRODUCTION

Milk is a complete food, containing proteins, fats, carbohydrates, vitamins and mineral salts (Park et al., 2007). Goat's milk is widely used for home consumption worldwide and to produce different cheeses and yoghurts (Pandya and Ghodke, 2007). The cheeses as several dairy products are obtained by fermentation and addition of starter cultures to raw or sterilized milk which convert its various elements into new molecules with new organoleptic, hygienic and even medical properties (Remeuf, 1992).

In Algeria, much of dairy products are manufactured by traditional methods, using raw cows or goats and also ewe's milk. El-Klila a traditional cheese is made from the raw cow or goat milk (Boubekri and Otha, 1996). The cheese fermentation, like many traditional fermenting processes, is spontaneous and uncontrolled and so involves several food microorganisms whose type are influenced by the environmental conditions of the area where the cheese is produced. Microorganisms which are responsible for the acid production in cheese making are lactic acid bacteria (LAB) (Boubekri and Otha, 1996; Cheriguene et al., 2006). They are extensively used in fermenting a large variety of food products (Cogan, 1980).

Lactic acid bacteria are widely distributed in the nature. They could be isolated from soils, waters, plants, silages, waste products, and also from the intestinal tract of animals and humans (Axelsson, 1998). They consist of a number of bacterial genera within the phylum Firmicutes. The genera *Carnobacterium*, *Enterococcus*, *Lactobacillus*, *Lactococcus*, *Lactosphaera*, *Leuconostoc*, *Melissococcus*, *Oenococcus*, *Pediococcus*, *Streptococcus*, *Tetragenococcus*, *Vagococcus* and *Weissella* are recognized as LAB (Jay, 2000).

Lactic acid bacteria (LAB) were first isolated from milk (Carr et al., 2002; Sandine et al., 1972) and have since been found in such foods and fermented products as meat, milk products, vegetables, beverages and bakery products (Gobbetti and Corsetti, 1997; Liu, 2003; O'Sullivan et al., 2002). LAB have been used as a flavoring and texturizing agent as well as a preservative in food for centuries and are now added as starters in food (Daly and Davis, 1998).

To our knowledge in Algeria, little information exists on lactic microflora in goat's milk. The objectives of this stu-

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Table 1. Microbiological profile of West Algerian goat's milk.

Media	Ranges of counts for all samples (CFU ml^{-1})	Ranges of counts for all samples (CFU ml^{-1})
PCA (Total microflora)	7.14×10^5 - 4.2×10^7	5.9×10^6
MRS agar (42°C) (thermophilic *Lactobacilli* and *Streptococci*)	3.2×10^4 - 5.1×10^6	7.12×10^5
MRS agar (35°C) (mesophilic *Lactobacilli* and *Leuconostoc*)	7.63×10^5 - 4.13×10^7	5.92×10^6
Rogosa agar (*Lactobacilli*)	4.37×10^5 - 2.28×10^6	6.16×10^5
M17 agar (*Lactococci*)	2.76×10^5 - 3.58×10^7	6.65×10^6
SF (*Enterococcus*)	2.11×10^2 - 3.48×10^4	4.57×10^3

dy were to collect a variety of milk samples from different areas of Western Algeria and to determine the predominant microbial groups (isolation and identification of microorganisms from goat's milk).

MATERIALS AND METHODS

Samples

The samples of goat's milk were collected from various areas of Western Algeria. They were obtained under good conditions from a healthy animal, to avoid any contamination which can influence the lactic flora. The samples were collected in sterile bottles and then transported quickly to the laboratory to be analyzed.

Enumeration and isolation of lactic acid bacteria

Before isolation of the lactic acid bacteria, the microorganisms (total flora and lactic acid bacteria) were enumerated by the plate count technique: PCA medium (PCA; Biokar Diagnostics, Beauvais, France) was used for enumeration and identification of total flora. For a specific isolation, the cultures were cultivated in the following media:

MRS (De Man et al., 1960) and incubation at 30°C and 42°C during 48 h for the enumeration of *Lactobacillus*, *Pediococcus* and *Leuconostoc*.

M17 (Terzaghi and Sandine, 1975) (Biolife, Milano, Italy) and incubation at 30°C for 48 h to count Lactococci.

Rogosa (Difco, USA) (Rogosa et al., 1951) and incubation anaerobically at 35°C for 48 h. This medium is specific for *Lactobacillus*.

SF (Biolife) and incubation aerobically at 37°C for 48 h. This medium is specific for *Enterococcus*.

The strains were then purified by streaking in their suitable media. The purified strains were stored at -20°C in skimmed milk (12.5%, w/v) with 15% glycerol (Sigma, St. Louis, MO) for a long conservation.

Identification of lactic acid bacteria

LAB strains were initially selected on the basis of Gram staining, morphology, and catalase test following the criteria of Kandler and Weiss (1986) Falsen et al. (1999) and Klein (2001). Gas production from glucose was determined in MRS-broth supplemented with 1% glucose and Durban tubes. Arginine dihydrolase was determined in MRS broth according to criteria of Harrigan and McCance (1976). Citrate production was determined according the method of Kempler and Mc Kay (1980). Also, all strains were tested for growth at 10ºC for 10 days, 45ºC for 48 h. For cocci strains, growth on SF broth medium and in the presence of 6.5% NaCl was also considered. These preliminary tests make it possible to classify the isolates in genus on the basis of characteristic and tests of identification mentioned by Harrigan and Mc Cance (1976), Hammes et al. (1992), Holzapfel and Schillinger (1992) and Dicks et al. (1993).

Identification of lactic acid bacteria to the species level

Fermentation of carbohydrates was determined according to the method described by Schillinger and Lücke (1987) using the miniplate method described by Jayne-Williams (1975) with bromocresol purple as an indicator. Carbohydrates tested were cellobiose, galactose, mannitol, (melizitose, melibiose, ribose, trehalose, xylose, glucose, lactose, saccharose, fructose and arabinose and sterile water were used as positive and negative controls (Sigma, St. Louis, MO, USA) (Samelis et al., 1994).

Some representative isolates (about 25%) of MRS (35°C, 42°C), M17 (30°C, 42°C) and Rogosa (35°C) were selected for identifycation to species level by determination of the enzymatic and carbohydrate fermentation patterns of strains using API 20 Strep and API 50 CHL galleries (bio Mérieux, Marcy l'Etoile, France). Tests were performed according to the manufactures instructions. The APILAB PLUS database (bio Mérieux, Sa) was used to interpret the results.

Characterization by SDS–PAGE analysis of the whole-cell proteins

Twenty five strains previously identified from their phenotypic characteristics were submitted to SDS-PAGE of whole-cell proteins to confirm the API results. Preparation of cell-free extracts and polyacrylamide gel electrophoresis were done as described by Pot et al. (1994). Identification of selected strains was performed by comparison of their protein patterns with a database of normalized protein fingerprints derived from reference strains for almost all known species of LAB of the Laboratory of Microbial Biochemistry of the University of Alexandria obtained from different culture collections.

RESULTS

Enumeration of microorganisms

Table 1 summarizes the microbial count obtained from the various collected samples. The total microflora as well as the other specific groups was counted on five dif-

Table 2. Physiological and biochemical characteristics of isolated strains.

Group	A				B		C		D	E
Nb of isolates	**24**	**22**	**14**	**4**	**25**	**05**	**04**	**03**	**05**	**02**
CO_2 from:										
Glucose	–	–	–	–	–	–	+	+	–	–
Acetone	ND	ND	ND	–	+	+	–	–	+	–
Dextrane	ND	ND	ND	ND	–	–	+	–	+	+
Croissance à:										
10°C	+	+	+	+	+	+	+	–	–	+
15°C	+	+	+	+	+	+	+	–	–	+
30°C	+	+	+	+	+	+	+	+	+	+
45°C	+	+	+	+	–	–	–	–	+	–
Growth in:										
3 % NaCl	+	+	+	+	+	–	±	±	–	+
4 % NaCl	+	+	+	+	+	–	±	–	–	+
6.5 % NaCl	+	+	+	+	–	–	–	–	–	+
Hydrolysis of:										
Arginine	+	+	+	+	+	–	–	–	–	–
Esculine	+	+	+	+	+	v	±	–	–	+
Citrate	+	–	–	+	–	–	+	–	–	+
Acid production from:										
Glucose	ND	ND	ND	ND	+	+	+	+	+	+
Galactose	+	+	+	+	+	–	+	+	+	+
Fructose	ND	ND	ND	ND	+	–	+	+	+	+
Mannitol	v	+	–	+	v	–	v	–	–	–
Lactose	+	+	+	+	+	+	±	+	±	+
Tréhalose	+	+	+	+	+	–	+	–	–	+
Cellobiose	ND	ND	ND	ND	+	+	v	–	–	+
Xylose	–	–	–	–	v	–	v	–	–	+
Raffinose	–	–	–	–	–	–	–	–	v	–
Sorbitol	–	v	–	+	–	–	–	–	–	–
Arabinose	+	–	–	+	–	–	–	–	–	–
Melibiose	v	–	–	–	–	–	+	+	–	–
Sucrose	v	+	–	+	v	v	+	+	+	+

All strains were Gram-positives, catalase-negatives and non spores forming.
±: More than 50% of strains were positive.
v: Variable.
ND: Not determined.

ferent media (Table 1). The enumeration of the lactic flora on MRS and M17 media gives respective mean values of 5.92 x 10^6 and 6.65 x 10^6 cfu.ml^{-1}, respectively; these values are more important than the mean of the total plate count (5.19 x 10^6 cfu.ml^{-1}), indicating the predominance of the lactic flora. These values also exceed those obtained with Rogosa medium (6.16 x 10^5 cfu.ml^{-1}). The mean of the thermophilic bacteria incubated at 42°C on MRS agar, 7.12 x 10^5 cfu.ml^{-1}, is also less than that of the mesophilic lactic flora (35°C) which is of 5.92 x 10^6 cfu.ml^{-1}. In addition, mean of enterococci (4.57 x 10^3 cfu.ml^{-1}) appeared lower than those obtained on the MRS agar (35 and 42°C), Rogosa agar, PCA the M17 agar.

Identification of the lactic bacteria

A total of 153 isolates of lactic bacteria was obtained. These isolates were identified to genus level based on their cellular morphology, gas production, growth at 10 and 45°C, in the presence of 6.5% NaCl and pH 9.6 according to Wood and Holzapfel (1995). All the isolates are Gram positive, catalase negative and non spores forming (Table 2).

The isolates were identified to species level according to the methods described by Schillinger and Lücke (1987) and Samelis et al. (1994). The first group (A) represented by the genus *Enterococcus* contains 68 species of

which 34 were identified using the API 20 Strep. This method confirmed that 80% of these isolates belonged to the genus and species identified by the physiological and biochemical tests. They were assigned to *Enterococcus faecium* with 24 isolates (15.68%), *Enterococcus faecalis* with 22 isolates (14.37%), *Enterococcus durans* with 14 isolates (09.15%) and *Enterococcus avium* with 04 isolates (02.61%). All these species were homofermentative, grew in SF medium, at 10°C and 45°C and in the presence of 6.5% NaCl, and are able to metabolize the majority of sugars. The other species were identified by API 50 CHL as well as SDS-PAGE technique.

The strains assigned to group B belonging to *Lactococcus* genus were identified as *Lactococcus lactis* subsp. *lactis* with 25 isolates (16.33%) and *Lc. lactis* subsp. *cremoris* comprising 05 isolates (03.26%). The species of this group are characterized by their incapacity to produce CO_2, to grow in SF medium, in the presence of 6.5% NaCl, at 45°C, but are able to grow at 10°C. With the exception of two isolates, all the strains belonging to this genus ferment glucose, galactose and cellobiose, but are unable to metabolize raffinose, sorbitol, arabinose and melibiose.

The strains belonging to group C were identified as *Leuconostoc* and are represented by 7 isolates (4.5%); they have oval cocci form, grow in pairs, produce gas from glucose and did not hydrolyze arginine. This group is subdivided in two sub-groups. The first sub-group contains 4 strains (2.6%) identified as *Leuconostoc mesenteroides* subsp. *mesenteroides*. Most of these strains produce dextrane from sucrose and ferment galactose, melibiose and trehalose. The isolates belonging to the second sub-group were affiliated to *Leuconostoc lactis* species including 3 strains (2%) and were identified thus for their incapacity to ferment mannitol, trehalose, cellobiose and to form dextrane.

The group D is represented by *Streptococcus thermophilus* comprising 5 isolates (3.26%). This species is characterized mainly by their thermophilic growth with an optimum around 42 to 43°C, their thermoresistance at 60°C for 30 min (Garvie, 1984), a fermentative activity generally reduced to some sugars and a strong sensitivity to NaCl. The group E includes two isolates belonging to *Pediococcus* genus. They were identified as *Pediococcus pentosaceus*. They are homofermentative, often associated in pairs and tetrad.

The strains identified as *Lactobacillus* are classified in three sub-groups (I, II and III) (Table 3) (Kandler and Weiss, 1986). The results of identification indicate that *Lactobacillus delbrueckii* subsp. *bulgaricus* is the dominant species in sub-group I represented by 7 species (4.5%), followed by *Lb. delbrueckii* subsp. lactis 3 (1.9%), *Lactobacillus helveticus* 2 (1.3%) and

Lactobacillus salivarius 1 (0.6%). These species are characterized by the transformation of hexoses exclusively by the homofermentative way, their important capacity of acidifying and a high optimal temperature of growth (42°C). All the strains belonging to *Lactobacillus bulgaricus* species are able to ferment ribose, galactose, fructose, maltose, cellubiose, raffinose, melibiose and trehalose. The lactobacilli belonging to group II are represented in majority by *Lactobacillus rhamnosus* with 9 strains (5.8%) that metabolize rhamnose, *Lactobacillus paracasei* subsp. *paracasei* with 4 isolates (2.6%), *Lactobacillus pentosus* containing 3 isolates (1.9%) and *Lactobacillus plantarum* with only one (0.65%) strain. These bacteria degrade hexoses in lactic acid by homofermentation and pentoses by heterofermentation. Their optimal growth temperature of growth ranges from 30 to 37°C. *Lactobacillus fermentum* represent the highest number of isolates 11 (7.18%) of sub-group III, followed by *Lactobacillus brevis* 4 isolates (2.6%). The bacteria belonging to this group ferment pentoses and hexoses by heterofermentation. Their temperature of growth differs according to species; it ranges from 30 to 45°C.

The identification based on biochemical tests or even by the API system leads sometimes to false results, or sometimes does not allow for identification of the strain. The use of SDS-PAGE makes it possible to determine the electrophoretic profile of the strains. This technique made it possible to confirm 75% of the results obtained. An example of electrophoretic profiles of some strains identified by SDS-PAGE technique in this work is illustrated in Figure 1.

DISCUSSION

In this study, our results indicate the predominance of the lactic acid bacteria compared to the total microflora. The results are in agreement with those of other workers, undertaken on the enumeration and isolation of the lactic acid bacteria from fermented milks. According to Beukes et al. (2001) and Savadogo et al. (2004), the number of lactic bacteria largely exceeds that of the other microflora of traditional fermented milk in South Africa and in Burkina Faso, respectively.

The high rate of the lactic acid bacteria can be explained by the selectivity of media used MRS, M17 and Rogosa for this type of bacteria (Reuter, 1985). Thus, the results revealed the presence of diversity in the lactic microflora isolated from goat's milk. This can be related to several factors. First of all, these species are frequently isolated from the animals such as bovines, sheep and caprines. The environment and the climate can play a very great role as indicated by Jenness (1980), Picque et al. (1992) and Remeuf (1992). Indeed, the samples were collected from different coastal like Oran and Mostaganem, arid or semi-arid areas like Chlef and Mascara knowing that the climate is different in these areas. In addition, the goats from which milk were collected belong to different races (Makatia, Makatia-

Table 3. Physiological and biochemical characteristics of isolated strains (*Lactobacilli*).

Group	(G.I)				(G.II)				(G.III)	
Nb of isolates	7	3	2	1	9	4	2	2	11	4
CO_2 from:										
Glucose	–	–	–	–	–	+	–	+	+	+
Acetone	–	–	–	ND	+	–	+	+	ND	ND
Dextrane	–	–	–	ND	–	–	–	–	ND	ND
Growth at:										
10°C	–	–	–	–	–	–	+	–	–	–
15°C	–	–	–	–	+	+	+	–	–	+
30°C	+	+	+	+	+	+	+	+	+	+
45°C	+	+	–	+	±	–	–	+	+	–
Hydrolysis of:										
Arginine	–	–	–	–	–	+	–	–	+	+
Esculine	–	–	–	ND	+	–	+	ND	ND	ND
Citrate	–	–	–	ND	+	–	+	ND	ND	ND
Acid production from:										
Glucose	+	+	+	+	–	+	+	+	+	+
Galactose	–	v	+	+	+	+	+	+	–	v
Fructose	+	+	v	+	+	+	+	+	+	+
Mannose	–	–	–	+	+	+	+	+	±	–
Mannitol	–	+	v	–	+	+	+	+	–	–
Lactose	+	+	+	+	+	+	+	+	–	–
Trehalose	–	+	v	+	+	+	+	+	–	–
Cellobiose	+	±	–	–	+	+	+	+	v	–
Xylose	–	–	–	–	+	–	–	–	–	+
Raffinose	–	–	–	v	–	+	+	+	+	v
Sorbitol	–	–	–	–	+	+	+	+	–	–
Arabinose	–	–	–	–	–	–	v	+	v	+
Melibiose	–	–	–	+	+	+	+	+	+	+
Sucrose	–	+	–	+	–	+	+	+	+	+

All strains were Gram-positives, catalase-negatives and non spores forming.
±: More than 50% of strains were positive.
v: Variable.
ND: Not determined.

Chamia, Arabia and Kabyle), and the difference in the variety can have a great impact on the concentration of the various components of the goat's milk (Remeuf et al., 1991).

Among the identified lactic microflora, *Enterococcus* appears dominant in the goat's milk (39.21%). 18.30% of cultures of this genus belong to the species *E. faecium* followed by *E. faecalis* which is represented by 17%. Enterococci were also isolated by other authors from various types of milk. Prodromou et al. (2001) showed that 60% of the strains isolated from “Orinotyri” a cheese manufactured from ewe's milk in Greece were enterococci. Enterococci grew under the hostile conditions, 6.5% NaCl and different temperatures (10 and 45°C), high pH (9) what would probably explain high amount in the raw goat's milk. Entrococci play an important part in cheese ripening, and more particularly those manufactured from ewe's or goat's milk. These cheeses present characteristic sensorial properties (Casalta et al., 1995). For example, *E. faecalis* takes part in the cheese ripening and it is sometimes regarded as a flavour starter contributing to a flavor pronounced in ripe cheese. Some *E. faecalis* strains are used as starter culture for the development of cheddar, mozzarella and labneh (Egyptian yoghourt) (Devriese et al., 1995).

Concerning Lactococci, more *Lc. lactis* subsp. *lactis* (16.33%) were obtained than *Lc. lactis* subsp *cremoris* (3.26%) in our samples. In other works, it was found that *Lc. lactis* subsp. *lactis* is more frequently isolated from raw milk samples (Moreno and Busani, 1990), of cheeses

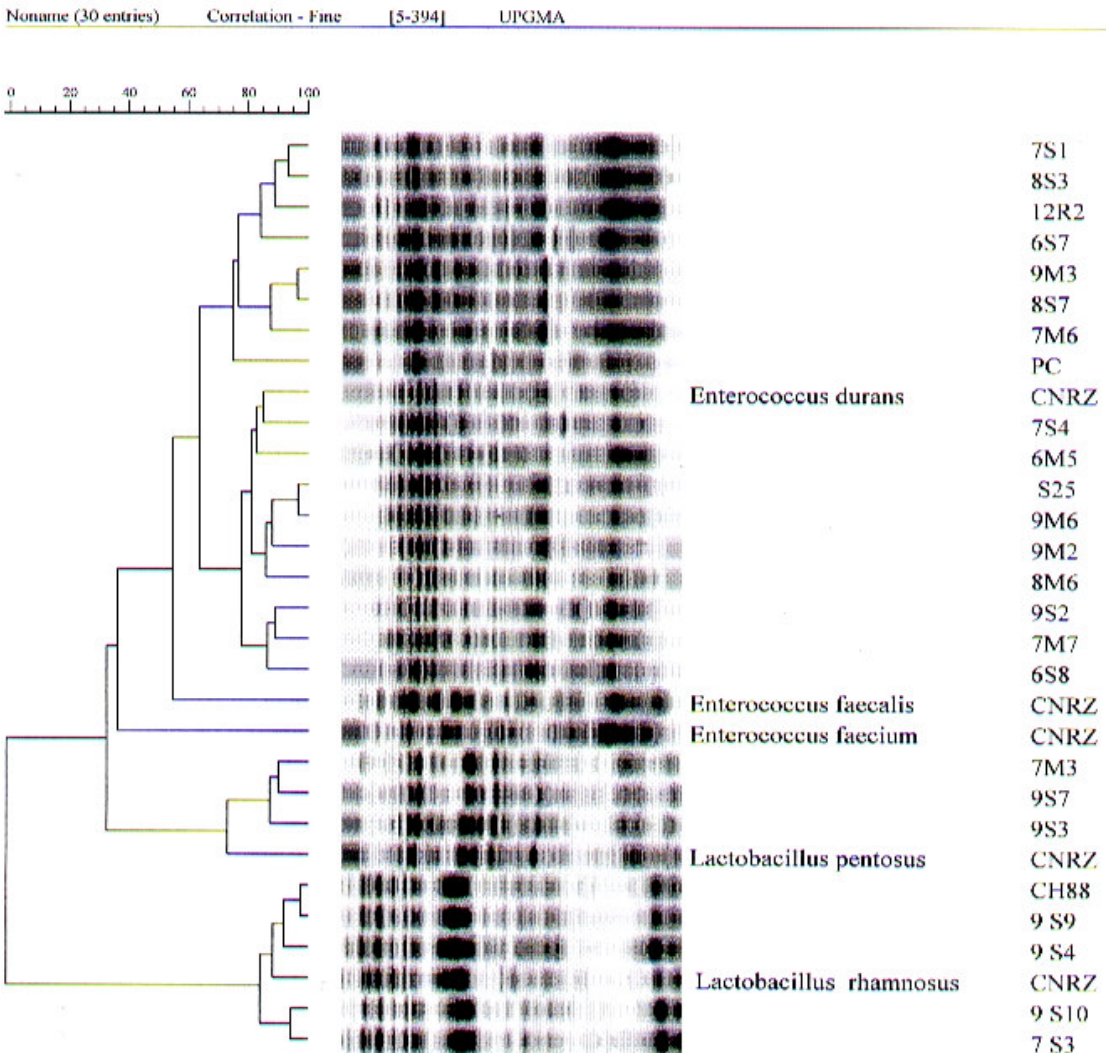

Figure 1. Dendogram calculated by the unweight average obtained using SDS-PAGE protein patterns of strains of the unknown isolated cultures, compared to a number of reference representative strains.

manufactured containing milk (Centeno et al., 1996) of Raib (Hamama, 1992) and from Dahi and butter samples in India (Padmanabha-Reddy et al., 1994). In addition, according to Holler and Steele (1995), *Lactococcus lactis* subsp *cremoris*, was isolated only from natural sources. In other work, Crow (1993) and Weerkamp et al. (1996) affirmed that the lactococci isolated from natural sources were often identified as *Lc. lactis* subsp. *lactis*, whereas the phenotype *Lc. lactis* subsp *cremoris*, which is common in industrial mixed strain starter cultures, was rarely isolated. The natural habitat of *Lc. lactis* subsp *cremoris* remains uncertain (Salama et al., 1995). It was also found that from 21 isolates identified from Amazi, fermented milk in Zimbabwe (Mutukumira, 1996) five strains belong to *Lc. lactis* subsp. *lactis* and four were *La. lactis* subsp. *lactis* biovar *diacetylactis*. Strains belonging to the phenotype *Lc. lactis* subsp. *lactis* biovar *diacetylactis* were not isolated in our work; this can be probably explained by the fact that the media used for the identification were not selective enough and the identification of a great number of strains in our study (50%) were based only on the phenotypical and physiological tests.

Also, the Leuconostoc form part of our collection, but with a small proportion (4.5%); 4 strains belonging to *L. mesenteroides* subsp. *mesenteroides* and 3 to *Lc. lactis*. The same observation was mentioned by Togo et al. (2002) who isolated a reduced number of leuconostocs from wine. The small proportion of these species among our isolates can be due probably to their incapacity for competition with the other lactic acid bacteria in mixed cultures (Teuber and Geis, 1981). *Leuconostoc* are usually found on plants as well as dairy products, wine, and liquids containing sugar. It is thus not surprising to

find *Leuconostoc* in the oral cavity and the digestive tract of the man or animals (Cai et al., 1998). *L. mesenteroides* subsp. *cremoris* was not isolated in this study. This could be also explained by the method of identification based on the morphological and physiological characteristics for a great number of samples, and probably by the fact that the characteristics of the citrate metabolism is encoded by plasmid DNA, which can be lost in some strains (Cogan, 1985). Studies on 182 representative strains of lactic acid bacteria isolated from raw milk in Brazil showed that *L. mesenteroides* subsp. *cremoris* is represented by only 1.1% of the total population (Holzapfel and Shillinger, 1992). It is also possible that the media used were not completely selective.

Five isolates (3.26%) were identified as *S. thermophilus*. They are generally isolated from fermented dairy products. The pediococci strains represent a very small proportion (1.3%) in our collection and identified as *P. pentosaceus*. This genus is rarely isolated from milk and from dairy products; it is often isolated from wine, crop products brines (Garvie, 1986).

Lactobacilli isolates represent a significant part among our isolates and are represented by *L. delbuuecki* subsp *bulgaricus*, *L. rhamnosus* and *L. fermentum*. These species are frequently isolated from raw milk and dairy products (Tsakalidou et al., 1994). In other works, Mathara et al. (2004) and Abdelgadir et al. (2001) isolated *Lactobacillus plantarum*, *Lactobacillus acidophilus* and *Lb. rhamnosus*, and *Lb. fermentum* from fermented products and showed that these species represent more than 60% of the isolated lactobacilli. In addition, Medina et al. (2001) showed that 8% of *Lactobacillus* isolated from ewe's milk and cheese in North from Argentina belonged to *Lb. acidophilus*.

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REFERENCES

Abdelgadir WS, Hamad SH, Möller PL, Jakobsen M (2001). Characteristics of the dominant microbiota of Sudanese fermented milk Rob. Int. Dairy J. 11: 63-70.

Axelsson L (1998). Lactic acid bacteria: classification and physiology. In: Salminen, S. & von Wright A (eds). Lactic Acid Bacteria: Microbiology and Functional Aspects 2nd Edition. New York: Marcel Dekker Inc 1-72.

Beukes EM, Bester BH, Mostert JF (2001). The microbiology of South African traditional fermented milks. Int. J. Food. Microbiol. 63: 189-197.

Boubekri K, Otha Y (1996). Identification of lactic acid bacteria from Algerian Traditional Cheese El Klila J. Sci. Food. Agric. 70: 501-505.

Cai Y, Benno Y, Takeda A, Yoshida T, Itaya T, Nakasa T (1998). Characterization of Leuconostoc species isolated from vacuum-packaged ham. J. Gen. Appl. Microbiol. 44: 153-159.

Carr FJ, Hill D, Maida N (2002). The lactic acid bacteria: A literature survey. Crit. Rev. Microbiol. 28: 281-370.

Casalta E, Zennaro R, Donini J, Maroselli MX (1995). Influence des ferments locaux sur les caractéristiques biochimiques et sensorielles du fromage de Venaco. Rapport de recherche dans le cadre du programme Interreg I, p. 10.

Centeno JA, Cepada AA, Rodriguez-Otero JL (1996). Lactic acid bacteria isolated from Arzua cow's milk. Int. Dairy J. 6: 665-678.

Cheriguene A, Chougrani F, Bensoltane A (2006). Identification and characterization of lactic acid bacteria isolated from Algerian goat's milk. Pak. J. Biol. Sci. 9(7): 1242-1249.

Cogan TM (1985). The Leuconostoc: milk products. In: Gilliland SE, (Ed.). Bacterial Starter Cultures For Foods. CRC Boca Raton FL, pp. 25-40.

Cogan TM (1980). Les levains lactiques thermophiles. Lait. 60: 397-425.

Crow VC, Coolbear T, Holland R, Pritchard GG, Martley, FG (1993). Starters as finishers: starter properties relevant to cheese ripening. Int. Dairy J. 3: 423-460.

Daly C, Davis R (1998). The biotechnology of lactic acid bacteria with emphasis on applications in food safety and human health. Agri. and food Sci. in Finland, 7(2): 251-264.

De Man J, Rogosa M, Sharpe E (1960). A medium for the cultivation of lactobacilli. J. Appl. Bacteriol. 23: 130-135.

Devriese LA, Pot B, Van Damme L, Kersters K, Haesebrouck F (1995). Identification of Enterococcus species isolated from foods of animal origin. Int. J. Food Microbiol. 26: 187-197.

Dicks LMT, Fantuzzi L, Gonzales FC, Du Toit M, Dellaglio M (1993). Leuconostoc argentinum sp.nov isolated from argentine raw milk. Int. J. Syst. Bacteriol. 43: 347-351.

Falsen E, Pascual C, Sjoden B (1999). Phenotypic and phylogenic characterization of a novel Lactobacillus species from human source: description of Lactobacillus in. sp.nov. Int. J. Syst. Bacteriol. 49: 217-221.

Garvie EI (1984). Separation of species of the genus Leuconostoc and differentiation of the Leuconostocs from other lactic acid bacteria. In: Bergan T. (Ed.). Methods in Microbiology. Academic press London, pp. 147-178.

Garvie EI (1986). Genus Pediococcus Claussen 1903, 68AL. In Bergey's Manuel of systematic bacteriology, 2:1075-1079.Williams, Wilkins, Baltimore.

Gobbetti M, Corsetti A (1997). Lactobacillus sanfrancisco a key sourdough lactic acid bacterium: a review. Food Microbiol. 14: 175-187.

Hamama A (1992). Morrocan traditional fermented dairy products. In: Ruskin FR (Ed) Applications of Biotechnology to Traditional Fermented Foods. National Academy Press Washington DC, pp. 75-79

Hammes WP, Weiss N, Holzapfel WH (1992). The genera Lactobacillus and Carnobacterium. In: Barlows AHG, Trüper M, Dworkin W, Harder KH, Schleifer (Ed.). The prokaryotes. Springer Berlin p: 1534-1593.

Harrigan WF, McCance ME (1976). Laboratory Methods in Food and Dairy Microbiology. Academic Press London.

Holler BJ, Steele JL (1995). Characterization of lactococci other than Lactococcus lactis for possible as starter cultures. Int. Dairy J. 5: 275-289.

Holzapfel WH, Schillinger V (1992). The genus Leuconostoc. In: Barlows A, Trüper HG, Dworkin M, Harder W, Schleifer KH (Eds.). The prokaryotes. Springer Berlin pp. 1509-1534.

Jay JM (2000). Fermentation and fermented dairy products pp. 113-130. In Modern Food Microbiology 6th edition. An Aspen Publication Aspen Publishers Inc. Gaithersburg USA.

Jayne-Williams DJ (1975). Miniaturized methods for the characterization of bacterial isolates. J. App. Bacteriol. 38: 305-309.

Jenness R (1980). Composition and characteristics of goat milk: J. Dairy Sci. 63: 1605-1630.

Kandler O, Weiss N (1986). Regular non-sporning Gram-positive rods. Genus Lactobacillus (Beijerink 1901.22). In Bergey's Manual of Systematic Bacteriology Vol 2, ed. Sneath. PHA, Mair NS, Sharpe ME and Holt JG, pp. 1208-1234.

Kempler GM, McKay LL (1980). Improved medium for detection of citrate-fermenting Streptococcus lactis subsp. diacetylactis. J. Appl. Environ. Microbiol. 39: 926-927.

Klein G (2001). International Committee of Systematic Bacteriology.

Subcommittee on the Taxonomy of Bifidobacterium, Lactobacillus and Related Organisms. Minutes of the Meeting. Int. J. Syst. Evol. Microbiol. 51: 259-261.
Liu SQ (2003). Review article: Practical implications of lactate and pyruvate metabolism by lactic acid bacteria in food and beverage fermentations. Int. J. Food Microbiol. 83: 115-131.
Mathara JM, Schillinger U, Kutima PM, Mbugua SK, Holzapfel WH (2004). Isolation identification and characterisation of the dominant microorganisms of kule naoto: the Maasai traditional fermented milk in Kenya. Int. J. Food Microbiol. 94: 269-278.
Medina R, Katz M, Gonzalez S, Oliver G (2001). Characterization of the lactic acid bacteria in ewe's milk and cheese from Northwest Argentina. J. Food. Prot. 64(4): 559-563.
Moreno I, Busani SFB (1990). Characterization of lactococci isolated from raw milk and commercial lactic starters. Coletanea do Instituto de Technologia de Alimentos. 20: 44-50.
Mutukumira AN (1996). Investigations of some prospects for the development of starter cultures for industrial production of traditional fermented milk in Zimbabwe. Doctor Scientiarum Thesis, Department of Food Science, Agricultural University of Norway.
O'Sullivan L, Ross RP, Hill C (2002). Potential of bacteriocin-producing lactic acid bacteria for improvements in food safety and quality. Rev. Biol. 84: 593-604.
Padmanabha-Reddy V, Habibulla-Khad MM, Purushothaman V (1994). Plasmid linked starter characteristics in lactococci isolated from dahi and buttermilk. Cult. Dairy Prod. J. 29: 25-30.
Pandya AJ, Ghodke KM (2007). Goat's and sheep milk products other than cheeses and yoghurt. Small Rumin. Res. 68: 193-206.
Park YW, Ju´arez M, Ramos M, Haenlein GFW (2007). Physico-chemical characteristics of goat and sheep milk. Small Rumin. Res. 68: 88-113.
Picque D, Perret B, Latrille E, Corrieu G (1992). Caractérisation et classification des bactéries lactiques à partir de la mesure de leur cinétique d'acidification. Lebensm. Wiss. U. Technol. 25: 181-186.
Pot R, Vandamme P, Kersters A (1994). Analysis of electrophoretic whole organism protein fingerprints. pp: 493-521, IN: M. Goodfellow and A.G. O'Dunnel (Eds.) Chemical Methods in Procaryotic Systematic. J. Wiley and Sons, Inc. Chichester, England.
Prodromou K, Thasitou P, Haritonidou P, Tzanetakis N, Litopoulou-Tzanetaki E (2001). Microbiology of "Orinotyri" a ewe's milk cheese from the Greek mountains. Food Microbiol. 18: 319-328.
Remeuf F **(**1992). Physico-chemical properties of goat milk in relation to processing characteristics. In: Proceedings of the National Symposium on Dairy Goat Production and Marketing Oklahoma City OK pp. 98-110.
Remeuf F, Cossin V, Dervin C, Lenoir J, Tomassone R (1991). Relations entre les caractéristiques physico-chimiques des laits et leur aptitude fromagère. Le Lait. 71: 397-421.
Reuter G (1985). Elective and selective media for lactic acid bacteria. Int. J. Food. Microbiol. 2: 55-68.
Rogosa M, Mitchell JA, Wiseman RF (1951). A selective medium for the isolation and enumeration of oral and fecal Lactobacilli. J. Bacteriol. 62: 132-133.
Salama SS, Musafija-Jeknic T, Sandine WE, Giovannoni SJ (1995). An ecological study of lactic acid bacteria: isolation of new strains of Lactococcus including Lactococcus lactis subspecies cremoris. J. Dairy Sci. 78: 1004-1017.
Samelis J, Maurogenakis F, Metaxopoulos J (1994). Characterization of lactic acid bacteria isolated from naturally fermented Greek dry salami. Int. J. Food. Microbiol. 23: 179-196.
Sandine WE, Radich PC, Elliker PR (1972). Ecology of the lactic streptococci. A review. J. Milk Food. Technol. 35: 176-185.
Savadogo A, Ouattara CAT, Savadogo AW, Ouattara AS, Barro N, Traore AS (2004). Microorganisms Involved in Fulani Traditional Fermented Milk in Burkina Faso. Pak. J. Nutr. 3(2): 134-139.
Schillinger U, Lücke FK (1987) Identification of lactobacilli from meat and meat product. Food. Microbiol. 4: 199-208.
Terzaghi BE, Sandine WE (1975). Improved medium for lactic streptococci and their bacteriophages Appl. Environ. Microbiol. 29: 807-813.
Teuber M, Geis A (1981). The family Streptococcaceae (non-medical aspects). In: Starr MM, Stolp H, Truper HG, Balows A, Schlegel HG, (Eds.) The Prokaryotes: A Handbook on Habitats Isolation and Identification of Bacteria vol. 2. Springer-Verlag Berlin pp. 1614-1630.
Togo CA, Sara B, Feresu SB, Mutukumira AN (2002). Identification of Lactic Acid Bacteria isolated from Opaque beer (Chibuku) for potential use as a starter culture. J. Food Technol Africa. 7(3): 93-97
Tsakalidou E, Manolopoulou E, Kabaraki E, Pot B, Karel K, Kalantzopoulos G (1994). The combined of whole-cell protein extracts for the identification (SDS-PAGE) and enzyme activity screening of lactic acid bacteria isolated from traditional dairy products. Syst. Appl. Microbiol. 17: 444-449.
Weerkamp AH, Klijn N, Neeter R, Smit G (1996). Properties of mesophilic lactic acid bacteria from raw milk and naturally fermented raw products. Neth. Milk Dairy J. 50: 319-322.
Wood BJB, Holzapfel WH (1995). The genera of lactic acid bacteria, Vol. 2 Glasg w: Blackie Academic and Professional.