



MOLECULAR CHARACTERIZATION OF THE BACTERIAL MICROBIOTA IN COTIJA CHEESE PRODUCED IN DIFFERENT REGIONS OF MEXICO

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Key words: Cotija cheese, PCR-DGGE, bacterial community

Introduction. Cotija cheese® is a Mexican artisanal dairy product made in a region between Michoacan and Jalisco (Jalmich). Although there is a Mexican Official Norm (1) that establishes its chemical characteristics and manufacturing process, there are several products in the market that are named the same way, but that do not comply with it.

In this work we compared, in terms of chemical properties and bacterial microbiota, by means of culture-independent methods, cheeses produced in Jalmich with the ones produced in Tonalá, Chiapas.

Methods. 12 cheese samples from Jalmich and 6 from Tonalá, Chiapas, were analyzed. Physicochemical analysis included: pH, acidity, [Cl⁻] and water activity (a_w). Proximate analysis included moisture, protein, fat, ash and carbohydrate content. For the study of the bacterial microbiota, PCR and denaturing gradient gel electrophoresis (PCR-DGGE) were used. The V3 16S rDNA (2) and the gene *rpoB* (3) were the targets for amplification. The dominant bands of each sample were excised and sequenced. The sequences were compared by BLAST and the identity was confirmed by the Neighbor-Joining model with bootstrap of 1,000 replicas. Samples were grouped by UPGMA analyses.

Results. As for the proximate and physicochemical analyses, there was statistical difference between samples from the two regions in all parameters, except a_w (Table 1).

Table 1. Average values of proximate and physicochemical analyses of the samples from the 2 regions

	Jalmich	Chiapas
pH	5.32 ± 0.11 a	5.62 ± 0.29 b
Acidity (%)	0.38 ± 0.06 a	0.29 ± 0.07 b
Cl ⁻ (%)	3.52 ± 0.49 a	5.56 ± 1.13 b
a _w	0.874 ± 0.02 a	0.865 ± 0.01 a
Moisture (%)	33.20 ± 3.97 a	40.85 ± 5.28 b
Protein (%)	23.76 ± 1.94 a	19.57 ± 1.43 b
Lipid (%)	31.76 ± 2.36 a	26.02 ± 7.61 b
Ash (%)	6.37 ± 0.5 a	8.26 ± 0.97 b
Carbohydrate (%)	0.16 ± 0.06 a	0.76 ± 0.43 b

Different letters mean statistical difference between values in a row, with α = 0.05.

Using PCR-DGGE, the band pattern of the bacterial microbiota for each cheese from the 2 regions was obtained (Fig.1). These were different for each one, regardless the target gene employed, though more diversity was found using *rpoB*.

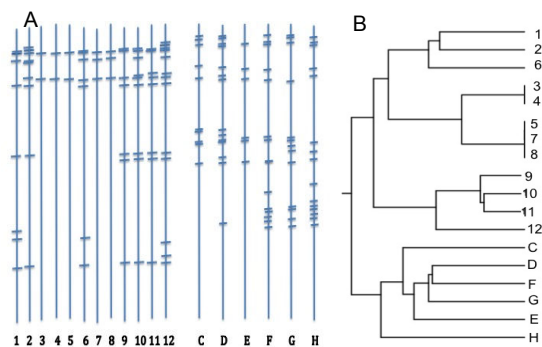


Fig. 1. A. Digital patterns of the amplicons using *rpoB* as target gene. B. Dendrogram of grouped samples constructed from digital patterns. Samples, numbers represent samples from Jalmich, letters from Chiapas.

Using 16S rDNA as target gene, we found the following bacteria as dominant in both regions: *Bacillus sp.*, *Virgibacillus pantothenicus*, *Bacillus vietnamensis* and *Vagococcus sp.* When the *rpoB* gene was the amplification target, we found *Enterococcus faecium* and *Streptococcus infantarius* in both the regions, *Staphylococcus equorum* and *Streptococcus parasanguinis* were exclusive of the cheeses produced in Jalmich, while *Streptococcus suis* was only present in the cheeses manufactured in Chiapas.

Conclusions. Cotija cheeses produced in Jalmich and Chiapas are different from each other, as for their proximate composition, physicochemical properties and bacterial population. These differences are caused by their raw materials used and, mainly, by their different ripening processes.

Acknowledgements. PAPIIT IN230511.

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