Title:

Distribution of antimicrobial resistant lactic acid bacteria along the production of a raw ewe milk-derived cheese.

Authors:

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Abstract:

Antimicrobial resistance (AMR) of microorganisms is defined as the ability to withstand or resist the action of one or more antimicrobial agents. AMR is widespread and the efficacy in treating certain life-threatening infections is already compromised. Thus, fermented products are considered notable reservoirs of antimicrobial resistant bacteria. Therefore, this work aimed to analyse the prevalence of antimicrobial-resistant lactic acid bacteria (LAB) throughout the production of a raw milk cheese (Idiazabal PDO).

Four artisanal dairies were selected and raw milk, whey, fresh and ripened cheese samples were collected. LAB isolates were obtained in MRS agar and identified by sequencing of the V1–V3 regions of the 16S rRNA. The minimum inhibitory concentration was evaluated by broth microdilution for the most commonly used antibiotics (amoxicillin, dihydrostreptomycin benzylpenicillin and polymyxin B). The resistance was interpreted according to the microbiological cut-off values proposed by the European Food Safety Authority and the European Committee on Antimicrobial Susceptibility Testing.

More than 170 LAB isolates were obtained throughout the cheese-making process. *Enterococcus faecalis* (36.4%) and *Bacillus thuringiensis* (13.6%) predominated in raw milk, while *Bacillus cereus* (15.8%) and *Bacillus* sp. (13.2%) did it in whey and *Bacillus* sp. (22.0%), *Enterococcus faecalis* (14.6%) or *Enterococcus hirae* (14.6%) in fresh cheeses. In ripened cheeses, instead, *Lactobacillus* sp. (20.8%) and *Lacticaseibacillus paracasei* predominated (16.7%). *Bacillus* and *Enterococcus* species showed intermediate-high resistance to all antimicrobials (average 64.0% and 75.0%, respectively), while *Lactobacillus* and *Lacticaseibacillus* were more sensible (34.0% and 36.0%).

In conclusion, these results indicate that a shaping effect occurs during the cheese-making process of a raw ewe milk-derived cheese, contributing to the minimization of AMRs.

Keywords:

lactic acid bacteria; antimicrobial resistance; antibiotic resistance; raw milk; whey; raw milk cheese; cheese-making; ripening