

TITLE: The bacteriocinogenic potential of bacteria isolated from Canastra cheese endogenous starter culture

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ABSTRACT:

Canastra artisanal cheeses are one of the typical dairy products of Minas Gerais and their manufacture is characterized by the use of natural starter cultures. Canastra cheese is produced using the backslopping method, where fermented whey from the previous production batch is used on the following cheese production cycle. Endogenous starter cultures are composed of lactic acid bacteria (LAB), as well as propionibacteria, coryneforms and yeasts. Acids produced by LAB play an essential role during cheesemaking, creating favorable conditions for curdling, as well as specific metabolism that can influence the cheese’s sensorial properties. Moreover, some of the LAB can produce antimicrobial molecules, such as bacteriocins. Here we investigate the presence of bacteriocins genes in 11 bacterial strains isolated from Canastra cheese endogenous starter cultures, identified through whole genome sequencing. The strains were obtained from the endogenous starter from 10 distinct Canastra cheese producers, using standard decimal dilutions and strain purification, and a diverse set of media and growth conditions to maximize the diversity of isolates. Sequences were obtained using the Illumina platform, genomes assemblies were obtained using SPAdes v3.14.1, and bacteriocin gene prediction/annotation using BAGEL4. The bacteriocinogenic strains were identified as *Enterococcus faecalis* (n=5), *Lactococcus lactis* (n=3), *Leuconostoc* sp. (n=2) and *Streptococcus* sp. (n=1). Almost all strains were shown to carry bacteriocins genes, with exception of one strain identified as *Enterococcus faecalis*. The most representative identified bacteriocins were enterocin, bicereucin and propeptin, but also were identified BlpD, enterolysin, lactococcin, lasso-peptide, listeriolysin, pneumolancidin, sactipeptides, streptide, subtilosin and thiopeptide. *In silico* analysis showed that the operon structure of most genes was complete, although it is still necessary to confirm their expression and activity *in vitro*. Based on this data we hypothesise that bacteriocins might play a major role in controlling the growth of susceptible bacteria during the production of Canastra cheese.

Keywords: bacteriocin, starter culture, “pingo”, artisanal cheese.

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