

EFFECTS OF CITRUS ESSENTIAL OILS ON THE MICROBIOLOGICAL SAFETY OF PRIMO SALE CHEESE



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Introduction

In the last years, the increasing demand for foods with no chemical preservatives added determined an interest by the food industries to use natural substances to be used as biopreservatives (Gaglio et al., 2017). Several essential oils from various species have been characterized for their antibacterial activities in order to select new biopreservatives (Oussalah et al., 2007).

Objectives

The aim of this work was to evaluate the organoleptic effect of citrus essential oils (EOs) to be applied in the processing of sheeps' milk "Primo Sale" cheese and their inhibitory effect towards the main bacterial pathogens of dairy interest. In order to better evaluate the antibacterial effect, the first approach was based on pasteurized milk, in view of the future application in raw milk cheese production.

Materials and methods

Three industrial citrus essential oils (EOs) extracted by cold pressing of the citrus peels were tested: Orange [*Citrus sinensis* (L.) Osbeck] Lemon [*Citrus limon* (L.) Osbeck] and Tangerine [*Citrus reticulata* Blanco]. Cheese making trials were carried out in a dairy pilot plant (Biopek, Gibellina, Italy) following the protocol of "pressed" cheeses (Fig. 1).



Fig. 1. Pressed cheese.

Sheep bulk milk was pasteurized for 30 min at 60°C (Fig. 2A). Each EO was tested at two different concentrations 100 and 200 µl/l of milk (Fig. 2B). For each dose, two cheese makings were made: one production was not added with pathogenic bacteria to evaluate the organoleptic impact of EOs on cheese, while the second production was added with 30 CFU/mL of *Listeria monocytogenes* 24B0 and *Salmonella typhimurium* 50431 and 10³ CFU/mL of *Escherichia coli* C45/1245 and *Staphylococcus aureus* PSS52. All cheeses were obtained by coagulation into plastic vats with 0.3 mL/L of microbial rennet (Fromase® 220 TL, DSM Bright Science Brighter Living, Heerlen, UK) (Fig. 2C) and acidification by means of the starter culture *Lactococcus lactis* CAG4 and CAG37 10 mL/L to reach a final concentration of 10⁶ CFU/ml in milk. The cheeses were salted in brine containing 20% NaCl (w/v) for 12 h (Fig. 2D) and air dried for 24 h (Fig. 2E). Four cheese makings without EOs were used as control trials and prepared as follows: only rennet; rennet plus pathogenic bacteria; rennet plus starters; and rennet plus pathogenic and starters.

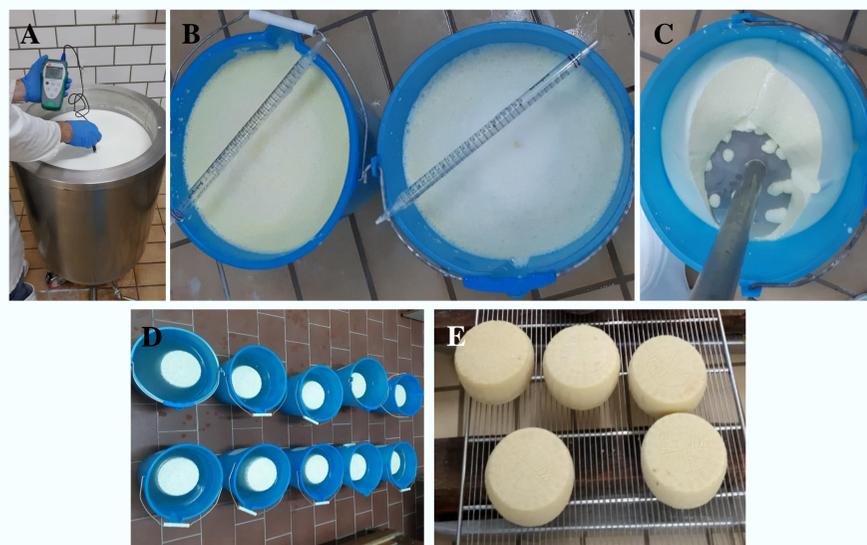


Fig. 1. Steps of cheeses production.

Cheese samples were homogenised with a stomacher (BagMixer® 400, Interscience, Saint Nom, France) in order to perform the decimal serial dilution. Microbial suspensions were plated and incubated as follows: coccus-shaped lactic acid bacteria on M17 agar, incubated anaerobically at 30°C for 48 h; *L. monocytogenes* on *Listeria* selective agar base incubated at 37°C for 48 h; *Escherichia coli* and *Salmonella typhimurium* were both detected on Hektoen enteric agar incubated at 37°C for 24 h; *Staphylococcus aureus* on Baird Parker and incubated at 37°C for 48 h.

Results

The microbial loads of experimental cheeses are reported in Figure 3. *Lactococcus lactis* strains were able to perform the rapid acidification of all productions, showing that EOs did not slow down the starter development.

Optimal results were registered against *L. monocytogenes* which was inhibited of almost 3 Log cycles by all EOs at both concentrations tested. *St. aureus* was not inhibited by tangerine EO, while 1-2 Log cycles lower than control (only pathogens) were found in orange and lemon EOs cheeses. Regarding *E. coli* and *S. typhimurium* the higher decrease was registered for lemon EOs at both concentrations. The inhibitory effect could also be due to the competitions for nutrients due to the most rapid growth of *Lc. lactis*.

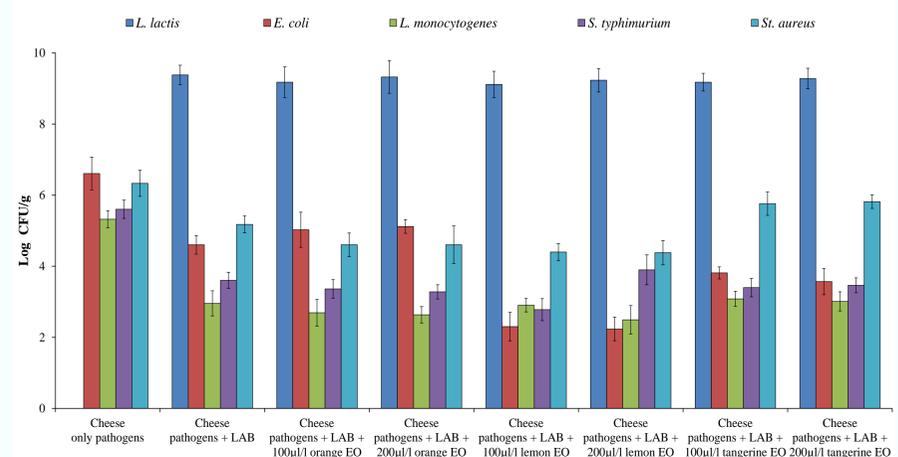


Fig. 4. Microbial loads of experimental cheese samples.

A sensory evaluation of the resulting cheeses, indicated that the cheeses processed with the addition of EOs were well appreciated by the judges.

Conclusions

The addition of industrial citrus EOs did not inhibit the grow of the four dairy pathogenic bacteria under the detection limits. However, the addition of EOs to milk determined the production of novel foods without influencing the fermentation activity of LAB.

Future perspective

Cheese productions with raw ewes' milk and EOs are in progress to enlarging the offer of raw milk processed products.

References

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Acknowledgments

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