

TITLE: Enterotoxigenic potential of *Staphylococcus aureus* isolated from artisanal cheese from Serra da Canastra, Brazil.

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ABSTRACT

Canastra cheese is a typical artisanal product from Brazil produced in Serra da Canastra region, in the Minas Gerais state. This cheese is made from raw milk, added with rennet, and “*pingo*” which is a natural starter culture extracted after the salting stage from the previous day's cheese production. Due to the artisanal manufacturing conditions, this product can present large variations from farm to farm. Additionally, since the cheese is made with raw milk, it may carry pathogenic microorganisms that cause foodborne diseases. The molecular characterization of bacterial species in food products is an important tool to ensure the safety of the final product. This study aimed to characterize the virulence potential of *Staphylococcus aureus* strains isolated from cheese produced in Serra da Canastra. A total of 151 *Staphylococcus aureus* were isolated from three dairies located in Serra da Canastra. The strains were submitted to biochemical tests, such as: coagulase, catalase, termonuclease, cumpling factor and Gram staining. The polymerase chain reaction (PCR) was performed to identify the genes *nuc*, *coa*, and *femA*, as well as the presence of genes encoding enterotoxins, such as *sea*, *see*, *seg* and *seo*. Strains with methicillin resistance (MRSA) were identified by amplification of the *mecA* gene. The clonal profile of the isolates was characterized by Random Amplification of Polymorphic DNA (RAPD) and *agr* typing. Our results showed that the most frequent identified enterotoxin genes were *seg* (16.5%) and *seo* (16.5%). The *sea* and *see* genes were not detected in these strains. The *agr* groups were determined by multiplex PCR and distributed as follows: *agr* I (29.8%), *agr* II (36.4%), *agr* III (2.6%), *agr* IV (0%), while 31,1% did not present an *agr* type. The *mecA* gene was detected in 3.9% of the strains. Our results demonstrated a diversified molecular profile of *S. aureus* depending on the farm by *agr* type, which show that there are multiple strain types in the Canastra region. So far, the characterization showed a virulence potential for the isolates that belong to *agr* type II, which carry *seg* and *seo* genes as well as some strains presented *mecA* gene. It is important to know whether the enterotoxin genes are expressed within the cheese production environment. Therefore, further studies are being done in order to fully characterize the virulence potential of these isolates.

Keywords: *Staphylococcus aureus*, Canastra cheese, virulence factors, genotyping, molecular typing.

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