# Maintenance of *Staphylococcus* spp. producing biofilms in a herd treated by homeopathy

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**Abstract**

*Staphylococci* present pathogenicity factors, such as the production of biofilms, and the possibility of these bacteria remaining in the mammary gland of cows treated with homeopathy is questioned, even after the use of these alternative therapies. This study investigated *Staphylococcus* spp in the milk of cows treated with homeopathy, and the *in vitro* formation of biofilms by these pathogens. Milk samples were collected to determine the somatic cell count (SCC), and duplicate pooled milk samples were collected over a 12-month period to identify *Staphylococcus* species. The cows were divided in two groups: treated and untreated. The biofilm was characterized phenotypically by the adhesion method on polystyrene plates. The results were analyzed using the Chi-square test to examine the association between treatment and biofilm production. The odds ratio was calculated to determine if one group of animals had a higher chance of producing biofilms than the other. *S. aureus* was the species with the highest occurrence in the untreated and treated groups during the study, i.e., 42.5% and 31.9%, respectively. In the untreated group, 2 (5.0%) bacteria presented strong adherence, *S. epidermidis* and *S. warneri*. The nine species (13.0%) identified in the cows treated with homeopathy were classified as strongly adherent, and 5 (55.6%) of them were *S. epidermidis*. The SCC of all the cows in which this species was isolated was higher than 990,000 cells/mL of milk. A significant association was found between homeopathic treatment and biofilm production in treated cows (P=0.013). The odds of isolating *Staphylococcus* biofilms in treated cows were 2.98-fold higher than in the untreated group (confidence interval: 1.24, 7.18). The capacity of biofilm production remained a pathogenicity factor in *Staphylococcus* strains isolated in the milk of cows with subclinical mastitis treated with homeopathy.

**Keywords: S**ubclinical mastitis, milk quality, bacteriology, mastitis treatment, bovine

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