

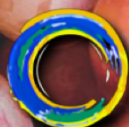
In vitro and in situ challenge on multi-resistant bacteria of food importance with traditional and innovative sanitizers

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The variety of pathogenic bacteria and their virulence and resistance factors makes microbiological control in the food industry challenging. Sanitizers are essential to prevent and control the spread of these pathogens, but the development of resistance makes it essential to implement innovative strategies. Our proposal included verifying the effect of ten different sanitizers on the control of the main causative agents of foodborne illness. The study was conducted with four strains from the poultry industry: *Campylobacter jejuni* (CJ), *Escherichia coli* APEC (ECA), *Salmonella* Choleraesuis (SC) and *Salmonella* Typhimurium (ST). We determined the Minimum Bactericidal Concentration (MBC), which established the minimum concentrations and times (one minute) required to stop the growth of the bacterial strains. The broth microdilution method was used for this. This was followed by an efficiency test, specific to the acceptance of new chemical agents. This was done by macrodilution, in which inoculums of 6 to 8 log CFU/ml were exposed to the compounds at concentrations (determined by the CBM) and for one minute, followed by counting on plates. The in situ test was carried out in facilities to check the antimicrobial effect of routine cleaning. The products tested included those traditionally used and natural (innovative) ones - Peracetic Acid, two types of quaternary ammonium, Biguanide, Chlorhexidine Digluconate (T1, T2, T3, T4, T5), Pine Oil, Neem Extract, Melaleuca Oil, Orange Oil and Lactic Acid (I1, I2, I3, I4, I5). Satisfactory results comprised a reduction of four log cycles in the initial count of microorganisms. The tests were carried out in triplicate and subjected to statistical analysis using Graph Pad Prism 8.0 software. The preliminary tests (CBM) showed that ECA and ST showed resistance to 50% of the agents tested, and CJ and SC to 40% of them. Peracetic acid, chlorhexidine digluconate and neem extract eliminated all the strains by CBM. After defining the target concentrations, we identified efficiency for all the traditional agents, with an average reduction of  $7.23 \pm 0.51$  log CFU, and for the neem extract (average of  $6.67 \pm 0.56$  log CFU) in controlling the four strains. Pine oil was not effective against any of the strains tested ( $p=0.9861$ ). The other compounds were also able to. With the exception of I1, the analysis by species showed that CJ was susceptible to all the other sanitizers, that SC and ECA were resistant to I5 and that ST was resistant to I3, I4 and I5. The in situ test showed that peracetic acid had the best effect, as there was no bacterial growth. However, the reduction of over 99.99% after using biguanide, neem extract, tea tree oil and orange oil highlights the possibility of using innovative alternatives for bacterial control on surfaces. remove more than 99.99% of the average value found for all the bacteria tested, equivalent to a reduction of 5.08, 5.22 and 5.79 log cycles for I3, I4 and I5, respectively. With the exception of I1, the analysis by species showed that CJ was susceptible to all the other sanitizers, that SC and ECA were resistant to I5 and that ST was resistant to I3, I4 and I5. The in situ test showed that peracetic acid had the best effect, as there was no bacterial growth. However, the reduction of over 99.99% after using biguanide, neem extract, tea tree oil and orange oil highlights the possibility of using innovative alternatives for bacterial control on surfaces.

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