TITLE: BIOFILM OF *SALMONELLA* TYPHIMURIUM ON DIFFERENT SURFACES COMMON IN THE FOOD PROCESSING ENVIRONMENTS.

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ABSTRACT:

Salmonella spp. is a pathogen strongly associated with outbreaks of foodborne diseases and has the ability to adhere and form biofilms on different surfaces present in food processing environments, becoming extremely resistant to antibiotics and disinfectants. Thus, the aim of this study was to evaluate the ability of *Salmonella enterica* Typhimurium ATCC 14028 to form biofilm on different surfaces that are common in the food processing environments. The ability of *S.* Typhimurium to form biofilm was evaluated by colony counts, biofilm total biomass, biofilm metabolic activity, and scanning electronic microscopy (SEM). The overnight culture of *S*. Typhimurium was diluted in TSB to yield 10^7 CFU mL^{-1}, placed in contact with polypropylene, stainless steel and polystyrene coupons and polystyrene microplate and incubated for 48 h at 35 ºC (two-day-old *S*. Typhimurium biofilm). After incubation, coupons were washed with 0.85% sterile saline solution and subjected to ultrasound at 25 Hz for 5 min. Serial dilutions were performed in 0.85% sterile saline solution, plated on Mueller Hinton Agar (MHA), and incubated at 35 °C for 24 h and results were expressed as $\log_{10}$ CFU cm^{-2}. For the biofilm total biomass determination the microplates of polystyrene with the two-day-old *S*. Typhimurium biofilm, were washed with 0.85% sterile saline, fixed with methanol PA for 15 min. and stained with 1% crystal violet for 20 min. After washing and drying, they were then solubilized with 95% ethanol and reading was performed with a microplate reader at 550nm. For the biofilm metabolic activity determination, 0.05% of MTT was added to the wells. After incubation for 1 h at 35 ºC, the MTT solution was replaced by isopropanol acid and mixed for 15 min, and the absorbance was measured at 550 nm. Number of *S*. Typhimurium cells recovered from polypropylene, steel surface and polystyrene were 8,27, 8,25 and 7,5 log CFU/cm^2 respectively. The biofilm biomass and biofilm metabolic activity showed a optical density of 2,28 and 0,1429 respectively. SEM images demonstrated the presence of microcolonies, cellular multilayer which are biofilm characteristics. These results prove the ability of *S.* Typhimurium to form biofilms on different surfaces. *Salmonella* spp. biofilms have attracted the attention of food processing industries because they are a continuing source of food contamination. For this, new strategies to biofilm control are necessary.

Keywords: Biofilm, polypropylene, polystyrene, *Salmonella* Typhimurium, stainless steel.

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