

TITLE: MICROBIOLOGICAL COUNTS OF COW TEATS SUBMITTED TO PHOTODYNAMIC INACTIVATION OF MICROORGANISMS WITH SAFRANIN-O AS POST-DIPPING

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ABSTRACT: Photodynamic Inactivation of Microorganisms (PDIM) consist of the generation of reactive oxygen species (ROS) that are toxic to microorganisms through the activation of photosensitizing compounds (PS) with a visible light photon. *In vitro* assays report that this technique is effective in inhibiting mastitis-causing microorganisms. The objective was to determine the microbiological count of teats from cows submitted to PDIM as post-dipping. The experiment was carried out at the Dairy Cow Sector of the Experimental Farm of Iguatemi, under the approval of the Ethics Committee on Animal Use of the State University of Maringá, process number 1287230920. The animals were randomly divided into two treatments: Ekomilk® lactic acid post-gel film (control) and Safranin-O Hydrogel (Sf) with 1 minute irradiation in each teat (green light emitting diode coupled to a conventional plastic teat, $\lambda = 520$ nm). Teat samples were collected with swab before milking, on days 0, 7, 14, 21 and 28, and submitted to microbiological analysis to count the total mesophilic aerobic, *Pseudomonas* spp. and *Staphylococcus aureus* by decimal dilutions and seeding on Plate Agar, Pseudomonas Agar and Mannitol Salt Agar, respectively. Plates were incubated at 35 °C for 48 hours, except *Pseudomonas* spp., incubated at 25 °C. There was no significant difference between treatments ($P > 0.05$). There was a significant difference for *Pseudomonas* spp. and *Staphylococcus aureus* ($P < 0.05$) over time. Counts of *Pseudomonas* spp. they were 3.94 log CFU/mL at the start of treatment (day 0) and 1.00 log CFU/mL after 28 days. For *Staphylococcus aureus*, the counts were 3.25 log CFU/mL at the beginning and 3.03 log CFU/mL at the end. PDIM performance depends on several factors, such as light dosage, PS concentration, presence of oxygen and interaction between PS and target cell and, therefore, there is a big difference between the results reported in the *in vitro* literature and the application of the technique on animals. For milk production systems, the main advantage of the method is the replacement of the use of antibiotics that cause bacterial resistance, as ROS cause nonspecific, immediate and irreversible damage. Safranin-O Hydrogel was as efficient as lactic acid when used as a post dipping for cows.

Keywords: dairy products, mastitis, milk, photodynamic therapy