Evaluation of Brazilian Boot Swab Matrix Interference on Loop-Mediated Isothermal Amplification (LAMP) Assay - Bioluminescent Collected From Farms at Sanitary Void and Pre-Slaughtering Moments

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Introduction: Brazil is one of the world\'s largest exporters of poultry products. To maintain safe production, sanitary measures such as the analysis of broiler litter for monitoring and controlling Salmonella spp. are necessary from farm to processor. These matrices, that can be highly alkalinized, can interfere at molecular biology reaction for the detection of pathogens. They are collected by dragging moistened boot swabs in the environment. Choosing a rapid and accurate detection method, that present low reaction interference, in primary production samples is crucial to obtain accurate results for monitoring intervention effectiveness and for taking action to avoid product contamination. Purpose: Determine the primary production boot swabs interference in LAMP-Bioluminescent assay reaction Methods: Boot swabs samples (n=132) from 2 Brazilian farms (sanitary void (n=60, from middle-west) and pre-slaughtering (n=72, from south)), were analyzed through Matrix Control kit. Sample composition was broiler litter (e.g., soil, feathers, feces, wooden shavings, and lime powder) and sterile boot swabs moistened with BPW ISO (10 mL). Fractional artificial contamination was used, blank and the strain (Salmonella Typhimurium ATCC 14028 for sanitary void and Salmonella Heidelberg for pre-slaughtering, both adapted to the matrix), was spiked in 2 levels, low (N1=0.2-2 CFU/test portion) and high (N1=5 CFU/test portion) for both groups. Samples were enriched with BPW ISO (360mL for the sanitary void and 225 mL for the pre-slaughtering), incubated (37°C/22hours) and analyzed with LAMP-Bioluminescent assay, by ISO 6579-1:2017 and by Matrix Control kit. Parameters required by ISO 16140-2:2016 for method comparison were determined. Results: The alternative LAMP-Bioluminescent presented sensitivity, RT, RLOD and AL of 94.7%, 96.7%, 1.30 for sanitary void and 94%, 97%, 1.15 being the rapid method fit for purpose. Matrix control kit revealed 100% for sanitary void and 97.2% of valid results for pre-slaughtering boot swabs. Significance: The Neogen®Molecular Detection assay 2 enables reliable and rapid pathogen detection in alkaline primary production sanitary void and pre-slaughtering boot swabs with none or very little interference.

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