Fluorescent Microspheres as Surrogates for *Salmonella enterica* serotype Typhimurium in Recovery Studies from Stainless Steel

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ABSTRACT

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To compare the optimum recoveries of an inoculation of *Salmonella enterica* serotype Typhimurium, fluorescent microspheres (1.0 µm diameter, carboxylate-modified, crimson FluoSpheres®, Molecular Probes, Eugene, OR), or a combination of both from stainless steel, three recovery methods, including a standard rinse, a one-ply composite tissue (Kimwipe®) or a sonicating brush were used. Findings were used to assess the effectiveness of fluorescent microspheres as surrogates for *S.* Typhimurium. For each method, ten coupons (304 grade, 2.5 x 8 cm) were inoculated with either 100 µl of a *S.* Typhimurium culture, or a solution of fluorescent microspheres, or both, at approximate concentrations of 10^6. After drying for one hour, coupons were sampled using either a rinse of 100 ml of phosphate buffered saline solution (PBS) for one min, a Kimwipe® tissue method, or submerged in PBS and subjected to a sonicating brush for one min. After treatments, PBS solutions were analyzed using duplicate plate counting (*Salmonella*) or hemacytometry (microspheres). For microspheres and *Salmonella*, recovery by sonicating brush > rinse > Kimwipe® method. Additionally, the retention of microspheres on the steel ranged from 16 to 25% (mean from five coupons each recovery method). Microspheres yielded a significantly higher recovery rate (11 – 60%) than *Salmonella* (~1%) for each

recovery method, therefore the microspheres used in this study, are not appropriate surrogates for *S.* Typhimurium for future recovery studies on stainless steel. However, due to their low standard deviations for their mean percent recovery, they hold the opportunity to provide better accuracy and reproducibility.