RESEARCH ABSTRACT

PORK SAFETY

Title: A Diagnostic for Rapid Detection of Pathogenic *Listeria monocytogenes* as an Indicator of Hygiene on Food Contact and Non-Food Contact Surfaces in the Pork Processing Environment - **NPB #06-178**

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Scientific Abstract

Listeria monocytogenes is a ubiquitous pathogen and a major cause of food related illness. Current methods of detection are slow, laborious, and require expensive equipment, or technical expertise. A rapid, easy to use test to detect *L. monocytogenes* was developed, that requires little to no equipment or technical training. The Phast Swab is a self-contained test device, containing a sampling tool (swab), growth media, immunomagnetic separation (IMS) beads, and a colorimetric substrate, which when cleaved by the enzyme phosphatidylinositol-specific phospholipase C (PI-PLC), forms a visible indigo reaction. To conduct the test, a surface (food, contact and non-contact surfaces) to be tested is swabbed, and the swab is returned to the test device. Follwing a 10-13 hour enrichment, any *L. monocytogenes* cells present in the growth media are isolated and concentrated by IMS and the growth media is removed. Bacterial lysis buffer is added, followed by the enzyme substrate, which reacts with the PI-PLC to form the indicative indigo color for positive identification. The indigo reaction had to be visually detected within 5 hours to be considered positive.

The Phast Swab was evaluated using artificially contaminated RTE meat and food contact and non-contact surfaces.

A five-strain cocktail of *L. monocytogenes* was prepared so that the final concentration of the cocktail was 10^9 CFU/ml. The cocktail was serially diluted (10-fold) in a dilution range from 10^0-10^{-9}, and individual slices of deli turkey meat (100mm diameter) were inoculated with 1ml of each dilution. The meat was allowed to dry, and then each piece was swabbed with an individual Phast Swab, and the test was conducted as described.

The Phast Swab was capable of detecting 10^2 cells on the deli meat within 15 hours. The test detected 55 of 57 *L. monocytogenes* tested and none of the 15 non- *L. monocytogenes* bacterial strains tested. In pure culture, as few as 10^7 CFU/ml were directly detected within 3 hours. The addition of 428 mM KCl to the growth media significantly increased the sensitivity of the assay.

For detection of *L. monocytogenes* on food contact and non-contact surfaces, a five-strain cocktail of *L. monocytogenes* was prepared so that the final concentration of the cocktail was 10^9 CFU/ml with 10X serial dilutions to 10^0 CFU/ml. Individual stainless steel, acrylic and ceramic tile coupons (4"×4") (100 cm^2) chosen to simulate food contact and non-contact surfaces, were inoculated with 250µl of the 10^7, 10^6, 10^5, CFU/ml dilutions. Plate counts showed that after each surface dried for five hours, the actual *L. monocytogenes* counts on the surfaces were 2 logs lower than the initial inoculums. The coupons were allowed to dry and were swabbed with an individual Phast Swab device. The devices were incubated for 13 h, and the test was completed as described.

The Phast Swab was capable of detecting as few as 10^1 CFU/cm^2 that was present on the coupons.

Collectively, these data show that the Phast Swab can detect *L. monocytogenes* on food samples (RTE meat) and food contact and non-contact surfaces in the food processing environment in a rapid and specific manner without the need for major instrumentation to read the test result.