**Introduction**

Boot swab sampling has proven to be a simple, safe, cost-effective and user-friendly environmental method for assessment of microorganism burdens in flocks. However, given high non-specific microbial populations and environmental contaminants, rapid, same-day analysis is challenging. Here we show the results from an environmental survey utilizing a newly developed Veriflow DNA-Signature Capture assay platform for the rapid detection of Campylobacter species from boot swab samples. The assay allows for same-day results in under 3-hours from sample processing, circumventing the need for enrichment or culture based methods. 142 boot swabs collected from broiler houses were assessed for total Campylobacter burden prior to live-haul, utilizing this method. Ultimately, this platform can enable real-time monitoring of Campylobacter levels in flocks and empower production teams to identify and mitigate the risk of cross-contamination prior to processing.

**Methods**

142 boot swab samples were collected from 48 poultry growth facilities during the Fall of 2016, encompassing turkey and chicken grow-out houses, hatcheries and breeders. Each set of samples were collected, coded (blinded study), and shipped at 4°C to the labs of Invisible Sentinel for immediate processing and testing. Each pair of boot swab were then rinsed in 400 mL Buffered Peptone water and processed according to Veriflow Campy Q Boot Swab procedures. Briefly, each sample was diluted in a proprietary treatment buffer system designed for one-step sample processing of poultry, cecal, and boot-swab samples. Samples were then analyzed on Veriflow and results were quantified using the Veriflow Boot Camp program on the Veriflow optical reader. Results were compiled and detection rates were compared between facilities.

**Results**

35% of all turkey grow house samples were positive (16/45) and 33% of chicken grow house samples were positive (19/57). Campylobacter levels above 300 cells/ml were not detected in boot swab rinsates from breeder and hatchery operations, indicating low incidence rates in these facilities and at these production stages. Positive results were clustered at specific ranch houses, allowing for identification of locations with relatively high levels or low levels of Campylobacter both in specific burden (cells per mL) or % sample positive rates. Quantitative results show an average range of log_{10} 2.6-3.9 per mL of rinsate in samples that were positive in this study. Results indicate that the Veriflow Campylobacter Q platform can provide rapid information regarding contamination levels from boot swabs, enabling site-specific assessment of relative flock contamination levels in under 3-hours from sample collection.

**Purpose**

The purpose of the study was to validate the utility of the Veriflow Campylobacter Q (Campy Q) assay to rapidly identify high burden flocks prior to processing via boot swab sampling. The Campy Q assay was designed to provide rapid, quantitative detection of Campylobacter species from various sample types including boot swabs. The assay produces results in under 3 hours, and provides a quantitative result (cells/mL) of the Campylobacter microbial burden found on the tested boot swab. The dynamic range of the Campy Q assay is approximately 300 cells per mL to 10,000 cells/mL of boot swab rinsate. Anything above 10,000 cells/mL would be considered high burden while anything below 300 cells/mL would be classified as low risk/low burden. This dynamic range was established in conjunction with industry partners.

Campy Q was built upon the AOAC approved Veriflow® Campylobacter species platform: a DNA Signature Capture based assay, where the results are garnered and visualized on a handheld cassette. The assay is designed to be easy-to-use, with no DNA purification or complex sample preparation.

**Significance**

*Campylobacter* is a commensal organism that naturally colonizes the digestive tract of poultry and can become a contaminant during processing as it is a leading cause of gastrointestinal disease in humans. Accordingly, poultry manufacturers employ multiple strategies to reduce the risk of *Campylobacter* species reaching the consumer including anti-bacterial treatments and managing cross-contamination during processing. The results of this study show that Veriflow Campylobacter Q can be employed as a valuable tool allowing for preemptive management of Campylobacter flock and facility cross-contamination, by providing same day information regarding the levels of Campylobacter in specific flocks. In this study, the Campylobacter Q assay identified correlates between specific houses and flocks, and provided production teams with the ability to stage high burden flocks and low burden flocks during processing. For example, this survey identified and delineated ranches with either high or low risk incidence rates, as shown in figure above. The advantage of obtaining same day, quantifiable results enables processors to stratify the risk of various poultry habitats and modify intervention strategies during production, providing a tool to manage the risk of cross-contamination during processing.