Technical Bulletin: Detection of *Salmonella* from Whole Peaches using the BAX® System PCR Assays

A paired validation was conducted to evaluate the performance of the BAX® System to detect *Salmonella* from whole peaches. Samples in this study were artificially inoculated with a low level (1.2 CFU/test portion) expected to produce fractional positive results and a high level (12 CFU/test portion) expected to produce all positive results. Samples were held for 72 hours and enriched using the U.S. Food & Drug Administration’s Bacteriological Analytical Manual (FDA BAM) reference method. Test results were analyzed using the probability of detection (POD), demonstrating equivalent performance between the BAX® System method and the reference method.

**Introduction**

Tree fruits are generally perceived as ‘low risk’ from becoming contaminated with pathogens since the fruits are not in direct contact with soil, irrigation water or fertilizers (1). However, mangoes, papayas and most recently peaches, have all been vehicles in foodborne disease outbreaks due to the presence of *Salmonella* (2). Since these products are usually consumed raw, microbial food safety hazards must be identified and reduced.

**Sample Preparation and Enrichment**

Whole peaches were inoculated with *Salmonella Enteritidis* to create 20 low-level samples and 5 high-level samples. Five samples were left uninoculated for negative controls. Additional samples were prepared for MPN analysis. All samples were held at room temperature for 72 hours to equilibrate the target organism in the matrix.

Samples were then enriched with approximately 500 mL of Universal preenrichment broth (UPB) which was sufficient volume to allow the peach to float. Samples were mixed by swirling and held at room temperature for 60 minutes before being incubated at 35°C for 22-26 hours.

See Figure 1 (note: sample enrichments in this study were not pH adjusted since measurements were within the correct range of 6.8 ± 0.2).

**Method**

**BAX® System Method**

All samples were processed following the procedures for Real-Time *Salmonella* (KIT2006) and *Salmonella* 2 (KIT2011) described in the BAX® System Q7 Users Guide.

**Reference Method**

All samples were culture confirmed regardless of BAX® System results following the FDA BAM Chapter 5 for *Salmonella*.

![Figure 1. Paired study using the FDA BAM reference enrichment method and BAX® System method for whole peaches.](image-url)
Results
Both BAX® System PCR assays (Real-Time *Salmonella* and *Salmonella 2*) returned positive results for 17/20 low spiked samples and 5/5 high spiked samples at 24 hours. When compared to culture, all BAX® System results were identical.

To compare the method performance, the BAX® System presumptive and confirmed results were analyzed using the probability of detection (POD). No significant difference was determined since the 95% confidence interval includes zero (Table 1).

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>CFU/Test Portion</th>
<th>N</th>
<th>BAX® System Presumptive</th>
<th>BAX® System Confirmed</th>
<th>dPOD&lt;sub&gt;cp&lt;/sub&gt;</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole Peaches</td>
<td>Control</td>
<td>5</td>
<td>0</td>
<td>0.00</td>
<td>0.00, 0.45</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>1.23</td>
<td>20</td>
<td>17</td>
<td>0.85</td>
<td>0.64, 0.95</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td>12.3</td>
<td>5</td>
<td>5</td>
<td>1.00</td>
<td>0.57, 1.00</td>
<td>5</td>
</tr>
</tbody>
</table>

N = Number of test portions  
X = Number of positive test portions  
POD<sub>cp</sub> = BAX® System method presumptive positive results divided by the total number of test portions  
POD<sub>cc</sub> = BAX® System method confirmed positive results divided by the total number of test portions  
dPOD<sub>cp</sub> = Difference between the BAX® System method presumptive result and BAX® System method confirmed result  
POD values  
95% CI = If the confidence interval of dPOD does not contain zero, then the difference is statistically significant at the 5% level

Conclusions
Overall, this study provides reliable and accurate results using the BAX® System PCR assays to rapidly screen whole peaches for the presence of *Salmonella* with 100% sensitivity and 100% specificity following the FDA BAM reference enrichment method:

- Enrich 1 whole peach with enough UPB for the peach to float (approximately 500 mL), swirl and let stand at room temperature for 60 minutes, adjust pH, if necessary, to 6.8 ± 0.2 and incubate at 35°C for 22-26 hours.

References
1. Alegbeleye, O. O., Singleton, I., Sant’Ana, A. S. (2018). Sources and contamination routes of microbial pathogens to fresh produce during field cultivation: A review. Food Microbiology. 73, 177-208.