Guacamole has increased in popularity over the last few decades. Made from raw avocado and often mixed with other raw ingredients including cilantro, onion, tomato, pepper and other produce, guacamole has emerged as a new concern for *Listeria monocytogenes* contamination.

In fact, in March 2019, the US Food and Drug Administration (FDA) announced a recall of whole California-grown avocados due to residues of *L. monocytogenes* on the products’ skins. Cutting through the skins could potentially transfer the bacteria to the food, although no illnesses were reported by either the FDA or the processing company.

As of October 2018, the FDA had tested 474 processed avocado or guacamole samples. The agency found 11 that were positive for *L. mono*. While a small percentage, there is a zero-tolerance policy for *L. mono*. Low levels of *L. mono* can cause harm or death, as they can survive refrigeration, freezing and other conditions that usually kill bacteria. As a result, proactive companies follow a “search and destroy” strategy for *L. mono*.

Meanwhile, the US Centers for Disease Control and Prevention created a new category of food-borne illness called SGA, short for salsa and guacamole-associated outbreaks. One out of 25 foodborne illness outbreaks are SGAs. Most lead back to restaurants.
The CDC estimates that about 1,600 people get sick from *Listeria* every year. *L. mono* can cause mild illness in many people, including fever and diarrhea, and may go unreported. But *Listeria* infections also have a high fatality rate. Of the 1,600 illnesses reported, about 260 result in death. *L. mono* is a particular problem for pregnant women, because the bacteria can cause miscarriage, stillbirth, premature delivery, or life-threatening infections to the newborn. Older people and those with compromised immune systems are also more susceptible to *Listeria* infection.

Because of these risks, Hygiena scientists designed a study to evaluate the performance of multiple BAX® System PCR assays to detect *L. monocytogenes* in guacamole. Real-Time Genus *Listeria*, Real-Time *L. monocytogenes*, standard Genus *Listeria* and standard *L. monocytogenes* PCR assays were tested on the BAX® System Q7 instrument, and Genus *Listeria* X5 and *L. monocytogenes* X5 PCR assays were tested on the BAX® System X5 instrument. Low and high levels of inoculated guacamole were enriched in two different primary enrichment mediums, 24 *Listeria* Enrichment Broth (LEB) Complete or Demi-Fraser, which both selectively enrich *Listeria* growth, in comparison to the FDA’s Bacteriological Analytical Manual (BAM) reference cell culture method.

Samples enriched in 24 LEB Complete media and then tested after a secondary enrichment in MOPS-BLEB returned positive results for *Listeria* in 17 low-spiked samples, and all 5 high-spiked samples tested with all six BAX® System *Listeria* assays. All results were identical to cell culture methods.

Samples enriched in Demi-Fraser broth and then tested after a secondary enrichment in MOPS-BLEB returned positive results for *Listeria* in 9 low-spiked samples, and all 5 high-spiked samples tested with all six assays. All BAX® results were identical to cell culture.

Samples enriched according to the FDA BAM reference method were also tested with the BAX® System after 48 hours, with positive *Listeria* results for 20 low-spiked samples and 5 high-spiked samples (Table 1).
No significant difference was seen between the BAX® System Method in 24 LEB Complete/MOPS-BLEB and the BAM reference method. However, a significant difference was seen between the BAX® System method in Demi-Fraser/MOPS-BLEB and the reference method—the reference method had more positives. It’s possible that certain components in the Demi-Fraser broth hindered the initial growth of *Listeria*.

Therefore, the study indicated that all of Hygiena’s Genus *Listeria* and *L. mono* BAX® System PCR assays on the Q7 and X5 instruments can accurately detect *Listeria sp.* and *L. mono*, respectively, in 25 g guacamole samples after a primary enrichment in 24 LEB Complete Media followed by a secondary enrichment in MOPS-BLEB equivalent to the reference method.

<table>
<thead>
<tr>
<th>Enrichment and method</th>
<th>Control samples (detected/total spiked)</th>
<th>Low-spiked samples</th>
<th>High-spiked samples</th>
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<tbody>
<tr>
<td>24 LEB Complete/MOPS-BLEB</td>
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<tr>
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<td>5/5</td>
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<tr>
<td>BAM reference method</td>
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<td>20/20</td>
<td>5/5</td>
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Table 1: Comparison of control and spiked samples using BAX® and reference methods