Bioburden Final Report

Study Number: 973554-S01.1 Amended
Test Article: ISO 80369-3 Connectors
ORANGE CONNECTORS (Brushed) - Lot #20160829
PURPLE CONNECTORS (Not Brushed) - Lot #20160826
Purchase Order: 061917
Study Received Date: 28 Jun 2017
Study Completion Date: 17 Jul 2017
Testing Facility: Nelson Laboratories, LLC, a Business Unit of Sterigenics International
6280 S. Redwood Rd.
Salt Lake City, UT 84123 U.S.A.
Test Procedure(s): Standard Test Protocol (STP) Number: 800-STP0036 Rev 14
Customer Specification Sheet (CSS) Number: 201704311 Rev 1

Summary: The sponsor performs any statistical analysis and determines the acceptable limits. Testing was performed in accordance with ANSI/AAMI/ISO 11737-1:2006/R2011. Testing was performed in compliance with US FDA good manufacturing practice (GMP) regulations 21 CFR Parts 210, 211 and 820.

Results: When bioburden results are calculated using a validated software program, manual calculations may differ slightly due to rounding. The counts determined on products are colony forming units and may not always reflect individual microorganisms.
**Test Method Acceptance Criteria:** If applicable, anaerobic controls are acceptable for the bioburden test results.

**Procedure:**

- **Positive Controls/Monitors:** *Bacillus atrophaeus*
  
- **Negative Controls/Monitors:** *Pseudomonas aeruginosa*

Extract Fluid: Peptone Tween®

Extract Method: Manual Shaking

Plating Method: Membrane Filtration

Agar Medium: Soybean Casein Digest Agar, Tryptic Soy Agar

Aerobic Bacteria: Plates were incubated 3 - 7 days at 30-35°C, then enumerated.

Anaerobic Bacteria: Plates were incubated under anaerobic conditions 3 - 7 days at 30-35°C, then enumerated.

<table>
<thead>
<tr>
<th>Unit Number</th>
<th>Test Article</th>
<th>Aerobic</th>
<th>Anaerobic</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Powder, Orange (Brushed)</td>
<td>TNTC</td>
<td>TNTC</td>
</tr>
<tr>
<td>2</td>
<td>Powder, Orange (Brushed)</td>
<td>$2.3 \times 10^3$\textsuperscript{a}</td>
<td>TNTC</td>
</tr>
<tr>
<td>3</td>
<td>Powder, Purple (Not Brushed)</td>
<td>27\textsuperscript{b}</td>
<td>TNTC</td>
</tr>
<tr>
<td>4</td>
<td>Express, Orange (Brushed)</td>
<td>TNTC</td>
<td>TNTC</td>
</tr>
<tr>
<td>5</td>
<td>Express, Orange (Brushed)</td>
<td>TNTC</td>
<td>TNTC</td>
</tr>
<tr>
<td>6</td>
<td>Express, Purple (Not Brushed)</td>
<td>$2.2 \times 10^3$\textsuperscript{a}</td>
<td>TNTC</td>
</tr>
</tbody>
</table>

**Averages**

<table>
<thead>
<tr>
<th></th>
<th>Aerobic</th>
<th>Anaerobic</th>
</tr>
</thead>
<tbody>
<tr>
<td>UTD\textsuperscript{c}</td>
<td>UTD\textsuperscript{c}</td>
<td></td>
</tr>
</tbody>
</table>

TN TC = Too Numerous to Count

UTD = Unable to Determine

Note: The results are reported as colony forming units per test article.

Note: Sample positive testing was performed using *Bacillus atrophaeus*. The test article was not inhibitory using this test method.

\textsuperscript{a}This value is considered an estimate since the raw count exceeds the statistically accurate range.

\textsuperscript{b}Spread. Count is considered a minimum estimate due to swarming of certain colonies on the membrane.

\textsuperscript{c}UTD due to a TNTC value.
Amendment Justification(s):

1 Amended: At the request of the sponsor, "ISO 80369-3 Connectors" was added to the test article section.