In-vitro growth characteristics of commercial probiotic strains and their potential for inhibition of Clostridium difficile and Clostridium perfringens

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Publication date:
2012

Document Version
Publisher's PDF, also known as Version of record

Link back to DTU Orbit

Citation (APA):
**IN-VITRO GROWTH CHARACTERISTICS OF COMMERCIAL PROBIOTIC STRAINS AND THEIR POTENTIAL FOR INHIBITION OF CLOSTRIDIUM DIFFICILE AND CLOSTRIDIUM PERFRINGENS**

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**Background**

C. difficile and C. perfringens are important animal pathogens. There are currently no preventative measures and therapeutics are limited to antibiotics. With antibiotic resistance on the rise new approaches are needed.

Probiotics have been evaluated in humans in relation to clostridial disease and results have been promising. There are no animal probiotics licensed for use against clostridial disease.

Many strains that show promising activity can not be commercially produced as they are not technically robust. This could be overcome by using strains already in commercial production.

**Objective and Hypothesis**

1) To assess the ability of selected commercial probiotic strains to inhibit growth of C. difficile and/or C. perfringens in vitro

2) To evaluate their ability to grow in the presence of oxygen, acid and bile

**Material and Methods**

Seventeen probiotic strains were used (Table 1).

**Inhibition of C. difficile and C. perfringens**

The effect of a cell free probiotic culture supernatant on the growth of C. difficile ribotype 078 and C. perfringens Type C was assessed. Supernatant was harvested and sterilized after 12, 24, 36, 48, and 72 hours and six days. One aliquot was adjusted to pH 7.4 (pH _adj_) the other aliquot was left at original pH (pH _orig_).

**Agar well diffusion assay**

The anti-clostridial activity was evaluated by agar well diffusion following addition of supernatant at pH _orig_ or pH _adj_.

**Results**

**C. perfringens**

<table>
<thead>
<tr>
<th>Strain</th>
<th>Trade Name</th>
<th>pH <em>orig</em> (3.8-4.9)</th>
<th>pH <em>adj</em> 7.4</th>
</tr>
</thead>
<tbody>
<tr>
<td>L. plantarum BG112</td>
<td>√</td>
<td>100</td>
<td>1</td>
</tr>
<tr>
<td>L. rhamnosus LRH19</td>
<td>x</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>E. faecium EF1</td>
<td>x</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>L. casei LC11</td>
<td>√</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>L. acidolin L62</td>
<td>x</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>L. rhamnosus SP1</td>
<td>√</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>B. animalis spp lactis</td>
<td>BLC1</td>
<td>100</td>
<td>0</td>
</tr>
</tbody>
</table>

**Agar diffusion assay**

**C. perfringens**

<table>
<thead>
<tr>
<th>Strain</th>
<th>Trade Name</th>
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<th>C. perfringens</th>
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<tbody>
<tr>
<td>L. plantarum BG112</td>
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<td>x</td>
<td>x</td>
</tr>
</tbody>
</table>

**Broth co-culture**

**S.17 probiotics inhibited C. perfringens and 10/17 inhibited C. difficile (Tab. 1)**

Inhibition was only seen with supernatant at pH _orig_.

**Growth characteristics**

All strains grew aerobically except B. animalis lactis.

None of the strains grew at pH 2. Growth at pH 4 ranged between 40-95%.

Growth ranged from 40-99% when bile was added.

**Contact Information**

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