The role of probiotics and prebiotics on the gut microbiota: an overview of the research

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Outline

- Definitions of probiotics & prebiotics
- Mechanisms of action

- Research insights for probiotics & prebiotics on:
  - the gut microbiota
  - digestive health
    - transit time, stool consistency & constipation
    - colon cancer
    - gut infections: AAD & C. Difficile
  - emerging areas of research
    - obesity/metabolic disease
    - the gut-brain axis
**Definitions**

**Probiotic**
Live microorganisms that, when administered in adequate amounts, confer a health benefit on the host.  
FAO/WHO (2002) updated

**Prebiotic**
A substrate that is selectively utilized by host microorganisms conferring a health benefit.  

- Occur naturally in our diet. E.g. garlic, onions, chicory, artichokes,
- Supplements: Inulin, Fructo-oligosaccharides (FOS), Galacto-oligosaccharides (GOS), Lactulose and Lafinose

**Synbiotic**
Synergistic combinations of pro- and prebiotics. The prebiotic compound must selectively favour the probiotic compound.
Identifying Probiotics

- Defined microbiota consortia
  - Probiotic drugs/medical foods/foods/dietary supplements
  - Probiotic infant formulas
  - Non-oral probiotics
  - Probiotic animal feeds

- Fermented foods with undefined microbial contents
  - Undefined consortia (e.g. FMT)

Probiotic

Not Probiotic

- **Yoghurt** starter cultures (*Lactobacillus delbreuckii* subspecies *bulgaricus* and *Streptococcus salivarius* subspecies *thermophilus*) were assessed for GI survival in a double-blind, cross-over study.
  - N=114 healthy young adults had 2-week consumption of live yoghurt (10^{11} bacteria) and 2-week consumption of pasteurised yoghurt.
  - Detection of yoghurt bacteria were consistently **negative in faeces**.

Del Campo et al. (2005) *Applied Environmental Microbiology* 71:547-549
Mechanisms of Action

- Increase numbers of beneficial bacteria in the gut
  - Increase competition for nutrients & adhesion sites

- Decrease intestinal pH
  - Create unfavourable habitat for most pathogens

- Modulate gut metabolic activities (host & flora)
  - ↓ carcinogens/toxins
  - ↑ short chain fatty acid production
  - Improving metabolism of nutrients

- Influence the intestinal epithelia
  - Normalising increased gut permeability
  - Maintaining gut barrier function
  - ↓ pathogen adhesion/colonisation

- Modulate the immune system
  - Innate & acquired

Mechanisms of Action

Widespread (among studied probiotics)
- Colonisation resistance
- Production of SCFAs
- Regulation of intestinal transit
- Normalisation of perturbed microbiota
- Increased turnover of enterocytes
- Competitive exclusion of pathogens

Frequent (species-level effects)
- Vitamin synthesis
- Pathogen antagonism
- Gut barrier reinforcement
- Bile salt metabolism
- Enzymatic activity
- Neutralisation of carcinogens

Rare (strain-specific effects)
- Neurological
- Immunomodulatory
- Endocrinological
- Production of specific bioactives

Adapted from:
Sanchez et al. (2017) Mol Nutr Food Res 61(1)
Impact on the Gut Microbiota

- An increased proportion of bifidobacteria and lactobacilli is thought to represent a “healthier” microbial composition
  - these bacteria are more likely to ferment carbohydrates and produce acids, and they generally lack potential toxicity

- **Probiotics:** The consumption of adequate doses of *Lactobacillus* strains often results in a measurable increase in the lactobacilli in the faeces, and in some cases there may be a decrease in unfavourable organisms such as staphylococci.

- **Prebiotics:** There is evidence from human subjects, including infants, that established prebiotics increase the proportion of bifidobacteria and sometimes lactobacilli present in the gut microbiota while having no measurable effects on other groups of bacteria.


science for health
Transit Time & Stool Bulking

- There is strong evidence that prebiotics and probiotics can influence gut function.

- **Prebiotics**
  - This effect for prebiotics is thought to be due to their fermentation in the colon, resulting in increased bacterial mass and osmotic water-binding capacity that contribute to increased stool weight, increased stool frequency and softer stools.
  - In some studies, prebiotics are reported to reduce symptoms of intestinal discomfort, such as bloating, abdominal pain and flatulence.

- **Probiotics**
  - Studies on certain strains of probiotic bacteria have demonstrated an impact on gut function, as revealed by normalisation of transit time and reduction of self-reported minor digestive discomfort symptoms.

Constipation

- Research from randomised-controlled trials, meta-analyses and Cochrane Reviews have shown effectiveness of a range of different probiotics;
  - **Bifidobacterium lactis**: increased stool frequency and transit time
  - **Lactobacilli and Bifidobacteria** are able to produce short chain fatty acids reducing intraluminal pH and promoting colonic peristalsis, which is beneficial for changing stool frequency
  - No adverse effect / safe
  - Alternative to using more medication

Effect of LcS on Microbiota & Bowel Habits

- Randomised placebo-controlled double-blind trial, n=72 elderly residents of facility for the elderly, aged 84-86 yrs
- Intervention: 6 months of a probiotic (*Lactobacillus casei* Shirota) or placebo beverage

In the probiotic group:

- Higher numbers of *Bifidobacterium* and *Lactobacillus*
- Lower numbers of destructive bacteria such as *Clostridium difficile*
- Higher total acidity
Effect of LcS on Microbiota & Bowel Habits

Table 2. Effect of long-term consumption of LcS-fermented milk on fever and bowel movements of elderly subjects

<table>
<thead>
<tr>
<th>Items</th>
<th>Measurement period</th>
<th>LcS-fermented milk</th>
<th>Placebo</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of days with fever (≥37°C)(a)</td>
<td>Before consumption of test beverage</td>
<td>1.6±2.2</td>
<td>1.9±3.6</td>
</tr>
<tr>
<td>Days/2 weeks</td>
<td>After 1 month of consumption</td>
<td>1.3±2.3</td>
<td>1.8±3.3</td>
</tr>
<tr>
<td></td>
<td>3rd month</td>
<td>1.2±1.8*</td>
<td>2.7±3.6</td>
</tr>
<tr>
<td></td>
<td>6th month</td>
<td>1.1±1.7*</td>
<td>2.5±3.8</td>
</tr>
<tr>
<td>Incidence of constipation(b)</td>
<td>Before consumption of test beverage</td>
<td>0.6±0.8</td>
<td>0.8±0.9</td>
</tr>
<tr>
<td>Days/2 weeks</td>
<td>After 1 month of consumption</td>
<td>0.6±0.7</td>
<td>0.8±0.9</td>
</tr>
<tr>
<td></td>
<td>3rd month</td>
<td>0.5±0.7</td>
<td>0.8±1.1</td>
</tr>
<tr>
<td></td>
<td>6th month</td>
<td>0.6±0.7*</td>
<td>1.0±1.1</td>
</tr>
<tr>
<td>Incidence of diarrhea(c)</td>
<td>Before consumption of test beverage</td>
<td>0.4±0.8</td>
<td>0.4±0.5</td>
</tr>
<tr>
<td>Days/2 weeks</td>
<td>After 1 month of consumption</td>
<td>0.4±0.8</td>
<td>0.5±1.0</td>
</tr>
<tr>
<td></td>
<td>3rd month</td>
<td>0.3±0.6*</td>
<td>0.7±1.0</td>
</tr>
<tr>
<td></td>
<td>6th month</td>
<td>0.3±0.5</td>
<td>0.5±0.8</td>
</tr>
</tbody>
</table>

- Conclusion: Long-term consumption of LcS fermented milk may be useful for decreasing the daily risk of infection and improving the quality of life among the residents.
Prebiotics: Colon Cancer

- Colon cancer has been linked to diets low in dietary fibre and thus the potential for prebiotics to reduce colon cancer risk has also been investigated.

- Results from animal studies and *in vitro* evidence suggest that prebiotics may reduce the risk of colon cancer.

- Potential mechanisms include:
  - changes in gut bacterial enzyme activities → modify fermentation products
  - up-regulation of apoptosis

Prebiotics: Colon Cancer

- A synbiotic study (SYNCAN project\(^1\)) in humans found:
  - an increase in lactobacilli and bifidobacteria, and a decrease in putrefactive bacteria such as *Clostridium perfringens* and coliforms
  - a reduction in DNA damage and a reduction in cell proliferation in colon biopsies.

- However, definitive evidence that certain prebiotics might reduce the risk of colon cancer in human subjects is lacking and requires more robust, multi-centre, prospective human trials.

Probiotics: Antibiotic Associated Diarrhoea & C. difficile

- Antibiotic-associated diarrhoea (AAD) develops in 5-39% of people taking antibiotics, either during or up to 2-3 weeks of finishing the course\(^1\)

- Up to 25% of cases are caused by *Clostridium difficile* (C.diff)
  - A spore forming bacteria normally present in 3% of people
  - Its growth is usually prevented by the gut microbiota
  - Antibiotics disrupt the microbiota, and other commensal organisms can grow unchecked, producing toxins that cause illness (diarrhoea → pseudomembranous colitis)

- Probiotics may be effective by preventing gut dysbiosis

“There is strong evidence of efficacy for probiotics in adults or children who are receiving antibiotic therapy”
*World Gastroenterology Organisation Global Guidelines 2017*

Probiotic administration can reduce the risk of developing *C. difficile* associated diarrhoea in patients receiving antibiotics\(^{(1)}\)

- Meta-analyses of 26 RCTs evaluating the use of probiotics in the prevention of CDAD (\(n = 7,957\): 4,124 received probiotic (*Lactobacillus, Saccharomyces*, or a mixture or probiotics) and 3,833 received placebo/no treatment)
- Probiotic administration was associated with a significantly lower risk of developing CDAD (\(RR = 0.395, p<0.001\))
- Sub-group analysis:
  - *Lactobacillus*: \(RR = 0.363, P<0.001\)
  - *Saccharomyces*: \(RR = 0.415, P=0.008\)
  - Mixed probiotic: \(RR = 0.418, P<0.001\)

*Lactobacillus casei* Shirota reduced recurrent CDI\(^{(2)}\)

- 66 patients (median age 70y/old) with *C. diff* infection
- 31 patients had taken antibiotics + LcS, 35 patients had taken antibiotics only
- During follow-up, LcS group had significantly lower recurring *C. diff* infection compared to control (3.2% Vs 20.0%, \(p=0.007\))
- Readmission for diarrhoea to hospital was lower in LcS group (19.4% Vs 35.1%)

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2. Lee et al. (2013) *International Journal of Probiotics & Prebiotics* 8:3-4
Obesity-Associated Metabolic Disorders

Background Evidence: Gut Microbiota Link

- Children born by caesarean-section are:
  - twice as likely to be obese at 3 years of age \(^1\)
  - 64% more likely to be obese than their siblings born by vaginal delivery \(^2\)

- Low bifidobacteria during infancy \(\Rightarrow\) ↑childhood obesity \(^3\)

- Increased gut permeability in type 2 diabetes mellitus (T2DM) \(^4\)

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\(^1\) Huh et al. 2012 Arch Dis Child 97(7): 610-616
\(^2\) Yuan et al. 2016 JAMA Pediatr 170(11):e162385
\(^4\) Bischoff et al. 2014 BMC Gastroenterology 14:189
Obesity-Associated Metabolic Disorders

Animal Studies

- Changes in gut microbiota controls metabolic endotoxemia, inflammation, and associated disorders by a mechanism that could increase intestinal permeability \(^1\text{-}^2\)

\[ \downarrow \text{bifidobacteria} \rightarrow \uparrow \text{gut permeability} \rightarrow \uparrow \text{endotoxaemia} \]

\[ \uparrow \text{metabolic disorders/diabetes} \leftarrow \uparrow \text{low grade inflammation} \]

- Studies based on gnotobiotic/germ-free animal models and faecal microbial transplants (FMT) have provided unequivocal evidence that perturbations in bacterial communities play a key role in the pathophysiology of obesity and insulin resistance \(^3\text{-}^4\)

1 Cani et al. 2007 Diabetes 56(7): 1761-1772
2 Cani et al. 2008 Diabetes 57(6): 1470-1481
3 Turnbaugh et al. 2006 Nature 444(7122): 1027-1031
Obesity-Associated Metabolic Disorders

Human Studies

- Not always in agreement with animal studies
- Metagenomic studies show difference between the gut microbiota of healthy people and those with T2DM (can even classify and predict risk of T2DM) \(^1\)
- Areas of probiotic research include: NAFLD, metabolic syndrome, insulin resistance, type 2 diabetes, obese subjects (to reduce girth and subcutaneous fat), cholesterol reduction, bariatric surgery (to reduce risk of small intestinal bacterial overgrowth) \(^2\)\(^-\)\(^3\)

\(^1\) Karisson et al. (2013) *Nature* 498, 99–103
\(^2\) Kobyliak et al. (2016) *Nutrition & Metabolism* 13:14
Probiotics & Insulin Resistance

- RCT of 17 healthy subjects (14 men, 3 women)
  - Control (n=9) vs. probiotic grp (n=8): LcS (2 x Yakult Light/d) for 4 wks
- Weeks 1-3: habitual dietary intake. Baseline OGTT performed on day 21.
- Week 4: high fat (65% energy), high energy (50% increase in energy intake) diet. OGTT repeated on day 28.

Main findings

- Control group:
  - Insulin sensitivity decreased by 27%
  - OGTT: 10% increase in glucose AUC value (P<0.05)
- Probiotic supplemented group:
  - Glycaemic control preserved and insulin action maintained
  - OGTT: no change

Concluded that LcS supplementation has the potential to prevent high-fat diet-induced insulin resistance in healthy human subjects.

Psychobiotics: A Novel Class of Psychotropic

Timothy G. Dinan, Catherine Stanton, and John F. Cryan

Here, we define a psychobiotic as a live organism that, when ingested in adequate amounts, produces a health benefit in patients suffering from psychiatric illness. As a class of probiotic, these bacteria are capable of producing and delivering neuroactive substances such as gamma-aminobutyric acid and serotonin, which act on the brain-gut axis. Preclinical evaluation in rodents suggests that certain psychobiotics possess antidepressant or anxiolytic activity. Effects may be mediated via the vagus nerve, spinal cord, or neuroendocrine systems. So far, psychobiotics have been most extensively studied in a liaison psychiatric setting in patients with irritable bowel syndrome, where positive benefits have been reported for a number of organisms including *Bifidobacterium infantis*. Evidence is emerging of benefits in alleviating symptoms of depression and in chronic fatigue syndrome. Such benefits may be related to the anti-inflammatory actions of certain psychobiotics and a capacity to reduce hypothalamic-pituitary-adrenal axis activity. Results from large scale placebo-controlled studies are awaited.
Summary

- Both probiotics and prebiotics may have a role in digestive health (and other areas..)
- Evidence can vary for different probiotic strains and prebiotic ingredients, and so specific evidence is important to consider

Dietary intake, drugs, ingredients impacting pH, inflammation, bile salts

Favourable conditions

Supplementing missing microbes

Adding substrate

Probiotics
Therapeutic microbes

Prebiotics
(Personalised) diets
Thank you for listening!
Any questions?

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