MICROBIOLOGY MEETS GYNECOLOGY:

THE APPLICATION OF PROBIOTICS TO WOMEN’S HEALTHCARE

Lisa Hanson, PhD, CNM, FACNM

Associate Professor, Marquette University College of Nursing, Associate Director Midwifery Program

Senior Staff Nurse-Midwife, Aurora Midwifery and Wellness Center
NOTHING TO DISCLOSE
OBJECTIVES

1. Critically analyze the state of the science concerning probiotics used in women’s health, including the benefits and gaps in knowledge.

2. Discuss the clinical uses of probiotics to prevent and treat a variety of women’s health problems.

3. Describe probiotic dietary supplements, including strains, dosages, and routes, in sufficient detail to make effective selection recommendations and prescriptive decisions.
YOGURT

Persian Bible Genesis 18:8
• “Abraham owed his longevity to the daily consumption of fermented milk products”. (Rijkers et al 2010)

Metchnikoff (1907)
• Began the scientific basis of probiotics
• Investigated lactic acid bacterial composition of yogurt as a protective factor for longevity
Microbiology Meets Gynecology: The Application of Probiotics to Women’s Healthcare

Dannon Yogurt may not help you live as long as Soviet Georgians. But it couldn’t hurt.

Bagrat Topagula, age 89.

There are two curious things about the people of Soviet Georgia. A large part of their diet is yogurt. And a large number of them live to be well over 100.

Of course, many factors affect longevity, and we are not saying Dannon Yogurt will help you live longer. But we will say that all-natural Dannon is high in nutrients, low in fat, reasonable in calories, and quite satisfying at lunch or as a snack.

Another thing about Dannon. It contains active yogurt cultures (many pre-mixed or Sevga style brands don’t). They make yogurt one of the easiest foods to digest and have been credited with other healthful benefits.

Which is why we’ve been advising this. If you don’t always eat right, Dannon Yogurt is the right thing to eat.

By the way, Bagrat Topagula thought Dannon was “ispahan kandga.” Which means he loved it.

Dannon Milk Products, 22-11 38th Ave., Long Island City, N.Y. 11101.
YOGURT NOW

Bifidus Regularis®
Bifidobacterium lactis DN-173 010
"Probiotics are live microorganisms, which when administered in adequate amounts confer a health benefit on the host" (FAO/WHO 2001)
PROBIOTIC PRODUCT INFORMATION:

- Most commonly available supplements contain:
  - Lactobacillus and/or
  - Bifidobacterium

  ...parts of normal human "microbiota"

- Not systemically absorbed in healthy individuals

- Commercially available probiotics are comprised of bacteria of human origin

- Active live culture in products is critical issue
  - Refrigeration is often recommended
  - Some products are freeze dried
BACTERIAL SPECIES USED AS PROBIOTICS

- *Lactobacillus* species
  - *L. acidophilus*
  - *L. bulgaricus*
  - *L. casei*
  - *L. crispatus*
  - *L. fermentum*
  - *L. gasseri*
  - *L. jonshonii*
  - *L. lactis*
  - *L. plantarum*
  - *L. reuteri*
  - *L. rhamnosus G.G*

- *Bifidobacterium* species
  - *B. adolescentis*
  - *B. animalis*
  - *B. bifidum*
  - *B. breve*
  - *B. infantis*
  - *B. lactis*
  - *B. longum*

Some genera and species have been omitted from this list.

Williams, 2010
MECHANISM OF ACTION: STIMULATES ACID PRODUCTION

• Produce acids on mucosal surfaces:
  • Lactic
  • Acetic
  • Others

• Effects:
  • Lowers pH
  • Impedes growth of pathogens
  • Some contribute anti-inflammatory properties (e.g., activate short chain fatty acids that enhance metabolic regulatory mechanisms)
MECHANISM OF ACTION: STIMULATES HEALTHY MUCOSA

- Produce numerous substances to maintain mucosal surface:
  - vitamins
  - bacteriocins
  - metabolites
  - enzymes
  - lipopolysaccharides
  - peptidoglycans
  - superantigens
  - biosurfactants

- Functions:
  - improve healthy microflora
  - alter surface tension
  - prevents adherence
  - displace harmful bacteria from the biofilm
MECHANISM OF ACTION: SUPPORTS IMMUNE RESPONSES

• Synergistically stimulate specific infection fighting substances of host immune system:
  • lymphocytes
  • cytokines
  • interleukins
  • IgG antibodies
  • IgA antibodies and secretory IgA
Probiotic Mechanisms of Action

Acidophilus and other probiotic bacteria secrete:
- Antiviral
- Antibacterial and antifungal chemicals.

Probiotics form a physical barrier to hinder invasion of bacteria and yeasts.

Probiotics like acidophilus create an acidic microenvironment which promotes iron and other mineral absorption.

http://theresourcefulmother.ca/2011/04/12/probiotics

[Adams, 2009; Reid, 2009; Karpa, 2003]
**PROBIOTIC RISK**

<table>
<thead>
<tr>
<th>Healthy individuals</th>
<th>Immunocompromised Adults</th>
</tr>
</thead>
<tbody>
<tr>
<td>• “Negligible”: Fewer than 1 per million users</td>
<td>• Best to avoid</td>
</tr>
<tr>
<td>• One review of 143 human trials of over 7,000 people over nearly 40 years found no adverse events</td>
<td>• vulnerable post surgical</td>
</tr>
<tr>
<td></td>
<td>• those undergoing chemotherapy or radiation</td>
</tr>
<tr>
<td></td>
<td>• High mortality in client with acute pancreatitis</td>
</tr>
</tbody>
</table>

(Madsen, 2001)

(Shane-McWhorter, 2010; Besselink, 2008)
ONGOING CONCERNS FOR CLIENTS

Positives for some
• See as CAM
• See as pharmacologic treatment, although misunderstanding
• See as gene therapy & genetic modification, which can also inspire fear

Bacterial DNA can always mutate by transfer mechanisms (e.g., drug resistance)

Non-traditional safety issues
• Mutated bacteria may behave unexpectedly in the host
• Genetically altered microbes may also behave unexpectedly
• Unanticipated interactions within the human host possible
• Novel bacteria could enter the external environment (e.g., wastewater can affect many ecosystems)

Careful informed consent needed

Sharp et al., 2009
GUT IS A KEY AREA

• Few overview facts:
  • Mammals’ gut: one of most densely populated ecosystems on planet
  • Multiple (2,000) microbial species present = \textit{microbiota}
  • Can contain 100 times the genes in the human genome = \textit{human microbiome}
  • GI tract has 400 square meters of surface area
  • Microbes ↑ in concentration as GI transit continues
    • Estimated 50% of feces are bacteria
    • With 1 trillion microbes per 1 gram of stool

Iannitti & Palmieri, 2010
THE HUMAN MICROBIOME

- Trillions of bacteria:
  Only 20% of species identified by laboratory culture techniques
- At least 2/3 of immune response is attributable to the gut:
  Dependent on predominance of healthy bacterial flora

Hart, 2002; Adams, 2009
GI EPITHELIUM COMPONENTS

http://www.youtube.com/watch?v=gnZEge78_78
GI LUMEN TO BLOOD & LYMPHATICS

http://www.youtube.com/watch?v=gnZEge78_78
CFUs are calculated with the following proportion:

\[
\frac{\text{# of colonies} \times \text{dilution of plate}}{\text{volume of culture on plate}} = \frac{\text{X}}{\text{ml}}
\]

The resulting CFU is then presented in scientific notation

\[10^9 = 1.0 \times 10 \times 10 \times 10 \times 10 \times 10 \times 10 \times 10 \times 10 = 1\text{billion}\]
PREBIOTICS DEFINED

- Contribute selectively fermented food ingredients **(not live organisms)**
- Used as food for growth by species of *Lactobacillus* and/or *Bifidobacterium*
- Confer health benefits upon host
- Inhibit growth of pathogenic bacteria
- Enhance acidic intestinal environment

**Examples include:**
- Banana
- Jerusalem artichoke
- Onion
- Garlic

(Gibson, 2004; Roberfroid 2007)
SYNBIOTICS

• Prebiotics and probiotics used in combination
• United Nations Food & Agriculture Organization (FAO) recommends “synbiotic” be used only if:
  • net health benefit is synergistic
  • prebiotic increases the population and/or function of the probiotic with which it is paired

• Examples:
  • Human breastmilk
  • Bifidobacteria and fructo-oligosaccharides (FOS)
  • Lactobacillus rhamnosus GG and inulins
  • Bifidobacteria or Lactobacilli with FOS or inulins or galacto-oligosaccharides (GOS)
YOGURT AND CULTURED MILK PRODUCTS

• Variation in probiotic live culture contents
• Refrigeration and freshness impact CFU
• 4 oz of a live cultured milk product may contain as much as $10^9$ (1 billion) probiotic microorganisms.
THE FEMALE MICROBIOME

GUT COMPOSITION

- Independent of hygiene, organisms that populate the vagina originate in the gut
- Bifidobacterium predominate in healthy gut

VAGINAL FLORA

- Healthy vaginal flora: Lactobacillus predominate
- Some strains produce $\text{H}_2\text{O}_2$
- Maintain acid vaginal pH: optimum=4.5
- Impacted by:
  - Sexual activity
  - Vaginal cleansing practices
  - Pathogens
Overall, you will see a pattern emerge that suggests that probiotics appear to be better at preventing than treating disease.
IMPORTANT CONSIDERATIONS:

- The benefit of one probiotic cannot necessarily be attributed to another
- Note the heterogeneity of the study samples, probiotic interventions, duration of treatment etc.
- Currently insurance does not cover probiotic therapy
JARISCH-HERXHEIMER REACTION OR “DIE OFF”

- When begin probiotics, may initially experience temporary bloating & flatulence that subsides with continued use

- Although controversial:
  - Referred to as a Jarisch-Herxheimer reaction = “die off”
  - Thought to occur when toxins from dying pathogens (viruses, bacteria, parasites, fungi, etc.) overwhelm body’s ability to clear them
  - More common with oral antifungals; less with probiotics
  - Temporary ↓ of dose and/or frequency may reverse symptoms

- Rare mention of “die off” in the probiotic scientific lit
Lisa Hanson, PhD, CNM, FACNM

Microbiology Meets Gynecology: The Application of Probiotics to Women’s Healthcare

<table>
<thead>
<tr>
<th>Author/yr</th>
<th>Study Type</th>
<th>Patients</th>
<th>Strain</th>
<th>Duration</th>
<th>RR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reid (1992)</td>
<td>RCT, placebo controlled</td>
<td>41 Premenopausal post abx tx</td>
<td>Intravaginal: L casei var, rhamnosis GR-1 1.6 X 10⁹</td>
<td>2x weekly for 2 weeks post antibx tx</td>
<td>No sig diff in UTI recurrence 0.45 (0.14-0.125) P=0.26 29% (probiotic) vs 41%</td>
</tr>
<tr>
<td>Bruce (1992)</td>
<td>Quasi-experiment</td>
<td>10 Pre/postmenopausal Post abx treatment (&gt;4UTI/yr)</td>
<td>Intravaginal: L casei var, rhamnosis GR-1, L fermentum B54 1.6 X 10⁹</td>
<td>Weekly for 12-16 weeks</td>
<td>No sig diff in UTI 0.64 (0.32-1.08) P=0.25</td>
</tr>
<tr>
<td>Baerheim (1994)</td>
<td>RTC, Placebo controlled</td>
<td>47 18-50 years ≥3 UT/yr</td>
<td>Intravaginal: L rhamnosis ≥7.5 X 10⁸ CFU</td>
<td>26 weeks</td>
<td>No sig diff in UTI (95% CI, 0.88-1.98)</td>
</tr>
<tr>
<td>Reid (1995)</td>
<td>RCT, single blind</td>
<td>55 Pre-menopausal (&gt;4UTI/yr)</td>
<td>Intravaginal: L casei var, rhamnosis GR-1; L fermentum B54 1.6 X 10⁹</td>
<td>Weekly for 12 mo</td>
<td>RR=0.42 (0.22-0.67) P1=0.001 RR=0.62 P2= &lt;0.05</td>
</tr>
<tr>
<td>Kontokiari (2001)</td>
<td>RTC, open-controlled</td>
<td>150 Pre-menopausal</td>
<td>Oral: Lactobacillus GG 4 X 10¹⁰ CFU/100 ml</td>
<td>5 d/week for 1 year</td>
<td>No sig diff in UTI 1.05 (0.63-1.76) P=0.5</td>
</tr>
<tr>
<td>Uehara (2006)</td>
<td>Quasi-experiment</td>
<td>9 Young women 2 UTI/12 mo</td>
<td>Intravaginal suppository L crispatus GA 98322 1X10⁹</td>
<td>Every 2 days for 1 year</td>
<td>No sig diff in UTI 1.28 (0.78-2.11) P=0.45</td>
</tr>
<tr>
<td>Beereport (2012)</td>
<td>RCT</td>
<td>252 postmenopausal women with recurrent UTI</td>
<td>Trimethoprim sulfamethoxazole 480 mg, daily Or oral 10⁹ L rhamnosis GR-1 and L reuteri RC-14 bid</td>
<td>12 mo</td>
<td>Probiotics ↓ recurrent UTI by 0.4/year (95% CI -0.4-1.5) No ↑ in antibiotic resistance</td>
</tr>
</tbody>
</table>

Certain probiotics appear to significantly ↓ UTI reoccurrence. More research needed.
PROBIOTIC BACTERIA AND VAGINAL FLORA

Childbearing age women

- *Lactobacillus plantarum* (LB931) Impregnated onto panty liners
- Double blind placebo controlled RCT with 191 women
- The number of *Lactobacillus* was significantly related to vaginal pH (p<0.001)
- 70% of women were carriers of specific *Lactobacillus* strains

(Ronnqvist, 2006)

Conclusion: Women with more *Lactobacillus* had:
less GBS (p=0.36) and lower pH
MENOPAUSE

- HRT: ↑ vaginal Lactobacillus
- ↑ Lacto associated with ↓ UTI and ↓ BV

- Oral _L. rhamnosus_ GR-1 has been shown to improve vaginal flora in postmenopausal women
- Gram Stain (Nugent score)
- Some studies of intravaginal probiotic administration route raise concerns about participant satisfaction (increased discharge and/or irritation)

(Petrusevic et al, 2008)
BACTERIAL VAGINOSIS TREATMENT (BV)

Cochrane Review: Probiotics for the treatment of BV (Senok, 2009)

- 4 RCTs meet inclusion and quality criteria
- Various preparations and administration routes
- Women allocated to the probiotic groups had significantly improved BV cures
  - Oral metronidazole/probiotic (OR 0.09 (95% CI 0.03 to 0.26)
  - Probiotic/estriol (OR 0.02 (95% CI 0.00 to 0.47)

Conclusion: Insufficient evidence for or against probiotics for the treatment of BV
BACTERIAL VAGINOSIS TREATMENT (BV)

Systematic Review (Abad & Safdar, 2009)
- 9 studies included
- 6 studies demonstrated a significant reduction in BV (by Amstel’s clinical criteria or Nugent’s Gram Stain score)

More recently, RCT (Ling, 2013)
- 60 Healthy women with BV
- Randomized to 10 days of L. delbrueckii subsp. lactis DM8909\textsuperscript{109} intravaginal suppositories OR 7 days of metronidazole
- Probiotics successfully treated the BV in comparison
- 10 days of probiotics as effective (88% cure) as 7 days Flagyl (83.3%)
- Metronidazole decreased the diversity of vaginal flora
- Probiotic maintained diversity of the flora

Probiotics appear to be an effective alternative treatment of BV with the added benefit of promoting diverse healthy vaginal flora
BACTERIAL VAGINOSIS RECURRENT PREVENTION

Findings: RCT (Ya, 2010)

- 120 Healthy Chinese women with recurrent BV
- Randomly assigned to 7 days on/7 days off for 60 days
  - Daily intravaginal capsule containing L. rhamnosus, L. acidophilus, & Streptococcus thermophilus
  - OR placebo
- Findings
  - Probiotic prophylaxis resulted in lower BV recurrence (15.8%) vs controls (45%) P<0.001
  - Lower recurrence sustained at 2 & 11 month follow-ups

Probiotics appear to be effective in the management of recurrent BV, although more research is needed
# STUDIES OF PROBIOTICS AND VAGINAL CANDIDIAS (VVC)

<table>
<thead>
<tr>
<th>Author/yr</th>
<th>Type of Study</th>
<th>Participants</th>
<th>Strain</th>
<th>Length of treatment</th>
<th>RR (95%-ile)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hilton (1992)</td>
<td>Prospective Crossover</td>
<td>33 Pre-menopausal H/O Chronic VVC</td>
<td>Oral: Lactobacillus yogurt 10^8</td>
<td>Daily x 6 mo</td>
<td>&lt;0.009</td>
<td>0.39 (0.17-0.7)</td>
</tr>
<tr>
<td>Pirotta (2004)</td>
<td>RCT, Placebo controlled</td>
<td>235 Health finished ab for gyn infection</td>
<td>Oral: L. hamnosus, Pessary: L. hamnosus, L. dekbrueckii, L. acidophilus, L. debrueckii Strep thermophilus</td>
<td>Daily x 4 days</td>
<td>0.8</td>
<td>0.20 (0.03-1.18)</td>
</tr>
<tr>
<td>Hilton (1995)</td>
<td>Quasi-experiment</td>
<td>28 h/o recurrent VVC &gt;5x/yr</td>
<td>Vaginal suppository: L. hamnosus GG 10^9</td>
<td>Twice daily x 7 days</td>
<td>0.19</td>
<td>0.20 (0.03-1.18)</td>
</tr>
<tr>
<td>Williams (2001)</td>
<td>RCT, Placebo controlled</td>
<td>164 HIV + Stratified by CD4 counts</td>
<td>Vaginal suppository: L. acidophilus</td>
<td>Weekly x 19 mo</td>
<td>0.14</td>
<td>0.54 (0.26-1.10)</td>
</tr>
</tbody>
</table>

Table from Abad & Safdar, 2009
VULVO VAGINAL CANDIDIA (VVC)

Prevention
- 4 studies
- Small samples
- Nonrandomized
- Goals varied
  - prevention vs treatment
- Include women with diverse healthcare needs
  - HIV positive
  - Menopause
- Show some efficacy in preventing recurrences

Treatment
- Only study demonstrated effective treat of VVC
- Utility of probiotics against VVC depends on adherence to vaginal epithelium
- Hormones, vaginal pH, glycogen content can impact adherence
- In vitro, Wood & colleagues (1985) found that yogurt-based sources of *Lactobacillus* had lower adherence to vaginal epithelium than other exogenous probiotic sources

*Lactobacillus* has potential to play a role in preventing VVC. More randomized controlled studies needed with larger samples & goal of prevention versus treatment.
Allergies

Perinatal programming
- Prenatal probiotics significantly reduce atopic disease in offspring
- Opportunity to make an impact diminishes with age

- 12 RCTs of probiotics against allergic rhinitis
- Various probiotics were used
- Symptoms were measured by self-report
- Probiotic groups
  - Lower reported symptoms
  - Less medication use

Ozdemir, 2012, Bjorksten, 2005

Probiotics appear to be effective in the management of allergic rhinitis
ECZEMA

• 3 Meta-analyses of probiotics as a treatment for eczema
  • No consistent evidence of benefit

• More recently, RCT of L salivarius LS01 in adults
  • Some improved quality of life measures following 16 weeks of treatment when compared to placebo
    (Drago et al, Int J Immunopathol Pharmol 2011; 24: 1037)
COMMON COLD SYMPTOMS

- 3 RCTs of probiotics against common cold
  - L. gasseri, B. Longum, B. Bifidum (2 studies)
  - L. casei (one study)
- 2 used vitamins/minerals in addition
- Spring/winter administration
- Probiotics significantly reduced the duration and severity of common cold symptoms
- No effect on incidence

(Lenoir-Wijnkoop et al., 2007)

Probiotics may reduce the duration and severity of common cold symptoms
ACUTE URI PREVENTION

• Recent RCT to explore prevention of the common cold (Berggren et al., 2011):
  • 272 adults (2/3 women)
  • 10°L. plantarum & L. paracasei
  • ↓ symptom duration: from 8.6 to 6.2 days (p<0.05)
  • ↓ incidence of ARIs: from 67% to 55% (p<0.05)
  • Probiotics reduced the risk of acquiring common cold

• Hao et al. (2011) Cochrane systematic review:
  • 10 studies, 3,451 participants
  • Various probiotics
  • Probiotics reduced
    • incidence of acute URI
    • antibiotics used

Fortney, 2013; *; Hao et al., 2011

Probiotics may prevent the common cold
ANTIBIOTIC ASSOCIATED DIARRHEA (AAD)

- AAD affects up to 30% of antibiotic users
- 3 meta-analyses of probiotics against AAD including up to 20 clinical trials each
- Probiotics significantly reduced the incidence of AAD by 42-66%
- No increase in adverse events
- Most studied strains (most included *Lactobacillus*)
  - *Saccharomyces boulardii* (yeast-based probiotic)
  - *Lactobacillus rhamnosus*, GG
  - Or a combination of the two

(Johnston et al 2012; Hempel et al 2012; Weichselbaum, 2010, Up to Date, 2014)

Probiotics appear to be effective in preventing antibiotic associated diarrhea
ANTIBIOTIC ASSOCIATED DIARRHEA: C. DIFFICILE (CDAD)

- C. Difficile is a serious form of antibiotic associated diarrhea also associated with fecal oral contamination via the hands of health workers and patients
  - Most at risk: elderly, immune suppressed, antibiotic exposed
  - Increase in CDAD infection worldwide due to hyper virulent strain
- Meta-analysis of 6 studies of probiotics used to prevent CDAD demonstrated a significant role in prevention (RR=0.59; 95% CI, 0.41, 0.85; P=0.005) (Avadhani 2011; Weichselbaum, 2010; Friedman, 2012, Ulbricht, et al 2011)

Prevention includes taking probiotics during the antibiotic course and then for 14 days afterwards
PROBIOTIC ADMINISTRATION DURING ANTIBIOTIC THERAPY

- Antibiotics can kill probiotic bacteria
- Negates the effect of taking probiotics

Separate antibiotic & probiotic doses by 2 hours
CELIAC DISEASE (CD)

• Probiotics show promise in management of autoimmune disorders such as CD
  • fermentation-derived metabolites
  • regulation of the epithelial cell barrier
  • modulation of the immune response
  • Inhibition of inflammatory markers of CD

• Example: *Bifidobacterium lactis* was shown to inhibit the toxicity produced by gliadin in epithelial cells (the gluten protein)

• Although this has not been studied in persons with CD

(Lindfors, 2008)

The impact of probiotics on CD have yet to be studied
TRAVELERS DIARRHEA (TD)

### Background
- Defined as: passing 3 or more watery stools per day, during or following travel
  - **Acute**: 3 or more loose stools within 24 hours (Most last 3-5 days)
  - **Dysentery**: with visible blood or mucus
  - **Persistent**: lasting 14 days or longer (5-10%)
- Cause: Ingestion of food or water contaminated with feces

### Prevention
- **Meta-analysis of 12 studies**
- Probiotics significantly prevented travelers diarrhea (RR=0.85; 95% CI: 0.79, 0.91; P<0.001)
- Significant efficacy
  - **Lactobacillus & B bifidum**
- **No serious adverse reactions reported**
- Prebiotic galacto-oligo-saccharide 5.5g [x 1 per day, 1 week prior to & during travel to a low risk area]
- Reduced TD incidence & duration

(Takahashi et al 2007; Bennett, 2012)

Probiotics appear to be effective in preventing travelers diarrhea
ACUTE INFECTIOUS DIARRHEA

- Meta-analysis (2010)
  - 63 RCTs
  - Different probiotics
  - Adults and children
  - Probiotic intervention resulted in:
    - 59% reduction in the risk of diarrhea lasting for 4 or more days (RR 0.41, 95% CI 0.32-0.53)
    - 25 hour average reduction in the duration of diarrhea (95% CI, 16-34 hr)

CHRONIC DIARRHEA

- Diarrhea of more than 4 weeks duration
- Related to:
  - Dysregulation of the intestinal homeostasis
  - Composition of gut flora
  - Chronic bacterial or parasitic infections
    - IBS, Celiac Disease (CD), small intestine bacterial overgrowth (SIBO), lactose intolerance
    - Other functional disorders
  - Probiotics of single or multiple species can modify the microbiota and result in clinically appreciable benefit
  - Studies focus on specific disease entities

(Scaldaferrri 2012)

Probiotics appear to be effective in the management of chronic diarrhea
Waizberg et al (2013) randomized 100 constipated Brazilian women (ages 18-75) to 2 daily doses (6g) of synbiotic or placebo for 30 days. Symptons were measured by self evaluation through daily records & standardized scoring tools. Synbiotic group women (beginning the 2nd & 3rd weeks) had significantly:

- increased BM frequency
- improved consistency
- no increase in abdominal symptoms

Koebnick et al 2003
- 70 adults (women & men) with chronic constipation
- Randomized to
  - 4 weeks of L casei OR placebo beverage
  - Improvement noted in second week of treatment
- Probiotic group showed:
  - No difference in bloating or gas
  - Significant improvement in stool consistency & symptom frequency ($p<0.001$)
  - Significant overall improvement in symptoms ($p=0.003$)

Probiotics appear to be effective in relieving constipation. More study is need about various strains.
Higashikawa et al (2010) studied effects of 3 types of Lactic Acid Bacteria (LAB) yogurts on GI symptoms, cholesterol and liver function.

- 68 healthy Japanese adults with either constipation or diarrhea were randomized to receive one of 3 (LAB) yogurts DB.
- Participants consumed 100g daily x 6 weeks
- Results
  - 2 yogurts resulted in dramatic improvements in BM frequency & consistency
  - 2 other yogurts decreased LDL cholesterol
  - One yogurt also improved liver function parameters

Marteau (2002) RCTS aimed to study transit time in women with "occasional irregularity"

- 36 Healthy women
  - consumed 125 g cups per day of probiotic milk product or placebo
  - 4 consecutive 10 day periods with washouts
  - Probiotic significantly shortened transit time of feces

*Japanese Dairy industry funding

Conclusion*: L Plantarum SN13T yogurt improved constipation, serum lipids & liver function
IRRITABLE BOWEL SYNDROME (IBS)

- Clinical diagnosis
  - IBS with constipation
  - IBS with diarrhea
  - Mixed
- Symptoms
  - Abdominal pain
  - Straining
  - Myalgia
  - Urgency
  - Bloating

- 2 meta-analyses of placebo controlled trials:
  - Nikfar, 2008 (8 trials)
  - McFarland, 2008 (20 trials)
- Probiotic use
  (Bidobacterium infantis 35624) was associated with modest improvement in IBS symptoms

(Trinkley et al, 2011; Ulbricht et al, 2011)

Probiotics appear to be effective in reducing symptoms of IBS
INTESTINAL GAS

- 61 otherwise healthy adults (1/2 females) with post prandial intestinal gas-related symptoms
- Randomized to either:
  - Bacillus coagulans GBI-30, 6086
  - Control
- Results:
  - Probiotic group participants had significant improvements in abdominal pain & total symptom score

(Kalman et al, 2009)

Probiotics may improve post prandial intestinal gas-related symptoms
INFLAMMATORY BOWEL DISEASE (IBD)

**Crohn’s disease**
- One systematic review Rolfe (2008), 7 trials
- One meta-analysis Rahimi (2008), 8 trials

**Ulcerative Colitis**
- Systematic Review Khimara, (2011), 4 trials
- Probiotics did not improve remission over mesalazine therapy

No evidence that probiotics prevent relapse

(Ulbricht et al, 2011)

Probiotics do not appear to be effective in Inflammatory Bowel Disease
LACTOSE INTOLERANCE

- GI (or other) symptoms after digesting lactose-containing foods
  - 15% Caucasians
  - >50% Mexican Americans
  - >80% African Americans
- Risk for calcium and protein deficiencies especially in developing countries
- Testing-Hydrogen breath test
  - 25-50 µg lactose challenge.
  - Intestinal bacteria metabolize carbs to generate hydrogen which is rapidly absorbed into blood
  - 20ppm or more = positive test
- Digest fermented milk products more easily
- Systematic Review 1966-2002
  - 10 studies met inclusion criteria
  - Overall, probiotics did not alleviate symptoms of lactose intolerance
    - Different probiotics and strains
    - Not all participants diagnosed with breath test
  - Some individual strains and concentrations may be effective
    - L bulgaricus
    - L acidophilus

Some probiotic stains & species may improve symptoms of lactose intolerance in individuals

Levri et al, 2005
DIABETES AND GLUCOSE METABOLISM

Dysbiosis may negatively impact production of gut hormones & substances that impact obesity & glucose metabolism

- ↓Glucagon-like peptide-1 (GLP-1) that promotes satiety and lower glucose levels
- ↓Insulin signaling
- ↓glucose transport
- ↑Inflammation
- ↑Triglyceride production
- ↑gut permeability
- ↑oxidative stress

Probiotics may prevent or reverse these

Probiotics and DM: Emerging science

- Probiotic increase insulin sensitivity in Type II DM (Andreason, 2010)
- Prenatal Probiotics reduced the incidence of gestational diabetes (Luoto, 2010)
- May be protective against macrosomia in infants of GDM mothers (NS trend found in Meta-analysis, Dugoua, 2009).

Shane-McWhorter, 2012
• Ejtahe d et al (2012)
  • 64 participants (30-60 years) with Type II DM were randomized to consume 300 grams of regular or enhanced yogurt for 6 weeks:
    • Experimental group:
      • *Lactobacillus acidophilus* La5 α & *Bifidobacterium lactis* Bb12
      • 15 Billion CFU total daily
    • Control group
      • Conventional yogurt FBS, 24 hour diet recall and anthropometric measures were collected at baseline and 6 weeks
  • Probiotic yogurt significantly ↓ FBS, HgA1-C & other measures, including total antioxidant status

**Probiotic may improve Type II DM measures**
• Double blind placebo-controlled trial
  • 48 male, 24 each group
  • 77 females, 38 probiotic
• Each group receive moderate calorie restriction for 12 weeks followed by 12 weeks of maintenance
• Body weight and composition measured at baseline, 12 and 24 weeks
  • Intervention: 2 capsules per day
    • L rhamnossis CGMCC1 3724 (LRP) 1.6 X 10^8 CFU with Oligofructose and inulin (prebiotics)
  vs
• Placebo

• Gender-based analysis was significant
• Among women the average weight loss was significantly higher in synbiotic vs controls
  • 4.4 kg at 12 weeks (p=0.02)
  • 5.2 kg at 24 weeks (p<0.05)
• Women in the synbiotic group continued to lose body weight and fat mass in the maintenance period

Sanchez et al 2014

Synbiotic supplementation may help women achieve weight loss goals
**ABDOMINAL ADIPOSE**

- Muti-center, RCT, double blind, placebo controlled trial
  - Japanese adults
  - 3 groups
  - 200 g *L. gasseri* SBT2055
    - $10^6$ CFU
    - $10^7$ CFU
  - Control

Abdominal viseral fat computed by tomography at 12 weeks

- Abdominal adipose significantly decreased in the probiotics groups by:
  - -8.5% in the $10^7$ group (95%, CI-11.9, -5.1; (p<0.01)
  - -8.2% in the $10^6$ group (95%, CI-10.8, -5.7; (p<0.01)

- BMI, waist and hip circumference were also significantly decreased in both groups compared with controls

- Cessation attenuated these effects

Kadooka, et al 2012

Probiotic supplementation appeared to lower abdominal adipose. Continued supplementation is needed to sustain the effect
FERMENTED MILK & BLOOD PRESSURE

- Meta-analysis of 14 placebo controlled RCTs
  - Studies done between 1996-2010
  - Included 702 participants: gender not specified

- Intervention
  - Fermented milk 100-450g/day
  - Controls received milk-based placebo product
  - 4-24 weeks duration (average 8 weeks)

- Findings: Fermented milk intervention significantly reduced blood pressure compared to control milk product consumption
  - Systolic \( \downarrow \) by 3.1 mm Hg (95% CI -4.64, -1.56)
  - Diastolic \( \downarrow \) by 1.09 mm Hg (-3.98-2.09)

Dong et al, 2013

Probiotic may improve blood pressure measures
In a double blind randomized cross over study,

- Randomized 32 healthy, moderately hypercholesterolemic women & men to 4 weeks of:
  - Probiotic drink containing $10^9 \text{L} \text{paracasei (LPC37)}$ AND
    - Bread enriched with calcium
    OR
    - Bread without calcium
  - After a 2 week placebo period, the groups were switched

Results: Probiotic + calcium significantly:

- decreased total cholesterol & LDL cholesterol
- Increased fecal $\text{L} \text{paracasei}$ & other $\text{Lactobacillus}$
- Increased bile acids in feces

Trautvetter et al, 2012

Probiotic may lower lipid parameters
### SPECULATION: ATOPY & COLON CA RISK

<table>
<thead>
<tr>
<th>Healthy individuals (n=15)</th>
<th>Allergic individuals (n=15)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt; Lacto &amp; Bifido</td>
<td>&gt; Clostridium &amp; Bacteroides [strict anaerobes]</td>
</tr>
<tr>
<td>↓ activity &amp; occurrence of CA-promoting enzymes &amp; metabolites</td>
<td>produce↑ metabolites &amp; problematic enzymes for a more genotoxic environment in gut lumen</td>
</tr>
<tr>
<td>Low levels: β-glucuronidase, azoreductase, &amp; nitroreductase</td>
<td>High level of these toxic enzymes</td>
</tr>
<tr>
<td>Lower colon pH generally</td>
<td>Less acidic colon pH may ↑ toxins</td>
</tr>
<tr>
<td>NS difference in pH (6.7-6.8)</td>
<td>pH significantly ↓ (7-6.6; P=.037)</td>
</tr>
</tbody>
</table>

Lisa Hanson, PhD, CNM, FACNM

Microbiology Meets Gynecology: The Application of Probiotics to Women’s Healthcare

**HPV VIRAL CLEARANCE**

**Exploratory pilot**
- Prospective controlled study
  - 54 women with LGSIL
  - Followed for 6 months
- 24 women received Intervention = Daily probiotic drink
  - Lactobacillus casi Shirota LeS (CFU not provided)
  - Commercially available in Belgium
- 27 controls

**Findings**
- Women in the probiotic group showed twice the HPV clearance of HPV infection related cytology compared to controls
  - 60 vs 31% (p=0.05)


In the future probiotic may provide a new option to manage HPV related cytology abnormalities
### SUMMARY OF EVIDENCE

<table>
<thead>
<tr>
<th>Strong evidence</th>
<th>Substantial evidence</th>
<th>Applications showing promise</th>
<th>Potential future applications</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute Diarrhea</td>
<td>BV Treatment</td>
<td>Prevention of C-Diff</td>
<td>Rheumatoid arthritis</td>
</tr>
<tr>
<td>Antibiotic</td>
<td>BV Prevention</td>
<td>Inflammatory bowel disease</td>
<td>Cancer (prevention)</td>
</tr>
<tr>
<td>Associated</td>
<td>UTI prevention</td>
<td>Diabetes</td>
<td>STD prevention</td>
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<tr>
<td>Diarrhea</td>
<td>Allergic Reactions</td>
<td>Blood pressure</td>
<td>HPV related cytology</td>
</tr>
<tr>
<td>Constipation</td>
<td>Atopic dermatitis</td>
<td></td>
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<tr>
<td></td>
<td>Irritable bowel</td>
<td></td>
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<tr>
<td></td>
<td>syndrome</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>Traveler’s diarrhea</td>
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</tr>
</tbody>
</table>

*Goldin & Gorbach, 2008*
SOCIOECONOMIC BENEFITS

- Probiotics themselves are relatively inexpensive, even with daily dosing.
- Associated with (but more research needed):
  - ↓ absences from work (&/or day care)
  - ↓ incidence of infections & recurrences
  - ↓ duration & severity of symptoms reported
  - ↓ length of stay if hospitalization needed
  - ↓ costs of antibiotics & other treatments
  - ↓ costs of provider visits through prevention
  - ↓ cost of alternative caregivers needed

Lenoir-Wijnkoop et al., 2007)
RESOURCES FOR CLINICIANS:

Natural Medicine DataBase
- Website contains a table that compares commercially available products including yogurts.

YouTube animation
- [Immunology in the Gut Mucosa](http://www.youtube.com/watch?v=gnZEge78_78)
- [http://www.youtube.com/watch?v=gnZEge78_78](http://www.youtube.com/watch?v=gnZEge78_78)
CONTACT INFORMATION

Marquette University
College of Nursing
Nurse-Midwifery Program
Milwaukee, WI 53201-1881

• lisa.hanson@mu.edu 414-288-3841