Gut Microbiota and Health:
Food Fad or Bacterial Therapy?

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Medical Director Hospital Nutrition Support
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Have We Now Moved From Disease to Dessert?

VRE growing on mitral valve  

Frozen yogurt *L. acidophilus*
Probiotics in Clinical Medicine: Two Schools of Thought!

Definition: “Microorganisms normally present in the human body that when delivered in adequate amounts, confer a health benefit on the host”

Quakery
- Claims to cure “everything”
- Why so many strains?
- How much is needed?
- Review articles vs original articles
- Inconsistent studies
- Few ITT studies with tangible outcomes
- Marginal statistics
- Growing suspicion of “holoistic” medicine

Therapy
- Differential support based on cultural and educational background (yogurt, kurd, kefir, kombucha)
- Aware of the differences in strains etc
- Understand the differences in study design
Probiotic literature ?
Science or Quackery

• Professional Literature
  • Few ITT studies available
  • Widely variable, heterogeneous groups
  • Meta-analysis not consistent
    – Questions of outcome
  • 2009 analysis of what is published probiotics vs antibiotics in PubMed Search
    – Probiotics > 5000 papers  ----- 28% were review articles
    – Antibiotics > 500,000 papers----8% review articles

• Lay literature
  • Recent lead articles NY Times 2012
  • Wall Street Journal  2012
  • Economist 2012
Some say we should be killing our bacterial!!
Gastroenterologist Survey of Probiotics

- Evaluate MD opinions regarding probiotics
- Large metropolitan area in midwest
- Results:
  - Safe for most patients 100%
  - 98% felt probiotics had a role in treating GI disease
  - 93% had patients currently taking probiotics
  - Most common bacteria used
    - Yogurt based, B.infantis (Align®), VSL#3,
  - Most common clinical diagnosis used
    - IBS, AAD, C.difficile
  - Most believed their practice was not supported by scientific data

Williams MD J Clin Gastro 2010
Nutritional Goals in Clinical Medicine Have Changed From Adjunctive Care to a “Therapeutic” Strategy

- **Previous goals**
  - Attempt to preserve lean body mass
  - Avoid metabolic complications

- **Current Goals: “Therapy not support”**
  - Attenuate metabolic response
  - Reverse loss of lean body tissue
  - Prevent oxidant stress
  - Favorably modulate immune response
    - Enteral feeding (GALT)
    - Appropriate macro and micronutrients
      - Glutamine, arginine, omega-3-FA, antioxidants
  - **Maintaining “normal “ commensal flora**
  - **Manipulating flora to host benefit**
    - SCFA, anti-inflammatory changes, decrease sepsis
Where “man meets microbe” a dynamic interplay

- Concepts are not new
  - Biblical references,
- 300 to 400 sq meter surface area
  - Surface area of a tennis court
- > 2 million genes in the bacterial genome vs 35,000 in the human
  - 100 trillion living bacteria in the human intestine
  - Over 500 species in human colon
  - Each individual with own bacterial fingerprint
- Significant “cross-talk” between bacteria and host
  - One bacteria species can turn on > 100 genes
  - Toll receptors on dendritic cells / macrophages
  - Gut contains complex neuroendocrine system
- Quorum sensing
  - Molecules secreted by bacteria: they partially explain bacterial community behavior and activation of virulence genes etc
Does the Mucosal Surface Environment Alter Function or Clinical Outcomes?

- Inflammatory changes
- Bacterial interrelationships
- Bacterial changes with host stress situations
  - Bacterial use environmental clues
    - pH, temperature, redox potential, osmolality
  - When energy supply is limited genes “switch on” virulence factors
  - Ex: E.coli can rapidly become virulent with host stress (epinephrine, cortisol etc)

Probiotics can *prevent*, *mitigate* and *treat* many of the current health crisis facing the western world

- **Cancer**
  - Multiple mechanisms
- **Heart disease**
  - Metabolic syndrome
  - Atherosclerosis
- **Depression**
- **Hepatic diseases**
  - NASH
- **Infectious disease**
- **Diarrheal diseases**
  - AAD
  - Bacterial
  - C. diff
  - Viral
- **Inflammatory diseases**
  - IBD
  - Allergy
  - Asthma
- **Autoimmune diseases**
- **Aging**
- **Obesity**
- **Critical Care / Surgery**
  - Trauma
  - Pancreatitis +/-
  - Transplantation
  - Sepsis
  - VAP prevention
  - C. difficile
Potential applications for probiotics

- **Metabolism**
- **Metabolism of dietary compounds**
  - in the gut lumen:
    - Lactose digestion
    - Lipid metabolism
    - Oxalate metabolism
- **Composition and metabolic markers of the gut microbiota**
  - Xenobiotics, phytochemicals
  - Indigestible dietary components
- **Metabolic activity of gastrointestinal mucosa and liver**
  - IBD and IBS
  - Inflammatory bowel diseases:
    - Crohn's disease
    - Ulcerative colitis
    - Pouchitis
    - IBS
- **Allergic diseases**
  - Eczema, atopic eczema
  - Allergic rhinitis
  - Asthma
- **Reduction of risk factors of infection**
  - Infectious diarrhea (acute and antibiotic-associated)
  - Traveler's diarrhea
  - Necrotozing enterocolitis (infants)
- **Helicobacter pylori**
- **Respiratory tract infections (adults and children)**
  - Ear, nose, and throat infections
- **Infectious complications in surgical critically ill patients**
Probiotics: Levels of Action

- Lumen
- Mucosal
- Submucosal
- Systemic
Colonization resistance
- Competitive exclusion

Maintain barrier function
- Reduce macromolecular permeability and bacterial translocation
- Maintain tight junctions (ZO-1, claudin1)

Metabolic effects
- Bacteriocins
- Decrease pH
- Quorum sensing

Modulation of signal transduction
- INFγ
- MAPK

Probiotics
- Enhance microbial flora
- Innate/Adaptive Immunomodulation
  - IgA, IgG, IgM
  - Increase mucin production
  - Enhance cytokines (IL-10, TGFβ)

Sherman NCP 2009, Wallace TC 2011
Mechanisms:

Colonization Resitence
Antimicrobial Factors

Mechanisms:
- Competitive inhibition
- Physical barrier (mucous)
- ↓ Adherence, attachment
- Produce bacteriocins
  Defensins, Trefoil
  Bind pathogens
- ↓ pH reduces growth
- Interfers quorum sensing
  ↓ Virulence expression
- Breaks up biofilms

Bacteria
- *Escherichia coli* (pathogenic)
- *Salmonella typhimurium*
- *Shigella* spp.
- *Campylobacter jejuni*
- *Streptococcus mutans*
- *Bacillus subtilis*
- *Clostridium perfringens*
- *Helicobacter pylori*
- *Staphylococcus aureus*
- *Listeria monocytogenes*
- *Pseudomonas fluorescens*

Fungi
- *Candida albicans*
- *Aspergillus flavus*
Schematic representation of endocrine cell-mediated signaling from enteric microbiota to host.
Protecting the mucosal lining:
“Soluble factors for Lactobacillus rhamnosus GG activate MAPKs and induce cytoprotective heat shock proteins in intestinal epithelial cells”

- Cell culture model
- DNA microarray methods, real-time PCR and electrophoretic mobility shifts studied

Studies confirm:
- L. GG modulates signaling pathways
- Activates via MAP kinase
- L.GG protects mucosa from oxidant stress via expressing HSP

Tao K, Drabik K, Waypa T
Am J Physiol Cell Physiol 290;1018-1030, 2006
Mechanisms:
Enhancing mucosal blood flow

Mechanisms: stimulation the immune system in the small intestine of healthy subjects

Before Reuteri intake

Resting CD4+ T-helper cells

After Reuteri intake

Activated CD4+ T-helper cells

SCFA = Fermentation end product of some probiotics (from prebiotics)

- Energy source;
  - colonic mucosa;
    - Stimulates cell proliferation, Promotes sodium and water absorption
  - Cardiac, Skel Mus, Brain
    - Acetate;
  - Propionate; gluceneogenesis
- Regulation of gene expression for ICAM-1 and E-Selectin on endothelial cells
- Decrease COX-2 expression
  - (butyrate and propionate)
- Prevention of neoplastic transformation
  - Inhibits histone deactylase by DNA hypermethylation to promote differentiation in cancer cell lines
- Enhances Leptin secretion
- Inhibition of pathogen overgrowth in gut lumen
- ROS scavenger
  - Pyruvate is anti-inflammatory and decrease NFKB expression
- Activation of polymorphonuclear cells
  - Both local and systemic
  - G-protein receptors on circulating PMN’s

Thangaraju M et al J GI Surg 2008
SCFAs, Fiber Fermentation and Butyrate Receptors

- Trophic effect, colonocyte fuel
- Anti-inflammatory
- Enhance WBCs, macrophage
- ↓ Adhesion molecules
- (↓ microvasc thrombosis)

Thangaraju M et al J GI Surg 2008
Ganapathy V 2011
Preventing Microvascular Thrombosis:
Regulation of gene expression for ICAM-1 and E-Selectin on endothelial cells

Probiotics (via SCFA) shown to decrease ICAM-1 and E-Selectin expression on endothelial cells
Clinical Use of Probiotics in the ICU

Where does the rubber meet the road?
Clinical Use: Sorting evidence from myths!

- Specific effects can be strain specific!

- **Level 1 evidence in:**
  - Infectious diarrhea (L GG)
  - Prevention of traveller’s diarrhea
  - Prevention of pouchitis after total colectomy for UC
  - Prevention of Ventilator Associated Pneumonia (VAP)
  - Prevention of Necrotizing fasciitis in neonates
  - Prevention of anti-biotic diarrhea

- **Level 2 evidence in:**
  - S.boulardii (with vanc) in preventing recurrent C.difficile
  - Prevention of post op infections in liver transplant
  - Prevention of post op infections in abdominal surgery
Can Probiotics be used for prevention of disease in “Healthy People”

**Sick days at home with short term gastro-intestinal or respiratory illness**

Placebo: 0.9 % sick days
2 days per individual and year

Reuteri: 0.4 % sick days
<1 day per individual and year **

**Number of people sick**

26% on placebo (23 persons)
11% on Reuteri (10 persons) p<.01**

*P < 0.01*

*Placebo*  
*Reuteri*  

*Tubelius P et al., Environ Health 2005*
Probiotics use in healthy nursery school children

- Children (4-10m) with increased risk for infection

12 weeks supplementation in baby formula

*P < 0.05 cf placebo & BB-12

**Weizman et al., Pediatr 115; 5-9 (2005)**
Gestational Diabetes

- Finland N=256 (3 groups)
- Strict definition of Gestational diabetes (GTT)
- Control, placebo, probiotics

Results:
- Control 36%
- Placebo 34%
- Probiotics 13%
- No change in pregnancy outcome
- No change in children at two years

Luoto R British J Nutrition 2010
### Areas of Critical Care Where Probiotics Have Reported Benefit

**Treatment:**
- Trauma
- Pancreatitis +/-
- Transplantation
- Sepsis
- NASH

**Prevention:**
- VAP
- Antibiotic associated diarrhea
  - C. difficile
- VRE colonization
Ventilator Associated Pneumonia

- One of most frequently occurring nosocomial infections in the ICU
- Current strategy not working
  - Antibiotics – increases resistant flora
  - Ventilator adjustments – variable success
  - Prokinetic agents – no influence
  - Medications – no influence
  - Surfactants – no influence
  - Mouth wash – variable
  - Etc etc etc
Oral probiotics and prevention of *P. aeruginosa* infections: a randomized, double-blind, placebo-controlled pilot study in intensive care unit patients

- **Hypothesis:** oral application of probiotics will prevent the secondary colonization with pathogens

- **PRDBPC trial**
  - Inclusion criteria
    - patients in ICU >48 hours
  - 807 eligible: 106 placebo vs 102 probiotic completed
  - $10^9$ L.*casei* BID started day 3 until discharge
  - Monitored gastric and oral bacteria cultures

- **Results:**
  - Delayed colonization of *P. aeruginosa* in respiratory tract

- Forestier C Critical Care 2008,12:1-10
Use of Probiotics to Prevent Ventilator Associated Pneumonia

- **Lactobacillus GG vs placebo (DBPCT)**
  - (2871 patients screened 146 met criteria)
  - On vent > 72 hours
  - Oral *and* via feeding tube
  - $1.0 \times 10^{10}$ BID to each site

- **Evaluated**
  - Oral flora pathogen vs normal flora
  - Gastric flora pathogen vs normal flora
  - Incidence of VAP

- **Results**
  - Less antibiotics used
  - Less C. difficile 5.8% vs 18.6% (*p*<.05)
  - Clinical VAP 35% vs 47% (*p*<.05)
  - Microbiologic VAP 19% vs 40% (*p*<.05)
  - Mortality 14% vs 24% (NS)

*Morrow S, Kollef M et al 2010 AJRCCM*
Not all Probiotics VAP studies positive:

• N = 259 ICU Mechanical ventilation > 72 h
• Probiotics delivered to GI via tube
  • With soluble fiber
• Results:
  • VAP w/ probiotics 9% vs 13 % in control (NS)
  • Mortality 27% in probiotics vs 33% in control (NS)
• Conclusion:
  • No significant improve in VAP or mortality

• (note: probiotics only given enterally, no oral / pharynx delivery)
Impact of administration of probiotics on VAP: Meta-analysis

• RCT with mechanical ventilation
• 5 RCT included
• Results:
  • Probiotics decrease VAP
  • Decrease in Pseudomonas colonization
  • No change in mortality
  • No change in ventilator days

I Siempos et al Crit Care Med 2010
H. pylori infects at least half of the world’s population. The prevalence among middle-aged adults is over 80% in many developing countries, as compared with 20% to 50% in industrialized countries. **WHO** classifies *H. pylori* as class one carcinogen.

*Suerbaum & Michetti NEJM 2002; 347:1175*

*Morowitz MJ Ann Surg 2011; 253:1094-1101*
Specific probiotics have surface proteins that inhibit the binding of \textit{H. pylori} in the stomach.

- \textit{H. pylori} attached to gastric cells
- \textit{L. reuteri} inhibits \textit{H. pylori} binding

\cite{Mukai2002}
# HP Eradication Therapy with and without Probiotics - Meta-analysis

<table>
<thead>
<tr>
<th>Outcomes</th>
<th># Trials / (n)</th>
<th>with (%)</th>
<th>w/o (%)</th>
<th>NNT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eradication Rates</td>
<td>11(1074)</td>
<td>85%</td>
<td>75%</td>
<td>11</td>
</tr>
<tr>
<td>Total Side Effects</td>
<td>7(625)</td>
<td>22%</td>
<td>38%</td>
<td>6</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>8(997)</td>
<td>6.1%</td>
<td>16%</td>
<td>11</td>
</tr>
<tr>
<td>Epigastric Pain</td>
<td>7(608)</td>
<td>16%</td>
<td>23%</td>
<td>14</td>
</tr>
<tr>
<td>Nausea</td>
<td>7(608)</td>
<td>16%</td>
<td>25%</td>
<td>12</td>
</tr>
<tr>
<td>Taste Disturbance</td>
<td>5(418)</td>
<td>14%</td>
<td>25%</td>
<td>5</td>
</tr>
</tbody>
</table>

Tong, A Pharm Therap 2007
Antibiotic Associated Diarrhea: preventable or inevitable?

- Hempel S et al JAMA 2012
- Meta-analysis 82 RCT met criteria for inclusion
- Probiotics strains were poorly documented
- N=11,811 participants (pooled data)
- Conclusion:
  - Probiotics confer significant decrease in AAD (p<.001)
  - # needed to treat N=13

Hempel S et al JAMA May 9, 2012
Rising Incidence of C. difficile

• Incidence of C. difficile by year

Pathogenesis of CDAD

Antibiotic therapy

Alteration in colonic microflora

*C. difficile* exposure and colonization

Release of toxin A and Toxin B

Colonic mucosal injury and inflammation

• Adapted from Kelly CP et al Ann Rev Med 1998;48:375-390
• Badger, VO et al JPEN 2012
Emergence of B1/NAP1 Strain

- Produces 16-23 times C. diff. toxins A and B in vitro, represented 50% of isolated strains between 2001-2003
  - Produces a 3rd binary toxin
- Increased risk of relapse
- Less responsive to standard therapies

Major Genes in the Pathogenicity Locus (PaLoc) of Clostridium difficile and Relation to the Genes for Binary Toxin

- McDonald NEJM 2005

Figure 2. States with the North American Pulsed Field Type 1 (B1/NAP1) strain of C. difficile confirmed by CDC as of November 15, 2005 (N=16).
The changing face of Clostridium difficile!
Use of probiotic Lactobacillus preparation to prevent diarrhoea associated with antibiotics:

- RDBPCT  N=135
- Age 64  all taking antibiotics
- 100 gm BID L. casei as drink
- Results:
  - Diarrhea: 7/57 (12%) vs 19/56 (34%)
  - 21% relative risk reduction, NNT 5
  - C.diff 0/57 vs 9/53 (17%)

Probiotic treatment of VRE: Randomized Controlled Trial.

- PRPCBT 27 VRE positive patients
- Yogurt (containing live Lactobacillus GG vs Pasteurized yogurt)
- 100 gm daily x 4 weeks
- Primary outcome measure: clearance of VRE
- Results:
  - L.GG group: 11/11 cleared VRE at 4 weeks, 3/11 reconverted + at 4 weeks
  - Control: 1/12 cleared
    - Allowed to crossover at 4 weeks 8/11 crossed over
    - 8/8 of the crossover group cleared in 4 weeks

PRPCBT = Prospective Randomized Placebo Control Blinded Trial
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<td>Katsumpasi</td>
<td>2007</td>
<td>N=65 Vent, multiple trauma</td>
<td>Synbiotics</td>
<td>Dec infection, SIRS, Sepsis, mortality</td>
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<td>2007</td>
<td>N=67 Whipple</td>
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<td>Decrease infections</td>
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<td>Alberda</td>
<td>2007</td>
<td>N=28 ICU</td>
<td>Probiotics VSL # 3</td>
<td>Enhance immune func</td>
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<td>Springer-vessel</td>
<td>2007</td>
<td>N=113 Trauma</td>
<td>4 groups, Synbiotics</td>
<td>Decrease infection, perm</td>
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<td>2007 (in press)</td>
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<td>Syn / pre/ TPN</td>
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## Pre and Probiotics in the ICU Setting

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Prebiotics – Probiotics or Synbiotics
Bringing the Science to Practice

Scientific American 2012
Probiotics are found in fermented foods and as additives to many foods.

Prebiotics can be found naturally in foods.

Probiotics + Prebiotics = Synbiotics
Probiotics and Prebiotics

• Probiotics
  – Food sources (most often in dairy products)
  – Capsules, tablets, powder or liquid form
  – Infant formulas

• Prebiotics
  – Occur naturally in food: Honey, wheat, onions, bananas, leeks, garlic
  – added as dietary ingredients: Fructo oligosacharides (FOS), inulin, galacto oligosaccharides, sugar alcohols
  – Enteral formulas containing fiber: Jevity with fiber, Replete with fiber, Specialized formulas (DM, ICU)
Probiotic Beverages

**Chilled dairy**
Yakult
Danactive / Actimel
Stonyfield
BioQ

**Chilled non-dairy**
ProViva
Good belly
Komboucha
Bravo Friscus

**Shelf stable**
Cocobiotic, Dong Quai, Innergy Biotic
Better probiotic delivery systems

- Keep it away from the liquid until ready to use!
  - Micro encapsulation
  - Packaging solutions
    - Bottle closures
    - Drinking straws
## Table 1. Common Probiotic Preparations Available in the United States

<table>
<thead>
<tr>
<th>Trade Name</th>
<th>Active Ingredient</th>
<th>Form</th>
</tr>
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<tbody>
<tr>
<td>Florastor</td>
<td><em>Saccharomyces boulardii</em> 250 mg</td>
<td>Capsules</td>
</tr>
<tr>
<td>Florastor Kids</td>
<td><em>S. boulardii</em> 250 mg</td>
<td>Powder</td>
</tr>
<tr>
<td>Align</td>
<td><em>Bifidobacterium infantis</em> 35264 (1 × 10⁹ CFU)</td>
<td>Capsules</td>
</tr>
<tr>
<td>DanActive</td>
<td><em>Lactobacillus casei</em> DN-114 001</td>
<td>Fermented milk</td>
</tr>
<tr>
<td>Activia</td>
<td><em>Bifidobacterium lactis</em> DN-173 010</td>
<td>Yogurt</td>
</tr>
<tr>
<td>Fem-Dophilus</td>
<td><em>Lactobacillus reuteri</em> RC-14, <em>Lactobacillus rhamnosus</em> GR-1</td>
<td>Capsules</td>
</tr>
<tr>
<td>Culturelle</td>
<td><em>L rhamnosus</em> GG (1 × 10¹⁰ CFU)</td>
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<tr>
<td>Culturelle for Kids</td>
<td><em>L rhamnosus</em> GG (1 × 10⁹ CFU)</td>
<td>Packets</td>
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<td>Sustenex</td>
<td><em>Bacillus coagulans</em> GBI-30, 6086 (BC30)</td>
<td>Capsules, chewies, and gummies</td>
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<td>Floranex</td>
<td><em>Lactobacillus acidophilus</em> (2 × 10⁶ CFU)</td>
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<td>Lactinex</td>
<td><em>L acidophilus</em> and <em>Lactobacillus helveticus</em> (<em>bulgaricus</em>)</td>
<td>Capsules and packets</td>
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<tr>
<td>Phillips Colon Health</td>
<td><em>Lactobacillus gasseri</em>, <em>Bifidobacterium bifidum</em>, and <em>Bifidobacterium longum</em></td>
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CFU, colony-forming units.
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<td>Phillips Colon Health</td>
<td>Lactobacillus gasseri, Bifidobacterium bifidum, and Bifidobacterium longum</td>
<td>Capsules</td>
</tr>
</tbody>
</table>

CFU, colony-forming units.
15.4 Billion US Market in 2008

Gray: number of publications
Black: Number of new probiotic products launched
What’s in a label?

• Marcobal et al tested 14 US commercial probiotic products:
  – 93% incorrectly labeled
  – 57% had contaminants
  – 36% did not list strains on the label

• Masco et al tested 58 different products from EU, UK, Asia, Japan, Canada:
  – Only 38% had the dose stated on the label
  – 29% did not contain strains on the label

Marcobal 2008 J of Pediatr Gastroenterol Nutr
Masco 2005 Int J Food Microbiol
Not all *lactobacilli* survive in the GI tract

12 dairy products off the shelf in UK stores

↓

8 with the “correct” bacteria

↓

35 strains of mainly *Lactobacillus* and *Bifidobacterium* isolated

↓

Stomach (pH, enzymes)

Duodenum (enzymes)

Ileum (bile)

Colon (competition)

It is all about “Risk vs. Benefit”
Probiotic Safety:
Generally Recognized as Safe (GRAS) USA
Qualified Perception of Safety (QPS) EU

• Can probiotic species transfer resistance genes?
• Lactobacillus bacteremia
  • 180 cases in 30 years
  • 69 cases of endocarditis in 30 years
    – (majority of L. rhamnosus)
• Several cases of liver abscess in immunocompromised hosts
• Hepatic Lactobacillus abscess in transplanted liver

• S. Boulardii
  • Recent data showing several outbreaks of S. Cervesiae fungemia when giving S. Boulardii
  • S. boulardii not true probiotic?

• Host risk factors
  • Immunocompromised
    » This is theoretical, clinical data would support use
  • Recent major dental work (theoretical anecdotal reports)

• Caution in severe pancreatitis (Lancet Feb 2008)

Sanders ME Ann NY Acad Science 2011
Probiotics in Pancreatitis: Randomized Prospective Multicenter Trial

- Multicenter RDBPC Trial 298 patients ITT analysis
- APACHE > 8, Imrie >3 or CRP > 150
- Assigned within 72 hours of symptoms
- Control N=145  Multispecies probiotic N=153
- 2 weeks of therapy
- Endpoints: Inf nec, BSI, pneumonia, urosepsis etc
- Results:
  - Infectious complications 30% vs 28%
  - Mortality 16% probiotic vs 6% in control

Probiotics in Pancreatitis?

- Majority of deaths were from bowel necrosis
  - No bacteremia with probiotic species
  - Necrosis patchy, not just at site of probiotic delivery

- What happened?
  - More organ failure in exp group at start (13% vs 4% in control)
  - Large number of bacteria (>10 billion)
  - Location of delivery D3 – D4
  - Bowel dysmotility “ileus”
  - Insoluble and soluble fiber in formula

- ? Localized fermentation, acidosis, necrotic bowel, poor randomization ????
Figure 3. The effect size (risk ratio) for gastrointestinal diseases and for probiotic species. (A) The effect size including the 95% confidence intervals for the total events of Antibiotic-associated diarrhea (AAD), Clostridium difficile disease (CDD), Helicobacter pylori positive (HPP), infectious diarrhea (ID), NEC and Pouchitis.

- Pouchitis: 0.17 (0.10-0.30)
- TD: 0.92 (0.79-1.05)
- NEC: 0.54 (0.23-1.24)
- ID: 0.35 (0.13-0.87)
- IBS: 0.77 (0.65-0.92)
- HPP: 0.70 (0.54-0.91)
- CDD: 0.60 (0.41-0.86)
- AAD: 0.43 (0.32-0.56)

- C. butyricum: 0.18 (0.09-0.37)
- E. faecium: 0.29 (0.13-0.64)
- L. acidophilus, B. infantis: 0.37 (0.17-0.83)
- L. acidophilus: 0.82 (0.47-1.43)
- B. lactis: 0.59 (0.38-0.92)
- LGG: 0.54 (0.39-0.75)
- L. casei: 0.42 (0.24-0.76)
- L. plantarum: 0.82 (0.65-1.04)
- B. infantis: 0.93 (0.78-1.10)
- S. boulardii: 0.46 (0.34-0.60)
- VSL #3: 0.17 (0.09-0.33)
Current Problems with “Probiotic”

- Extravagant claims without research
  - Still perceived as “quackery” by many

- ? of good manufacturing practice
  - Quality assurance
    » Additional species and devoid of label common
  - Label vs content
  - Viability of bacterial species
    » Strain variation, SNP changes ?

- Validate biomarkers for assessing function and activity

- Improve the reliability and ease of taxonomic classification of pre and probiotic
  - Fermentation index
  - FISH (fluorescent in situ hybridization)
  - 16S rRNA
  - Pulse-field gel electrophoresis
  - Amplified fragment-length typing
  - Multilocus sequence typing

- No specific guidelines currently
  - USA far behind EU in regulation
Probiotics: So many questions, so few answers!!!

- Monostrain vs multistrain?
- Pre, pro or synbiotic?
- Will cell free extracts work?
- Quantity and quality of probiotic needed for desired effect?
  - Most studies “doses” range from $10^7$ to $10^{12}$
  - What dose in Peds?
- How best to assess the activity/viability?
- Probiotic safety?
- Which Probiotics remain viable in GI tract?
- When are probiotics contraindicated?
- Resistant patterns?
- Immunocompromised host?
# Probiotic Protocols

## OHSU Protocol for Synbiotic Use in Hospitalized Adult Patients

<table>
<thead>
<tr>
<th><strong>Indications</strong></th>
<th>Patients at risk for developing AAD, CDI (broad spectrum antibiotics, ex: fluoroquinones)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Contraindications</strong></td>
<td>Immunosuppressed patients (ex: BMT) (neutrophil count &lt;500)</td>
</tr>
<tr>
<td><strong>Route &amp; Dosage</strong></td>
<td></td>
</tr>
</tbody>
</table>
| **Oral** | 4 ounces Nancy’s Yogurt or Kefir BID  
1 pack Benefiber QID |
| **Feeding Tube** | 80 ml Nancy’s Kefir + 1 pack Benefiber + 60ml sterile water TID |
# Probiotic Protocols

## OHSU VAP Prevention Protocol for Adults

<table>
<thead>
<tr>
<th>Indications</th>
<th>Ventilated patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Contraindications</td>
<td>Immunosuppressed patients (neutrophil count &lt;500)</td>
</tr>
</tbody>
</table>

### Route & Dosage

<table>
<thead>
<tr>
<th>Route</th>
<th>Dosage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oropharyngeal</td>
<td>Swabbed with Nancy’s Kefir BID (following oral care)</td>
</tr>
<tr>
<td>Feeding Tube</td>
<td>80 ml Nancy’s Kefir + 1 pack Benefiber + 60ml sterile water TID</td>
</tr>
</tbody>
</table>

![Nancy's Kefir Logo](image1.png)

![Nutrition Facts](image2.png)
<table>
<thead>
<tr>
<th>Indications</th>
<th>Patients at risk for developing AAD (broad spectrum antibiotics, ex: fluoroquinolones)</th>
</tr>
</thead>
<tbody>
<tr>
<td>* Critically ill patients will be assessed by RD for appropriateness</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Contraindications</th>
<th>Pancreatitis, Neutropenic precautions, AIDS (T-Cell count &lt;200)</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Route &amp; Dosage</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral Feeding</td>
<td>8 ounces Nancy’s Kefir daily</td>
</tr>
<tr>
<td>Feeding Tube – gastric only</td>
<td>80 ml Nancy’s Plain Yogurt + 200ml water daily</td>
</tr>
<tr>
<td>For patients with dairy intolerance ??</td>
<td>Culturelle LGG 1 pill BID taken 1 hr before or after antibiotics</td>
</tr>
</tbody>
</table>
# Portland VAMC NFS Probiotic Protocol for Hospitalized Patients

## Indications
Patients at risk for developing AAD, CDI (broad spectrum antibiotics, ex: fluoroquinones)

## Contraindications
Neutropenic precautions

## Route & Dosage

<table>
<thead>
<tr>
<th>Route &amp; Dosage</th>
<th>100 ml DanActive® + 60 ml water BID</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral Feeding</td>
<td>100 ml DanActive® BID</td>
</tr>
<tr>
<td>Feeding Tube</td>
<td></td>
</tr>
<tr>
<td>Product</td>
<td>Type of Bacteria</td>
</tr>
<tr>
<td>-----------------</td>
<td>-------------------------------------------------------</td>
</tr>
<tr>
<td>Nancy’s Yogurt</td>
<td><em>L. acidophilus</em>, <em>L. casei</em>, <em>B. bifidum</em>, <em>L. rhamnosus</em>,</td>
</tr>
<tr>
<td>Nancy’s Kefir</td>
<td><em>L. acidophilus</em>, <em>L. casei</em>, <em>B. bifidum</em>, <em>L. rhamnosus</em>, Prebiotic - inulin</td>
</tr>
<tr>
<td>Culturelle LGG</td>
<td><em>L. GG</em></td>
</tr>
<tr>
<td>Danactive</td>
<td><em>L. casei</em></td>
</tr>
</tbody>
</table>
General Guidelines for Use of Probiotics

- Critically evaluate and use only when data supports
  - Base choice on molecular typing, metabolic characteristics and interaction in the environment
  - Caution with meta-analysis, heterogeneity is key
- Do not extrapolate from one strain to another
- Identify optimal strain, insoluble fiber and commercially available product
  - ~Probiotic: $10^{9-11}$ viable cells per day ?
  - ~Prebiotic: 20-30 gm/day ?
- Continued intake of probiotic be required to maintain benefits
- Prebiotic are an excellent option to modify flora on long term basis
  - Persistent levels require continuous intake!
Concepts the clinical team need to understand regarding probiotics!

- **NO** single probiotic meets the need in all patients
  - Effects are often strain specific

- **Consider the disease process:** prevention vs treatment

- **Decision should depend upon:**
  - Metabolic insult or expected insult
  - Timing of delivery; pre, post, or both
  - Severity of condition
  - Expected duration of need
  - Tolerance
  - Function of GI tract remaining
  - Strain by strain assessment

- Base decision on scientific evaluation of the data
Future Trends: Probiotics in Clinical Medicine

- Understanding of inflammation relationship
- More data on specific strains of probiotics
- Better acceptance by “public and scientific community”
- New attention to gut / microbe mutualism
- Probiotics as drug delivery tools genetically engineered
  - “Designer probiotics”
Probiotics as drug delivery tools!

**Lactococcus lactis**

**IL-10**

**DSS**

**Histologic Score**

- Not treated
- Treated

- p = 0.01

**IL-10 knockout**

**Histologic Score**

- Not treated
- Treated

- p = 0.02

Science 2000; 289:1352-5 (mice)
Clin Gastroenterol Hepatol 2006;4:754-759 (humans)
**L. stimulans**  Probiotic – balancing skin microflora

Stimulation of *S. epidermidis*, but not *S. aureus* is seen after 6 h of co-culture

- **S. epidermidis**
- **S. aureus**

<table>
<thead>
<tr>
<th>Cell Count / ml after 6 h*</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.00E+00</td>
</tr>
<tr>
<td>2.00E+07</td>
</tr>
<tr>
<td>4.00E+07</td>
</tr>
<tr>
<td>6.00E+07</td>
</tr>
<tr>
<td>8.00E+07</td>
</tr>
</tbody>
</table>

- **L. stimulans**
- **L. stimulans**
L. anti-pylori
Probiotic - the gentle alternative

LB anti-pylori is a natural isolate which acts as a “probiotic“

- *H. pylori* loses mobility by co-aggregation with
- *L. anti-pylori*
- *H. pylori* no longer adheres to the mucosa
- Aggregated *H. pylori* are carried out of the stomach

Lang et al 2010
Ongoing Trials: Probiotics

- **Neurologic disorders**
  - Pain control, ADHD, Tourette syndrome
- **Inflammatory diseases**
  - Aging, IBD, arthritis, asthma, diabetes
- **Use on non-GI surfaces**
  - Burns, tracheostomy sites, skin in ICU, wounds, STSG, Vagina, respiratory tree
- **AIDS prevention**
  - Changing the pH of the vagina alters HIV receptors
  - Gene transfer HIV receptor into probiotics
    - Already done for L. jensenii
- **Cancer prevention**
  - Multiple mechanisms
    - Dietary procarcinogens by commensal bacteria
    - Histone deacetylase inhibitor
Is it time for a paradigm shift regarding bacteria?

Are we making a leap of faith?

Supply viable beneficial bacteria or a substrate which enhances these specific beneficial bacteria instead of trying to eliminate the pathogen?

“Bioecological control”