How do the microbiota and pro/prebiotics influence nutritional status?
# Discussion group members

Karen Scott, chair  
Nathalie Delzenne, co-chair

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<thead>
<tr>
<th>Discussion group members</th>
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<tbody>
<tr>
<td>James Bunn</td>
<td>Delphine Saulnier</td>
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<tr>
<td>Gregor Reid</td>
<td>Maciej Chichlowski</td>
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<tr>
<td>Esther Nova</td>
<td>Saskia van Hemert</td>
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<td>Eduardo Schiffrin</td>
<td>Kerstin Holmgren</td>
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<td>Paul O’Toole</td>
<td>Natalie Lamb</td>
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<td>Connie Weaver</td>
<td>Tomoyuki Sako</td>
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<td>Howard Jenkinson</td>
<td>Lori Lathrop Stern</td>
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<td>Andrew Serazin</td>
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<td>Paul Sheridan</td>
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<td>Laure Bindels</td>
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Main topics discussed

Role of pre/probiotics in improving nutritional status in

- Malnourished children
- Malnourished mothers
- Malnourished elderly

Role of microbiota in undernourishment associated with non communicable diseases

Can probiotics and/or prebiotics improve nutritional and micronutrient uptake?

AIM Can pro and prebiotics be used to overcome situations where malnourishment has a detrimental effect on health
Definition — Malnutrition versus undernutrition

When a person is

1. not getting enough food or
2. not getting the right sort of food

http://www.wfp.org/hunger/malnutrition

3. Not digesting and absorbing nutrients efficiently

Consensus
We have to reach optimal nutrition status for an individual in a specific context
Immunity and Malnutrition

• Malnutrition is associated with a significant impairment of:
  – cell-mediated immunity
  – phagocyte function
  – immunoglobulin A concentrations
  – cytokine production

• Leads to a high risk of infection, and often to episodes of diarrhoea, hence aggravating the nutritional status further
Gregor Reid
Role of pre/probiotics in improving nutritional status in malnourished children

- Undernutrition in developing world
- Undernutrition in Western countries
- Overnutrition (not discussed)

Questions:

• How do we identify undernutrition in these different contexts?
• When do we intervene (before or around birth? Until 2-3 years of life? when a disease happens?)
• Seasonality
• Do we need prevention or treatment?
Identification: consequences are often irreversible, lifelong, and transgenerational.

Childhood Undernutrition Associated:

- 2x increase in risk of severe diarrhea or pneumonia
- Diminished vaccine response
- Major cognitive delays (15+ IQ pt drop)
- Several-fold increase in risk of adult adiposity, coronary heart disease, and Type II diabetes
- Decreased adult wages (30% decrease in adult wages)
### Healthy Growth risk factors, causes, pathways, and outcomes

#### Potentiating Risk factors
- Low income / education level
- Maternal and childcare practices
- Poor sanitation
- Genetic predisposition
- Pollution, chemicals, xenobiotics
- Early child bearing
- Food insecurity and access
- Exposure to infectious diseases e.g. malaria

#### Mechanistic Pathways

<table>
<thead>
<tr>
<th>Causes</th>
<th>Insufficient Intake</th>
<th>Reduced Absorption and Transfer</th>
<th>Altered Utilization and Control of Growth</th>
<th>Increased Demand</th>
<th>Unknown</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Deficient maternal nutrient stores</td>
<td>Placental insufficiency</td>
<td>Genetic \ Epigenetic</td>
<td>Increased cellular turnover (e.g. RBC, enterocyte)</td>
<td></td>
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<tr>
<td></td>
<td>Decreased breast milk quality / quantity</td>
<td>Damage to gut function</td>
<td>Cell / tissue alterations</td>
<td>Immune activation, febrile illness</td>
<td></td>
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<tr>
<td></td>
<td>Deficient appetite</td>
<td>Microbiome dysfunction</td>
<td>Hormonal dysregulation</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>Deficient infant and child diet</td>
<td>Parasitic infection</td>
<td>Signaling from env. exposures</td>
<td></td>
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</tbody>
</table>

#### Structural and Regulatory Factors (e.g. GH-IGF, Etc, and Unknown)

- Bone
- Lean
- Fat
- Organs

#### Outcome

- Intrauterine Growth Restriction (IUGR)
- Stunting & Wasting
  - Immune System Dysfunction
  - Cognitive Development
  - Unknown

*Causes and pathways are likely to influence tissues and cell types differently*
Probiotics and prebiotic functional foods in the treatment of severe acute malnutrition in Malawi. James Bunn, Natalie Lamb

SYNBIOTIC 2000 Forte (Medipharm, Sweden)

4 PROBIOTICS
(lactobacillus casei sp paracasei; lactobacillus plantarum; leuconostoc; pedicoccus)

Dose: \( >10^{10} \) colony forming units bacteria / day

4 PREBIOTICS: (betaglucans; inulin; pectin; resistant starch)

MIXED IN:

READY-TO-USE THERAPEUTIC FOOD
(= WHO approved standard diet for severely malnourished children)
- composition: peanut butter; milk powder; oil; sugar; micronutrients
<table>
<thead>
<tr>
<th>PRIMARY OUTCOME:</th>
<th>Synbiotic (n=399)</th>
<th>Control (n=396)</th>
<th>relative risk or mean difference (95% confidence interval)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) NUTRITIONAL CURE (total)</td>
<td>215/399 (53.9%)</td>
<td>203/396 (51.3%)</td>
<td>1.06 (0.93 to 1.21)</td>
<td>0.40</td>
</tr>
<tr>
<td>HIV seropositive cures</td>
<td>66/170 (38.8%)</td>
<td>71/192 (37.0%)</td>
<td>1.05 (0.81 to 1.37)</td>
<td>0.71</td>
</tr>
<tr>
<td>HIV seronegative cures</td>
<td>145/203 (71.4%)</td>
<td>131/190 (68.9%)</td>
<td>1.04 (0.91 to 1.18)</td>
<td>0.59</td>
</tr>
<tr>
<td>SECONDARY OUTCOMES:</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2) DEATHS (TOTAL)†</td>
<td>108/399 (27.1%)</td>
<td>119/396 (30.0%)</td>
<td>0.90 (0.72 to 1.12)</td>
<td>0.31</td>
</tr>
<tr>
<td>3) OUTPATIENT DEFAULTERS or WARD ABSCONDEES</td>
<td>27/399 (6.8%)</td>
<td>36/396 (9.0%)</td>
<td>0.74 (0.46 to 1.20)</td>
<td>0.23</td>
</tr>
<tr>
<td>4) FAILURES OF NUTRITIONAL TREATMENT</td>
<td>14/399 (3.5%)</td>
<td>14/396 (3.5%)</td>
<td>0.99 (0.48 to 2.05)</td>
<td>0.98</td>
</tr>
<tr>
<td>5) READMISSIONS</td>
<td>27/399 (6.8%)</td>
<td>16/396 (4.0%)</td>
<td>1.67 (0.92 to 3.06)</td>
<td>0.08</td>
</tr>
<tr>
<td>6) Other: (transfers out; final outcome unknown)</td>
<td>8/399 (2.0%)</td>
<td>8/396 (2.0%)</td>
<td>1.12 (0.44 to 2.86)</td>
<td>0.81</td>
</tr>
<tr>
<td>7) Rate of weight gain (mean g/kg/day ± SD)</td>
<td>4.18 ± 4.0</td>
<td>4.14 ± 4.1</td>
<td>0.04 (-0.53 to 0.61)</td>
<td>0.65</td>
</tr>
<tr>
<td>8) Length of stay in programme (median days to cure ± IQR)</td>
<td>37 ± 14</td>
<td>38 ± 13</td>
<td></td>
<td>0.42</td>
</tr>
</tbody>
</table>
Interpretation of the data

• How to choose a candidate probiotic?
• Definition of the health outcomes is crucial
• We need to know the mechanism of action (target population and outcome)
• Does the diet influence the response?
• Use a model to test for the mechanism, for the efficacy of a probiotic-prebiotic approach
Diet – Microbiota Experiments in the Context of Malnutrition in Children in Malawi

A

\[\% \text{ of starting weight}\]

Time (days)

Malawian diet

RUTF

Malawian diet

\[
\begin{array}{cccccccccccc}
0 & 7 & 14 & 21 & 28 & 35 & 42 & 49 & 56 & 63 \\
\end{array}
\]

Healthy

Kwashiorkor

B

PC1 (27%)

Time (days)

Malawian diet

RUTF

Malawian diet

\[
\begin{array}{cccccccccccc}
0 & 7 & 14 & 21 & 28 & 35 & 49 & 56 & 63 \\
\end{array}
\]

Amino acids

Carbohydrates

Fatty acids

Nucleotides

TCA cycle

Urea cycle

Malawi Diet 1, Day 16

Healthy

Kwashiorkor

[Data Table and Graphs]
3 (or 5) main topics discussed:

Role of pre/probiotics in improving nutritional status in

Malnourished children

Malnourished mothers

Malnourished elderly

Role of microbiota in undernourishment associated with non communicable diseases

Can probiotics and/or prebiotics improve nutritional and micronutrient uptake?

AIM: Can pro and prebiotics be used to overcome situations where malnourishment has a detrimental effect on health
The effect of micronutrient supplemented probiotic yogurt on malnutrition (under and obese) in pregnant women in Africa

In the next ten years, an estimated 2.5 million maternal deaths, 2.5 million child deaths and 49 million maternal disabilities will occur in Africa alone.

Yogurt: 250mL daily, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Streptococcus thermophilus* and *Lactobacillus rhamnosus* GR-1. 17.09 g of powdered Moringa is added to each litre of yogurt.

Patients: age 19-40; before 24 weeks pregnant; Placement of the subjects based on an assessment by a clinician, BMI and anthropometric (mid-upper arm circumference measurements).

Under-nourished – 10 subjects
Under-nourished receiving Moringa supplemented probiotic yogurt -10 subjects
Obese – 10 subjects
Healthy -10 subjects

Recruitment since August, 2012
10 undernourished
2 obese
25 healthy (nourished)
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Procrustes: Microbiota & diet correlate, &
by community location

Unweighted UniFrac PCoA vs. FFQ PCA

Weighted UniFrac PCoA vs. FFQ PCA

FFQ

Community Day Hospital Rehab Long-stay

Paul O’Toole
Intervention in elderly

- Microbial diversity
- Inflammation
  - Probiotics (IL6, TNFa)
  - Prebiotics
- Frailty
  - Appropriate (diverse) diet
  - (sarcopenia, body weight...)

Eduardo Schiffrin
• Cancer cachexia: disabling loss of muscle and fat mass
• Frequent (50% cancer patients; 40% associated with acute non-lymphocytic leukemia and chronic myeloid leukemia)
Main topics discussed

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AIM: Can pro and prebiotics be used to overcome situations where malnourishment has a detrimental effect on health
Non communicable diseases: prebiotic and/or probiotic intervention studies

• Periodontal Disease-Systemic Health-Infectious Disease Axis
• Severe damages in the GI tract and in systemic immune system
  – Preterm infants
  – Short bowel syndrome
  – Severe congenital anomaly
  – Severe systemic inflammatory response syndrome (SIRS)
  – Biliary cancer surgery
  – Patients having mechanical ventilation support and enteral feeding

Howard Jenkinson
Tomoyuki Sako
Yogurt in the nutritional rehabilitation of malnourished anorexia nervosa patients

- Refusal to eat
- Very underweight or emaciated.
- Amenorrhea.
- Subtypes: restrictive/ binge-purging

Adaptive mechanisms
- Biochemical abnormal laboratory findings are rare
  - Leukopenia with relative lymphocytosis
  - Depleted T cell numbers
  - Surprisingly free from infections

• Yogurt in the nutritional rehabilitation of malnourished anorexia nervosa patients
• According to the nutritional marker CD4+/CD8+ ratio, yogurt is an advantageous food choice to include in the refeeding therapy of AN patients.

Esther Nova Rebato
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Prebiotics/probiotics on micronutrients availability

• Prebiotics (soluble corn fiber) increase calcium availability and bone density (end-point)
• Probiotics increase iron absorption (check)
• Most data from animal studies (mechanism?); general lack of human studies...
• Combination of probiotics and micronutrients to target diarrhea?

Connie Weaver, Kerstin Holmgren
### Effects of probiotics and calcium on diarrhea and respiratory infections: randomized trial in Indonesian children

<table>
<thead>
<tr>
<th>Outcome</th>
<th>Low Ca 124</th>
<th>Regular Ca 126</th>
<th>Casei 120</th>
<th>Reuteri 124</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>WHO-defined diarrhea (≥3 loose/liquid stools in 24-h)</strong></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Mean incidence</td>
<td>0.91</td>
<td>0.86</td>
<td>1.05</td>
<td>0.67</td>
</tr>
<tr>
<td>Number of episodes</td>
<td>0.40 ± 0.81</td>
<td>0.38 ± 0.78</td>
<td>0.47 ± 0.87</td>
<td>0.30 ± 0.56</td>
</tr>
<tr>
<td>Adjusted RR (95% CI)</td>
<td>1.00 (ref)</td>
<td>0.99 (0.62-1.58)</td>
<td>1.21 (0.76-1.92)</td>
<td>0.76 (0.46-1.23)</td>
</tr>
<tr>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td><strong>All diarrhea (≥2 loose/liquid stools in 24-h)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean incidence</td>
<td>1.73</td>
<td>1.86</td>
<td>2.04</td>
<td>1.28</td>
</tr>
<tr>
<td>Number of episodes</td>
<td>0.73 ± 1.14</td>
<td>0.77 ± 1.38</td>
<td>0.87 ± 1.32</td>
<td>0.56 ± 0.77</td>
</tr>
<tr>
<td>Adjusted RR (95% CI)</td>
<td>1.00 (ref)</td>
<td>1.10 (0.77-1.59)</td>
<td>1.06 (0.74 ± 1.53)</td>
<td>0.65 (0.46-0.93)</td>
</tr>
</tbody>
</table>

Shown: mean ± SD
Adjusted for: living area, sex, age, illness before study start, hh expenditure, weight-for-height
Describing (potential) health benefit properties: Synthesis of essential nutrients and vitamins

Both strains (*L. reuteri* 55730 and 6475 (genome comparison)) have the predicted ability to produce:
- Essential amino acids (e.g. lysine)
- Vitamin B12
- Folate

A complete pathway for thiamine (Vit B1) biosynthesis was predicted in *L. reuteri* 55730/17938

- Produce acetate and lactate (D/L in different ratio for DSM17938)

Saulnier et al, 2011. Plos One
Key messages/questions

• Probiotics/prebiotics may influence nutritional status through improved digestive and intestinal (barrier) function, rebalancing gut microbiota, immunity/modulation of infection, production/interaction with nutrients, others (change in brain/cognitive function?)

• Initiative “far away from home” are expensive, and not transferable in western countries: who is paying (attention)...?

• How to convince the clinicians of the interest of probiotic-prebiotic approaches in the management of severe diseases (HIV, cancer...)?