Gut Microbial Metabolism of Food Constituents: Modulating Human Dietary Exposures

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Relationship of Diet and the Gut Microbiome to Health and Disease

- Dietary constituents
- Fuel availability

Energy imbalance

Gut bacteria

Disease Risk
- Cancer
- CVD
- Diabetes
Outline

- What are the gut microbes doing with our food?
- What is the effect of the gut microbiome on host dietary exposures?
- How might this influence disease risk?
- Gaps, needs, and challenges
The human diet is complex.

- 1000s of compounds
- Variety of methods of food preparation
  - Structure and particle size
  - Bioavailability to host
Gut Microbial Metabolism -- Designed to make the most of the situation

- Fermentation
- Reduction -- nitrate, sulfate
- Esterification
- Aromatic fission
- Hydrolysis/deconjugation -- glycosides -- glucuronide conjugates
Distribution of Metabolic Pathways in the Gut Microbiome

- Xenobiotic biodegradation
  - phytochemicals
  - pyrolysis products
  - drugs

Fermentation of Carbohydrates

- Acetate
- Propionate
- Butyrate

Microbial Metabolism of Proteins & Amino Acids

Proteins
Peptides

hydrolysis

Aromatic
Amino acids

α, β elimination

Phenols
and indoles

Ammonia
NH$_3^+$/NH$_4^-$

Amines

H$_2$, CO$_2$, CH$_4$

Organic acids

Sulfur
Amino acids

deamination

deamination & fermentation

decarboxylation

Adapted from Nyangale et al. J Proteome Res, 2012
Aromatic Amino Acid Metabolism:
Conversion of L-Tryptophan to Indole

- Concentration in human and rodent lumen – 0.1 to 4 mM
- Modulates expression of pro- and anti-inflammatory genes
- Strengthens epithelial cell barrier properties
- Decreases pathogen colonization

Bansal T et al. *PNAS* 2010
Slide courtesy of R Alaniz, Texas A&M
Sulfur Amino Acid Metabolism: Generation of Hydrogen Sulfide (H$_2$S)

Produced by gut bacteria:
- Fermentation of sulfur-containing amino acids (methionine, cysteine, cystine, and taurine)
- Action of sulfate-reducing bacteria on inorganic sulfur (sulfate and sulfites)

- Toxic to colonocytes both in vitro and in vivo
- Contributes to inflammation (UC and colon cancer)
Fecal sulfide concentrations increase with increased protein intake in a controlled feeding study

- 5 male volunteers
- Randomized cross-over study of 5 protein doses for 10 days each:
- 0 – 600 g meat /d
- Measured fecal sulfide excretion

Conversion of Choline to Trimethylamine

- Microbial metabolism important in production of TMAO.
- Levels of TMAO and choline and betaine increased after a phosphatidylcholine challenge (2 eggs and [d9]-phosphatidylcholine).
- Plasma TMAO suppressed after antibiotics and reappeared after antibiotic withdrawal.

Tang et al. NEJM, 2013
Major Adverse Cardiovascular Events Increase by Quartile of Plasma TMAO

- 4007 adults undergoing elective diagnostic cardiac catheterization
- 3-y F/U for major adverse CVD events.
- Increased plasma TMAO associated with increased risk of CVD event.

Tang et al. NEJM, 2013
Dietary Bioactive Phytochemicals

- **Phenolics**
  - Phenolic acids
  - Stilbenes
  - Curcuminoids
  - Chalcones
  - Lignans
  - Flavonoids
  - Isoflavones

- **Terpenoids**
  - Phenolic terpenes
  - Carotenoids
  - Saponins
  - Phytosterols

- **Organosulfurs**
  - Thiosulfinates

- **N-containing compounds**
  - Glucosinolates
  - Indoles

Adapted from Scalbert et al, J. Agric. Food Chem. 2011, 59, 4331–48
Isothiocyanates from Glucosinolates in Cruciferous Vegetables

S-D-Glucose

R–Cʻ\(\text{N–O–SO}_3^-\)

Glucosinolate

Glucose

Thioglugcosidase (Myrosinase)

矿泉水

R–Cʻ\(\text{N–O–SO}_3^-\)

HSO\(_4^-\)

R–N=C=S Isothiocyanate

Yuesheng Zhang, Roswell Park Cancer Institute, Buffalo, NY
Inverse association between urinary ITC excretion and aflatoxin-DNA adducts – Interindividual variation in ITC bioavailability

- N=200, Qidong, China
- Randomized, parallel arm, 2-week trial
- 400 umol glucoraphanin/d vs. placebo
- Urinary ITC recovery 1-45% of dose

Kensler et al, Cancer Epidemiol Biomarkers Prev, 14:2605, 2005
Isothiocyanate Recovery in Urine Ranged from 1 to 28% with 200 g Cooked Broccoli

% ITC excreted in urine after 200 g broccoli

Li et al., Br J Nutr, 2011
Fecal Bacterial Degradation of Glucosinolates In Vitro Differs by ITC-Excreter Status

- Low- and high-ITC excreters identified with standardized broccoli meal
- Fecal bacteria incubated with glucoraphanin for 48 h

Li et al., Br J Nutr, 2011
Microbial Production of Equol and ODMA

80-90% of individuals produce O-Desmethylandangolensin

20-60% of individuals produce Equol

Daidzein \rightarrow \text{Dihydrodaidzein} \rightarrow \text{Cis/Trans-isoflavan-4-ol} \rightarrow \text{Equol}
Urinary Equol Excretion with Soy Challenge

Lampe et al., *PSEBM* 217:335-339, 1998
Soy Interventions
Equol-Producing Capacity Associated with:

- Greater lengthening of menstrual cycle follicular phase.
  

- Lower estrone, estrone-sulfate, testosterone, DHEA, DHEA-sulfate, androstenedione, and cortisol, and higher SHBG and mid-luteal phase progesterone
  

- Improved bone mineral density in post-menopausal women.
  

- Differential gene expression in peripheral lymphocytes of equol producers and non-producers.
  
Equol-Producing Capacity and Health: Observational Studies

- Positively associated with 2-OH/16αOHE1 ratios in premenopausal and postmenopausal women.
  

- Mammographic density 39% lower in equol producers.
  

- Plasma equol concentrations inversely associated with prostate cancer risk in Japanese men.
  
  Akaza et al., *Jpn J Clin Oncol* 32:296, 2002

- Significant interaction between soy intake and equol-producer status in predicting breast density in postmenopausal women.
  
  Fuhrman et al., *Cancer Epidemiol Biomarkers Prev* 17:33, 2008
What Human Gut Microbes Produce S-(-)Equol?

Daidzin ➤ Daidzein ➤ Dihydrodaidzein ➤ Equol

Daidzein ➤ Equol
- Adlercreutzia equolfaciens
- Bacteroides ovatus
- Bifidobacterium
- Eggerthella sp YY7918
- Enterococcus faecium
- Finegoldia magna
- Lactobacillus mucosae
- Lactococcus garvieae
- Ruminococcus productus
- Slackia sp HE 8
- Streptococcus intermedius
- Veillonella sp

Daidzin ➤ Dihydrodaidzein
- Clostridium-like bacterium

Dihydrodaidzein ➤ Equol
- Eggerthella sp Julong 732

Microbial Metabolism of Dietary Components

Summary

- Gut microbial metabolism modifies a variety of dietary components.
- Differences in gut microbial community capacity to handle substrates is detectable as metabolic phenotypes.
- Diet as consumed is not necessarily that experienced by the host.
- The gut microbiome needs to be considered in context of host diet to understand its impact on metabolism and disease risk.
Gaps, Needs and Challenges: More Specific to Nutrition

- **Challenge**: Testing causality of gut microbiome’s contribution to health and disease in humans.

- **Need**:
  - Prospective cohorts with repeated measures of exposure (i.e., diet, etc) and samples for gut microbiome characterization.
  - Well-controlled dietary interventions to understand inter-individual variation in bacterial metabolic phenotypes in the context of diet.
  - Accurate model systems of human dietary metabolism and associated microbiota.
Gaps, Needs and Challenges: Broader Considerations

- To facilitate transdisciplinary research to allow for integrated breadth and depth of knowledge.
- Methods of assessing composite functionality of the gut microbiome and integration of the structure and function of microbial systems.
- Computational methods to integrate high-dimensional microbiome and metabolome data.
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