Incorporating Food and Gut Microbiota into 21st Century Risk Analysis

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COLEMAN SCIENTIFIC CONSULTING

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Invited Lecture (University of Liverpool, Institute for Risk and Uncertainty)

Outline

1. Introduction to Human Superorganisms and Risk Analysis
2. Health and Disease: Gut, Gut-Lung Axis, Respiratory System
3. Managing our Microbes for Health and Protection from Disease
4. Evidence Maps on Benefit-Risk Analysis for Mammalian Milks
5. Incorporating Food and Gut Microbiota into 21st Century Risk Analysis

Section 1.

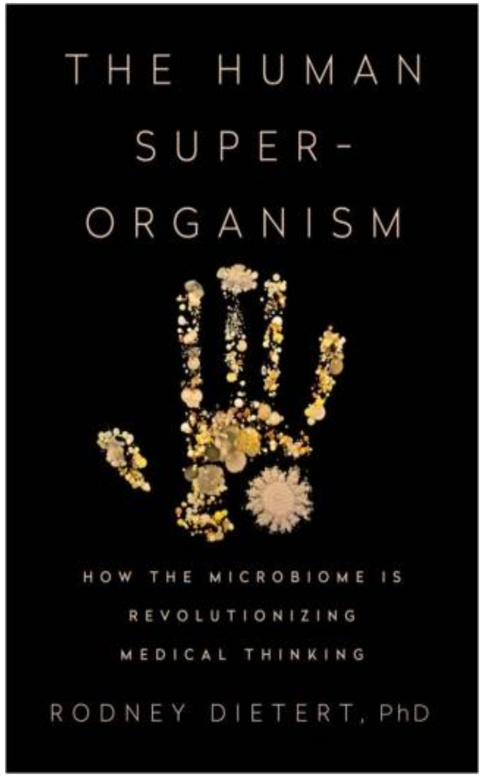
Introduction to Human SUPERORGANISMS and Risk Analysis

Current Knowledge:
Sterility is NOT Healthful, Actually Harmful

Rodney Dietert (Cornell U. Professor Emeritus of Immunotoxicology)
2017 SRA webinar, *Protecting the Human Superorganism*

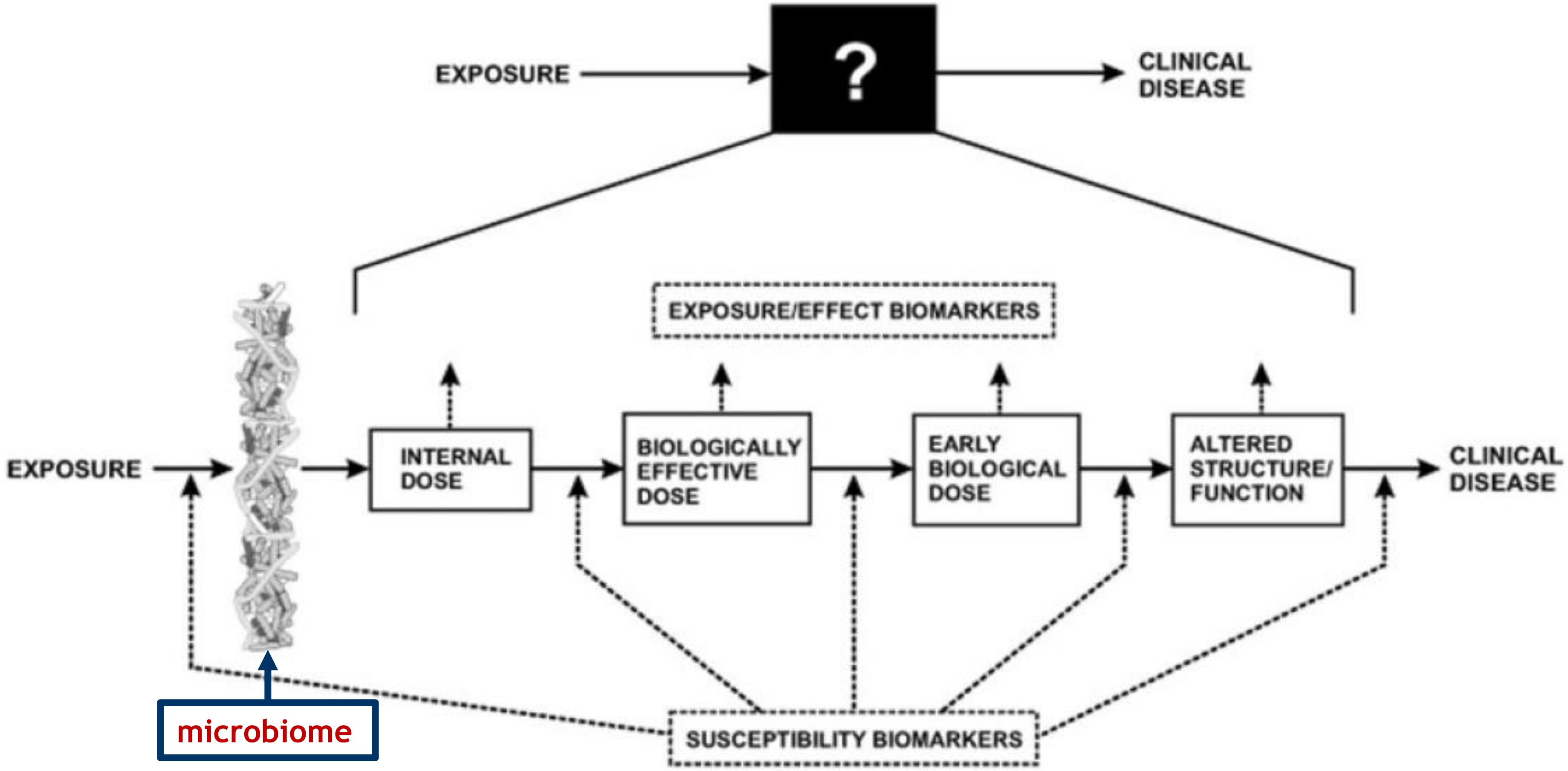
- ***Homo sapiens* + microbiota = human 'superorganism' (holobiont, 'supraorganism')**

- New medical landscape emerging in **21st century**, with **microbial ecology of superorganisms** challenging assumptions about health and disease, with emerging paradigm shift to '**managing our microbes**'



Dietert, 2016. The Human Superorganism: How the Microbiome is Revolutionizing Medical Thinking.

Chemical (AND Microbial) Risk Assessment in Human Superorganism, not *Homo sapiens*



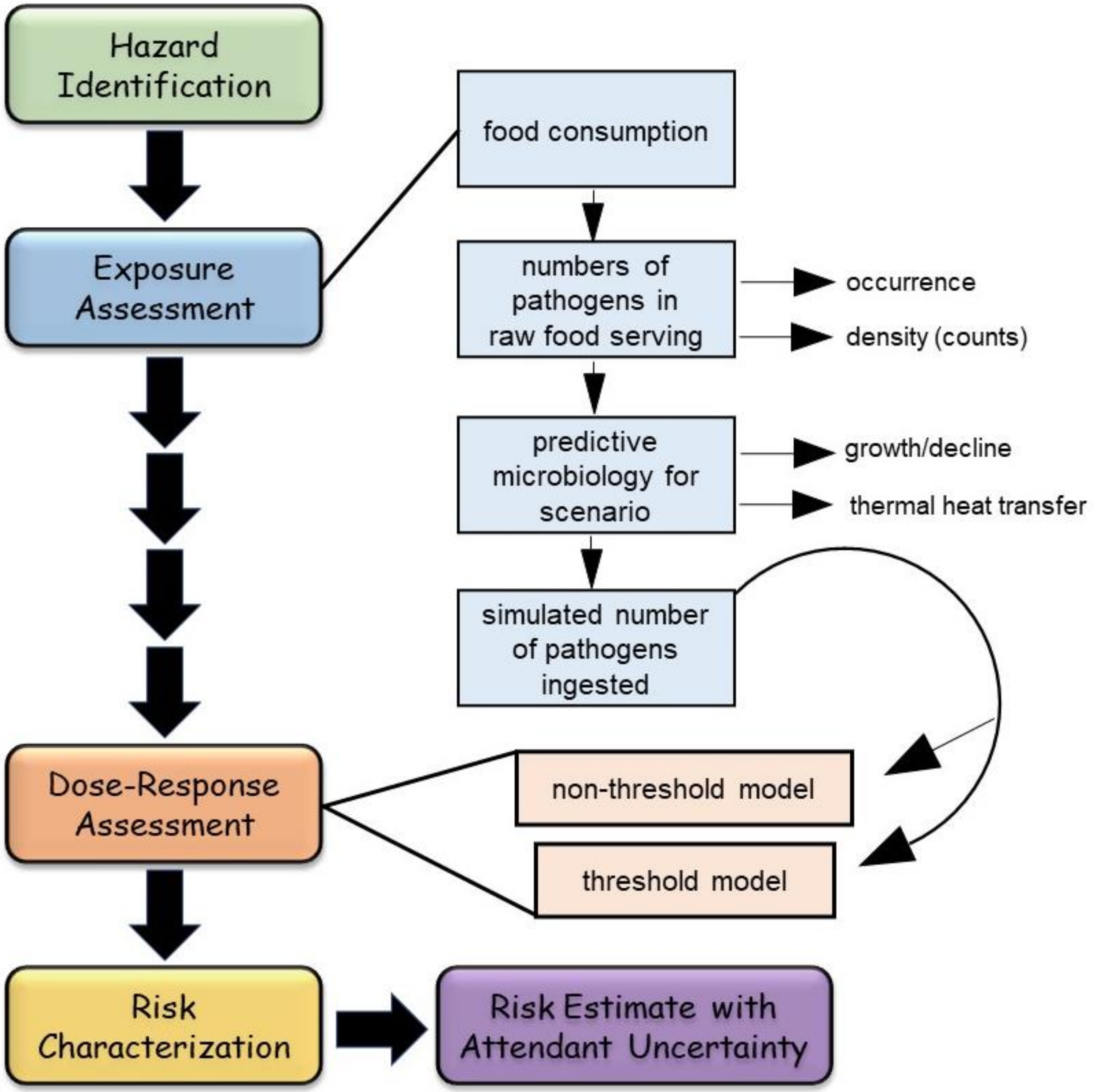
Dietert and **Silbergeld**, **2015**. Biomarkers for 21st Century: Listening to the Microbiome.
Toxicological Sciences

Traditional Framework for Microbial Risk Assessment

(Marks et al., 1998; Coleman et al., 2021b)

Perceptions in Food Safety

- 20th century: manage presence or detection of pathogens (genera including pathogens)
- 21st century: account for effects of natural microbiota in milk and healthy gut microbiota driving resistance to low doses of pathogens
    - Evidence for thresholds challenges past default assumption that single pathogen cell causes disease in healthy humans



Motivation: Evidence Map Approach to Promote Transparency for Evidence on the State of Science

Evidence Maps: Communicating Risk Assessments in Societal Controversies: The Case of Engineered Nanoparticles

- Promote **openness** and **transparency** for evaluating ambiguous and conflicting scientific evidence for applications in risk analysis
- Provide **structured graphical representation** of the **evidence basis**, drawing attention to both **pro-** and **contra-** arguments, with **supporting** and **attenuating** data
- Assist **risk analysts** in **avoiding 'traps** such as **confirmation bias'** that may **distort** judgments about **weighing and synthesizing evidence** from **multiple disciplines**
- Facilitate **constructive dialogue** between diverse perspectives/opinions of all **stakeholders**, including decisions makers and educated public
- Assist **diverse experts** and **non-experts** to acknowledge the full body of scientific evidence, the **evidence basis**, as well as **quality of evidence** and **uncertainty**

Wiedemann et al., **2011**. Evidence Maps: Communicating Risk Assessments in Societal Controversies: The Case of Engineered Nanoparticles. *Risk Analysis*

Evidence Map Template from *Applied Microbiology* Paper

Coleman et al., 2021b. Enhancing Human Superorganism Ecosystem Resilience by Holistically 'Managing our Microbes' (motivated by Wiedemann et al., 2011)

Pro Supplemental Studies on Mechanisms

- **Vertical and Horizontal (Environmental) Transfer of Microbes** from foods or environment to humans
  - Study$_A$
  - Study$_B$
  - Study$_C$
- **Microbial Ecology** of foods or environment
  - Study$_A$
  - Study$_B$
  - Study$_C$
- **Microbiome: Immune System Crosstalk**, indirect colonization resistance for humans
  - Study$_A$
  - Study$_B$
  - Study$_C$

supporting

- Evidence attributing benefits of microbes or microbial consortia in specific foods or the environment to humans
- Other

Pro-Argument on Health Benefits

1. Extent and consistency of the evidence for dose-dependent protective effects against incidence and severity of infectious diseases

2. Extent and consistency of the evidence for dose-dependent protective effects against incidence and severity of non-communicable diseases

attenuating

- Evidence reducing benefits of microbes or microbial consortia in specific foods or the environment to humans
- Other

Contra Supplemental Studies on Mechanisms

- **Foodborne pathogen susceptibility to innate defenses including microbiota,** direct colonization resistance
  - Study$_A$
  - Study$_B$
  - Study$_C$

supporting

- Evidence on infectious disease causation, dose-dependency
- History of use of foods or exposure in environments
- Other

Contra-Argument on Risks of Enteric Infections

Extent and consistency of the evidence for dose-dependent effects linked to infectious (and/or non-communicable) diseases

attenuating

- Assessment of predictive value of pathogen presence/absence and levels in foods and environment for human health and illness
- Other

Evidence Basis

- Numbers of benefit-risk, systematic review, meta-analysis, and clinical studies in the body of evidence
- Number of other supporting or attenuating studies
- Number of Supplemental Studies with evidence on plausible mechanisms

Conclusions

1. Overall biological benefits with assessment of limitations and strengths, clarity , consistency, and cohesion of the body of evidence, with supplemental studies on plausible mechanisms
2. Overall biological risks with assessment of limitations and strengths, clarity, consistency, and cohesion of the body of evidence, with supplemental studies on plausible mechanisms

Remaining Uncertainties

- Uncertainty$_A$
- Uncertainty$_B$
- Uncertainty$_C$

Section 2.

Health and Disease: Gut, Gut-Lung Axis, Respiratory System

Common Risk Management Worldview in 1990s:
Eliminate Bacteria in Foods

Current decade, some shifts in food safety management and education

- **USDA FSIS** monitors by **Whole Genome Sequencing** since 2015; high variability in virulence profiles for foodborne pathogen isolates

- **USDA, FDA, Partnership for Food Safety Education** promote use of **thermometers** in **refrigerators** to maintain food temperatures **unlikely to permit pathogen growth** (40° F / 4.4° C)

- **EFSA (2015).** Scientific Opinion on the Public Health Risks Related to the Consumption of Raw Drinking Milk.

Superorganisms Differ in Susceptibility, Resistance, Severity

- **Typical 20th century assumptions:** neonates, infants, young children highly susceptible to all infectious diseases

- **UNTRUE** for ***C. difficile*** **infection (CDI):**
  - consistent body of evidence from nearly 10 decades of observations of **high rates of asymptomatic colonization**, **very rare symptomatic CDI** in neonates and infants (Smith et al., 2020) versus
  - **disproportionately severe affects for elderly** in nursing home environments (Haran et al., 2021)
  - rising incidence, severity (~500,000 annual US cases, 1/5 severe with recurrence)

- **TRUE** for ***Staphylococcus aureus*** in **hospitalized** neonates, infants**:**
  - early life dysbiosis in nasal, gut, lung microbiota set up immune system for inflammation in the lung and chronic conditions including asthma (Khamash et al., 2018)

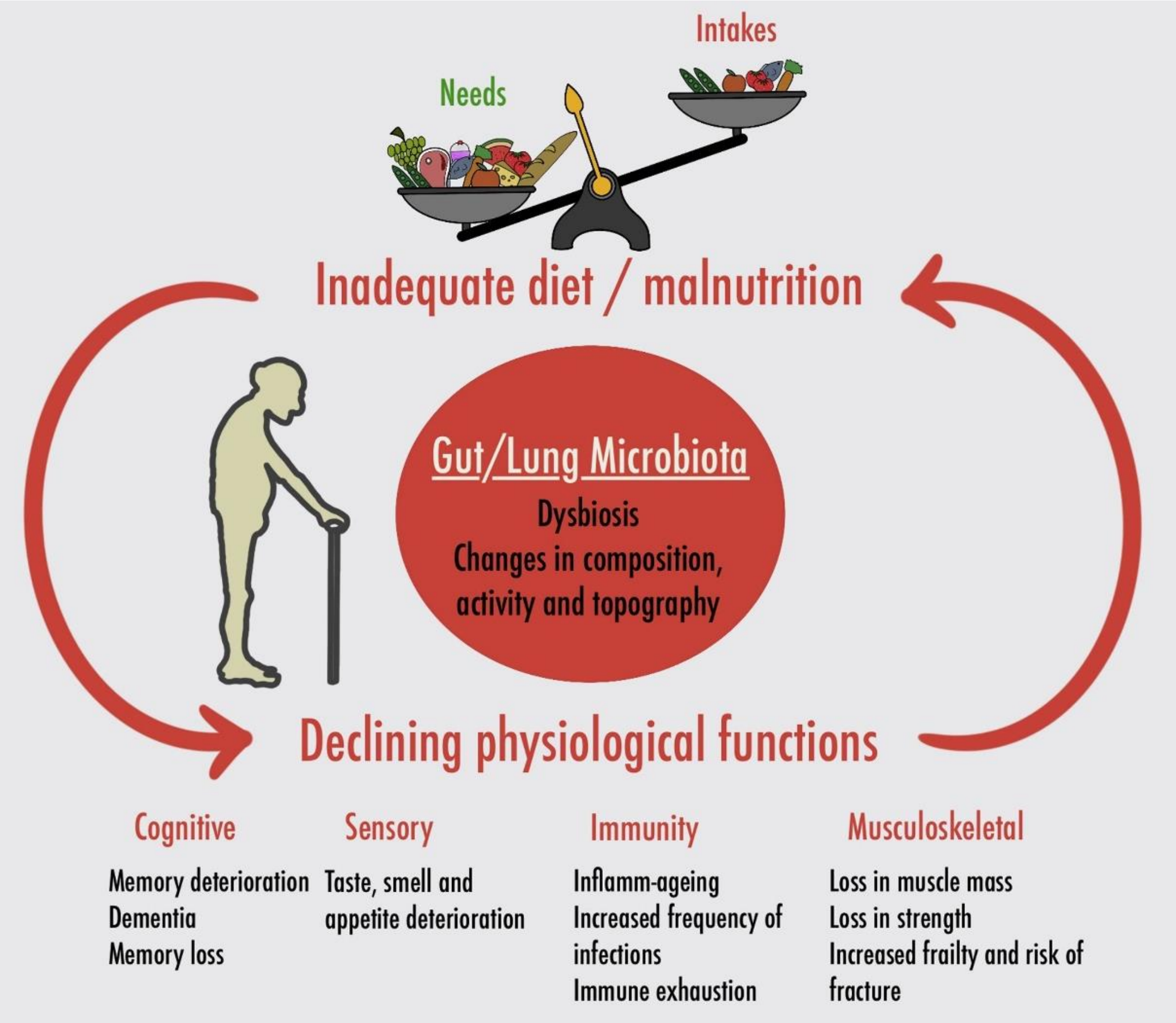
Consider Pathogen-Microbiota-Host-Environment-Specific Dose-Response Models

Primary Risk Factors for Hospital Acquired Diarrhea (HAD), *C. difficile* Infection (CDI)

1. Exposure to **antibiotics**, particularly broad spectrum**, poly-pharmacy** (i.e., antibiotic + proton pump inhibitors), **chemotherapy**
2. Duration **hospitalization** (risk increases each day)
3. Advanced **age**
4. Underlying **co-morbidities**
5. Vitamin D deficiency
6. GI tract manipulation (i.e., GI tube insertion, GI surgery)

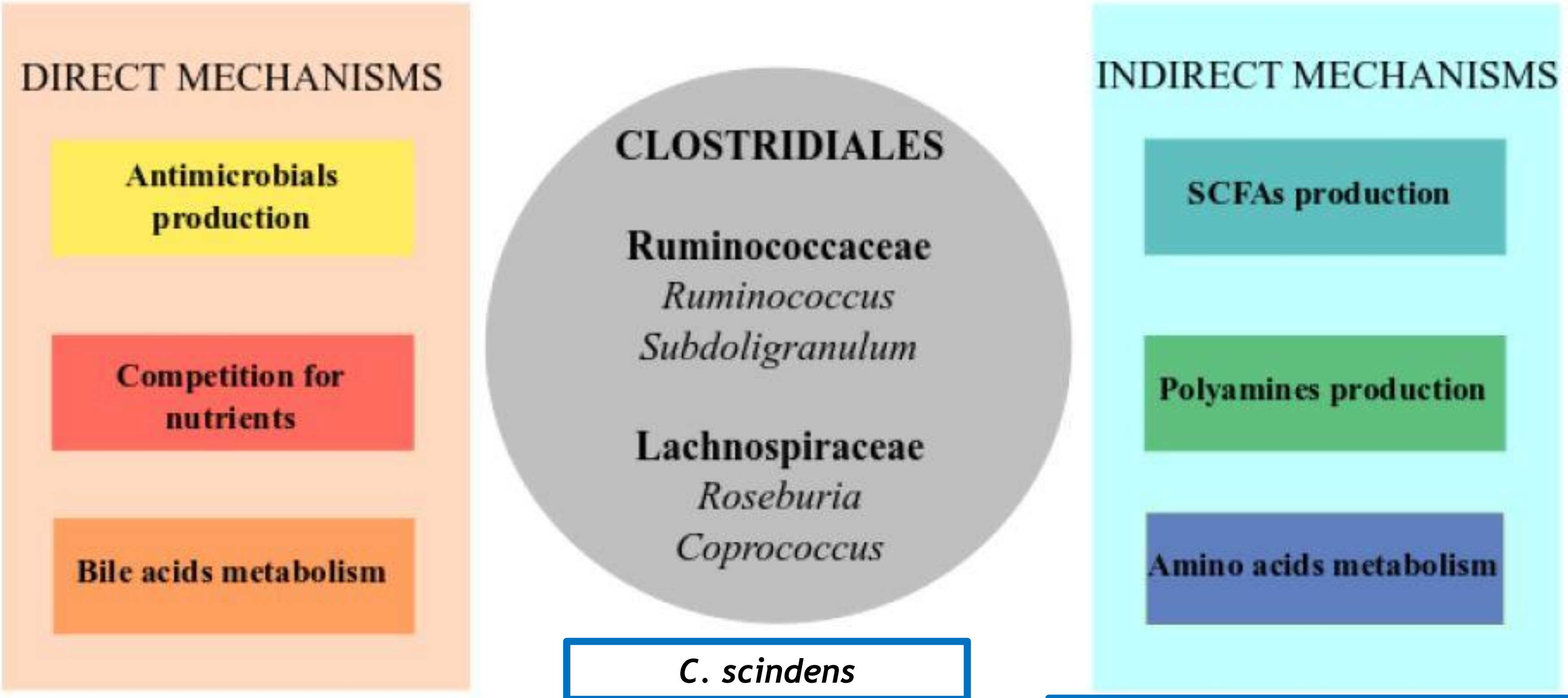
Durham et al., **2020**. Navigating changes in Clostridioides difficile prevention and treatment *Journal of Managed Care + Specialty Pharmacy*

Interconnections of Diet and Dysbiosis Contribute to Physiological Declines with Aging



Saint-Criq et al., **2021**. Dysbiosis, malnutrition and enhanced gut-lung axis contribute to age-related respiratory diseases. *Ageing Research Reviews*

Colonization Resistance to CDI: Related Commensals

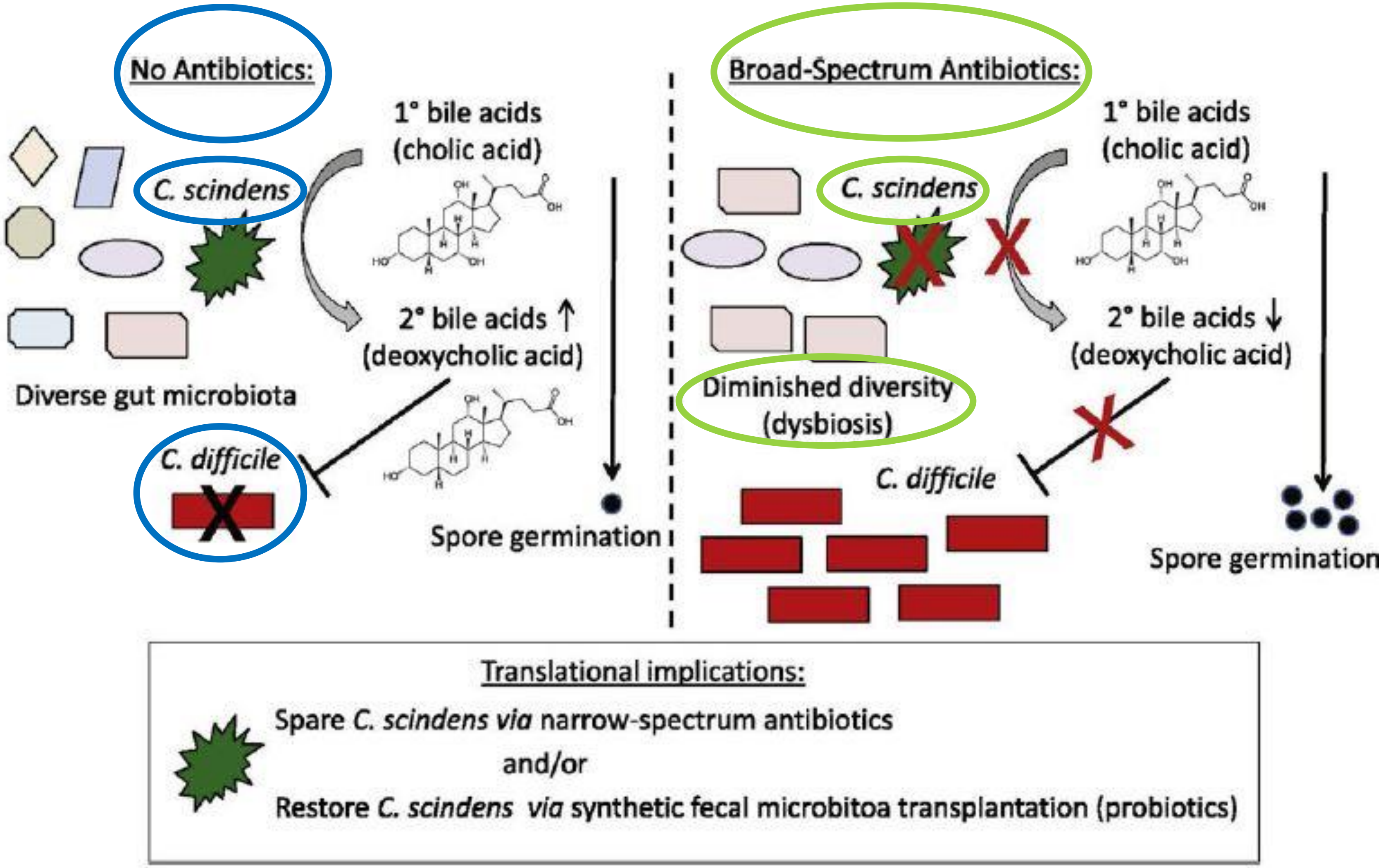


C. scindens

SCFAs: Short Chain Fatty Acids
(e.g., butyrate, propionate)

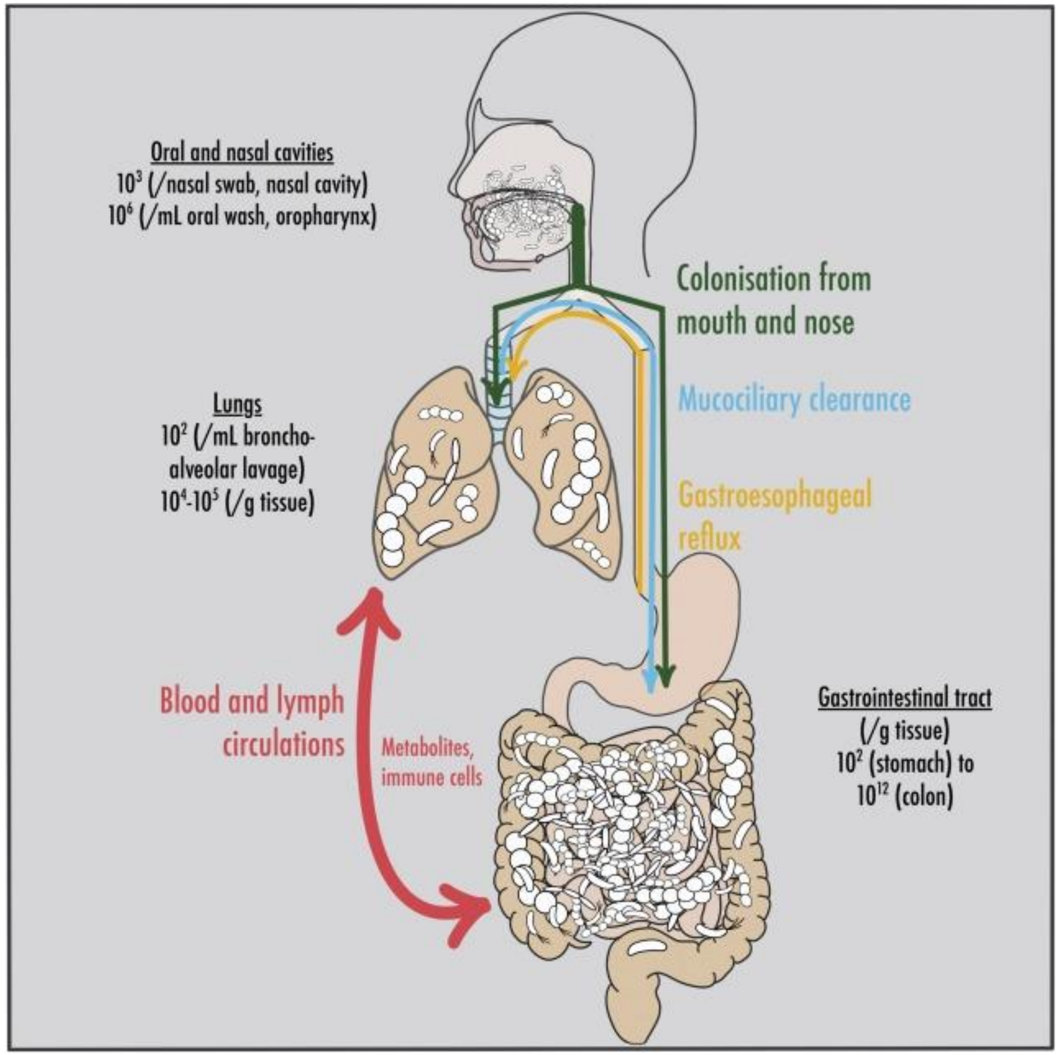
Perez-Cobas et al., **2015.** Colonization Resistance of the Gut Microbiota against Clostridium difficile.
Antibiotics

More on Related Commensals



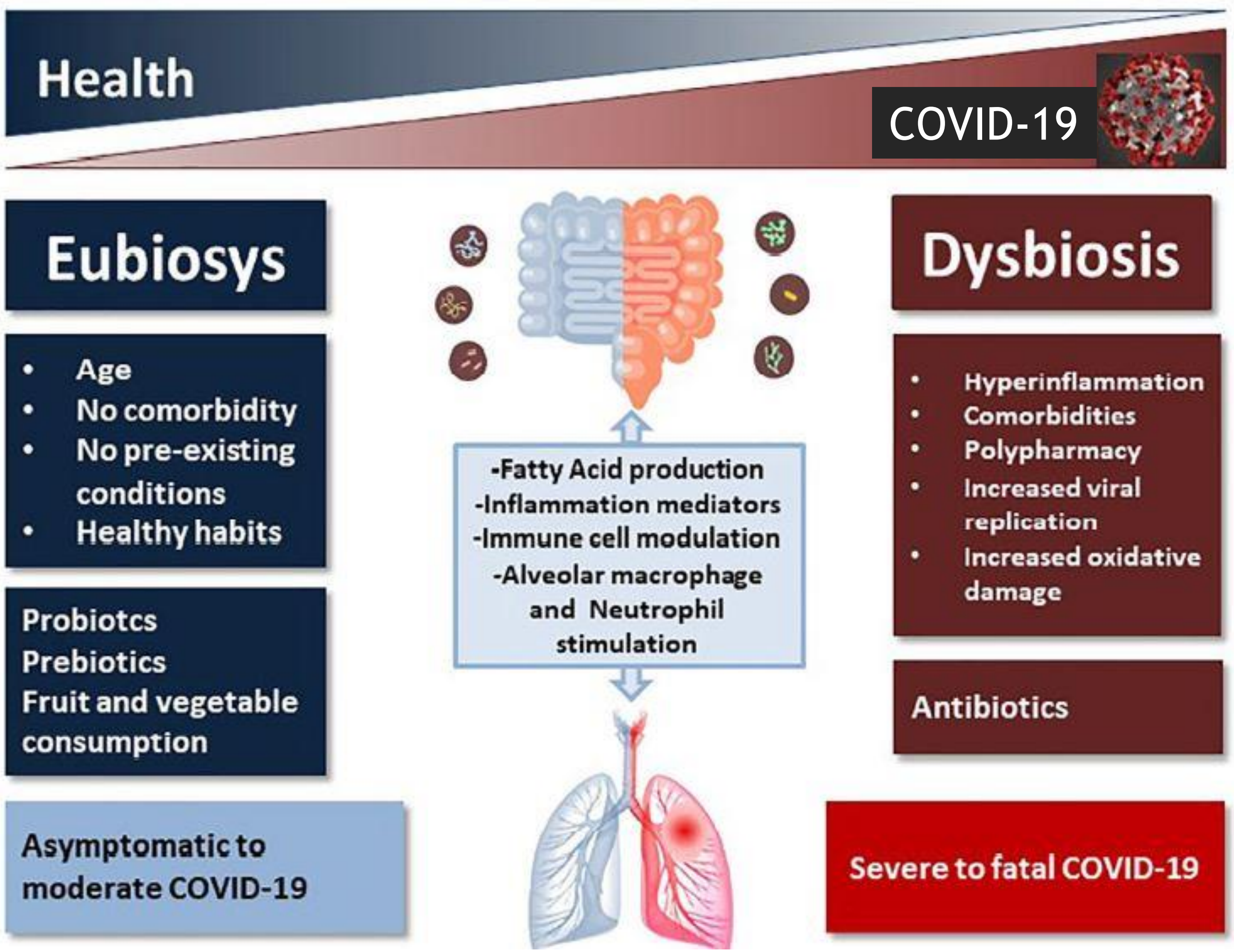
Greathouse, 2015. Dysfunctional Families: Clostridium scindens and Secondary Bile Acids Inhibit the Growth of Clostridium difficile. *Cell Metabolism*

Understanding Gut-Lung Axis



Saint-Criq et al., **2021**. Dysbiosis, malnutrition and enhanced gut-lung axis contribute to age-related respiratory diseases.
Ageing Research Reviews

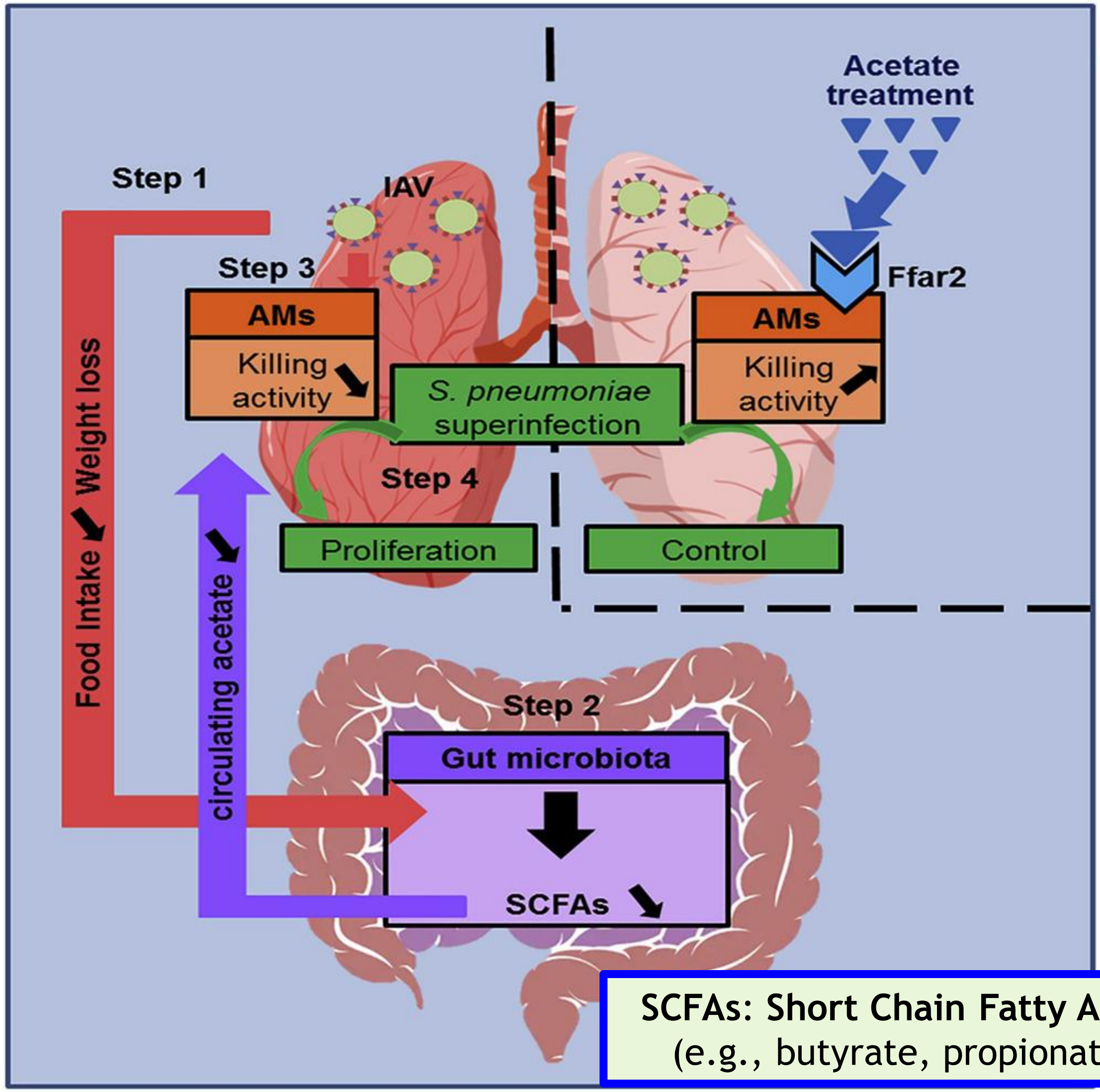
Building on Gut/Lung Microbiota Studies



Long COVID???

Zeppa et al., **2020**. Gut Microbiota Status in COVID-19: An Unrecognized Player?
Frontiers in Cellular and Infection Microbiology

Gut Dysbiosis, Influenza, Pneumonia



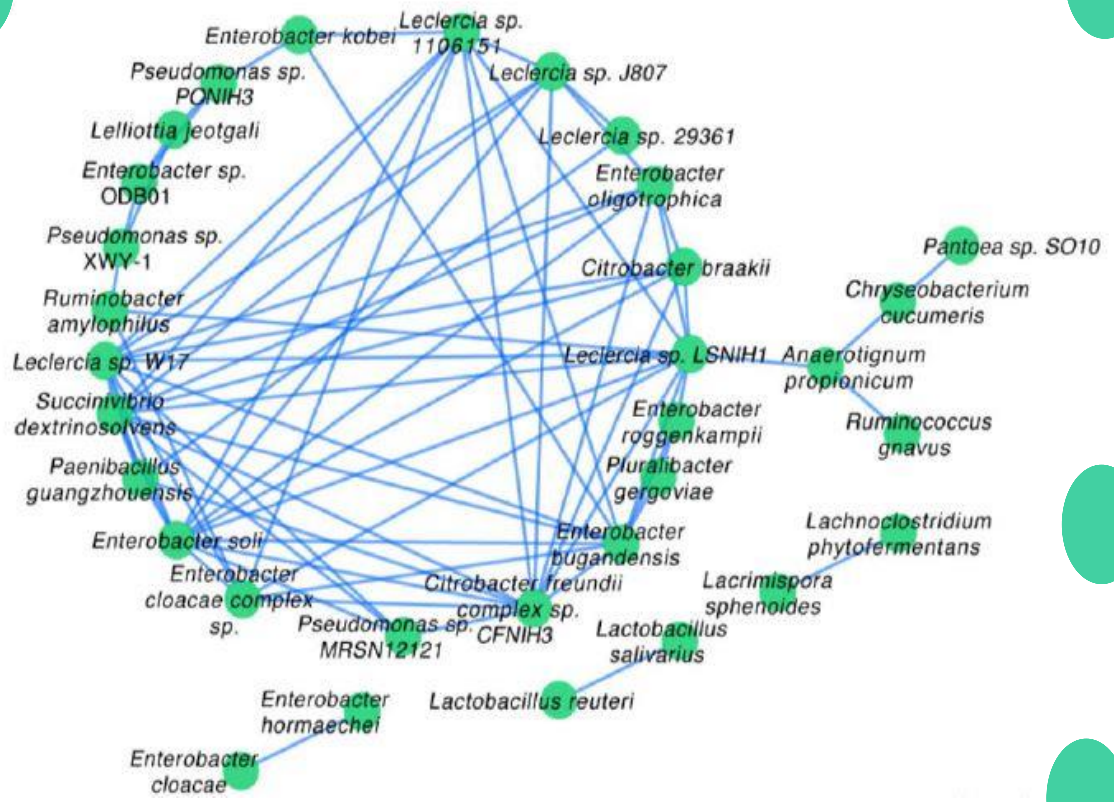
SCFAs: Short Chain Fatty Acids
(e.g., butyrate, propionate)

Sencio et al., **2020.** Gut dysbiosis during influenza contributes to pulmonary pneumococcal superinfection through altered SCFA production. *Cell Reports*

Significant Differences in Lung Microbiota
23 Healthy Controls, 19 COVID Cases

- **lung bacteriome (99 species)**
  + and - correlations
- **lung virome (18 viruses)**

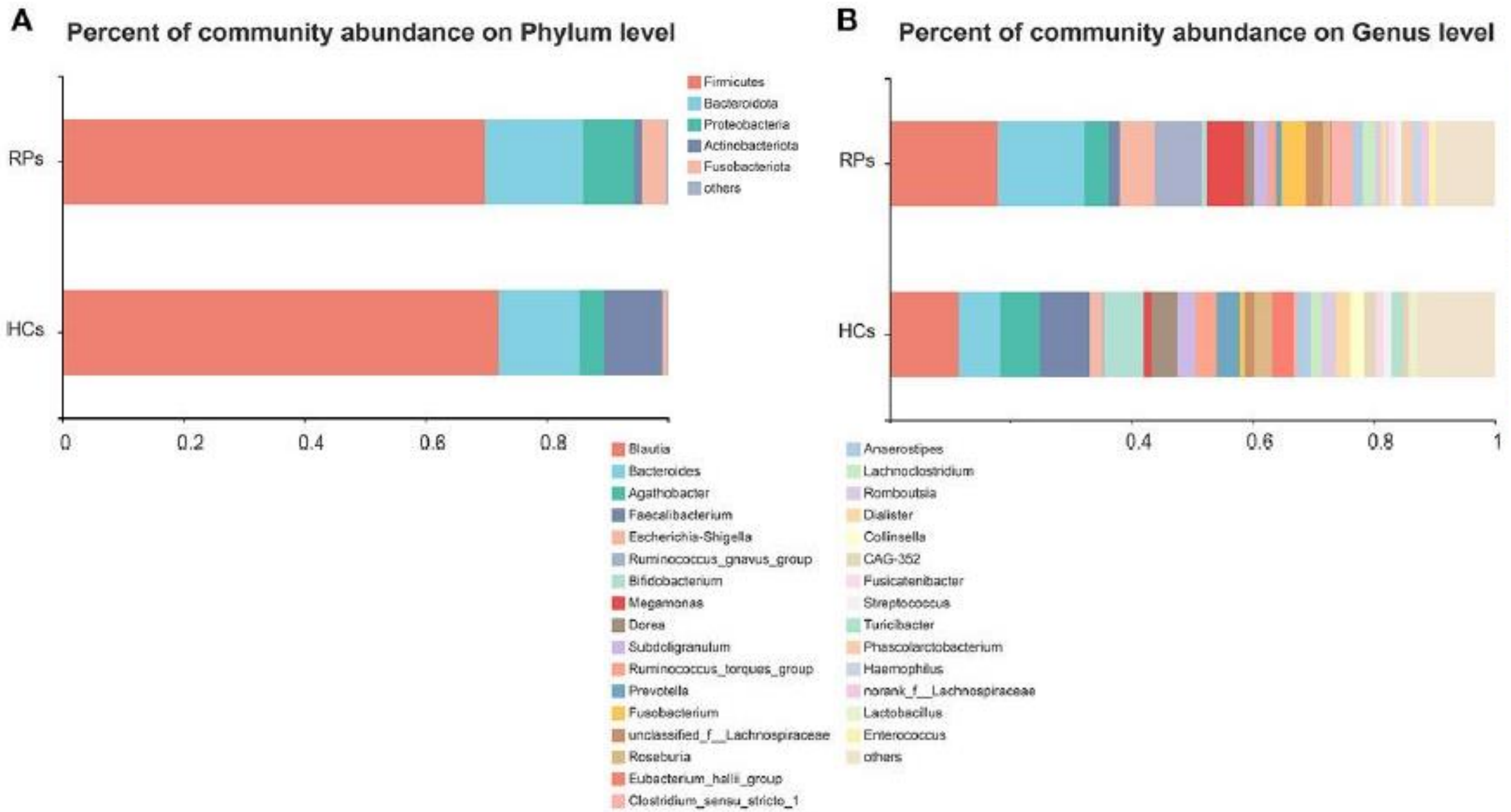
Correlations suggest **lung microbiota** merits further studies on **mechanisms** for interactions, temporal and spatial dynamics, and **causation** to identify priorities for **'managing our microbes'** in the respiratory tracts of COVID patients



Han et al., **2020.** The active lung microbiota landscape of COVID-19 patients. *medRxiv*

Delayed Recovery of Gut Microbiota

7 Healthy Controls, 7 COVID Cases



Tian et al., **2021**. Gut Microbiota May Not Be Fully Restored in Recovered COVID-19 Patients After 3-Month Recovery. *Frontiers in Nutrition*

Prevention: Severity? Long COVID?

Review > J Clin Med. 2021 Dec 16;10(24):5913. doi: 10.3390/jcm10245913.

A Systematic Review of Persistent Symptoms and Residual Abnormal Functioning following Acute COVID-19: Ongoing Symptomatic Phase vs. Post-COVID-19 Syndrome

Glenn Jennings [1 2], Ann Monaghan [1 2], Feng Xue [1 2], David Mockler [3], Román Romero-Ortuño [1 2 4 5]

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ann. behav. med. (2021) XX:1–16
https://doi.org/10.1093/abm/kaab081

SYSTEMATIC REVIEW

Fatigue Symptoms Associated With COVID-19 in Convalescent or Recovered COVID-19 Patients; a Systematic Review and Meta-Analysis

Sanjay Rao, MBBS, MD, FRCPsych, FRCPC, MBA[1] · Tarek Benzouak BA[2] · Sasha Gunpat BA[2] · Rachel J. Burns PhD[2] · Tayyeb A. Tahir MBBS, MD, FRCPsych[3] · Stephen Jolles MD, MRCP, FRCPath, PhD[4] · Steve Kisely MD, PhD, DMedRes[5]

> Sci Rep. 2021 Jun 28;11(1):13414. doi: 10.1038/s41598-021-92717-8.

Follow-up of COVID-19 recovered patients with mild disease

Alina Kashif [1], Manahil Chaudhry [2], Tehreem Fayyaz [1], Mohammad Abdullah [3], Ayesha Malik [2], Javairia Manal Akmal Anwer [1], Syed Hashim Ali Inam [1], Tehreem Fatima [2], Noreena Iqbal [4], Khadija Shoaib [1]

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4. Medicine, Milton Keynes University Hospital, Milton Keynes, UK.

Version 2. medRxiv. Preprint. 2020 Nov 29.
doi: 10.1101/2020.11.24.20238261

PMCID: PMC7709187
PMID: 33269366

This article is a preprint.
Preprints have not been peer reviewed.
To learn more about preprints in PMC see: NIH Preprint Pilot.

The Kids Are Not Alright: A Preliminary Report of Post-COVID Syndrome in University Students

Julie Walsh-Messinger, PhD,[1,2,*] Hannah Manis, BS,[1] Alison Vrabec, BA,[1] Jenna Sizemore,[1] Karyn Bishof, BS,[3] Marcella Debidda, PhD,[4] Dolores Malaspina, MD MSPH,[5] and Noah Greenspan, DPT[4]

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Section 3.
Managing our Microbes for Health and Protection from Disease

Graphical Abstract: Applied Microbiology

Building Resilient Gut Ecosystems to 'Manage our Microbes'

Defining Exposures

- Diet ↑ in whole fresh foods with beneficial microbes, prebiotic fibers
- RDAs for vitamins and microbes?

Healthy Human Superorganism

Expected Outcomes

- Healthy gut microbiome;
  ↑ colonization resistance;
  ↓ pathogen blooms;
  ↑ immune defenses to clear pathogens;
  ↓ health risks
- Well-primed immune system balances inflammatory responses to microbes in environment

Defining Exposures

- Diet ↑ in processed foods, ↓ in beneficial microbes, prebiotic fibers
- Pharmaceuticals
- Open niches for opportunistic pathogens

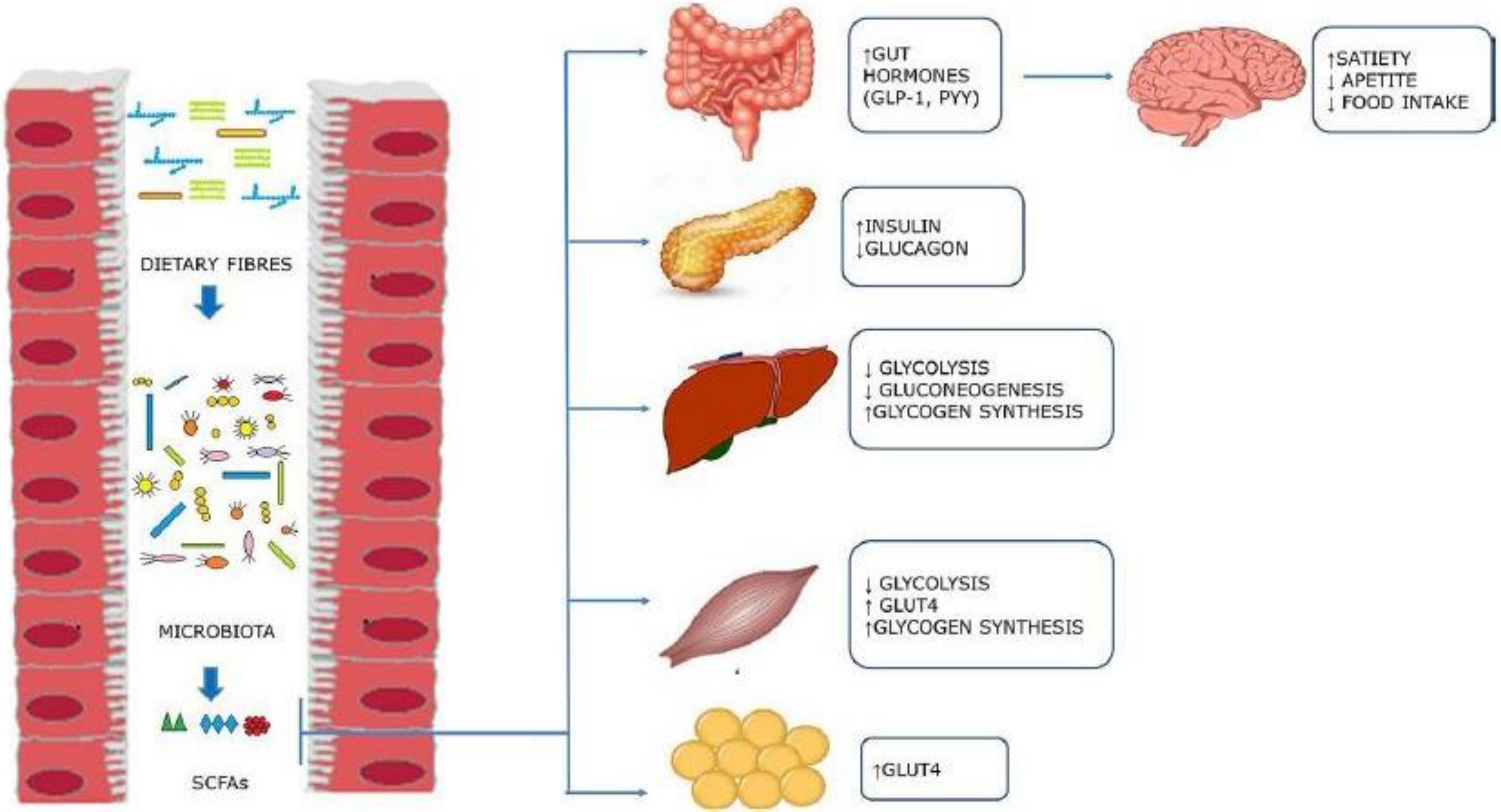
Dysbiotic Human Superorganism

Expected Outcomes

- Dysbiotic gut microbiome;
  ↓ colonization resistance;
  ↓ capability to assist immune defenses to limit, clear pathogens;
  ↑ pathogen blooms; ↑ health risks
- Imbalanced immune system;
  ↑ inflammatory responses;
  ↑ susceptibility to pathogens;
  ↑ damage host

Coleman et al., 2021. Enhancing Human Superorganism Ecosystem Resilience by Holistically 'Managing Our Microbes'. *Applied Microbiology*

Diet Affects Whole Superorganism:
Understanding Gut-Lung-Brain Axis



Short Chain Fatty Acids
(butyrate, propionate)

Mechanistic Data linking Fiber, SCFAs to Superorganism Health

Salamone et al., **2021.** The relationship between gut microbiota, short-chain fatty acids and **type 2 diabetes** mellitus: the possible role of dietary fibre. *Ageing Research Reviews*

Cross-Talk Between Gut Microbiota, Immune System, Lungs

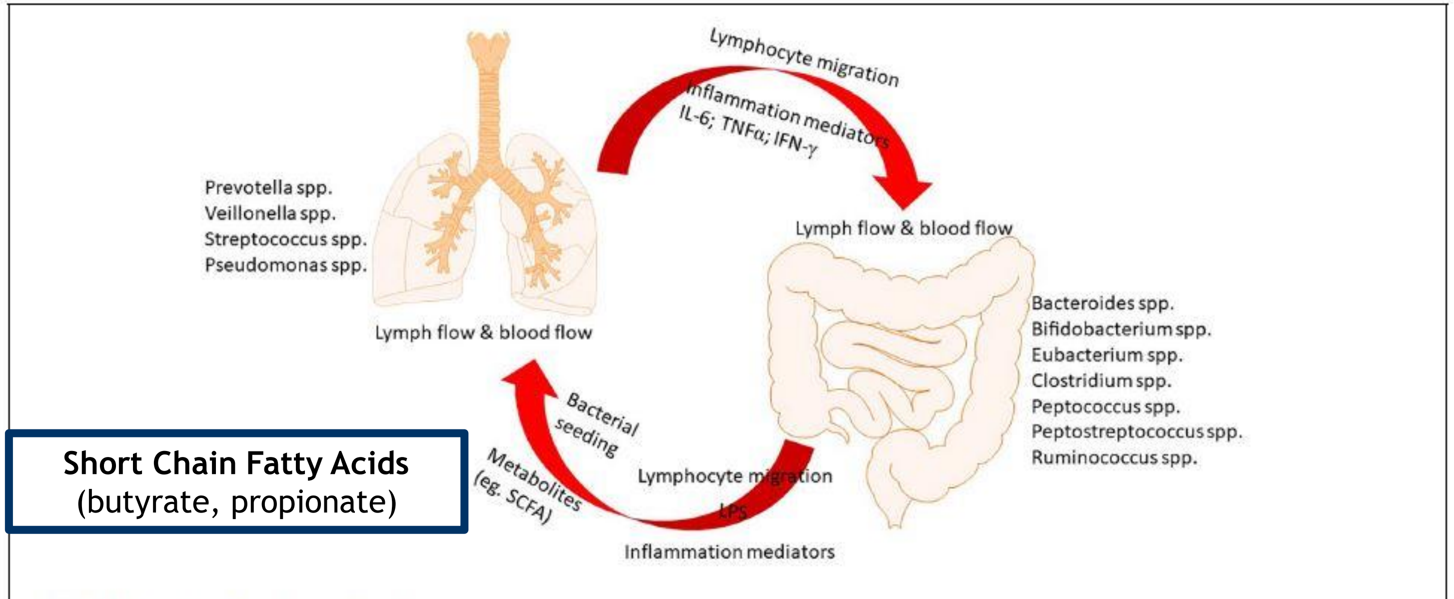


FIGURE 1 | The cross-talk between gut and lung.

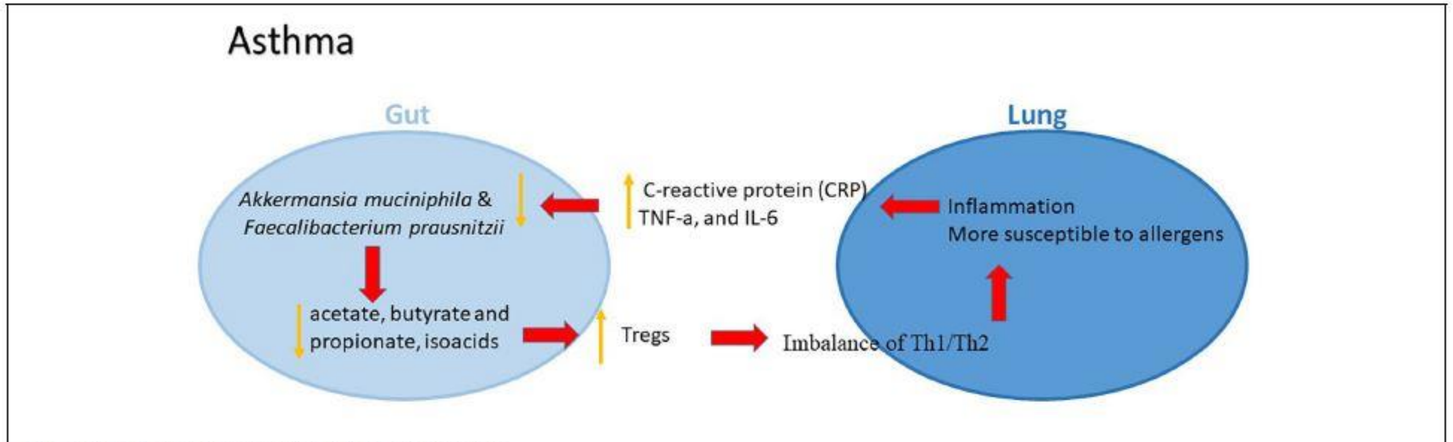
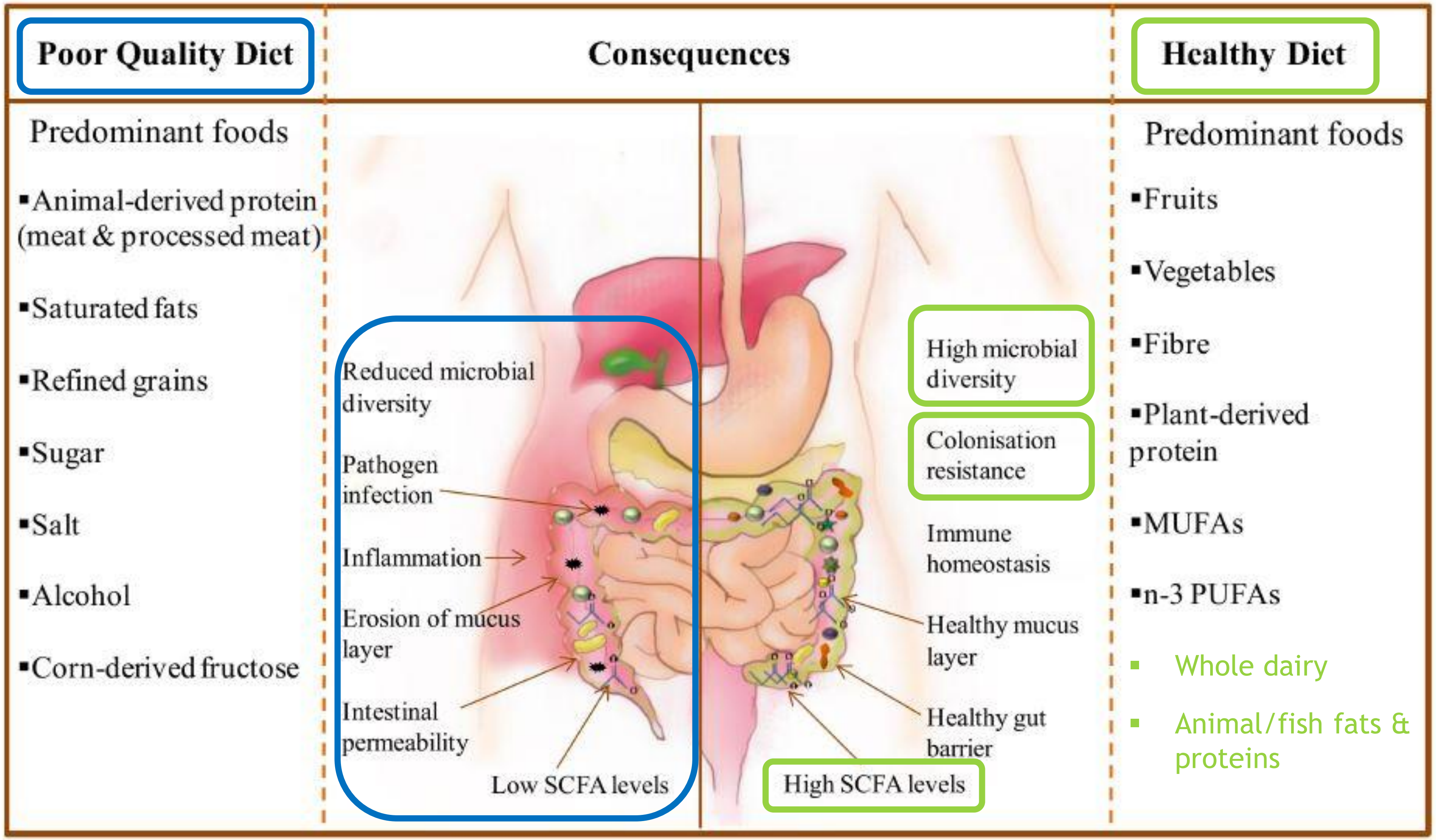


FIGURE 2 | The potential role of gut–lung axis in asthma.

Zhang et al., **2020**. The Cross-Talk Between Gut Microbiota and Lungs in Common Lung Diseases. *Cell Reports*

‘Managing our Microbes’ for Health and Resilience



Mills et al., **2019**. Precision Nutrition and the Microbiome, Part I: Current State of the Science. *Nutrients*

Resilience of Healthy Gut Ecosystem

- Ability of the system/ecosystem to sustain or restore its basic functionality following a perturbation, challenge, or stressor

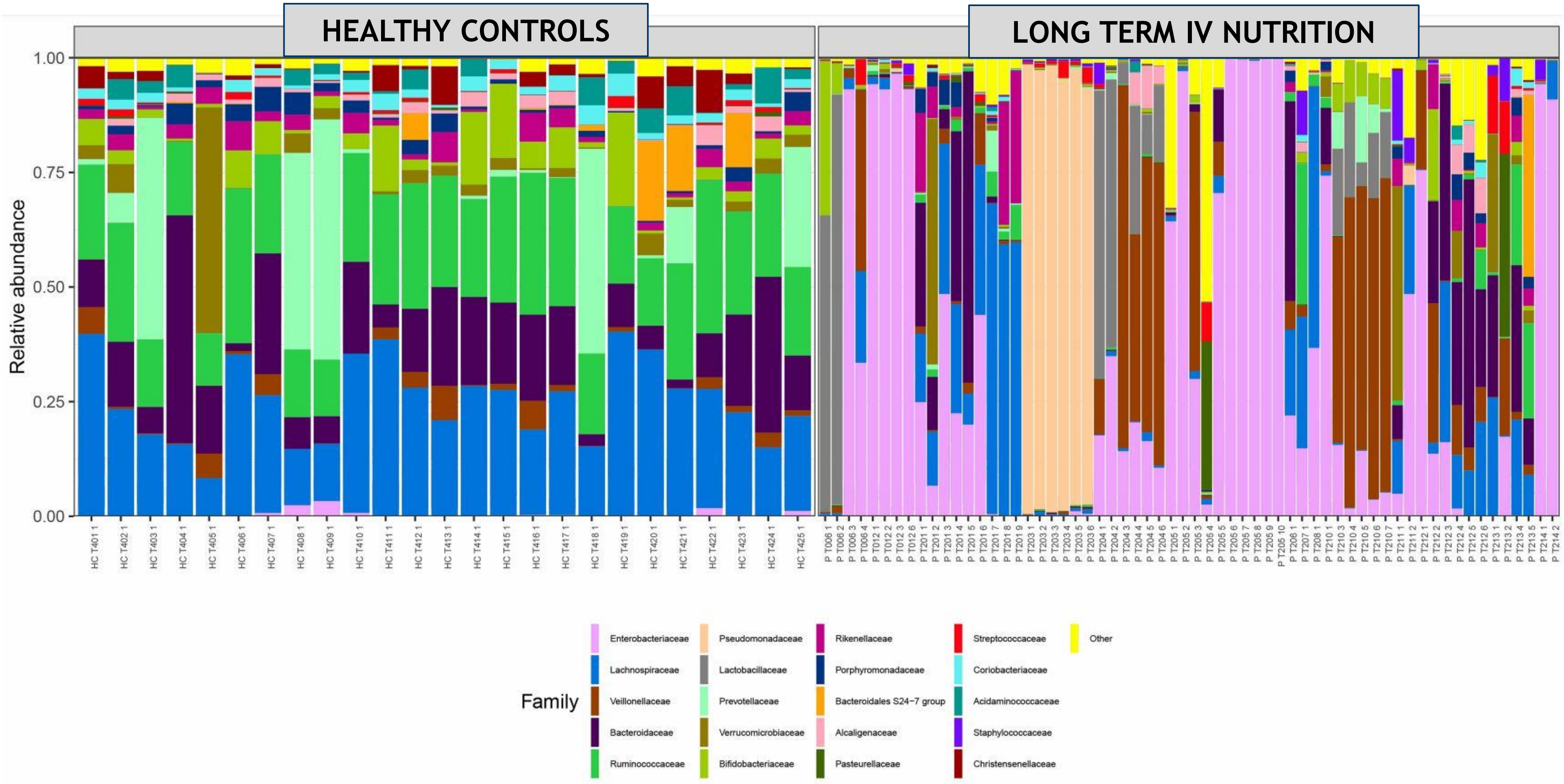
- Healthy **gut ecosystem** particularly complex, **high diversity and richness** (over 2,000 species representing 12 phyla; 9 dominant genera) and **functional redundancy**, stable to small perturbations (Perez-Cobas et al., 2013; Anwar et al., 2021)

- Stressors for gut resilience include pharmaceuticals, malnutrition, and diets low in fiber and enriched in processed foods

Pharmaceutical stressors: antibiotics, laxatives, NSAIDs, proton pump inhibitors (PPIs), AND polypharmacy

Diverse Gut Microbiota in Healthy Children, Blooms in Pediatric Disease

(profound dysbiosis)

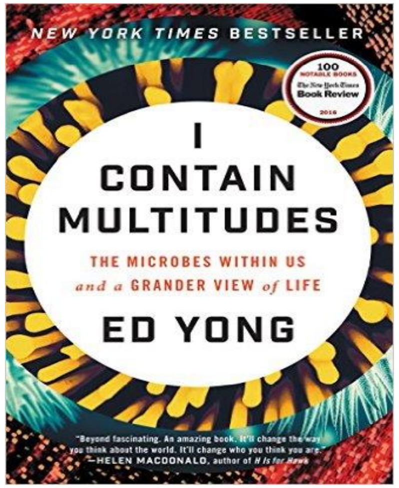


Neelis et al., **2021**. Gut microbiota and its diet-related activity in children with IF [intestinal failure] on long-term parenteral nutrition [intraveneous].
Journal Parenteral Enteral Nutrition

Section 4.
Evidence Maps on Benefit-Risk Analysis for Mammalian Milks

Milk: A Mammalian Innovation

200 Million-Year-Old 'Superfood' (Yong, 2016)



Human milk

- Emphasis on human milk waxed and waned over recent centuries, but now **maternal milk** recommended from birth and for two years or more
- Wet nursing ancient practice in many cultures (Code of Hammurabi from 2250 BC)

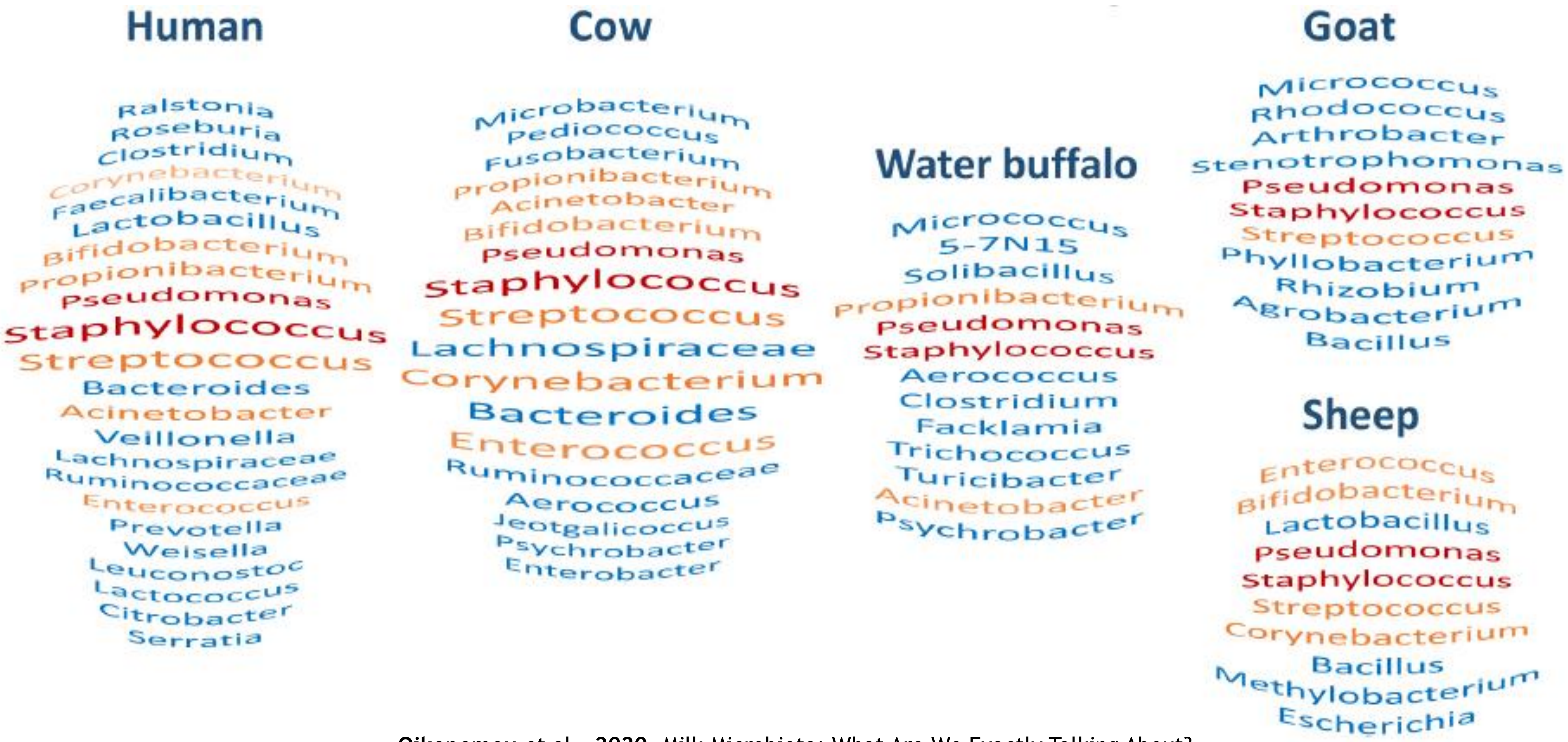
World Health Organization recommends exclusive breastfeeding for first 6 months of life (WHO, UNICEF, 2003)

Breastfeeding reduces **frequency** AND **duration** of **respiratory** and **diarrheal** illness in infants <6 months age (Lopez-Alarcon et al., 1997)

Exclusive breastfeeding protects against common infections during infancy and lessens the **frequency** AND **severity** of infectious episodes (Ladomenou et al., 2010)

Recent Milk Microbiota Study

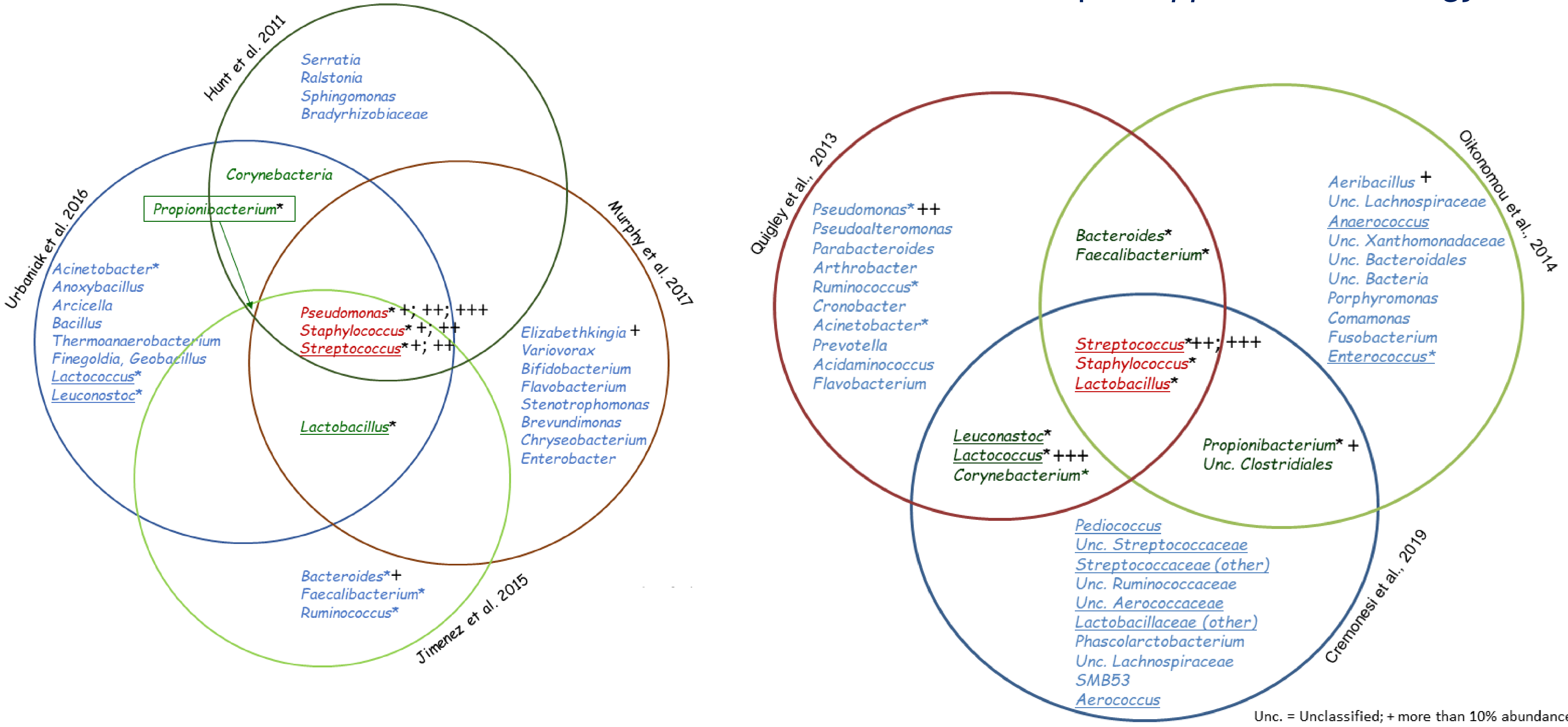
University Liverpool Colleague George Oikonomou



Oikonomou et al., **2020**. Milk Microbiota: What Are We Exactly Talking About?
Frontiers in Microbiology

Milk Microbiota Similarities

Dietert, Coleman et al., 2021c. Nourishing the Human Holobiont to Reduce the Risk of Non-Communicable Diseases: A Cow's Milk Evidence Map Example. *Applied Microbiology*



Unc. = Unclassified; + more than 10% abundance;
++ more than 25%; +++ more than 50%;
Underlined genera members of Lactic Acid Bacteria (LAB);
* Dominant genera in common for breastmilk and bovine milk

Graphical Abstract from *Applied Microbiology* Paper

Coleman et al., 2021a. Examining Evidence of Benefits and Risks for Pasteurizing Donor Breastmilk

Benefits and Risks of Raw and Pasteurized Breastmilk

Raw Breastmilk

photo by Kyle Nieber on Unsplash

- ↑ diversity of gut microbiota
- ↑ colonization resistance
- ↓ infectious and noninfectious diseases
- ↓ risk of childhood and maternal obesity
- ↑ developing nervous system
- ↑ cognitive development
- ↓ chronic disease

Pasteurized Donor Milk

photo by Lucy Wolski on Unsplash

- ↓ diversity gut microbiota
- ↑ dysbiosis
- ↓ colonization resistance
- ↓ weight gain and growth
- ↑ risk of necrotizing enterocolitis
- ↑ risk of mortality
- ↑ risk of infectious and noninfectious diseases
- ↑ cost
- ↓ cognitive development
- ↑ chronic disease

General View for Human Milk Bank Policies

- Rigorous donor screening methods similar to blood donation
- Some screen donor milk for other potential pathogens and indicators of contamination
- Some limits for pathogens/indicators (counts per mL) in donor milks (Omarsdottir et al., 2008)
  - <100,000 *Staphylococcus aureus*
  - <100 Enterobacteriaceae
  - 0 (below limit of detection) for potential pathogens

    Listeria monocytogenes, *Salmonella*, Group B/α-hemolytic *Streptococcus*, coagulase-negative *Staphylococcus*
- Most **pasteurize** donor milk (NOT Germany, Japan, Norway)

Assumption: *Pasteurization Minimizes Risks for NICU Infants*

Benefits AND Risks for Vulnerable Population

Human Milk Banks

provide **pasteurized** human donor milk to hospitalized preterm infants and sick/high risk infants

Holder pasteurization (heating to 62.5°C for 30 minutes) is required due to **perception**: possible presence of potential pathogens perceived as **'risky'**

Yet Loss of Benefits for Pasteurized Milks in Clinical Studies around the World!

- **Ford et al., 2019:** 74 preterm infants raw, 43 past donor (US, TX)
- **Sun et al., 2019:** 98 very preterm infants raw, 109 past donor (China)
- **Squires, 2017**: 302 low birth weight infants (US, WA)
- **Cossey** et al., **2013**: 303 very low birth weight infants (Belgium)
- **Strand** et al., **2012**: 335 infants and toddlers (Nepal)
- **Montjaux-Regis** et al., **2011**: 55 premature infants (France)
- **Schanler** et al., **2005**: 243 extremely low birth weight infants (US, TX)
- **Narayanan** et al., **1984**: 226 high risk, low birth weight infants (India)

Evidence Map for Breastmilk Ecosystem

Pro Supplemental Studies on Mechanisms

- **Vertical and Horizontal (Environmental) Transfer of Microbes** from maternal diet to gut, mammary tissues, milk, infant
  - Gregory 2016
  - Sawh 2016
  - Murphy 2017
  - Toscano 2017
  - de Andrés 2018
  - Ojo-Okunola 2018
  - Moossavi and Azad 2019
  - Van Deaele 2019
  - Wang 2020
- **Microbial Ecology**
  - Arroyo 2010
  - Fernandez 2016
  - Cacho 2017
- **Microbiome: Immune System Crosstalk**, indirect colonization resistance, recent reviews
  - Ward 2013
  - Chong 2018
  - Dietert 2018
  - van den Elsen, 2019

Contra Supplemental Studies on Mechanisms

- **Pathogen susceptibility to innate defenses including microbiota**
  - Cacho and Lawrence 2017
  - Dietert 2018
  - Le Doare 2018
  - Ojo-Okunola 2018

supporting

- ***B-RA* (*Meltzer 2016*), *SR* (Miller 2018), *SR/MA* (Villamor-Martinez 2018), *CSs* (Sun 2019; Ford 2019)** demonstrated loss of benefits (protection against mortality, NEC, sepsis, other) for pre-term infants fed pasteurized donor milk or formula.

Pro-Argument on Benefits of Raw Breastmilk

1. Extensive consistent evidence for dose-dependent protective effects compared to formula (or pasteurized donor milk) against incidence and severity of infectious diseases: ear and upper respiratory infections, diarrhea.
2. Extensive evidence for protective effects against non-communicable diseases: convincing for obesity; probable for asthma, celiac, Crohn's, diabetes, eczema, high blood pressure, ulcerative colitis, wheezing.

attenuating

- No studies identified that attribute benefits to specific raw milk microbes or microbial consortia.
- ***SR/MA*** of observational studies demonstrated pasteurized donor milk reduced bronchopulmonary dysplasia compared to formula; effect not observed in randomized trials (**Villamor-**Martínez **2018**).

supporting

- **CS (Bapistella 2019)** demonstrated lower CMV infection rates for mother's own breastmilk treated with short-term pasteur-ization than historical controls fed raw breastmilk.
- Policy paper on infectious diseases associated with mothers' & donors' breastmilk (**American Academy of Pediatrics, 2017**).

Contra-Argument on Risks of Enteric Infections from Potential Pathogens in Raw Breastmilk

Limited evidence for normal breastmilk from healthy mothers causing infectious diseases in infants.

attenuating

- No **B-RA** or **QMRA**, ***SRs***, ***MAs***, or ***CSs*** identified estimating risks of infectious disease transmission by breastmilk; review (**Gribble and Hausman, 2012**).
- ***CS*** (**Schanler, 2011**) demonstrated pathogen presence in breastmilk not predictive of illness in preterm infants.
- Long history of use of raw donor breastmilk in Norway (**Grøvslien and Grønn, 2009; Grøvslien 2020**).

Evidence Basis

- 1 ***Benefit-Risk Assessment*** **(Meltzer 2013/16)**, 1 ***SR*** **(Miller 2018)**, 1 ***SR/MA*** **(Villamor-Martinez 2018)**, 2 ***CSs*** **(Sun 2019; Ford 2019)** citing extensive, consistent evidence
- 5 Other supporting or attenuating studies
- 18 Supplemental studies with evidence on plausible mechanisms

Conclusions

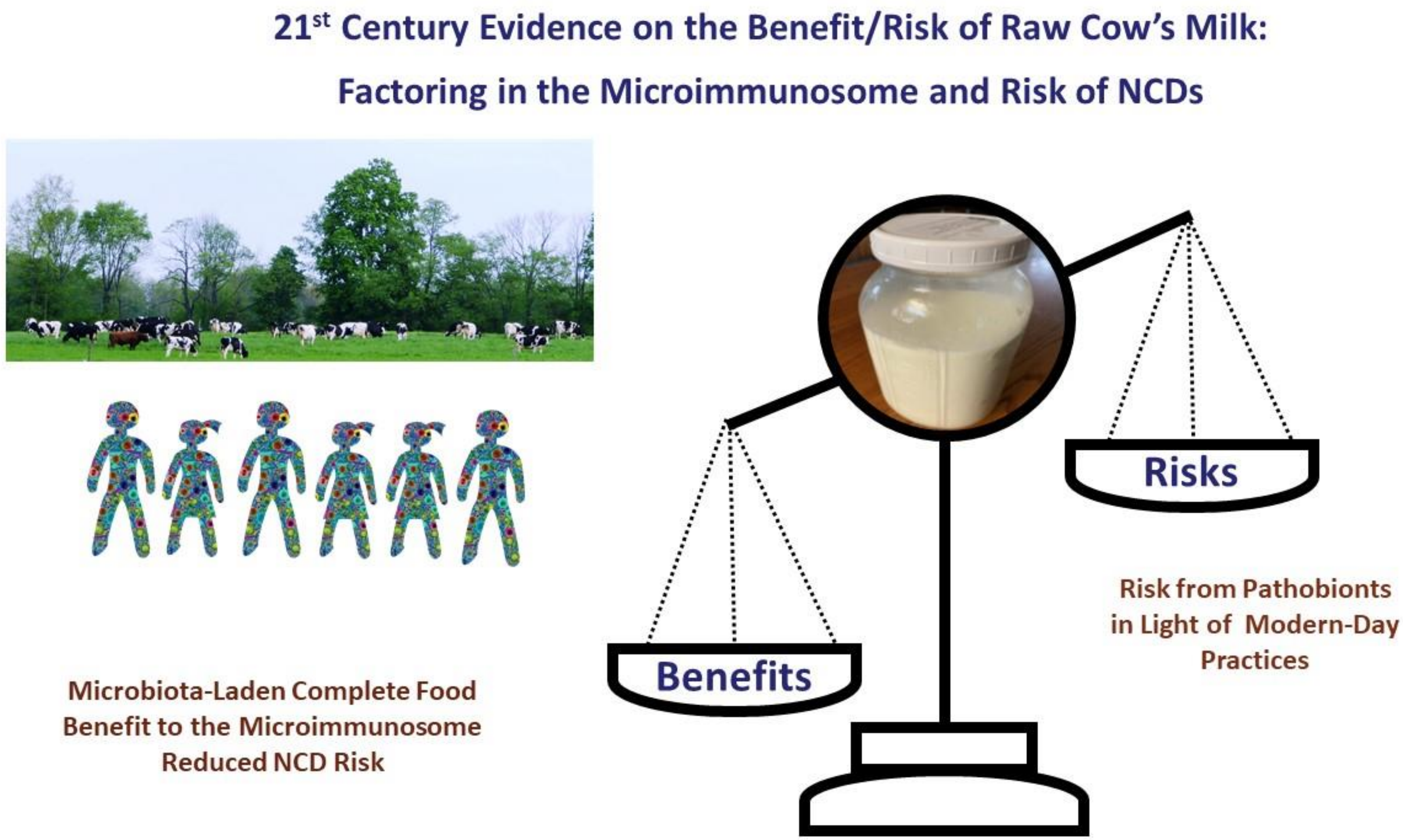
1. Overall biological benefits associated with breastmilk clear, convincing, and conclusive, with supplemental studies on plausible mechanisms attributed to biologically active raw breastmilk
2. Evidence for assessing risks of pathogen infections in infants fed breastmilk from moms and donors limited and inconclusive

Remaining Uncertainties

- How do milk microbiota function in protection against infectious and non-communicable diseases in infancy and later in life?
- Are presence or levels of potential pathogens in breastmilk predictive of illness in infants or mothers?
- Are there health benefits to pasteurizing donor milk for preteen or ill infants?

Graphical Abstract 2 from *Applied Microbiology* Papers

Dietert, Coleman et al., 2021c. Nourishing the Human Holobiont to Reduce the Risk of Non-Communicable Diseases: A Cow's Milk Evidence Map Example



Evidence Map for Bovine Milk Ecosystem

Pro Supplemental Studies on Mechanisms

- Murine models, experimental systems for microimmunosomes, indirect colonization resistance
  - Kääriö 2016
  - Melnik 2016
  - Von Mutius 2016
  - Abbring 2017
  - Boudry 2017
  - Mezouar 2018
  - Müller-Rompa 2018
  - Perdijk 2018
  - Abbring 2019
  - Frei 2019
  - Butler 2020
  - Franco-Lopez 2020
  - Hufnagl 2020
  - Quinn 2020
  - Radosavljevic 2020
  - Van Esch 2020
  - Wang 2020
  - Abbring 2021

Contra Supplemental Studies on Mechanisms

- Pathogen susceptibility to innate defenses including microbiota, direct colonization resistance
  - Pricope-Cicolacu 2013
  - McCarthy 2015
  - Buchanan 2017
  - Dietert 2017
  - Schroder 2017
  - Coleman 2018
  - Perdijk 2018
  - Benmoussa & Provost 2019
  - Li 2019
  - Lima 2019
  - Melnik & Schmitz 2019
  - Wu 2019

supporting

- *MA* (**Brick 2020**) and *CSs* (**Loss 2015; Brick 2016; Schroder 2017; Müller-Rompa 2018**) Loss of protection against asthma, allergies, gut, respiratory diseases for children, adults consuming boiled or pasteurized milk; (**Butler 2020**) increased richness gut-brain modules, decreased anxiety (**Wyss 2018**); increased pulmonary function
- CSs (**House 2017; Schröder 2017; Abbring 2019; Sozańska 2019**) Raw milk, raw milk whey proteins, farm/rural environments protect against allergies, asthma, wheezing
- CS (**Wyss 2018**) Higher pulmonary function in raw milk consumers

Pro-Argument on Benefits of Raw Bovine Milk

Extensive evidence from large cohort studies on protective effects

1. Raw versus boiled or pasteurized milk, reducing incidence and severity of infectious diseases of gut and respiratory systems
2. Extensive evidence for protective effects against non-communicable diseases, including asthma, atopy, eczema, wheezing and improved pulmonary, gut, immune system functioning

attenuating

- No studies identified that attribute benefits to specific raw milk microbes or microbial consortia

supporting

- **QMRAs (FDA/FSIS 2003; EFSA 2015; Giacometti 2015a,b, 2017)** Estimate risks for raw milk associated with *Campylobacter*, enteropathogenic *E. coli*, *Listeria monocytogenes*, *Salmonella*
- **Ois (Jaros 2008; Whitehead & Lake, 2018)** Raw milks associated with outbreaks, illnesses, deaths
- **Exp** studies from Canada, Finland, Germany, Poland, US, UK (Table 1)

Contra-Argument on Risks of Infectious Diseases with Raw Bovine Milk

One early systematic review and recent QMRA reassessments and outbreak investigations

attenuating

- **QMRAs (FDA/FSIS 2003;** reassessments **Latorre 2011, Stasiewicz 2014, Buchanan 2017)** and **Rev (EFSA 2015; Berge and Baars 2020)** Limited evidence, pathogen levels, growth in milks, dose-response
- **QMRA (FDA/FSIS 2003) Oi (Whitehead & Lake, 2018) CS (Loss 2015)** Illness, mortality rates for raw milk not higher than pasteurized or boiled milks
- **Ois (Jaros 2008; Whitehead & Lake, 2018; Hanson 2019)** Pasteurized milk associated with outbreaks, illnesses and deaths

Evidence Basis

- 1 **MA;** 9 ***CSs*** including 1 experimental ***DBHPP; 7 QMRAs;*** 3 ***Ois*** including 1 ***SR;*** 2 **Rev,** 9 **Exp:** Cite consistent evidence
- 30 supplemental studies on plausible mechanisms for effects

Conclusions

1. Overall biological benefits associated with raw milk consistent across multiple large ***CSs*** and a ***DBHPP***, with supplemental studies on plausible mechanisms
2. Limited evidence for milk-borne risks of infectious diseases in children and adults for both raw and pasteurized milks

Remaining Uncertainties

- Are levels of pathogens in raw milk (and milk microbiota) predictive of risks (and benefits)?
- Is risk to children higher than adults based on current evidence and analysis?
- Is 'zero tolerance' for pathogens in raw milk scientifically, economically, ethically defensible?
- Who benefits from access to raw and pasteurized milks?
- What level of risk reductions are achievable for HACCP, cold chain, other?

Cohort of 983 Children

Raw Milk Protects Against Common Respiratory Diseases

1. Consumption of breastmilk and raw cow milk provided **comparable protective effects** against **respiratory and other infections**

2. Controlling for breastfeeding, **raw cow milk** consumption provided **protective effects** against:
   - Rhinitis ($p=0.015$)
   - Respiratory Tract Infections ($p=0.045$)
   - Otis ($p<0.001$)
   - Fever ($p=0.058$)

3. Commercial pasteurized milk was protective against fever, and Ultra High Temperature (UHT) milk and formula not protective against infections

4. **No clear associations reported for diarrhea and milk consumption**

Loss et al., **2015**. Consumption of unprocessed cow's milk protects infants from common respiratory infections. *Journal Allergy and Clinical Immunology*

Pilot Study in 11 Allergic Children

Reduced Allergenicity for Raw Milk; Likely Mechanisms

TABLE 1 Organic raw cow's milk tolerated by cow's milk allergic children

			Skin		Serum		DBPCT	
Patient	Gender	Age (y)	SPT (mm)	APT (class)	Total IgE (kU/L)	Specific IgE (kU/L)	Raw milk (mL)	Shop milk (mL)
1	M	2.65	10	++	322.0	26.3	50.0	2.0
2	M	3.52	4	++	123.0	4.2	50.0	10.0
3	M	0.55	7	+++	37.5	8.4	50.0	0.5
4	F	0.96	12	++	66.8	5.6	50.0	50.0
5	M	1.59	3	+++	nd	nd	50.0	1.0
6[a]	M	1.65	0	+	nd	nd	50.0	50.0
7[a]	M	1.09	0	+	nd	nd	50.0	50.0
8	M	0.96	5	++	98.6	12.4	50.0	0.5
9	F	0.83	7	+++	44.2	5.5	50.0	10.0
10	F	1.28	4	++	nd	nd	50.0	2.5
11	M	1.10	8	+++	nd	nd	50.0	1.0
Mean		1.49	6.7	2.4	115.4	10.4	50.0	8.6**
SEM		0.32	1.0	0.2	43.4	3.4	0.0	5.3

Note: Shown are gender, age, skin prick test, atopy patch test and serum total and cow's milk-specific IgE levels of 11 cow's milk allergic children before oral provocation as well as their level of tolerance to organic raw cow's milk and conventional shop milk during oral provocation.

Abbring et al., **2019.** Milk processing increases the allergenicity of cow's milk—Preclinical evidence supported by a human proof-of-concept provocation pilot. *Clinical Experimental Allergy*

Function of Gut Microbiota in Children:

Correlations of Phyla with Levels of SCFAs, Lactic Acid

Short Chain Fatty Acids (SCFAs; e.g., butyrate, propionate)

Children with intestinal failure (IF)

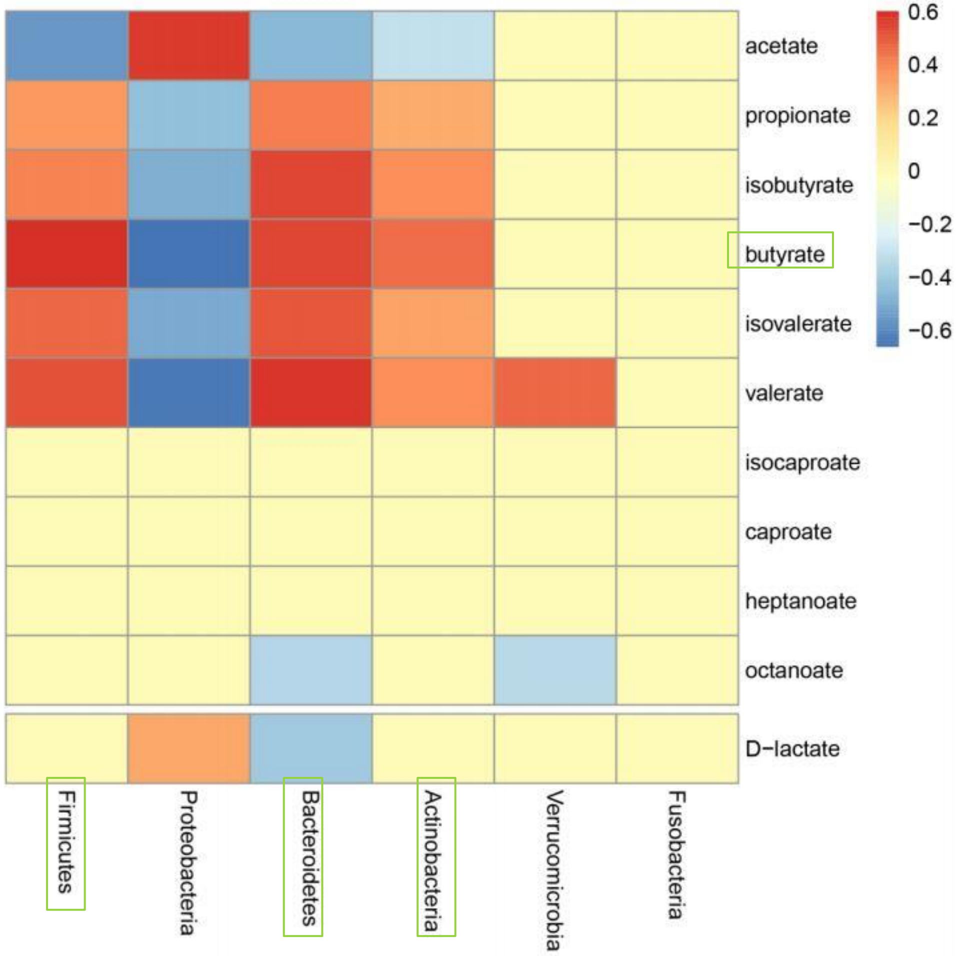
- significantly less propionic and butyric acid
- significantly more D and L-lactate

Underappreciated corrolary:

- Broadly diverse functional redundancy for gut bacteria producing SCFAs

One strategy for **'managing our microbes'** might be to replace antibiotic administration with daily doses of a **Synbiotic**

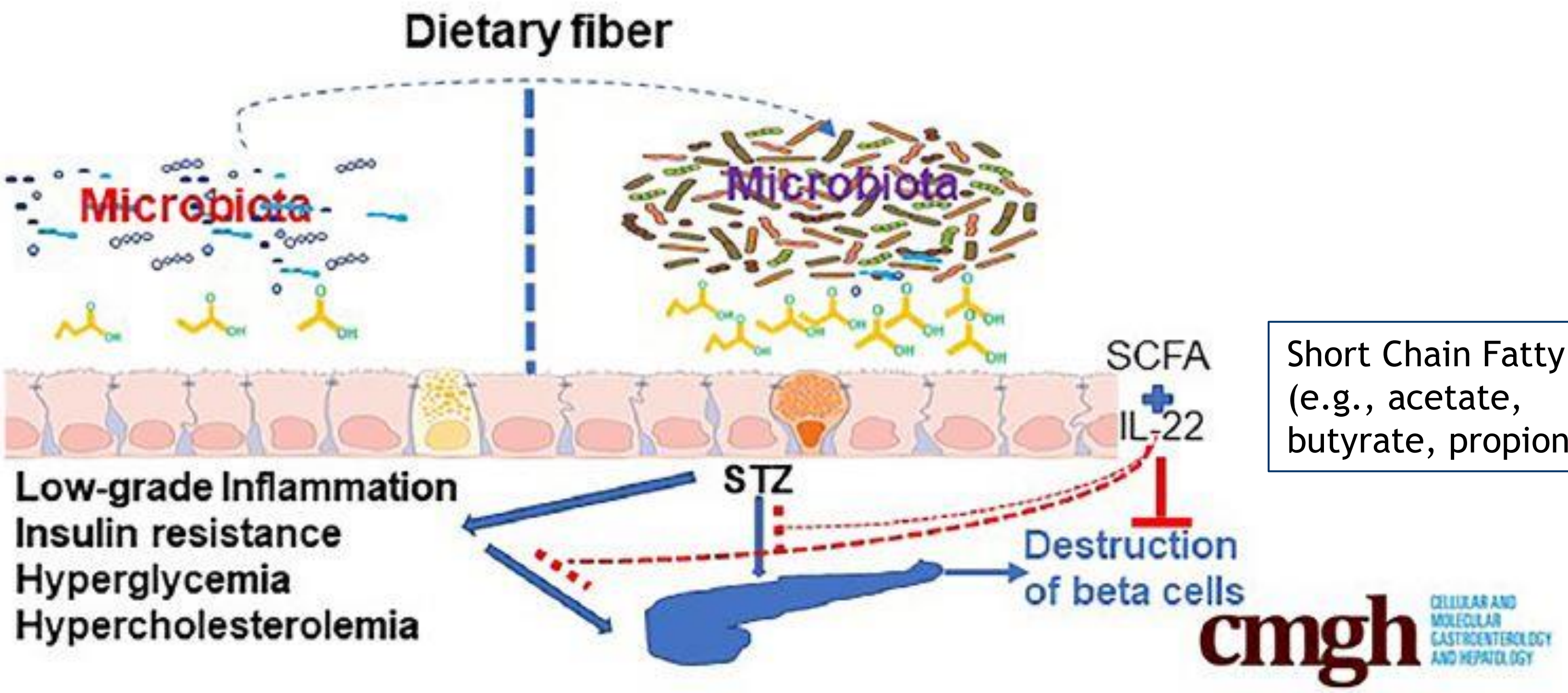
- prebiotic nutrients for SCFA-producers PLUS
- probiotic strains that effectively metabolize those prebiotics in the gut



Supplementary Figure 2. Heatmap of correlations between the main 6 phyla of the gut microbiota and short-chain fatty acids and D-lactate (both per gram dry feces).

Neelis et al., **2021.** Gut microbiota and its diet-related activity in children with IF on long-term parenteral nutrition. *Journal Parenteral Enteral Nutrition*

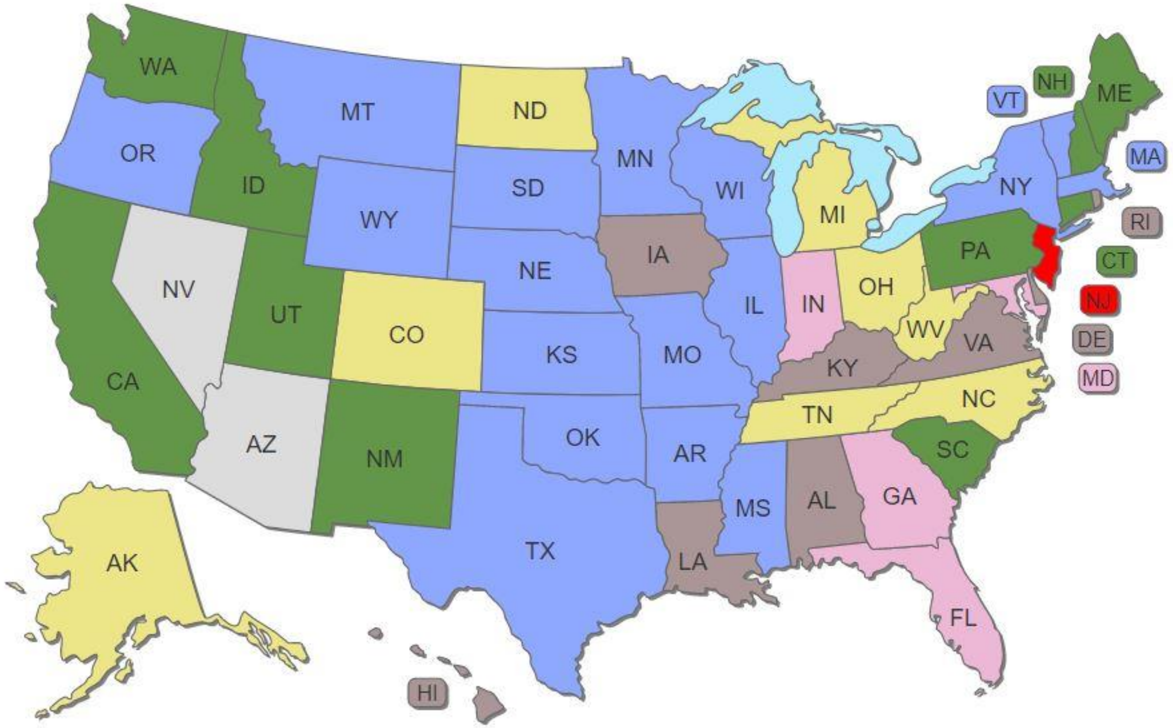
Gut Microbiota, Dietary Fiber, Inflammation, Metabolic Diseases



Short Chain Fatty Acids (e.g., acetate, butyrate, propionate)

Zou et al., **2021**. **Inulin** fermentable fiber **ameliorates type I diabetes** via IL-22 and **SCFAs** in experimental models.
Cellular and Molecular Gastroenterology and Hepatology

Daily Consumption of Raw Milks



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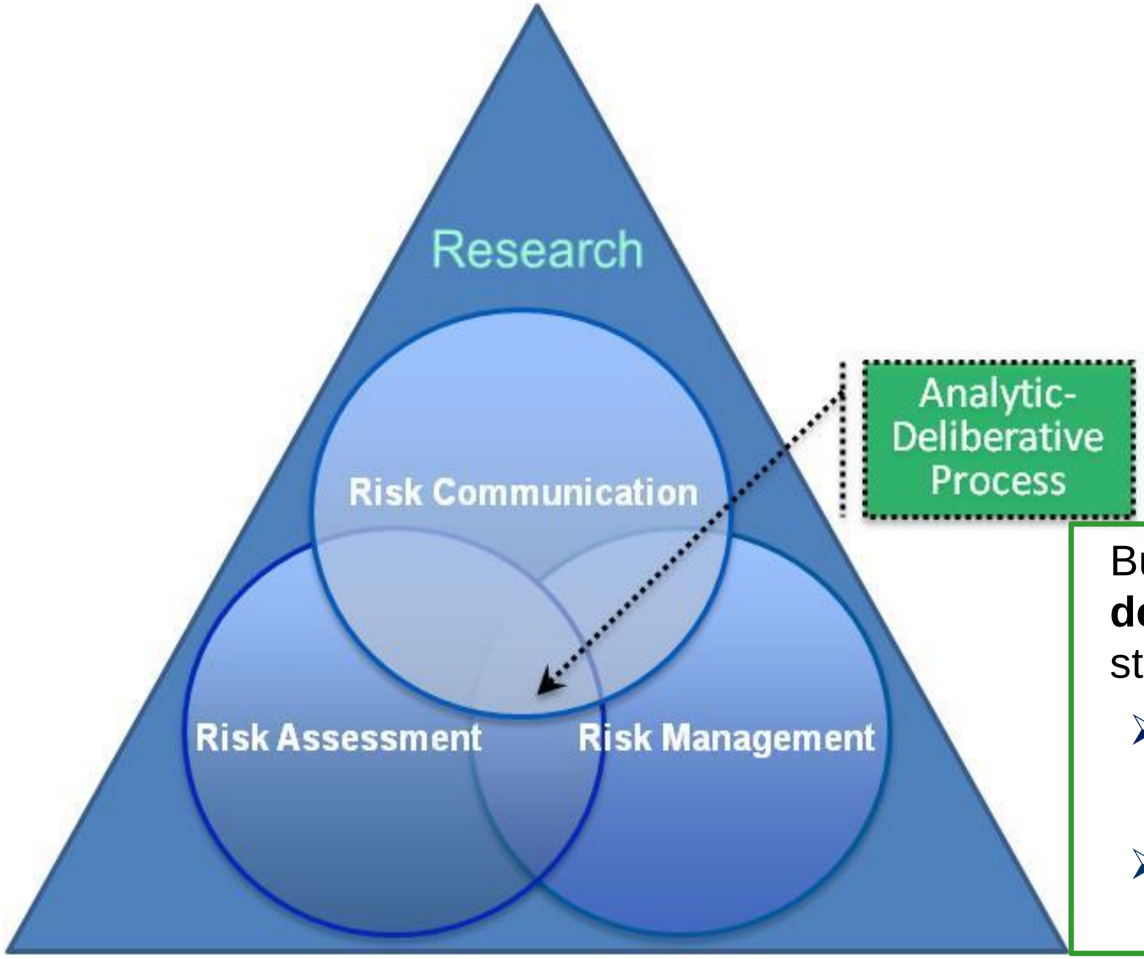
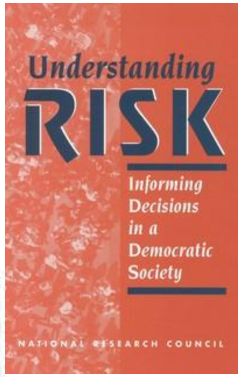
Section 5.

Incorporating Microbiota into 21st Century Risk Analysis

Dogmas from 20th Century Science, Risk Analysis, and the 'Microbiome Revolution'





Free download at National Academies Press
https://www.nap.edu/catalog/5138/understanding-risk-informing-decisions-in-a-democratic-society

Builds in cycles of **research, analysis**, **deliberation**, and **interpretation** with stakeholders on

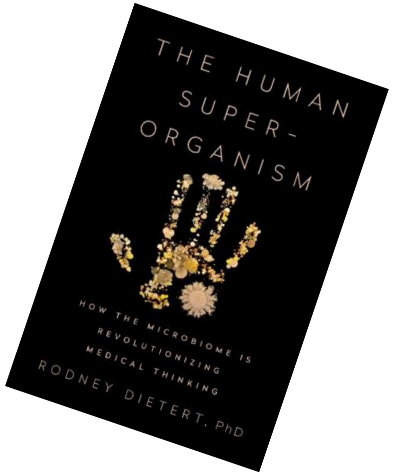
- **what goes in** (data, assumptions)

AND

- **what comes out** of risk models (**estimates of risk, uncertainty**).

Dogmas (assumptions, opinions, or perceptions) about risks that don't match up with scientific evidence warrant analytic-deliberative process.

Updating Glossary for Risk Analysts



Evolution Fueling 'Microbiome Revolution'

•***colonization resistance*** – protection of hosts with healthy microflora/microbiota against pathogens, with dose- and time-dependencies (Van der Waaij et al., 1971; Brugiroux et al., 2016)

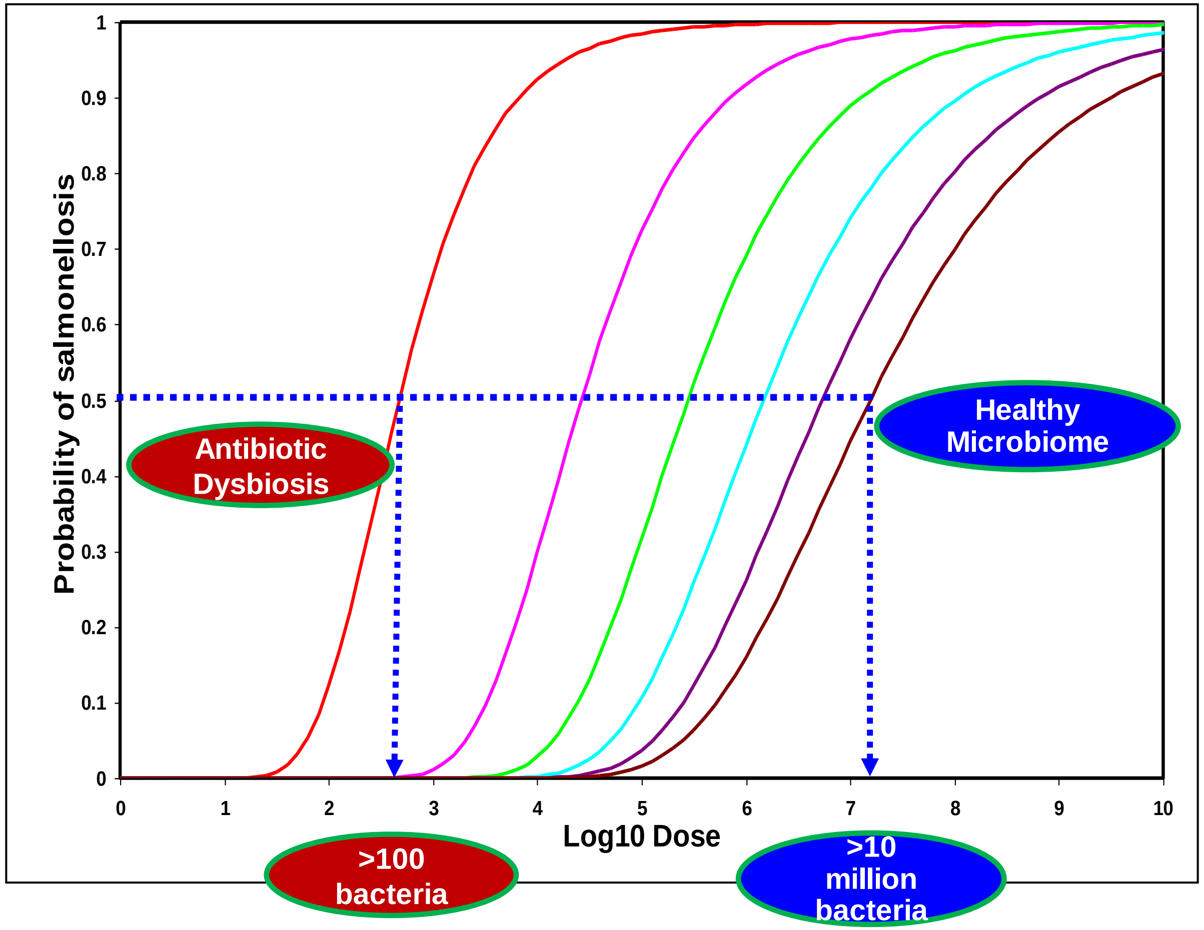
•Human Microbiome Project and Unified Microbiome Initiative beginning in 2007 and 2015, respectively, to study earth's diverse and connected microbial ecosystems

•***superorganism*** - a hybrid consortium of human and microbial communities that together, synergistically and cooperatively, regulate health and disease (Turnbaugh et al., 2007; Dietert, 2016)

Coleman et al., **2018.** Microbiota and Dose Response: Evolving Paradigm of Health Triangle. *Risk Analysis*

Gut Microbiota Impacts Dose-Response

- ID_{50} healthy volunteers after dosing with **~10^7** (ten million) *Salmonella* bacteria (**brown line**)
- ID_{50} for antibiotic dysbiosis after dosing with **~10^2** (>100) (**red line**)
- Microbiota recovers over time (2 days, pink line; 3 days, green, 4 days, aqua line, 5 days navy line)
- Indirect evidence of **10^5** (**100,000 x**) protection as magnitude of effects for **colonization resistance** (mouse and human data)



Coleman et al., **2018**. Microbiota and Dose Response: Evolving Paradigm of Health Triangle. *Risk Analysis*

Highlights of Ongoing Project on Milk Microbiota Benefits and Risks

Joint Project, Upstate NY Society for Risk Analysis (SRA), partners in **Australia/New Zealand, New England,** and **UK** on the **Natural Microbiota of Raw Milks** of human, bovine superorganisms

- **2017**: **SRA webinar series**, beginning with record-setting webinar by Rod Dietert, ***Protecting the Human Superorganism***, closing with ***Preparing to Deliberate the Evidence on Benefits and Risks*** by collaborators Warner North & Peg Coleman

- **2017-2019:** SRA round table panel symposia, presentations on evidence, data/analysis, pasteurization policies for human donor breastmilk and bovine milk

- **2019-2021**: prepared companion manuscripts on epidemiology, immunology, microbiology, and decision science for breastmilk and bovine milk

- **2021:** prepared invited manuscripts for special collection in *Applied Microbiology*

- **2022**: seeking partners for developing international workshops to deliberate evidence/knowledge gaps for **BENEFITS and RISKS** of raw milks

What Is Evidence that Pasteurizing Human Donor and Cow Milks is Beneficial to Health?

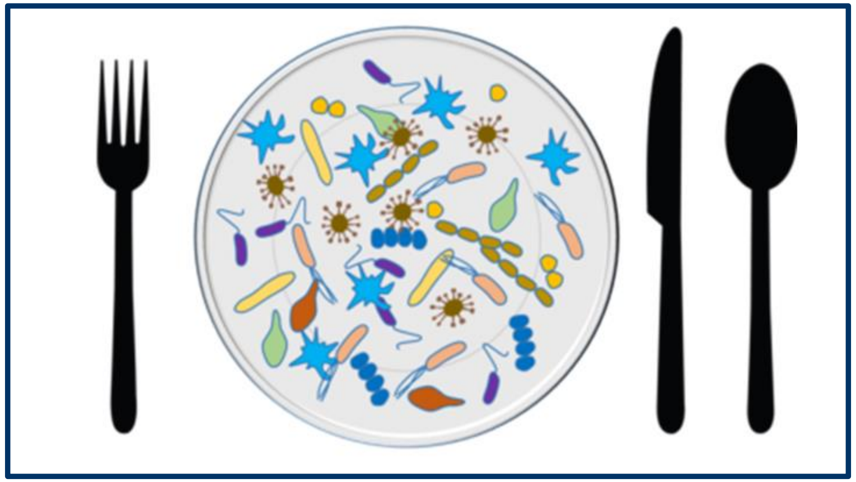
- **Human donor milk banks** pasteurize breast milk from donors because of the assumption that pathogens may be present.

- Similarly, some fear **fresh unprocessed (raw) cow milk** because pathogens may be present.

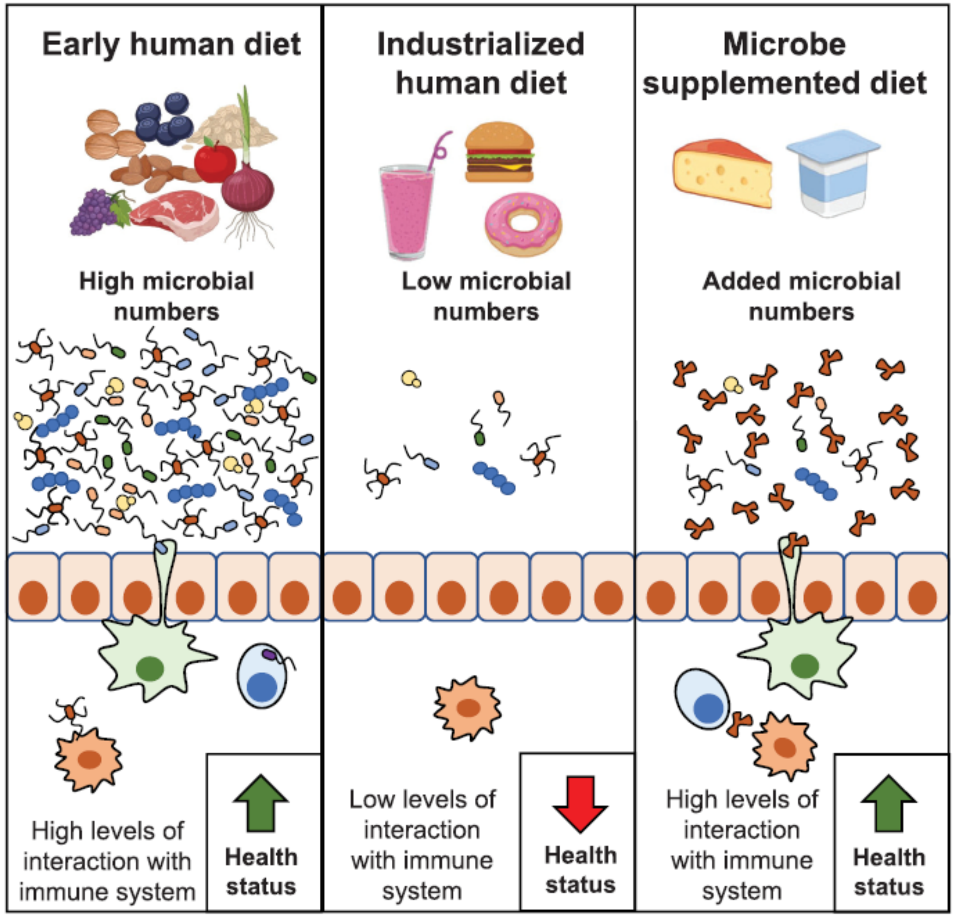
- However, **natural, beneficial microbes** (microbiota) dominate milk from cows as well as humans.

- The dense and diverse natural microbiota **outcompete pathogens, protect against illness** (provide colonization resistance), and **contribute to healthy gut, immune, neural, and respiratory systems**.
  Benefits are **lost** with **pasteurization**!

Consider *Recommended Daily Allowances for Microbes* (*RDA_M*) as for Vitamins



(Hill, 2018)



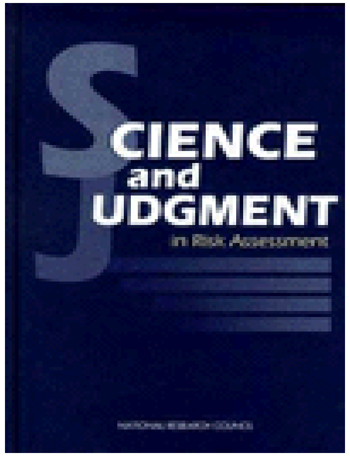
(Marco et al., 2020)

Whole Foods Contribute More than Nutrients for Human Cells

‘Managing our Microbes’ for Health and Resilience

- ## Introduce commensal competitors to pathogens (*C. difficile)*
  - Fecal Microbiome Transplant (FMT) from healthy hosts to restore colonization resistance, health, and resilience of gut ecosystem (**Durham** et al., **2020**)
  - *Clostridium scindens* (**Parkar** et al., **2021**) or other probiotics as biological detoxification tools to suppress *C. difficile* germination and toxic effects, **increase microbiome diversity** and **gut** and **immune health**
- ## Modify gut ecosystem functionality and integrity
  - Enhance **butyrate-producing microbiota** to enhance protection against GI disease (**Neelis** et al., **2020**) and **respiratory viral infections** (**Haak** et al., **2018**)
- ## Modify diet to increase gut diversity and richness and shift commensal-pathogen competition for nutrients
  - **High-fiber diets** and **prebiotics** increased production of secondary bile acids and/or SCFAs and reduced **obesity** and metabolic diseases including **type 2 diabetes** (**Zhang** et al., **2020**)

Science? Opinion? Outdated Dogma?



Fact?

Fiction?

Possibility?

Judgment?

Evidence from 21st century science challenges opinions, outdated dogma and misinformation

- **Presence** of bacteria alone **insufficient** to predict **responses** (**beneficial OR adverse**)
- **Doses** (amounts) of **beneficial AND pathogenic** bacteria ingested **matter**
  - **Dose - Response** curves **simulate** foodborne illness
  - **Effects** (**beneficial** and **adverse**) increase with increasing doses (**natural microbiota** and **pathogens**)
- **Microbiota matters**, protects against pathogens (**colonization resistance**)

Suggested Reading

Margaret E. (Peg) Coleman and Colleagues, *Risk Analysis*, *Applied Microbiology*, and More

- **1996**: National Research Council (NRC) Understanding Risk: Informing Decisions in a Democratic Society.
- **1998**: Marks, H.M.; Coleman, M.E.; Lin, C.T.J. Topics in Microbial Risk Assessment: Dynamic Flow Tree Process. Risk Anal. 18, 309–328.
- **2011**: Wiedemann, P.; Schütz, H.; Spangenberg, A.; Krug, H.F. Evidence Maps: Communicating Risk Assessments in Societal Controversies: The Case of Engineered Nanoparticles. Risk Anal. 31, 1770–1783.
- **2018**: Coleman, M., Elkins, C., Gutting, B., Mongodin, E., Solano-Aguilar, G., Walls, I. Microbiota and Dose Response: Evolving Paradigm of Healt Triangle. Risk Anal. 38, 2013–2028.
- **2020**: North, D.W. Risk Analysis, Decision analysis, causal analysis, and economics: A personal perspective from more than 40 years experience. Risk Anal. 40, 2178–2190.
- **2021a**: Coleman, M.E., North, D.W., Dietert, R.R., Stephenson, M.M. Examining Evidence of Benefits and Risks for Pasteurizing Donor Breastmilk. Applied Microbiology 1(3):408-425. https://doi.org/10.3390/applmicrobiol1030027.
- **2021b**: Coleman, M.E., Dietert, R.R., North, D.W., Stephenson, M.M. Enhancing Human Superorganism Ecosystem Resilience by Holistically 'Managing Our Microbes'. Applied Microbiology. 1(3): 471-497. https://doi.org/10.3390/applmicrobiol1030031 .
- **2022**. Dietert, R.R., Coleman, M.E., North, D.W., Stephenson, M.M. Nourishing the Human Holobiont to Reduce the Risk of Non-Communicable Diseases: A Cow's Milk Evidence Map Example. Applied Microbiology. 2(1):25-52. https://doi.org/10.3390/applmicrobiol2010003.
- **2022**. North, D.W., Coleman, M.E., Hull, R.R. Need for International Workshops to Deliberate Evidence of Benefits and Risks of Raw Milks. Accepted in Corpus Journal of Dairy and Veterinary Science.

Rodney R. Dietert, Collaborator and Emeritus Professor of Immunotoxicology, Cornell University

- **2016**: Dietert, R.R. The Human Superorganism: How the Microbiome Is Revolutionizing the Pursuit of a Healthy Life; Dutton: New York, New York.
- **2015**: Dietert, R.R.; Silbergeld, E.K. Biomarkers for the 21st Century: Listening to the Microbiome. Toxicol. Sci. Off. J. Soc. Toxicol. 144, 208–216.
- **2017**: Dietert, R.R. Safety and Risk Assessment for the Human Superorganism. Hum. Ecol. Risk Assess. 23, 1819–1829.
- **2018**: Dietert, R.R. A Focus on Microbiome Completeness and Optimized Colonization Resistance in Neonatology. NeoReviews 19, 78–88.
- **2021**: Dietert, R.R.; Dietert, J.M. Twentieth Century Dogmas Prevent Sustainable Healthcare. Am. J. Biomed. Sci. Res. 13, 409–417.

Trans-disciplinary Risk Analysis in 21st Century: Holobiont Theology?

- Recent dissertation from Duke Divinity School challenges our knowing, what we think we know, and what ‘**troubles**’ us as risk analysts
- **Aminah Al-Attas Bradford** (2021)
  - **Symbiotic Grace: Holobiont Theology in the Age of the Microbe**
  - **The microbiome**
    - ‘**troubles**’ modern **Anthropology**
    - ‘**troubles**’ modern **Theology and Philosophy**
      - Thomas Aquinas, theologian and Scholastic philosopher; method of learning by dialectical reasoning to extend knowledge by inference and to resolve contradictionsism; much of modern thinking ‘developed or opposed his ideas on ethics, natural law, metaphysics, and political theory’
    - also ‘**troubles**’ modern **Risk Analysis!**

Questions? Comments? Interested Partners?

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