Microbes, microbiota and microbiomes; their role in humans, animals, plants and the environment.

Stanley Brul @ Swammerdam Institute for Life Sciences; University of Amsterdam

Acknowledgements: Jianbo Zhang, Meike Wortel, Age Smilde, Marten Smidt (Science Faculty UvA), Hilde Herrema, Max Nieuwdorp, Wouter de Jonge, Anja Lok (Amsterdam UMC), Hauke Smidt (WUR)
Human host more bacterial cells than human cells?

- Human body is composed of ten trillion \( (10^{13}) \) cells
- There are hundreds of trillion \( (10^{14}) \) bacterial cells in and on our bodies
- Collectively, these bacteria have 100X more genes than all the genes in the human genome \((\sim 25000)\)
Open questions:

Where are the microbes and what do they do?

Are there common elements between Human, animal, Plant and environmental consortia?

How do we analyse the complex microbial consortia?
Where are the microbes and what do they do?

Are there common elements between Human, animal, Plant and environmental consortia?

How do we analyse the complex microbial consortia?
Where are microbes in and on our body?

Some microbes are native, normally found in the body:
- Nose
- Mouth
- Skin
- Gastrointestinal
- Urogenital

Some microbes are introduced, suddenly arriving at a new residence in the body:

Throughout your life, microbes secrete compounds that regulate immune cells (T cells)
All these trillions of bacteria in the human gut are together with the other microorganisms (including viruses) called our microbiota.

Joshua Lederberg (Nobel Price Medicine, 1958) first introduces the term ‘microbiome’ in 2001.

‘collection of genomes of microbiota living in a specific niche’

Humans consist of multiple organisms

Up to 2kg per person!
For an 80kg person, this is 2.5% of total body weight

At least 1000 different bacterial species currently known
Many are yet to be identified
Interindividual differences are large

Gut bacteria express over 2.5 million genes
100 times more than human genes

Based on DNA content, we are as much (or more?) microbial as human

Figure adapted from Magnus Simrén, Gut, 2013
Gut microbiota: crucial symbionts that constitute an endocrine organ

- **Increase energy harvest from diet**
  - Secrete carbohydrate degrading enzymes

- **Produce metabolites with host metabolic effects**
  - Short-chain fatty acids (SCFA’s) → Regulate gut hormones
  - Trimethylamine (→ TMAoxide in liver with pro-atherosclerotic effects)

- **Produce vitamins**
  - (biotin, B12 and vitamin K)

- **Metabolize** bile acids, sterols and xenobiotics*
  - * Drugs such as antidiabetic drug metformin

- **Train host immune system**
  - Protect from pathogen invasion
  - Maintain healthy gut barrier function

*Paradigm Shift in Medicine: No Bacteria or Not The Right Consortium May Cause Disease.*
  *Potential for Diagnostics & Therapy.*

Gut microbiota in human health and disease

Gut microbiota implicated in host health and development of disease

Microbiota research is moving from an associative to a translational/ causative science (C. diff. is the best proven case)

Multidisciplinary endeavor
- Wide range of expertise and skill sets (Big Omics Data Analysis!)
Gut microbiota and human disease; Association or Causality?

Causality in infectious diseases are described by Koch’s postulates:

- Micro-organism should be associated with disease(s)
- Micro-organism should be found in a sick person
- Cultured micro-organism introduced in animal drives disease
  - or eradication prevents disease

Fecal microbiota transplantation (FMT) as tool to study causality of gut microbiomes
Insulin sensitivity response and bacterial colonization after FMT

SNV (single-nucleotide analysis) = method to distinguish and trace donor- and recipient-specific strain populations

- No ‘super-donors’
- Enhance colonization?
- Lower diversity at baseline more likely to benefit
- Predict success of FMT using computational approaches
- Note that the small intestine is exposed to fecal/colonic microbiota that it normally never sees!
Better understanding of the gut ecosystem microbial class diversity might increase FMT efficacy (and other microbiome-targeted interventions)

- Bacteria
- Archaea
- Viruses
  - Eukaryotic viruses (1%)
  - Bacteriophages (99%)
- Fungi
- Parasites
- Spores
- Metabolites
“All Disease Begins in The Gut” (?) A statement by Hippocrates 460-377 bc.

Role microbiome:

Causal

- Course of disease
- Responders-non responders
- Influence of genetics and diet
- Multiple FMT or real failures
- Concomitant medication

Disease modifier

or...consequence of disease?

- C. difficile
- Multidrug resistant bacteria
- IBD/IBS
- Insulin resistance/T2DM
- Autoimmune diseases (T1DM /hypothyroidism)
- Gut-brain axis diseases (anorexia /autism)

Longitudinal integrated cohort studies needed to get clues towards temporal events in health perturbation.

In Amsterdam the HELIUS cohort exists which was started from a multiethnic perspective on health.

http://www.heliusstudy.nl/
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Where are the microbes and what do they do?

Are there common elements between Human, animal, Plant and environmental consortia?

How do we analyse the complex microbial consortia?
Intestinal microbiota are influenced by a broad range of diet & environmental factors!
Are there common elements among the different microbes? Look at us and our environments as ONE system, ONE health!

A Growth fund proposal in the making:
The HoloMicrobiome
Leveraging the microbiome for economic growth

Home-en | Holomicrobiome (holomicrobioom.nl)

Joining forces between existing research groups and research programs in the Netherlands (UvA, WUR, RUG, UMCs TNO, Companies etc........)
Knowledge and Innovation are the connecting flywheel for the realization of our mission and is an overlapping activity for all domains.

Partnership Industry is crucial for creating impact, valorization and realization from Knowledge and Innovation.

Partnership Policy guarantees the implementation of a policy regarding the realization of a healthy, safe and sustainable food chain for the Netherlands.
Separate microbiomes

The Holomicrobiome
Four linked example projects

**Human**
- Gut-microbiome (disease)
- Environmental microbiome
- Health increase
- Lower costs
- Lower use of antibiotics

**Animals**
- Dietary strategies
- Farm animal health
- Reduced emission of Methane and N2
- No use of antibiotics

**Plant**
- Farming strategies
- Soil microbiome
- Plant root microbiome
- Higher yield, lower costs
- Lower use of pesticides etc.

Linking the domains:
- Building a new supercohort with stored information on nutrition and food related microbes in all different domains
- Transfer of microbiome (and related chemicals)
- Causal Models for strategic interventions

**PIG-PARADIGM International research team to combat antibiotic resistance in pigs.**
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How do we analyse the complex microbial consortia?
Gut microbiome from association to mechanisms associated with health and disease

Combine forces, share knowledge

How? – Microbiota Center Amsterdam

Study design → Sample processing → Sequencing → Data analysis + interpretation

What can we learn from this?
- Predictive components?
- Signal identification
Microbiota Center Amsterdam (MiCA)

*Expertise and equipment in-house*

- **Research question/study design**
- **Patient folders**
  (Equipment located at EVG, CEMM, Tytgat and LVGA)
- **Sample processing**
- **Sequencing**
  (MiSeq EVG, located at LVGA)
- **Data processing, analysis, interpretation**
  (EVG)
- **Anaerobic chamber**
  (Located CEMM tower)
Microbiota Center Amsterdam (MiCA)
“powered by Amsterdam UMC location AMC”

Joost Wiersinga  Wouter de Jonge  Max Nieuwdorp  Hilde Herrema

Theo Hakvoort  Jorn Hartman  Mark Davids  Evgeni Levin

Iris Admiraal  Xanthe Verdoes
Host-Microbiome Interactions in the laboratory and beyond

How to study their influence?

- **Clinical studies**
  - (e.g. case-ctrl, FMT)

- **Animal models**
  - (e.g. Germ-free mice)

- **Simulator of fermentation**
  - (e.g. PolyFermS; SHIME)

- **Physiomimetic systems**
  - (e.g. gut-on-a-chip; HMI; GuMI)

Understanding by recreating Host-Microbiome (Microbiota) interactions in a miniature human gut

Dr. Jianbo Zhang (SILS-UvA formerly ETH and MIT)
The Wish List (Roadmap)

- Controlled ANAEROBIC
- apical flow

- microbiome = super strict anaerobic / commensals + pathogens (imaging)

- Primary Epithelial – stromal-immune tissue
- crypt / lumen structure
- Lymph drainage

- Perfusable microvascular network with circulating immune cells
Gut-MIcrobe (GuMI) physiomimetic system

Zhang, Huang et al, Med, 1: 1-25. 2021. in collaboration with Dr. David Trumper @MIT
Workflow to study microbial impact on gut inflammation

Zhang et al. Nat. Protocols, 2021
Bacterial growth in the GuMi model system

Super oxygen-sensitive bacteria grew

GuMI maintains barrier function and supports bacterial growth

Faecalibacterium prausnitzii growth reached the plateau after 24 h.
Epithelial barrier function is maintained despite the presence of highly dense bacterial cells.
Progress in understanding the molecular basis of host-microbe interaction: Short chain fatty acids/ amino acids
Progress in understanding the molecular basis of host-microbe interaction; tryptophan metabolism and serotonin
Progress in understanding the molecular basis of host-microbe interaction; proline metab., glutamate and GABA

DOI: https://doi.org/10.1016/j.cmet.2022.04.001 (Cell Metabolism)
Bacterial spores; the good……& the bad

Culturing of ‘unculturable’ human microbiota reveals novel taxa and extensive sporulation

Hilary P. Browne1*, Samuel C. Forster1,2,3*, Blessing O. Anonye1, Nitin Kumar1, B. Anne Neville1, Mark D. Stares1, David Goulding4 & Trevor D. Lawley1

Visualication of SpoVAEa Protein Dynamics in Dormant Spores of Bacillus cereus and Dynamic Changes in Their Germinosomes and SpoVAEa during Germination

Yan Wang,* Norbert O. E. Vischer,* Demi Wekking,* Alessandra Boggian,* ©Peter Setlow,* ©Stanley Brul4

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Organization and dynamics of the SpoVAEa protein and its surrounding inner membrane lipids, upon germination of Bacillus subtilis spores

Juan Wei1, Norbert O. E. Vischer1, Arndt L. de Vos1, Erik M. M. Manders1, Peter Setlow1 & Stanley Brul4

https://systemsbiology.amsterdam/
Sporulation cycle of *Bacillus cereus*

Germination and outgrowth:

Sporulation:
ANTIMICROBIAL RESISTANCE (AMR)

The group linked to the NVWA chair @ SILS-UvA (Benno Ter Kuile) works on:

- **Plasmid-mediated** → transfer of antibiotic resistance genes carried on plasmids (horizontal gene transfer)

- **Antibiotic-induced** → sub-lethal antibiotics exposure leads to mutagenesis with acquisition of resistance (vertical gene transfer)

Can be extended to real life (microbiome/ microbiota) settings!
Model development on individual level and its impact on a biological system; example at the Centre for Urban Mental Health @ UvA (Van der Wal et al. Lancet 11, 991, 2021)

Figure 2: Conceptual framework of the relationship between factors in the urban environment and CMD outcomes

This framework conceptualises urban mental health from a complex systems perspective. Meta factors (grey box) are considered to have a dynamic effect on the urban environment and its inhabitants. Factors are categorised as urban factors (e.g., air pollution or built environment), social factors (e.g., social cohesion), or individual factors (e.g., individual demographic, psychological, or neurobiological characteristics). Arrows between the factors and CMD symptoms represent the different temporal scales across which factors can assert their effect. CMDs are represented as symptom clusters connected by overlapping symptoms. Feedback arrows represent the possibility of feedback from CMD symptoms to explanatory factors, which can also occur over different timescales. CMD = common mental disorder.
Final conclusions

- Causality of gut microbiota in many human (autoimmune) diseases is not known.

- Fecal microbiota transplantation studies suggest a role for specific bacterial strains in disease including diabetes, inflammatory bowel disease and under- or overweight

- *Future:* personalized diet (based on gut microbiota composition) in combination with novel probiotic-medication (synergy) to improve human (autoimmune) diseases.

- The gut harbors an ecosystem with (spore forming) adapted microbes! Study of (transkingdom) interaction between its members is important to understand microbiota-related pathologies and to design effective interventions.

- Miniature Gut models can recreate important physiologically relevant environments to help understand the function of gut microbiota.

- Unique and extensive physiological knowledge on AMR and bacterial spore formers, prime inhabitants of the Gut and highly represented in the gut microbiota is available.

- Extensive modelling knowledge infrastructure needed at different time and length scales. @UvA an example is available at the Centre for Urban Mental Health.
Discussion and queries
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Timeline as of April 2022:

- Pre-proposal: Oktober 2022
- Full application: January 2023
Task Force; Consortium core; Extended consortium

Marten Smidt (UvA, FNWI, SILS): Project leader
Harro Bouwmeester (UvA, FNWI, SILS): Plant/root domain
Hauke Smidt (WUR): Animal microbiome
Age Smilde (UvA, FNWI, SILS): Modeling, data analysis
Stanley Brul (UvA, FNWI, SILS): General microbiology and fermentation
Gerard Muijzer (UvA, FNWI, IBED): Environmental microbiology
Max Nieuwdorp (AUMC): Human Microbiome, health
Hilde Herrema (AUMC): Human Microbiome, health

“External”
Helen Bergman Quartermaster
Peter Vermij Scientific writer

Extended Consortium has a great number of representatives from UMCs, universities, Knowledge Institutes, industries, Ministries.
Task Force; Consortium core; Extended consortium

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