

## Long-covid, post-acute infectious syndromes (PAIS) and the microbiome

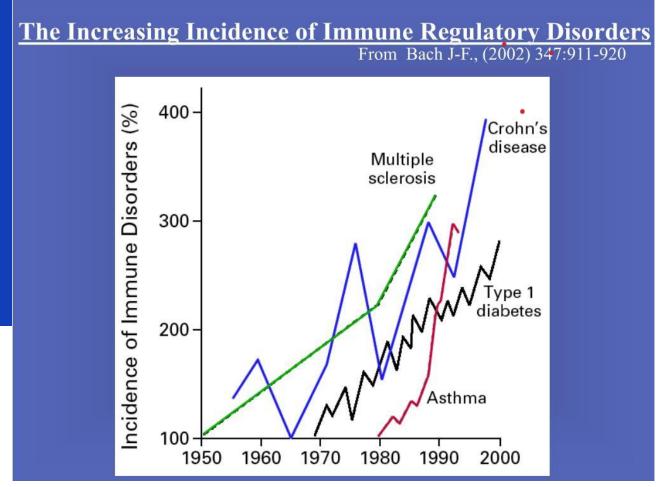
There is sufficient reason to treat

## Gut microbe-host interactions in Long-COVID

Roy Montijn



### **History of Chronic Diseases**



Bach JF. The effect of infections on susceptibility to autoimmune diseases and allergic conditions. *N Engl J Med*. 2002

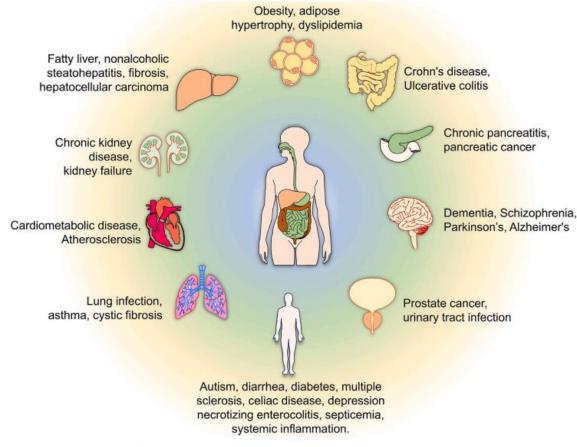


Fig. 1. Human chronic diseases with direct or indirect intestinal etiology.

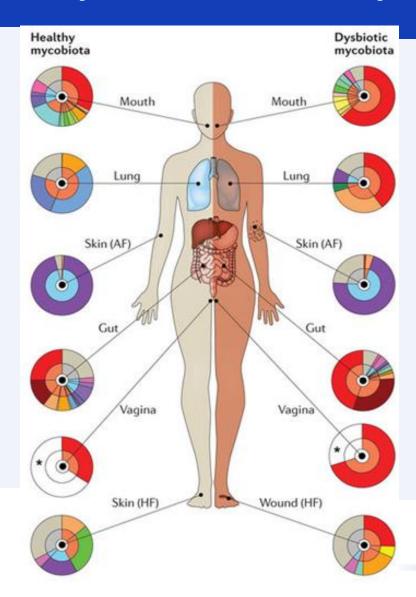




### **Healthy Microbiome vs Dysbiosis**

### A healthy microbiome is balanced and self-regulating:

- High diversity of healthy bacteria
- Many important functions present
- Interaction with organs
- Good alignment with the immune system
- Ability to control pathogenic microorganisms



#### Dysbiosis is a disruption of the balance of the composition and function of the microbiome, characterized by:

- Fewer types of healthy bacteria, leading to loss of healthy functions
- Disturbed immune responses
- Disrupted interactions with organs
- Overgrowth of pathogenic microorganisms
- Association with chronic diseases



### Functions of the Microbiome

- Food metabolism (SCFA, vitamins, neurotransmitters)
- Protective functions
   (bacteriocins, competition)
- Development of structural functions (villi, crypts, tight junctions)
- •Immune-modulating functions



Food metabolism



Epithelial protection



Modulation of the nervous system





Immune development



Metabolism of therapeutics



Protection against pathogens



### **Causes of Dysbiosis**



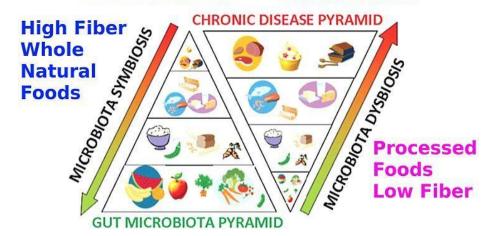
Lifestyle





**Diet** 

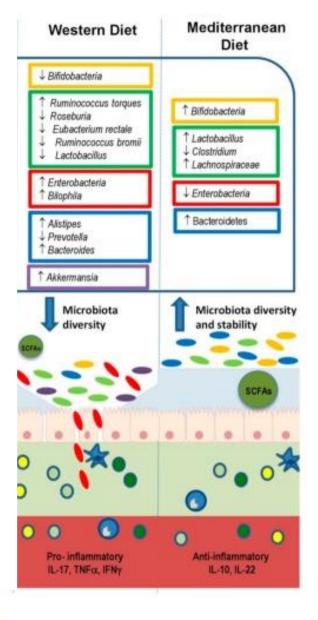
#### Mediterranean vs Western Diet



### **Environmental Exposures**

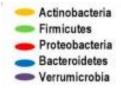






Effects of Different Types of Nutrition on Gut Microbiota, Mucus Layer, and Immune Cells

Rinninella E, et al. Nutritional components and dietary habits: keys for a healthy gut microbiota composition. Nutrients. 2019









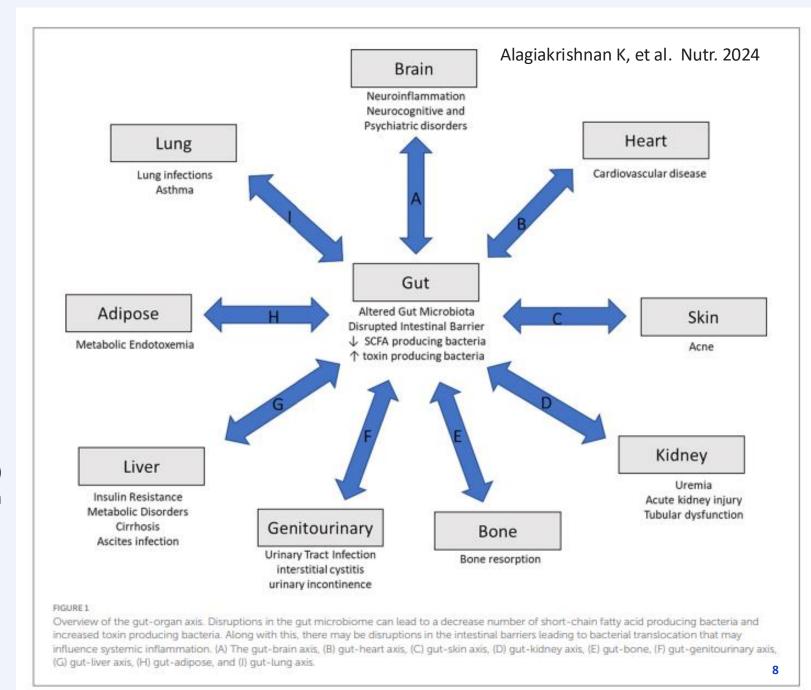


Fecal variations microbiota of healthy adults following GFD over one month.

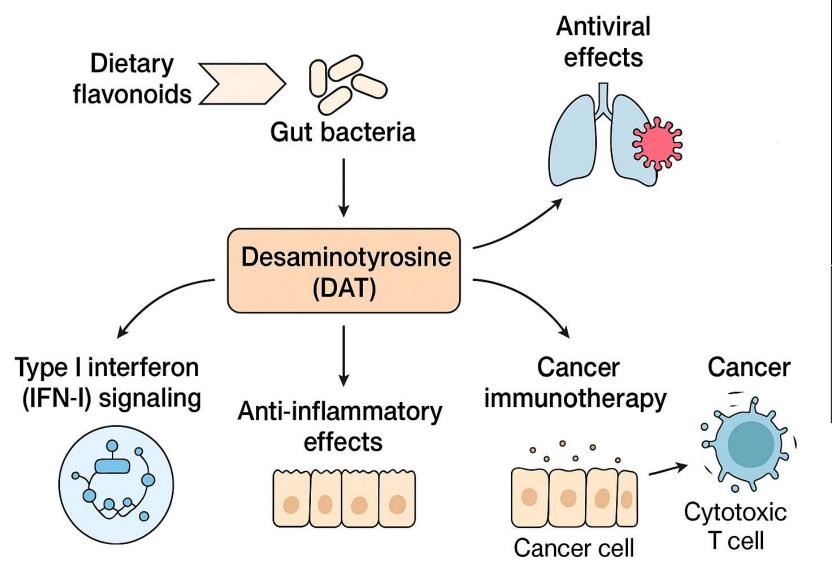
### **Gut-Organ Axis**

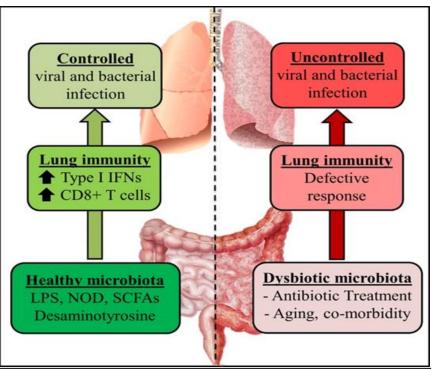
### beneficial gut metabolites:

- Short-chain fatty acids (SCFAs): Acetate, Propionate, Butyrate
- Amino acid-derived metabolites: Indoles, phenols, amines, branched fatty acids
- Lipid-derived metabolites: Glycerol derivatives, choline metabolites (e.g., TMAO)
- Vitamins and cofactors: B vitamins (e.g., B12, biotin, folic acid)
- Polyphenol metabolites (e.g. desaminotyrosine)
- Gas metabolites: Hydrogen, methane, hydrogen sulfide
- Neuroactive substances: GABA, Serotonin precursors, tryptamine
- Secondary bile acids



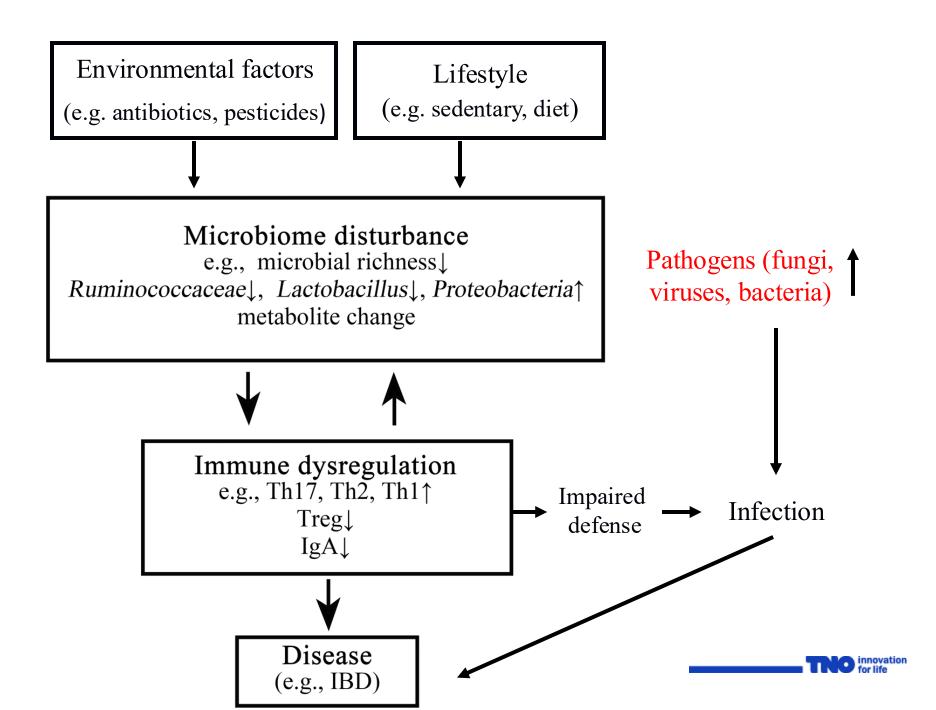
### **The Gut-Lung Axis**



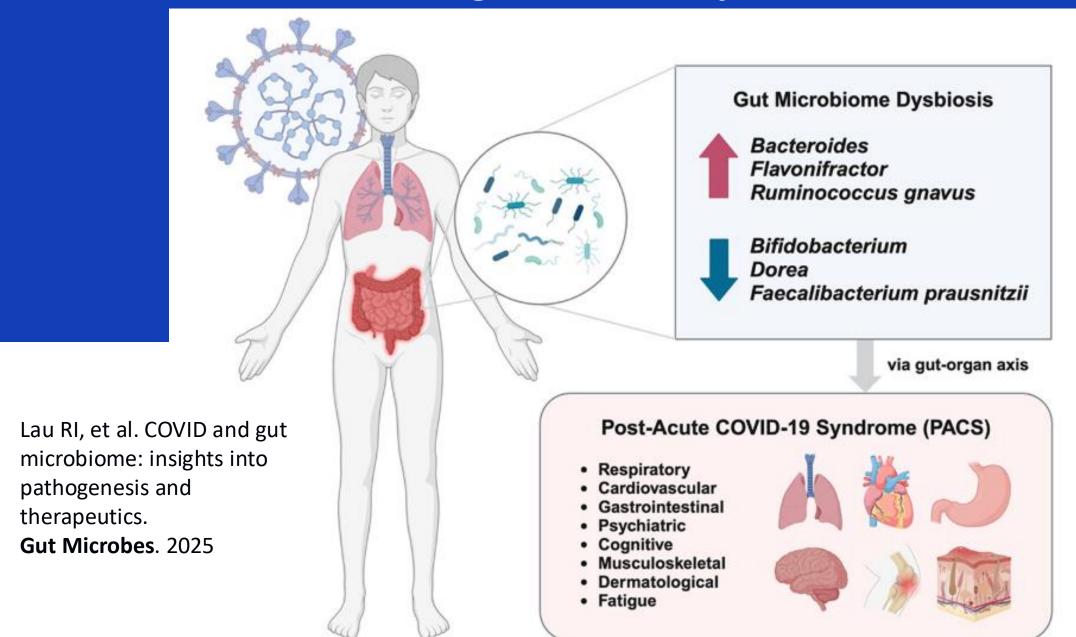


Göttert S, et al. Nat Commun. (2025)
Sencio et al. Mucosal Immunol (2021)
Steed et al. Science (2017)
Rosshart et al. Cell (2017)
Ichinohe et al. PNAS (2011)

## Dysbiosis and Chronic Diseases



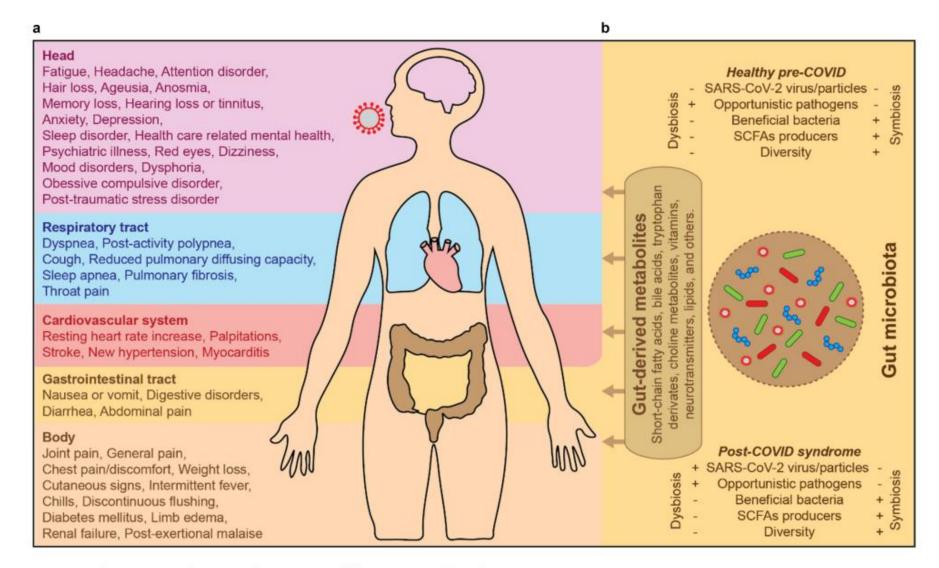
### **Long-Covid and Dysbiosis**





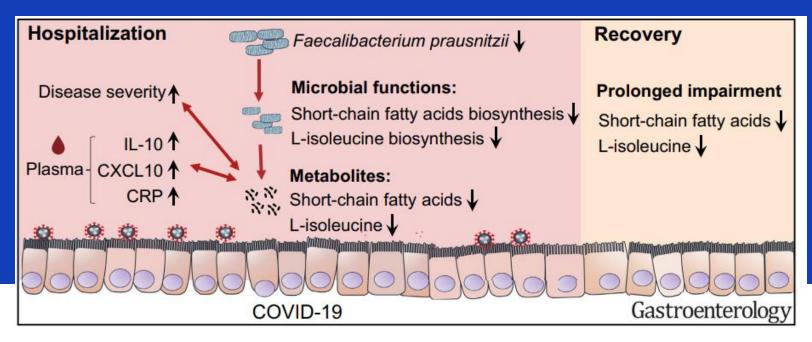
#### Gut microbe—host interactions in post-COVID syndrome: a debilitating or restorative partnership?

Torsten P. M. Scheithauer, Roy C. Montijn, and Arnout Mieremet. *Gut Microbes*. September 2024





### Production of SCFA and L-Isoleucine is low in Long-Covid





Prolonged Impairment of Short-Chain Fatty Acid and L-Isoleucine Biosynthesis in Gut Microbiome in Patients With COVID-19.

Fen Zhang et al., Gastroenterology 2022



### **Health Benefits of Butyrate**

#### 1. Digestive System Health:

- It serves as the primary energy source for the cells lining your colon, meeting approximately 70% of their energy needs
- supporting the gut barrier function

#### 2. Immune System Support:

- Butyrate signals to your immune system that the gut bacteria are within the desirabl range.
- When butyrate levels are low due to inadequate fiber intake, the body may trigger a inflammatory reaction

#### 3. Reducing Inflammation:

 Butyrate has been described as a potent pro-resolution molecule. It helps regulate inflammation in the gut and beyond.

### 4. Cognitive Function and Insulin Resistance:

butyrate contribute to better cognitive function and lower rates of insulin resistance

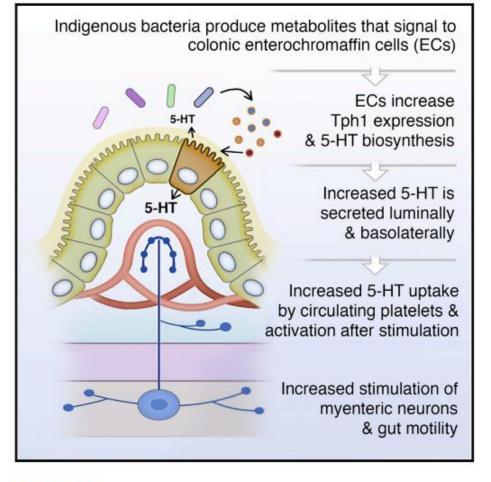
### The gut microbiome bacteria regulate serotonin (5-HT) levels

• Indigenous Bacteria from the Gut Microbiota Regulate Host Serotonin Biosynthesis.

Jessica M Yano et al. April 9, 2015 Cell

Serotonin reduction in post-acute sequelae of viral infection.

Wong et al., Cell October 26, 2023



#### Highlights

- Gut microbes regulate levels of 5-HT in the colon and blood
- Spore-forming bacteria modulate metabolites that promote colon 5-HT biosynthesis
- Microbiota-dependent changes in 5-HT impact GI motility and hemostasis
- Altering the microbiota could improve 5-HT-related disease symptoms

### Serotonin; a hormone and a neurotransmitter

- 1. Mood Regulation
- 2. Sleep
- 3. Digestive System
  - Most of the body's serotonin resides in the digestive system.
  - It affects bowel function and controls bowel movements.
- 4. Blood Clotting:
  - Blood platelets use serotonin to aid in wound healing.
  - It encourages blood clot formation and narrows tiny arteries.
- 5. Appetite regulation.
- 6. Bone Health
- 7. Sex Drive
- 8. Learning and Memory
- 9. Cognition



### Healthy Levels of Butyrate and Serotonin

BENEFIT	EFFECT
Gut Health	Improved integrity and reduced inflammation
Immune System	Strengthened and anti-inflammatory response
Metabolic Regulation	Cholesterol control, blood sugar stabilization
Mental Health	Better mood, sleep regulation, cognitive support
Weight Control	Appetite regulation and weight management

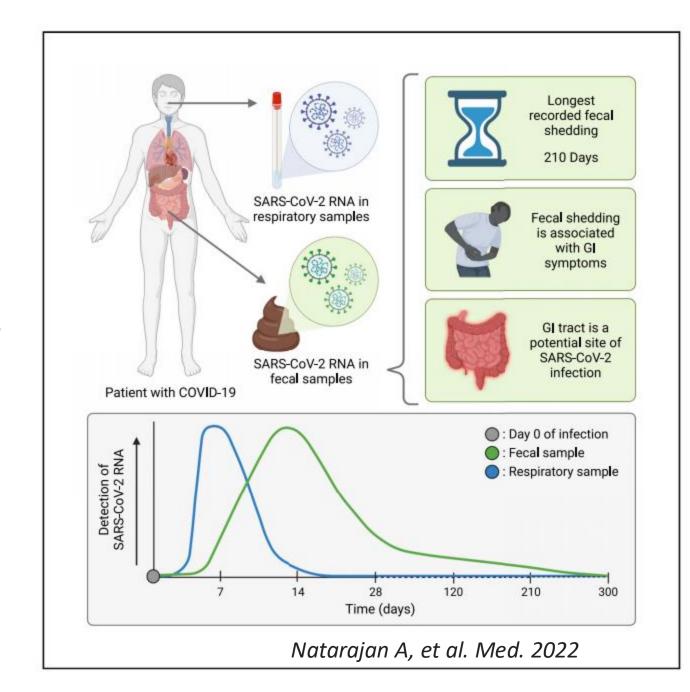
### Symptoms Linked to Dysbiosis and Low Butyrate and Serotonin levels

SYMPTOMS	POSSIBLE CAUSE
Fatigue	Low energy substrate availability
Concentration Problems	Reduced serotonin affecting cognitive function
PEM	Metabolic imbalance and inflammation
Muscle Pain & Weakness	Inflammatory response and poor nutrient absorption
Sleep Problems	Serotonin dysregulation
GI Complaints	Compromised gut barrier and dysbiosis

### SARS-CoV-2 can persist in the gastrointestinal tract

- Gastrointestinal symptoms (like diarrhea) in some COVID-19 patients.
- Prolonged fecal shedding of viral RNA, suggesting the virus remains active or detectable in the gut for an extended period.
- This implies the gut could act as a reservoir for infection, which may influence transmission, disease severity, and long-term effects.

Neurath, et al. (2021). **Gut** Nagai, M., et al. (2023). **Nature Communications** El-Khoury, et al. (2025). **Frontiers in Microbiology** 



### Intestinal *Candida albicans levels* are high in Long-Covid

Candida makes a lasting impression in COVID-19 Lagree K & Chen P. Nat Immunol. 2023 Nov

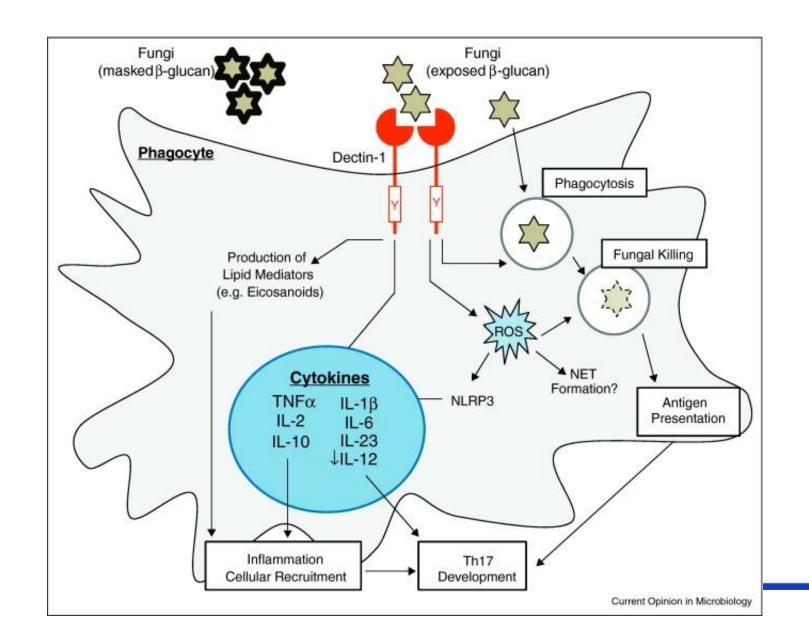
Severe COVID-19 is marked by excessive inflammation that can persist after infection. The commensal yeast *Candida albicans* is now implicated in the acute and chronic immunopathology of COVID-19

Fungal microbiota sustains lasting immune activation of neutrophils and their progenitors in severe COVID-19. Kusakabe T, et al. Nat Immunol. 2023 Nov

Gut fungal pathobionts contribute to immune activation during inflammatory diseases, offering potential **mycobiota-immune therapeutic strategies** for sCOVID-

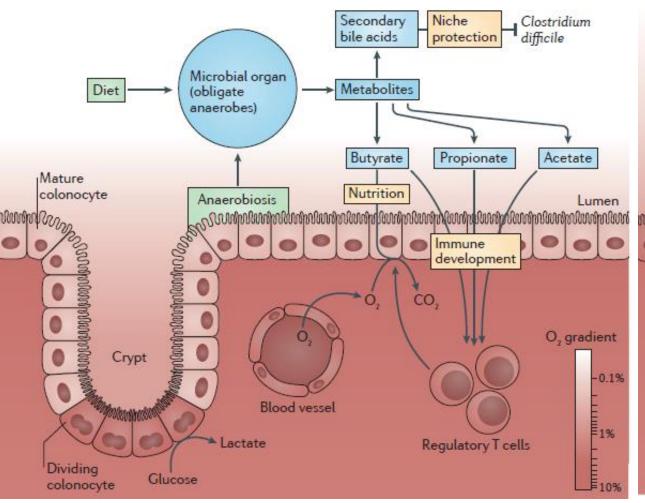
19 with prolonged symptoms.

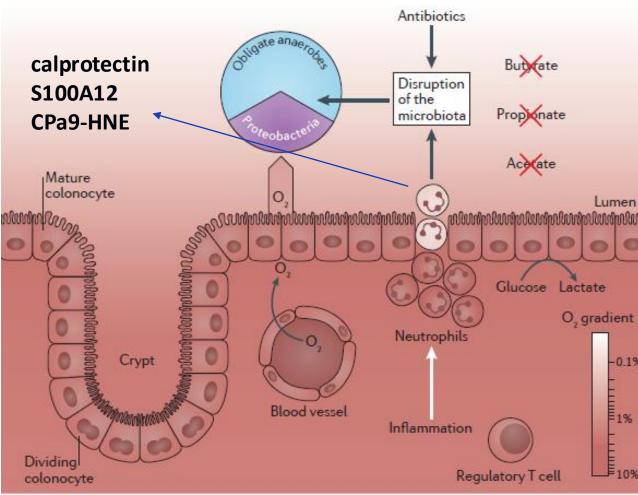
### Macrophage recognition of fungal infections



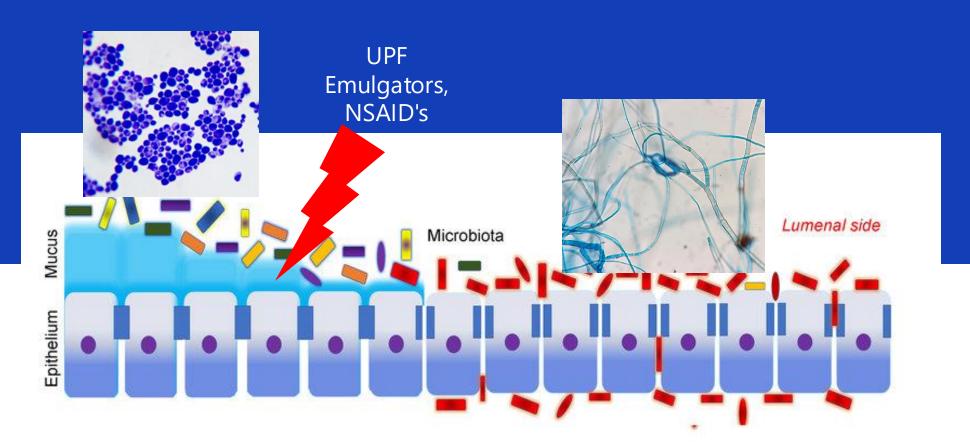
### **HOMEOSTASIS**

### **DYSBIOSIS**





# Loss of anaerobiosis and mucus in dysbiosis and invasion of Candida albicans

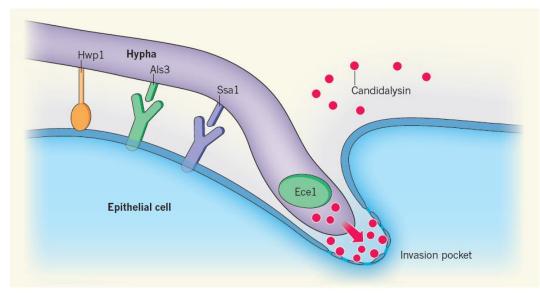


- -Normal microbiota
- Intact mucus and TJs
- -Dysbiosis
- Loss of mucus and TJs
- -Bacterial adherence
- Activation of inflammatory pathways (NFkB, STAT3)
- Bacterial translocation
- DNA damage (ROS, genotoxins, secondary bile)

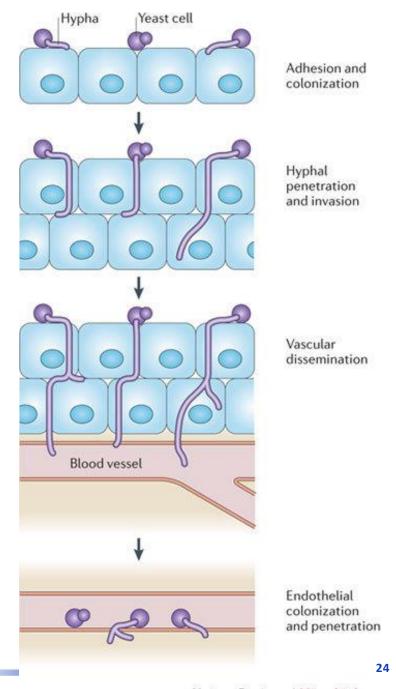
Lamina propria



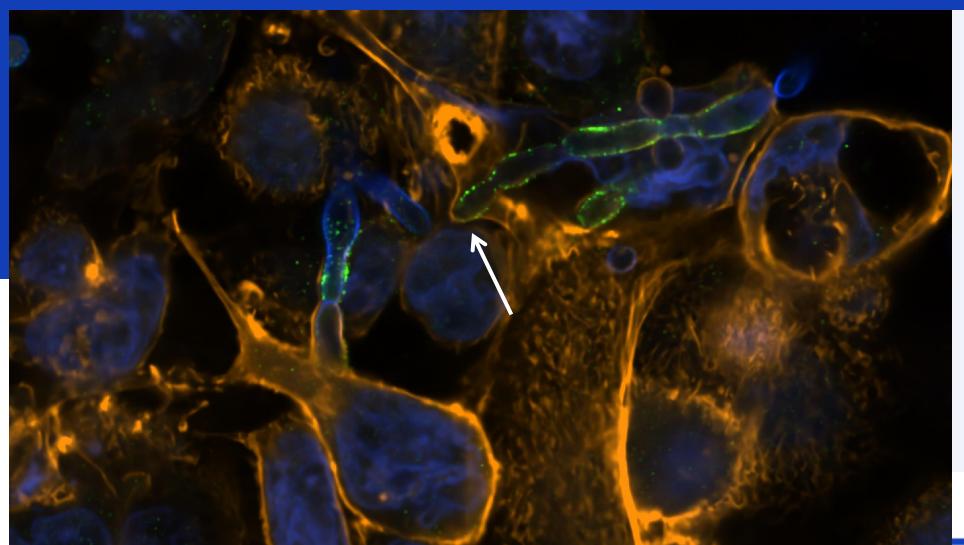
### Candida hyphal growth = pathogenicity



**Figure 1** | **A toxic relationship.** The pathogenic fungus *Candida albicans* infects its host by forming filamentous structures called hyphae. Proteins on the hyphal surface — the adhesin Hwp1, the invasin Als3 and its partner Ssa1 — make contact with the host cell directly or through receptor proteins to promote adhesion and engulfment of the hypha by the host. Moyes *et al.*<sup>2</sup> report that the protein Ece1 is secreted from the hypha as eight short peptides. One of these, Candidalysin, acts as a toxin that accumulates in the invasion pocket and attacks the host-cell membrane, leading to membrane permeabilization and the induction of host defences.



### Dysbiosis: invasion of intestinal cells by Candida



Candida albicans germination tubes secreting candidalysin on intestinal cells. Stained by candidalysin Immunofluorescence (Alexafluor 488 green) Phalloidin (yellow), Calcofluor white (blue) and DAPI (purple-blue). The white arrow indicate an invasion pocket. (photo: TNO & Nikon Europe)

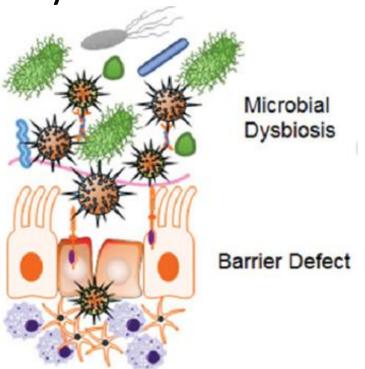


### Microbiome Restoration for Treatment Chronic Diseases

**Probiotic** 

**Prebiotics** 

Long-Covid Dysbiosis

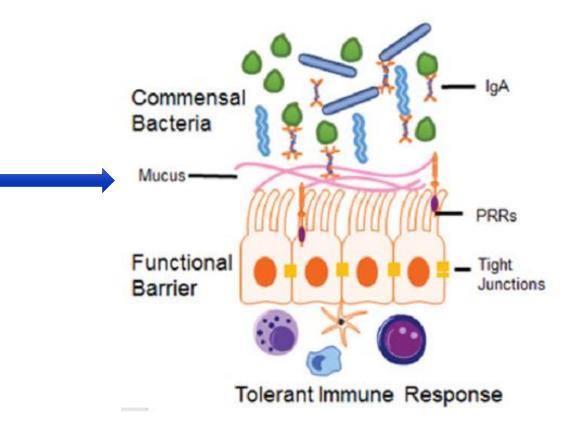


Dysregulated Immune Response

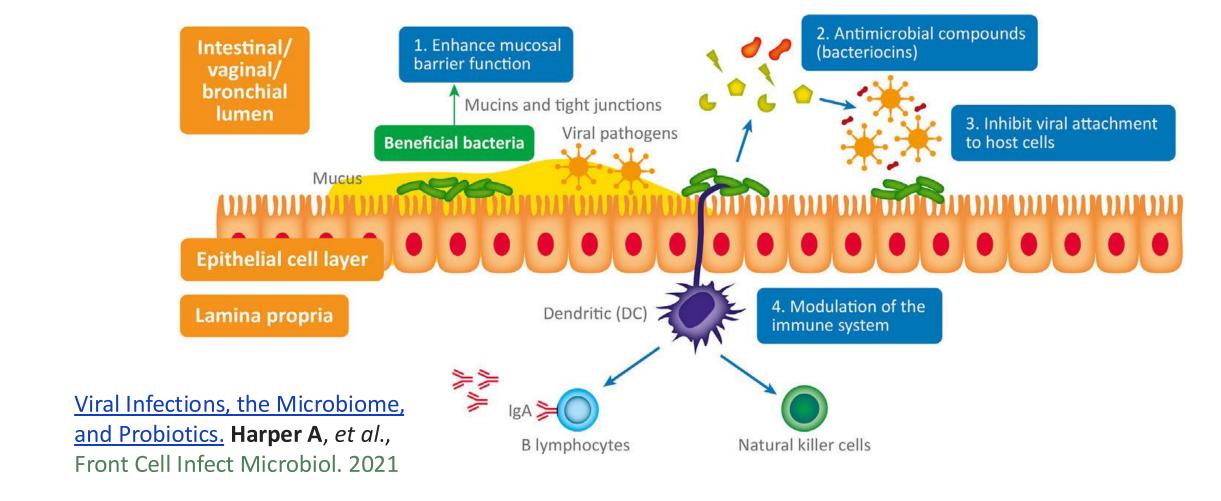
Low production of SCFA & Serotonin

High levels of Pathogens

**Gut Homeostasis** 



### **Anti-viral Activities of Probiotics**







### Pilot project QFS







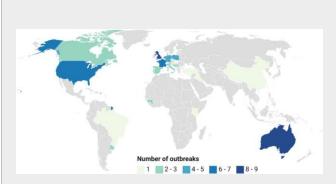


### What are Q fever and QVS again?



#### **Q-fever**

- Zoönosis: bacteria Coxiella burnetii (Cb)
- Widespread worldwide
- Large animal reservoir:
  - Small ruminants
  - Wild, domestic, farm animals, birds, ticks
- 2007-2010: Largest epidemic ever: in NL
  - 50,000 to 100,000 infections

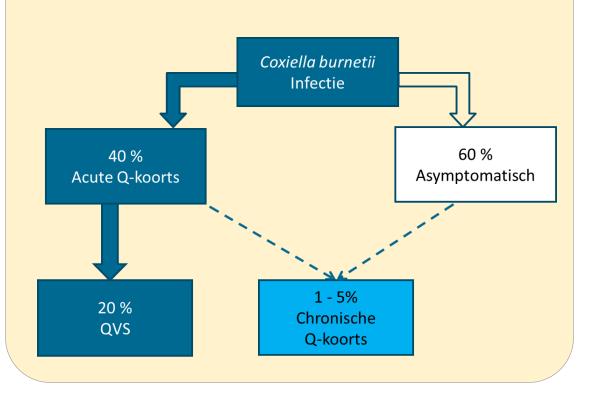


Tan eeta al. A systematic review of global Q fever outbreaks. One Health 2024. https://doi.org/10.1016/j.onehlt.2023.100667.



### Q-fever vfatigue syndrome (QFS)

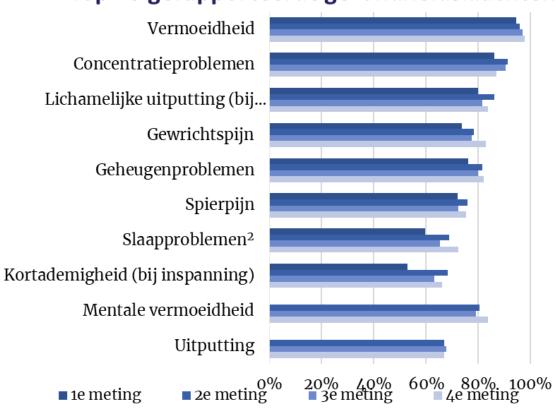
- Follows after infection
- No more live bacteria detectable
  - Different from chronic Q fever



### QVS – typical symptoms



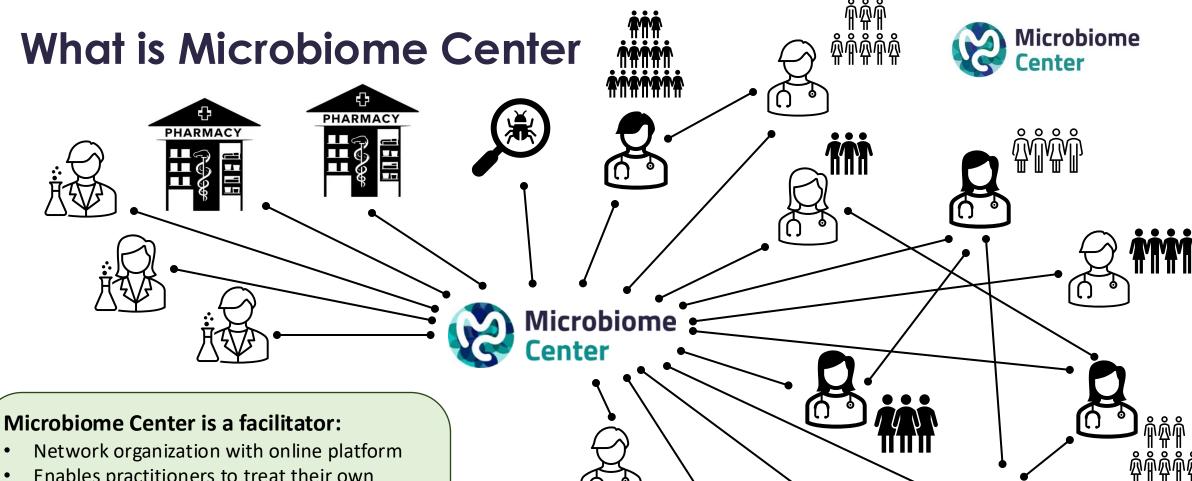




0.2 -0.0 Diagnosis -0.4 -0.0 0.2 -0.2 0.4 PCo<sub>1</sub>

Stemerdink et al., Vier jaar QVS database onderzoek. Erasmus MC en Q-support. 24 november 2025

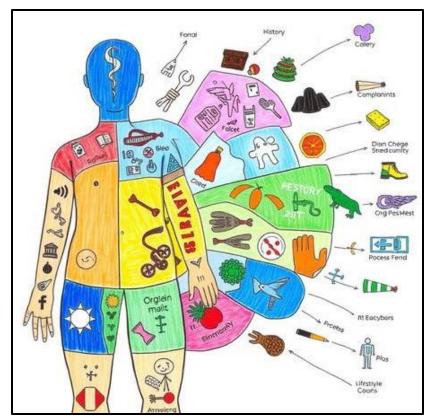
Raijmakers RPH, Roerink ME, Jansen AFM, et al. Multi-omics examination of Q fever fatigue syndrome identifies similarities with chronic fatigue syndrome. J Transl Med 2020; 18: 448.



- Enables practitioners to treat their own patients with personalized microbiome treatment.
- Truly personalised: each prescription is prepared by independent pharmacists
- Knowledge center for microbiome treatment

### Why personalized?

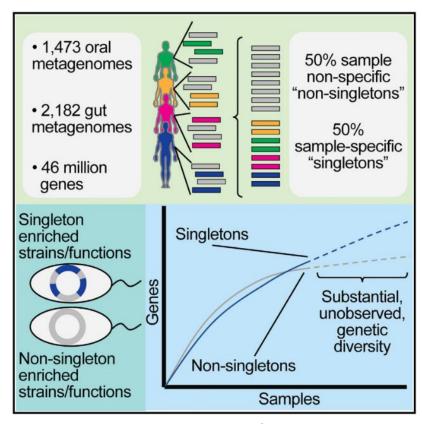




deepai.org

### **Every person is unique in:**

- Pattern of complaints
- Medical background
- Microbiome
- Diet
- Lifestyle
- Movement
- Stress
- Sleep patterns
- History
- Etc.



Tierney BT, Yang Z, Luber JM, et al. The Landscape of Genetic Content in the Gut and Oral Human Microbiome. Cell Host & Microbe 2019; 26: 283-295.e8.



# Pilot project Set-up and results









### Project design





In 2024 Q-support invites patients with QFS and GI complaints for the project



Between
September 2024
and August 2025,
85 patients
received
reimbursement
from Q-support



Received fecal analysis



Doctor visit with anamnesis.

Start lifestyle treatment with MyOwnBlend



Follow lifestyle with MyOwnBlend for 2-3 months. One doctor's visit. Questionairre every 3 weeks.



Final doctor's
visit. Standard
service call from
Microbiome
Center about
effects of the
treatment

On average these patients have been ill for 16 years

#### Data retrospectively analyzed:

- Indications during anamnesis
- Course of complaints
- Electronic patient dossier

### Common complains reported by QFS patients before start of treatment



Common complains reported by 82 patients



N	Indication score > 0
77	Low levels of butyrate
74	Fatigue
68	Intestinal permeability
51	Bloating
50	Sad mood
45	IBS
44	Lactose intolerance
40	Pathogenic bacteria overgrowth
39	Diarrhea
34	Inflammation
33	Allergic conditions
29	Constipation
20	Yeast overgrowth

### **Evaluation of Patients' Improvement Over time**

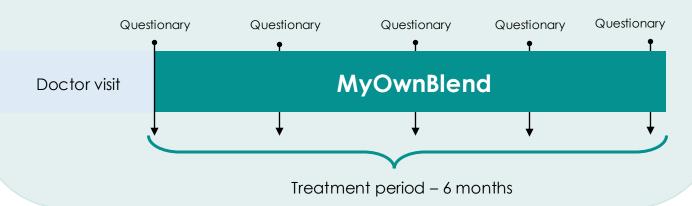


Online scoring over time data analyzed for 69 QFS patients



Questions that patients can answer multiple times to monitor their progress:

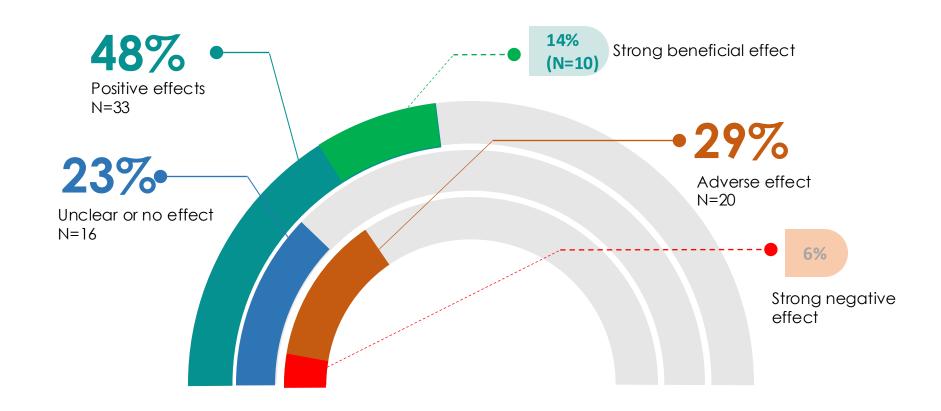
- "How have you been (in the past week/weeks)?" Scale 0–10
- "Have you used your MyOwnBlend in the past week/weeks?"
   Not/Hardly Occasionally Yes/Mostly
- Standard medical complaints:
  Constipation, diarrhea, bloating, flatulence, abdominal pain, cognition, low mood, stress, anxiety/tension, fatigue, sleep quality, vaginal complaints, acne
- Any other personal complaints?



#### Overall well-being over time



Results based on 69 participants with QFS
Results based on the question: "How have you been doing (in the past week/weeks)?"



#### Overall effect on medical complaints



Results based on 79 participants with QFS Focus on standard medical complaints

(constipation, diarrhea, bloating, flatulence, abdominal pain, cognition, low mood, stress, anxiety/tension, fatigue, sleep quality, vaginal complaints, acne)

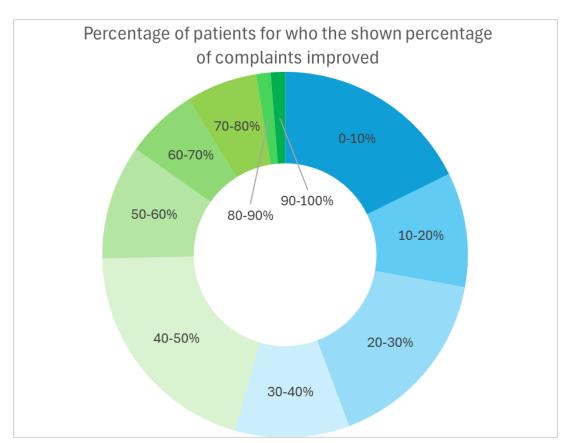


The combined effect on all complaints is categorized as a score from -4 (strongly negative) to +4 (strongly positive) Very unfavorable (-1.5 - -2.5)0% (N=0) Fairly unfavorable (-1.5 - -2.5)3% (N=2) Slightly unfavorable (-1.5 - -0.5)-10% (N=8) No effect (-0.5 - +0.5)30% (N=24) 37% (N=29) Slightly favorable (+0.5 - +1.5)15% (N=12) Fairly favorable (+1.5 - +2.5)Very favorable (+2.5 - +3.5)4% (N=3)

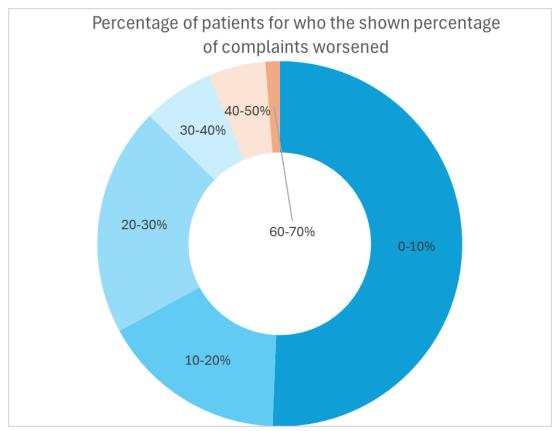
#### Overall effect on medical complaints



## In almost half of the participants (36/79), more than 40% of their symptoms improved



## In more than half of the participants (40/79), less than 10% of their symptoms worsened

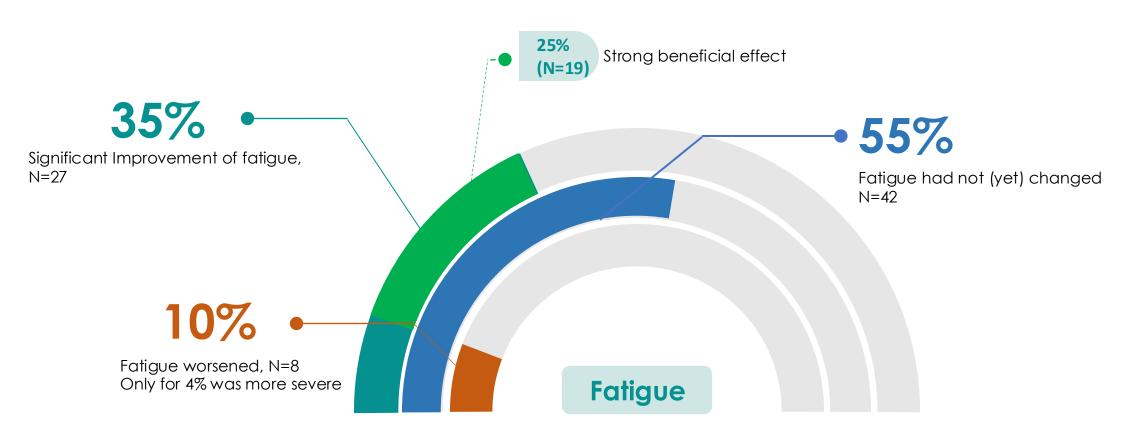


#### Improvement of fatigue



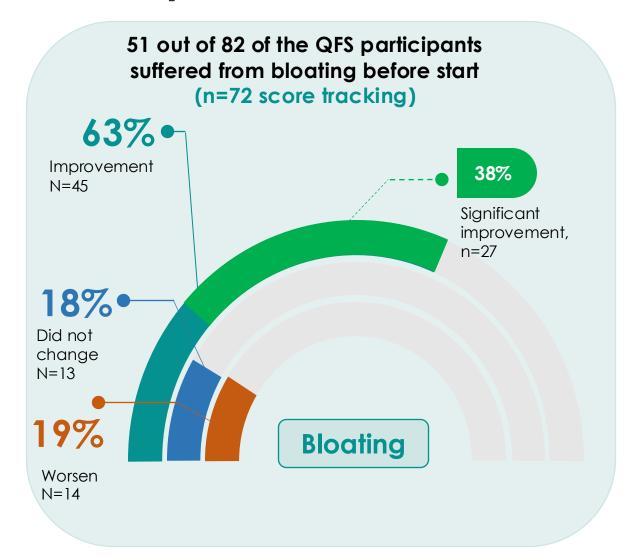
#### Fatigue is the most commonly reported complaint by 74 out of 82 QFS participants

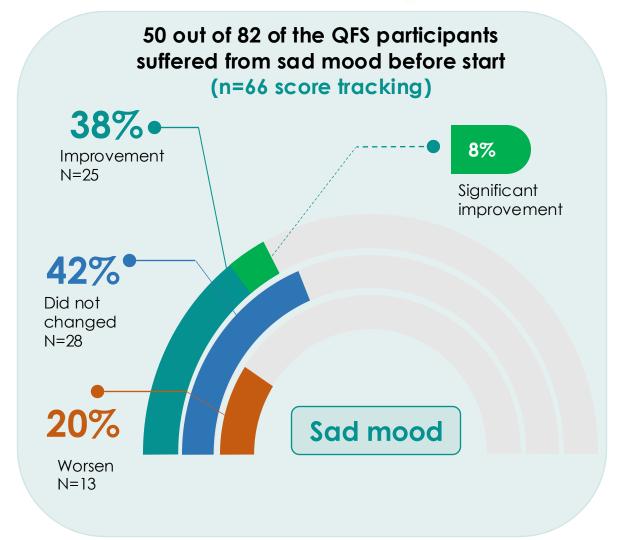
For 77 patients the score tracking data allows conclusions



#### Improvement of other common complains



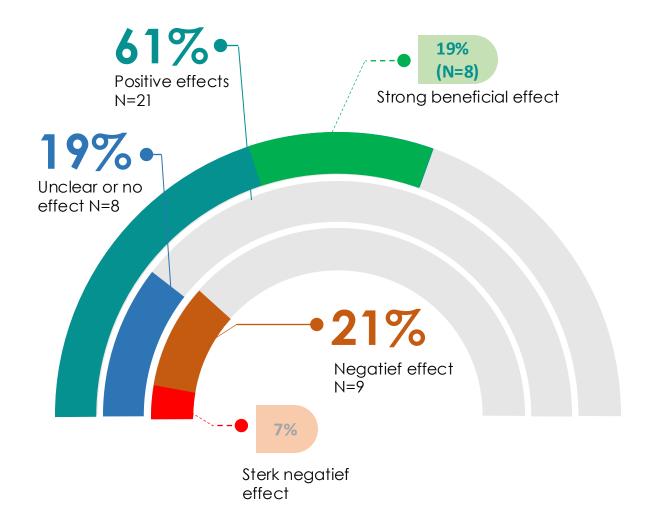




#### Other PAIS results

Microbiome Center

- In literature, other Post-Acute Infectious Syndromes (PAIS) are also related to dysbiosis
- PAIS examples:
  - ME/CFS
  - QFS
  - Post-Covid
  - Post-Pfeiffer
- An initial analysis of 43 patients in which Post-Covid is listed showed that more than 60% improved





# Example case 1: Ingeborg, 61 year



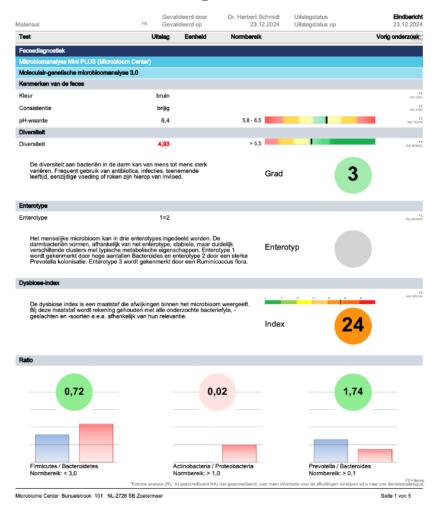


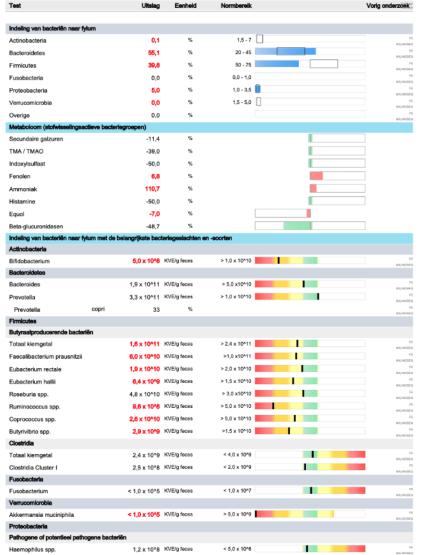






- Ingeborg, 61 year
- 2009 complaints of flu, lasted for months, finally with pneumonia in ZH where Q fever was detected.
- When she was two already suffering from joints and kept that whole life, in addition many unexplained physical complaints.
- Sitting at home since June 2023, fatigue, forgetful, rehabilitation process without result, poor condition, does not move.
- Since Q fever suffers from intestines, does not dare to leave the house because of IBS, attacks of urgency and then sometimes too late to the toilet. If she has to go away for a day, she doesn't eat anything.
- 2dd bread meal, can easily be done without a snack
- Microbiome analysis









Test	Ultslag	Eenheld	Normberelk	Vorig onderzoek
Acinetobacter spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^6	PE NA MODO
Proteus spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^6	FE. NA MOSSO
Klebsiella spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^7	PE NAMES O
Enterobacter spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^6	FE NAM MOREO
Serratia spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^7	FE NAMOSEO
Hafnia spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^8	rs.
Morganella spp.	< 1,0 x 10^5	KVE/g feces	< 1,0 x 10^6	NA MODEQ FE
Citrobacter spp.	< 1,0 x 10^5	KVE/g feces	< 5,0 x 10^8	NA MOSEQ FE
Pseudomonas spp.	< 1.0 x 10^5	KVE/g feces	< 5,0 x 10^7	NAJ MOSEG FE
Providencia spp.	< 1.0 x 10^5		< 5.0 x 10^7	NY MOSEQ FE
H2S-vorming	- 1,0 11 10 0			NA WORKO
Sulfaatreducerende bacteriën (SRB)	1.6 x 10^9	KVE/g feces	< 2.5 x 10^9	
Desulfovibrio piger	< 1.0 x 10^5	KVE/g feces	< 1,0 x 10^9	NA MOSSICE FE
Desulfomonas pigra	< 1.0 x 10^5	KVE/g feces	< 1,0 x 10^9	NAI MOSEO FE
Bilophila wadsworthia	< 1.0 x 10^5	KVE/g feces	< 2,0 x 10^9	NA WOODS
Immunogeniciteit / mucine vorming				NAJ MOSEO
Immunogeen werkende bacteriën				
Escherichia coli	< 1,0 x 10^5	KVE/g feces	10*6 - 10*7	FE. NA MOSEO
Enterococcus spp.	8,70 x 10^6	KVE/g feces	10^6 - 10^7	FE NV MOSEO
Lactobacillus spp.	6,5 x 10^6	KVE/g feces	10*5 - 10*7	TE NAMONO
Mucine vorming / slijmvliesbarrière				
Akkermansia muciniphila	< 1,0 x 10^5	KVE/g feces	> 5,0 x 10^9	PE NAMED OF THE PERSON OF THE
Faecalibacterium prausnitzii	6,0 x 10^10	KVE/g feces	>1,0 x10*11	FE NA MOSEO
Archaea				
Methanogenen				
Methanobrevibacter spp.	< 1,0 x 10^5	KVE/g feces	< 5,0 x 10^8	FE. NA MOSCO
			Opmerking: Het nieuwe OmicSnap matrix maken een nog effectievere vooral bij grampositieve bacteriën. Dit resulteert in lichte verschulvings	monsteranalyse mogelijk,

Mycobioom: relevante gisten				
Candida albicans (CA)	<1,0 x 10^3	KVE/g feces	<1,0 x 10^3	NA PCF
Candida krusei (CK)	<1,0 x 10^3	KVE/g feces	< 1,0 x 10^3	P1 NAO PCE
Candida glabrata (CG)	<1,0 x 10^3	KVE/g feces	< 1,0 x 10^3	FI NAV. PCE
Candida dubliniensis (CD)	<1,0 x 10^3	KVE/g feces	< 1,0 x 10^3	FI NAO PCE
Candida parapsilosis (CP)	<1,0 x 10^3	KVE/g feces	< 1,0 x 10^3	FI NA PCE
Candida tropicalis (CTp)	<1,0 x 10^3	KVE/g feces	< 1,0 x 10^3	FI NA) PCF
Candida lusitaniae (CL)	<1,0 x 10^3	KVE/g feces	< 1,0 x 10^3	FI NAO PCZ
Vertering				
Vetgehalte	4,30	g/100g	< 3,5	FI No. PHO
Stikstofgehalte	0,30	g/100g	< 1,0	NA PHO
Suikergehalte	5,50	g/100g	< 2,5	FI No. FHO
Watergehalte	78,30	g/100g	75 - 85	PI NAU PHOT
Extra parameter(s)				

Calprotectine

T4	I III alaa	English	Normberelk	Mode on do-Notice
Test	Ultslag	Eenheid	Normbereik	Vorig onderzoek
Alfa-1-antitripsine	6,8	mg/dl	< 27,5	FC A) CLOA
Constale leaves and builton &	4007.4	unimi	510 - 2040	A) LUIAN.
Secretoir Immunoglobuline A	1267,1	µg/ml	310 - 2040	A) 8U8A
Zonuline	78,42	ng/ml	< 55	27 ADLIAN
Speciale gastro-enterologische diagnostiek				
Giuten-sensitieve enteropathie / coeliakie				
Anti-gliadine antilichamen in feces	36,50	UA	< 100	FE A) SUBA
Anti-transglutaminase antistoffen in feces	54,44	UA	< 100	415.004



PHGG	4
L. rhamnosus GG	2
Bacillus coagulans Unique IS-2	2
DJ repair	3
Akkermansia muciniphila, gepasteuriseerd	1
L fermentum ME-3	1
Acaciavezels	2

- MyOwnBlend
- Eating sourdough spelt bread
- Three weeks later clear effect: 1 time daily bowel movement and no more diarrhea attacks
- Has gone for longer walks with the dog
- Quality of life back







## Example case 2: Caroline, 40 year











- Caroline , 40 year
- 13 years ago Q fever contracted at petting zoo, watched goat give birth
- High fever, difficulty breathing, muscle pain immediately tested and Q fever came out.
- Treated with AB but complaints persisted: fatigue, muscle pain, shortness of breath, painful joints.
- Can work 2 days, sports often don't work, has a positive outlook on life.
- Already many abdominal complaints before Q fever, known with IBS
- Since Q fever even more complaints: bloating, constipation, abdominal pain
- Nutrition:
  - B: 2 slices of bread with dark chocolate spread (LV)
  - S: cereal bar
  - L: 6 spoons of soy yogurt with some fruit, flaxseed
  - S: cookie or candy
  - D: varied, pasta/rice/potato, mainly meat or vegetarian.
- Microbiome analysis: no abnormalities
  - Div 8, DI 12, bifi= buty= pathogenes= canida= inflammation= zon=



Bacillus coagulans Unique IS-2	2
Bifidobacterium lactis HN019	2
L. plantarum P-8	1
L fermentum ME-3	2
L. rhamnosus SP1	1
Enterococcus faecium + Bacillus subtilis	1
L. rhamnosus GG	1
PHGG	3
Acaciavezels	2

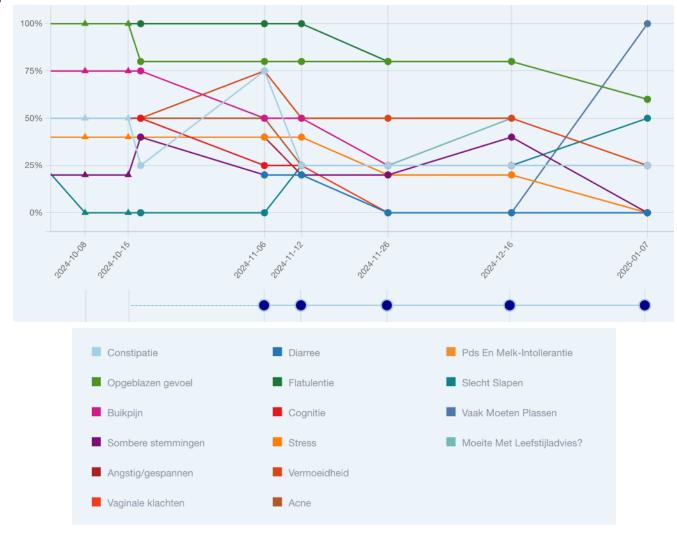
Started with sourdough spelt bread, no more stomach ache in the morning after breakfast.

In the evening large portion of healthy which makes you feel full, relieved feeling inside. More energy, better in body.

"What a fantastic stuff this is, so very happy with it."

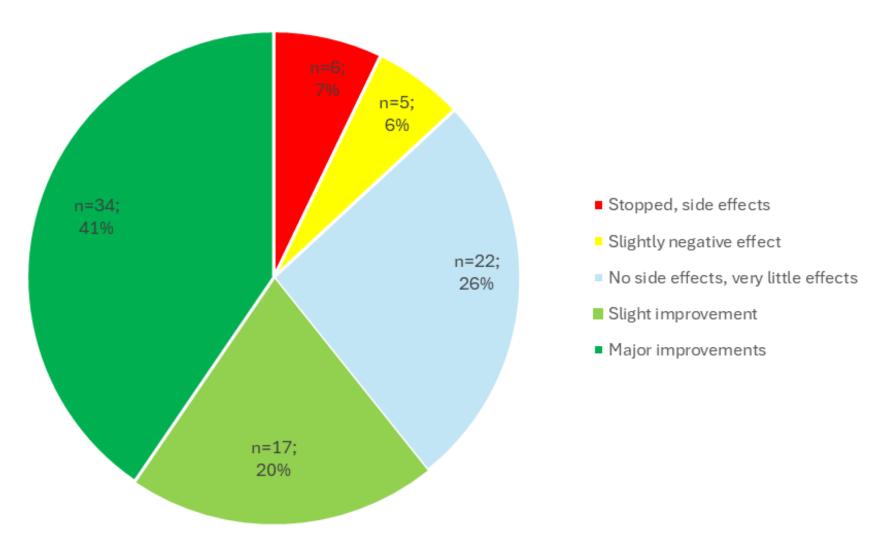
Feeling that probiotics have made the biggest difference, more than food, but food does help.





#### **Experiences patient practice/EPD**







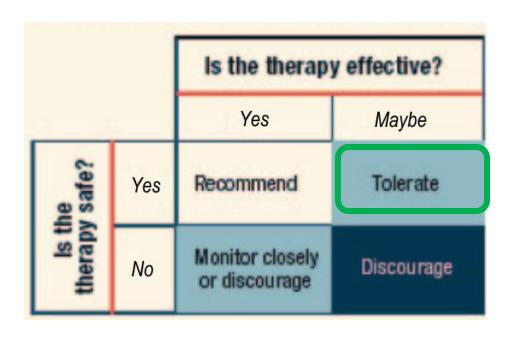
## The next step?



#### Is there sufficient reason to start treatment??



#### → Ethical framework provides guidance



## Question the patient can ask themselves

- Wat are my possibilities?
- What are the pros and cons of these?
- What does this mean for my situation?

Medische Oncologie | 'Integratieve zorg is een aanvulling, geen vervanging'.
https://medischeoncologie.nl/artikelen/2022/maart/integratieve-zorg-is-een-aanvullinggeen-vervanging (accessed Dec 9, 2025).

Kemper KJ, Vohra S, Walls R, the Task Force on Complementary and Alternative Medicine, the Provisional Section on Complementary, Holistic, and Integrative Medicine. The Use of Complementary and Alternative Medicine in Pediatrics. Pediatrics 2008; 122: 1374–86.

#### Do you have any questions?





