‚Gut-brain-axis dysfunctions: What probiotics can do for us‘

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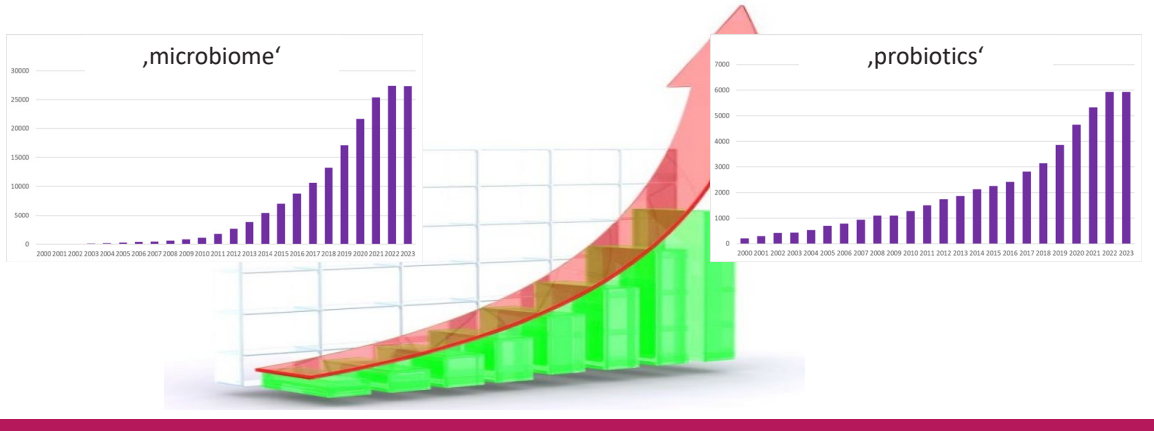
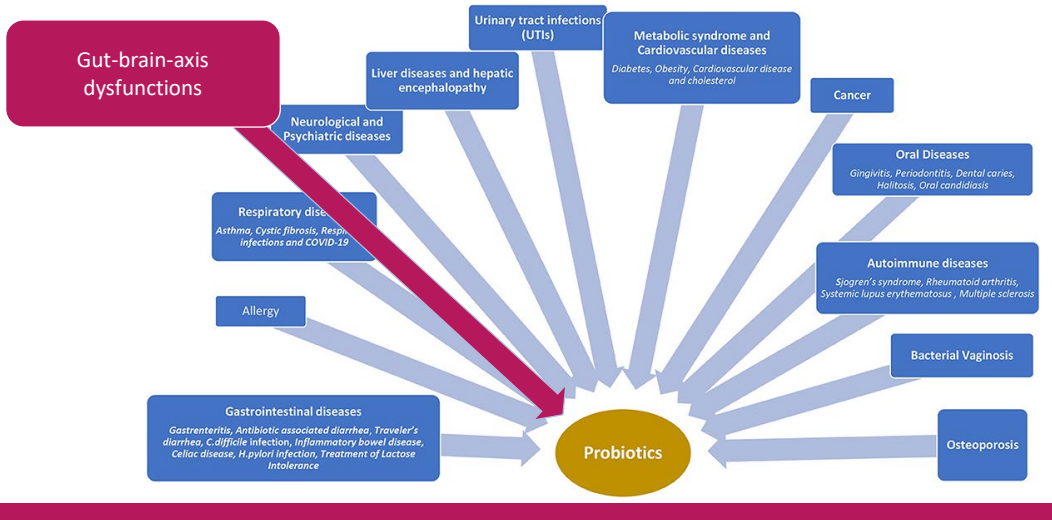
Observational Trial

Probiotics 1991

Probiotics 2024

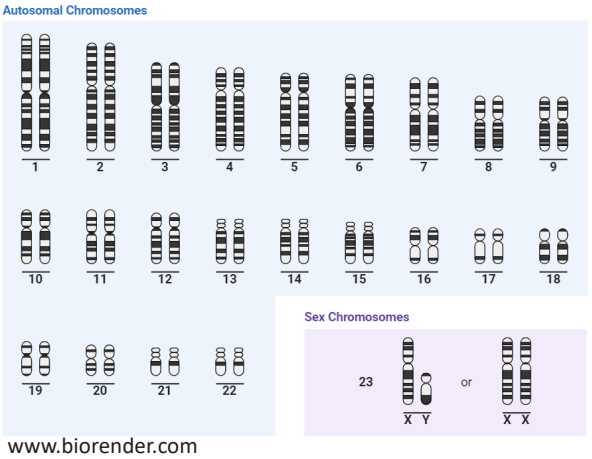


Use of indication specific Probiotics



https://doi.org/10.3389/fimmu.2020.02192

Neuronal dysfunctions



www.biorender.com

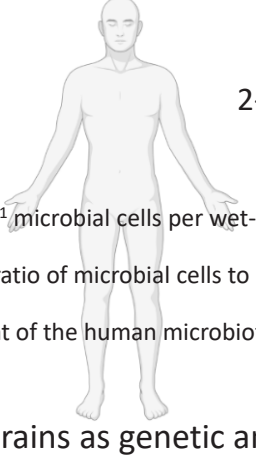
- specific events that cause neuronal dysfunction remain poorly understood
- neurons progressively accumulate somatic mutations
- disease-associated oligodendrocyte signature
- ...

Are we looking at the right place?

doi.org/10.1038/s41593-022-01104-7

Human genes – microbial genes

20.000 human genes



2-20 million microbial genes

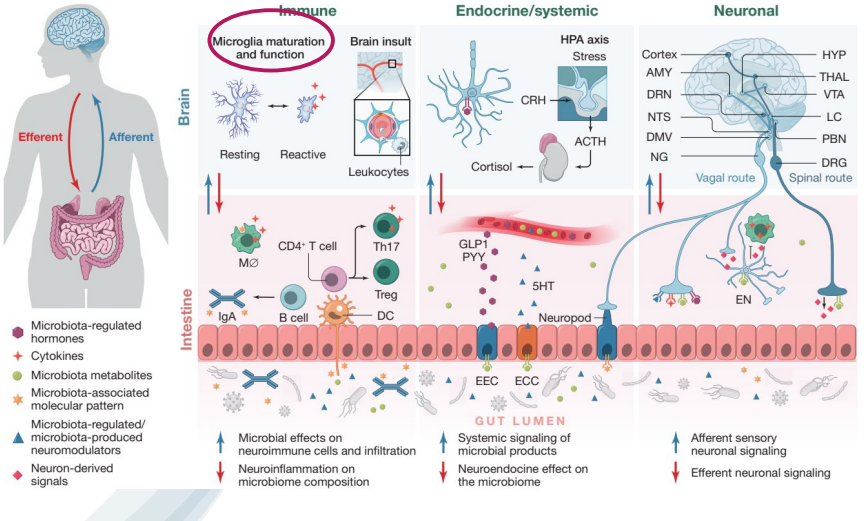
- 10^{10} and 10^{11} microbial cells per wet-weight gram of feces
- 1:1 cell ratio of microbial cells to human host cells
- total weight of the human microbiota is less than 500 g

Probiotic bacterial strains as genetic and metabolic ressource.

https://doi.org/10.1038/s41564-023-01426-7

Gut-Brain-Axis

'Nothing in the gut stays in the gut....'



https://doi.org/10.1016/j.molmed.2020.05.003; 2524 Cell 184, April 29, 2021 © 2021 Elsevier Inc. DOI https://doi.org/10.1016/j.cell.2021.03.022

Quality Standards for Probiotics

frontiers in Microbiology

Spotlight on the Compositional Quality of Probiotic Formulations Marketed Worldwide

*Diletta Mazzantini[1], Marco Calvigioni[1], Francesco Celandroni[1], Antonella Lupetti[1] and Emilia Ghelardi[1,2]**

[1] Department of Translational Research and New Technologies in Medicine and Surgery, University of Pisa, Pisa, Italy, [2] Research Center Nutraceuticals and Food for Health-NutraFood, University of Pisa, Pisa, Italy

- many incongruences in the compositional quality of some probiotic formulations
- sufficient amount of the indicated living microbes
- number of living organisms until product expiry
- pathogen-free, free of antibiotic resistance, safe
- requirement for **more stringent quality controls by manufacturers**
- standardized production process
- **Quality Standards are missing but required**

ipa EUROPE INTERNATIONAL PROBIOTICS ASSOCIATION

IPA Europe wants to…
…establish probiotic standards and guidelines
…establish a harmonized legal status for probiotics in the EU
…participate actively in the EFSA consultation process
…engage dialogue with all relevant stakeholders to increase awareness and knowledge of probiotic foods

REVIEW article, Front. Microbiol., 20 July 2021, Sec. Food Microbiology, https://doi.org/10.3389/fmicb.2021.693973

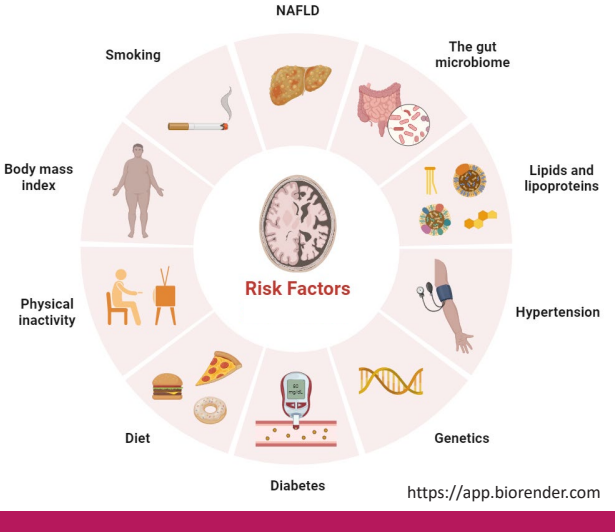
Quality Standards for Probiotics

1. Safety
2. Bacterial count
3. Activity
4. Ability to reproduce
5. Gastric and bile-acid resistance
6. Stability at room temperature
7. Milieu optimization
8. Multispecies formulation of symbiotic strains
9. Synergy of the strains
10. Efficacy Studies
11. Matrix
12. Free from sugar, gluten and animal protein

according to WHO-, WGO-, FDA- and OePROM-criteria

Neurodegenerative diseases: risk factors

- Age
- Genetics
- Environmental factors
- Lifestyle factors
- Traumatic brain injury (TBI)
- Neuroinflammation
- Metabolic disorders
- Hormonal factors
- THE GUT MICROBIOME
- …



https://app.biorender.com

Clinical trials…

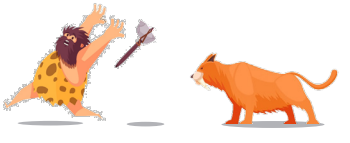
…indication specific multispecies probiotics

…gut-brain-axis dysfunctions

STRESS RESPONSE

stress affects the digestive organs

- **puts strain on the gastrointestinal tract**
- **reduces blood flow to the intestines**



‚Leaky Gut‘

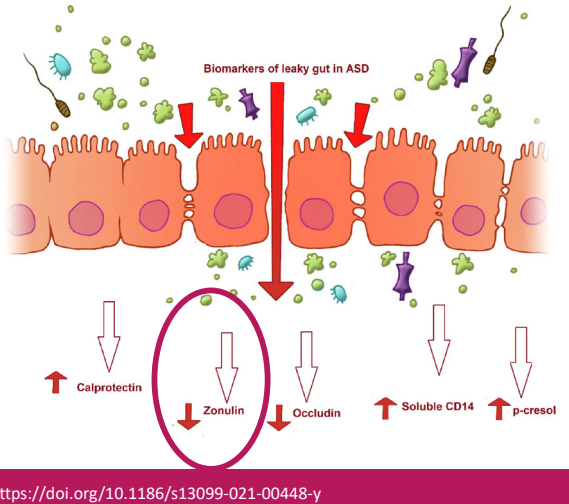
Inflammation

‚Leaky Brain‘ Neurological changes, Brain imflammation

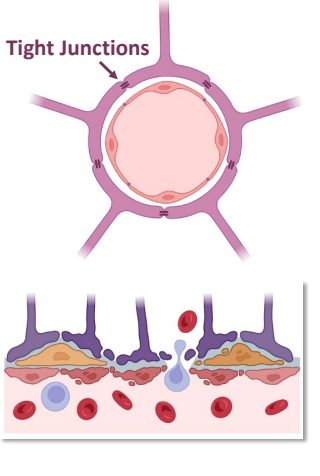
toxins, pathogens and allergens enter the body

pathogens can multiply unhindered

Leaky gut – brake down of the barrier



- Zonulin



https://doi.org/10.1186/s13099-021-00448-y

The importance of the gut-brain axis in dementia

RESEARCH ARTICLE

Probiotic Supplementation in Patients with Alzheimer's Dementia – An Explorative Intervention Study

Friedrich Leblhuber[1], Kostja Steiner[1], Burkhard Schuetz[2], Dietmar Fuchs[3,*] and Johanna M. Gostner[4]

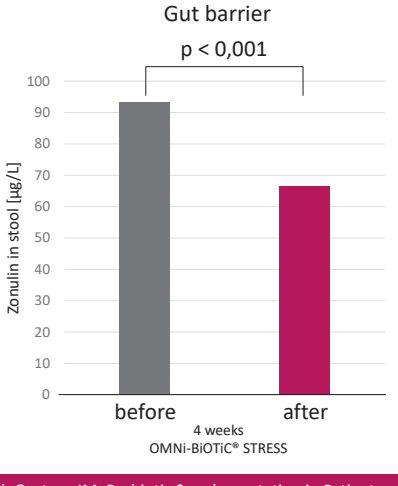
[1]Department of Gerontology, Neuromed Campus, Kepler University Clinic, Linz, Austria; [2]Biovis Diagnostik MVZ GmbH, Limburg, Germany; [3]Division of Biological Chemistry, Biocenter, Medical University of Innsbruck, Innsbruck, Austria; [4]Division of Medical Biochemistry, Biocenter, Medical University of Innsbruck, Innsbruck, Austria

STUDYDESIGN

probands	people with Alzheimer's disease (age 78 ± 8,5)
design	OMNi-BiOTiC® STRESS once daily for 28 days
	usual medication and vitamin D3 administration constant for duration of probiotic supplementation
Parameters	Zonulin, calprotectin, S100A12 in stool samples, neopterin, serum kynurenine

Leblhuber F, et al. Gostner JM, Probiotic Supplementation in Patients with Alzheimer's Dementia - An Explorative Intervention Study. Curr Alzheimer Res. 2018;15:1106–13.

OMNi-BiOTiC® STRESS STRENGTHENS THE INTESTINAL BARRIER



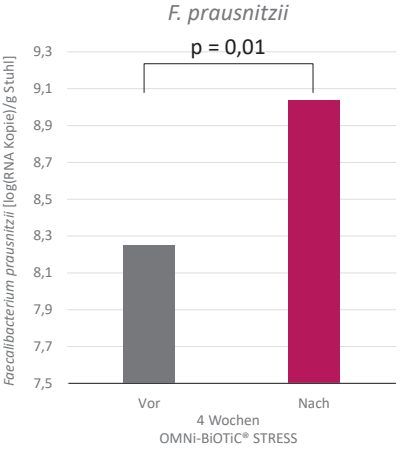
Zonulin: Parameter for

- the opening of the tight junctions and associated leaky gut
- Zonulin reduced (p<0,001)

Inflammation and leaky gut decreased

Leblhuber F, et al. Gostner JM, Probiotic Supplementation in Patients with Alzheimer's Dementia - An Explorative Intervention Study. Curr Alzheimer Res. 2018;15:1106–13.

Increase of *F. prausnitzii*



Faecalibacterium prausnitzii

- important **Butyrate prodcuer**
- releases anti-inflammatory metabolites

Significant increase in *F. prausnitzii* after 4 weeks of probiotic intervention (p=0,01)

anti-inflammatory effect

Leblhuber F, et al. Gostner JM, Probiotic Supplementation in Patients with Alzheimer's Dementia - An Explorative Intervention Study. Curr Alzheimer Res. 2018;15:1106–13.

RESULTS

Successful use of OMNi-BiOTiC® STRESS in Alzheimer's disease:

- Strengthening the intestinal barrier and reducing leaky gut (zonulin levels in the stool)
- Significant increase in *F. prausnitzii* (anti-inflammatory properties)
- Tryptophan metabolism in serum positively influenced

Leblhuber F, et al. Gostner JM, Probiotic Supplementation in Patients with Alzheimer's Dementia - An Explorative Intervention Study. Curr Alzheimer Res. 2018;15:1106–13.

First probiotic with proven multiple effect on intestinal barrier function:

Advances in Microbiology, 2013, 3, 212-221
doi:10.4236/aim.2013.32032 Published Online June 2013 (http://www.scirp.org/journal/aim)

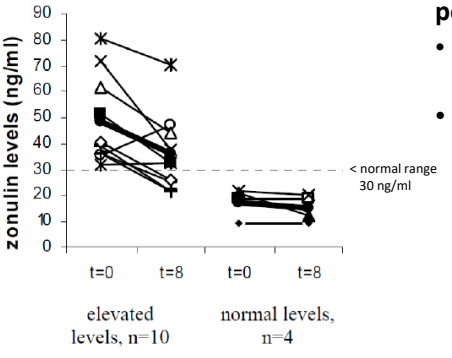
Scientific Research

Clinical Studies Evaluating Effects of Probiotics on Parameters of Intestinal Barrier Function

Saskia van Hemert[1*], Jurre Verwer[1], Burkhard Schütz[2]
[1]Winclove Probiotics, Amsterdam, the Netherlands
[2]Biovis Diagnostik MVZ GmbH, Limburg-Offheim, Germany
Email: [*]Saskiavanhemert@winclove.nl

Received March 6, 2013; revised April 4, 2013; accepted May 4, 2013

OMNi-BiOTiC® STRESS reduces *leaky gut*



Zonulin
(marker for increased gut permeability)

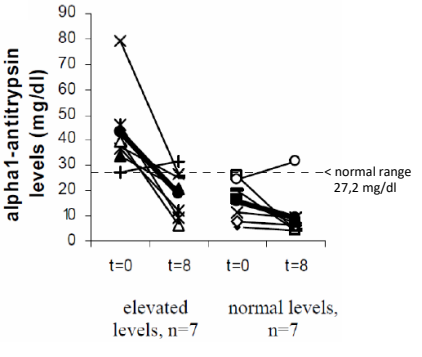
- modulates the intercellular *tight junctions*
- is associated with a ***leaky gut***

Zonulin levels were significantly reduced after 8 weeks ($p<0.01$)

Inflammation and *leaky gut* regressed

van Hemert, Schütz et al, Clinical studies evaluating effects of probiotics on parameters of intestinal barrier function, Advances in Mikrobiology, 2013

OMNi-BiOTiC® STRESS reduces intestinal inflammation



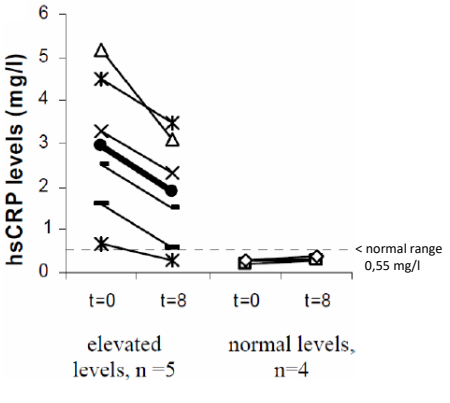
α-1-antitrypsin:

Parameters for **inflammatory processes** in the intestine

α-1-antitrypsin levels were significantly reduced in 12 out of 14 patients ($p<0.01$)

van Hemert, Schütz et al, Clinical studies evaluating effects of probiotics on parameters of intestinal barrier function, Advances in Mikrobiology, 2013

OMNi-BiOTiC® STRESS reduces inflammation throughout the body



C-reactive protein (CRP)

Parameter for **systemic inflammation**

The serum hsCRP concentration could be lowered significantly

van Hemert, Schütz et al, Clinical studies evaluating effects of probiotics on parameters of intestinal barrier function, Advances in Mikrobiology, 2013

Conclusion: OMNi-BiOTiC® STRESS demonstrates potent anti-inflammatory properties

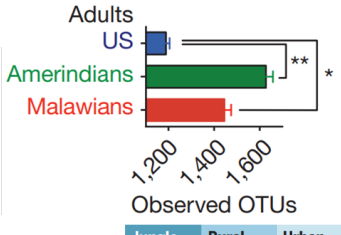
Supplementation for 8 weeks:

- **significantly reduced inflammation parameters**
  - both **systemic** and **mucosal**
- and reinforced the **intestinal barrier function**.

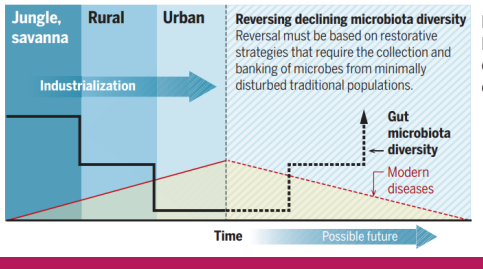
van Hemert, Schütz et al, Clinical studies evaluating effects of probiotics on parameters of intestinal barrier function, Advances in Mikrobiology, 2013

Missing Microbes – Missing Functions

loss of diversity, loss of functions



Yatsunenko, T., Rey, F., Manary, M. et al. **Human gut microbiome viewed across age and geography**. Nature 486, 222–227 (2012). https://doi.org/10.1038/nature11053



Bello, M. G. D., Knight, R., Gilbert, J. A., & Blaser, M. J. (2018). Preserving microbial diversity. Science, 362(6410), 33–34. doi:10.1126/science.aau8816

A multi-species probiotic positively affects irritable bowel syndrome (IBS)

European Journal of Nutrition
https://doi.org/10.1007/s00394-018-1826-7

ORIGINAL CONTRIBUTION

CrossMark

Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome

Adrian Mathias Moser[1,2] · Walter Spindelboeck[1,2] · Bettina Halwachs[2,3] · Heimo Strohmaier[4] · Patrizia Kump[1,2] · Gregor Gorkiewicz[2,3] · Christoph Högenauer[1,2]

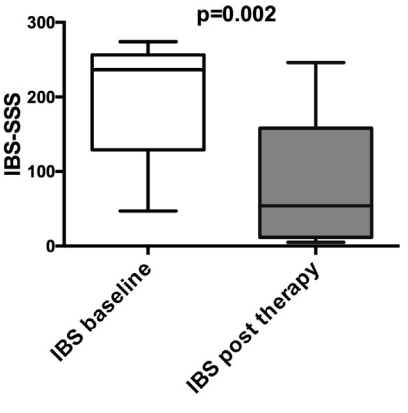
Received: 21 February 2018 / Accepted: 18 September 2018

STUDYDESIGN

Subjects	diarrhea-predominant irritable bowel syndrome (IBS-**D**) median score of 236 (of 300) on the IBS-SSS (*severety scoring system*)
Methods	Multispecies probiotic twice a day for 4 weeks
Strains	*Lactobacillus casei W56, Lactobacillus acidophilus W22, Lactobacillus paracasei W20, Bifidobacterium lactis W51, Lactobacillus salivarius W24, Lactococcus lactis W19, Bifidobacterium lactis W52, Lactobacillus plantarum W62, Bifidobacterium bifidum W23*
Samples	**Biopsy mucosal** samples, Stool
Read out	microbial diversity, short chain fatty acids, fecal zonulin, disease severity (IBS-SSS), mucosal immune cell, mucosal cytokine levels

Moser et al. Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr. 2019 Oct;58(7):2767-2778. doi: 10.1007/s00394-018-1826-7

IBS - Symptom severity is reduced by therapy with a multispecies probiotic

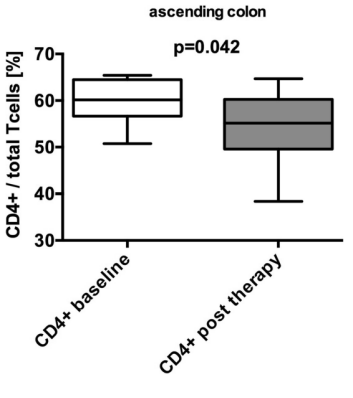


IBS-SSS:
assessment of severity of IBS-symptoms
(IBS: > 75; max. 300)

Highly significant improvement of severity of symptoms
(p = 0.002)

Moser et al. Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr. 2019 Oct;58(7):2767-2778. doi: 10.1007/s00394-018-1826-7

Multispecies probiotic modulates number of mucosal immune cells in IBS

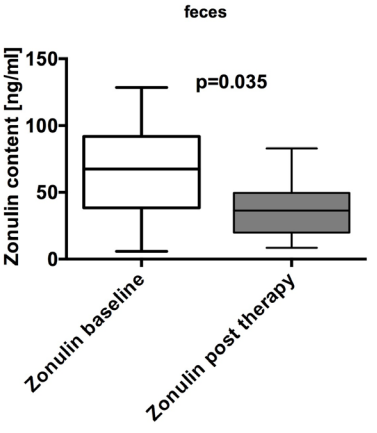


ISB-D patients have **elevated numbers of T-cells** in jejunal and colonic biopsy specimen compared to healthy individuals

<u>4 weeks of probiotics:</u>
anti-inflammatory modulation of colonic mucosa
(p = 0.042)

Moser et al. Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr. 2019 Oct;58(7):2767-2778. doi: 10.1007/s00394-018-1826-7

Multispecies probiotic reduces leaky gut marker content in IBS

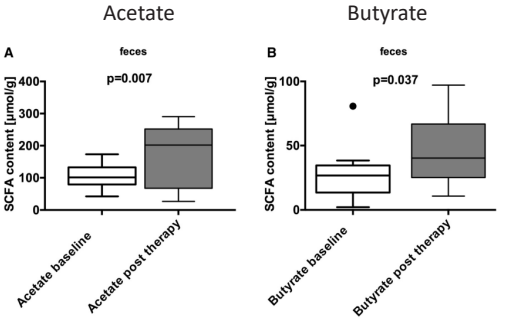


Zonulin:
reliable marker for increased gut permeability

Significantly improved mucosal barrier function
(p = 0.035)

Moser et al. Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr. 2019 Oct;58(7):2767-2778. doi: 10.1007/s00394-018-1826-7

Multispecies probiotic increases levels of *short chain fatty acids* (SCFAs)



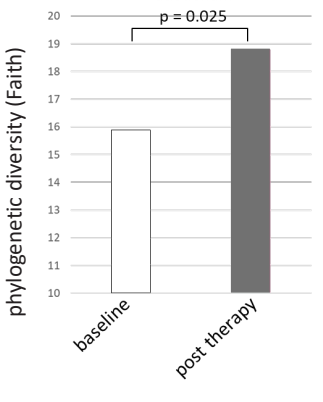
4 weeks of probiotics:

→ **butyrate** increased 1,5-fold

→ **acetate** increased 2,0-fold

Moser et al. Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr. 2019 Oct;58(7):2767-2778. doi: 10.1007/s00394-018-1826-7

Multispecies probiotic increases diversity in the upper GIT



4 weeks of probiotics:
significant elevation of phylogenetic diversity
(duodenum: p = 0.025)
gastric corpus (p=0.08)

Moser et al. Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr. 2019 Oct;58(7):2767-2778. doi: 10.1007/s00394-018-1826-7

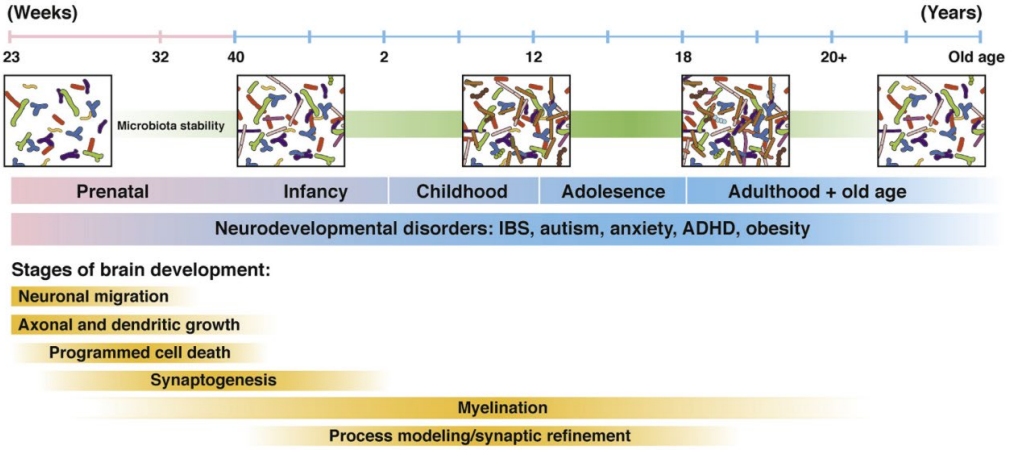
RESULTS

OMNi-BiOTiC® STRESS positively influenced mucosal immune system and microbiota

- **reduction** of **proinflammatory immune cells**
- **strengthening of intestinal barrier** (decrease of fecal zonulin)
- **elevation** of fecal **SCFAs** (butyrate and acetate)
- **increased phylogenetic diversity in the upper GIT** (gastrointestinal tract)

Moser AM, et al. Högenauer C, Effects of an oral synbiotic on the gastrointestinal immune system and microbiota in patients with diarrhea-predominant irritable bowel syndrome. Eur J Nutr 2018, doi:10.1007/s00394-018-1826-7.

Mechanisms of action



https://doi.org/10.1016/j.cgh.2018.10.002

OMNi-BiOTiC® STRESS improves brain functions and emotional memory

Gut Microbes
Taylor & Francis
ISSN: 1949-0976 (Print) 1949-0984 (Online) Journal homepage: http://www.tandfonline.com/loi/kgmi20

Probiotics drive gut microbiome triggering emotional brain signatures

Deepika Bagga, Johanna Louise Reichert, Karl Koschutnig, Christoph Stefan Aigner, Peter Holzer, Kaisa Koskinen, Christine Moissl-Eichinger & Veronika Schöpf

To cite this article: Deepika Bagga, Johanna Louise Reichert, Karl Koschutnig, Christoph Stefan Aigner, Peter Holzer, Kaisa Koskinen, Christine Moissl-Eichinger & Veronika Schöpf (2018): Probiotics drive gut microbiome triggering emotional brain signatures, Gut Microbes, DOI: 10.1080/19490976.2018.1460015

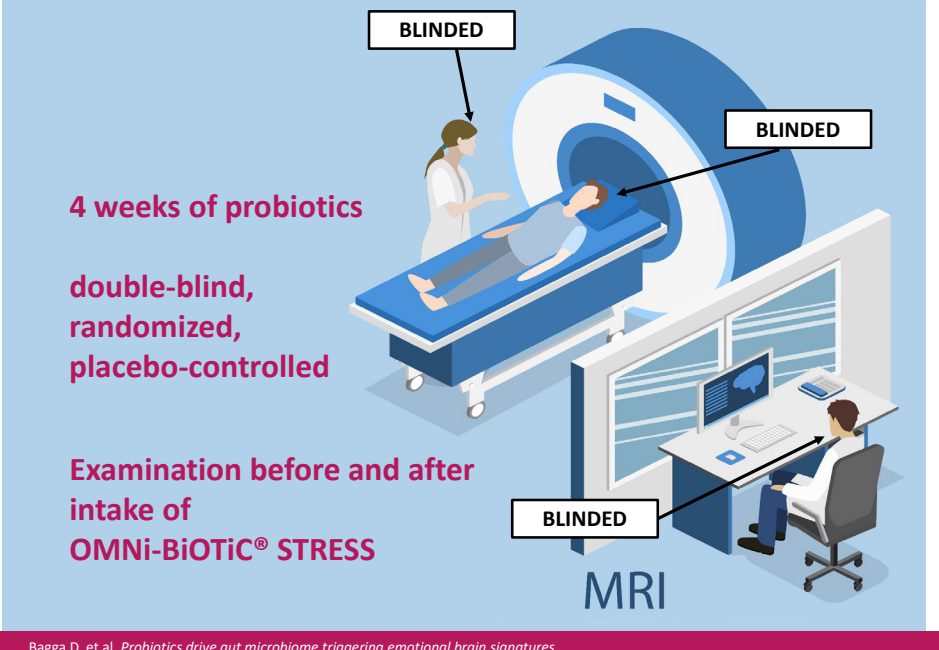
To link to this article: https://doi.org/10.1080/19490976.2018.1460015

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

STUDYDESIGN

Subjects	45 healthy volunteers, age 20 - 40
Methods	OMNi-BiOTiC® STRESS once daily for 4 weeks Double-blind, randomized, placebo-controlled 3 groups: Probiotics Placebo No intervention

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)



Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

In this study the answers to the following questions were sought:

Does OMNi-BiOTiC® STRESS influence…

… behavior

… brain signatures

… functional connectivity

?

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

Part 1: Behavior

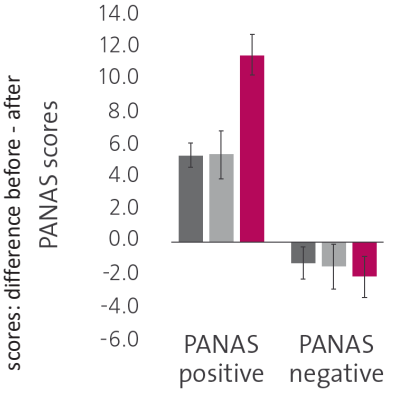
Does OMNi-BiOTiC® STRESS influcence affect and cognitive reactivitiy?

→ Examination with scientific behavioral tests based on self-assessment:

1. PANAS-Score
2. LEIDS-R questionnaire

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

OMNi-BiOTiC® STRESS enhances positive mood



PANAS-score (positive):

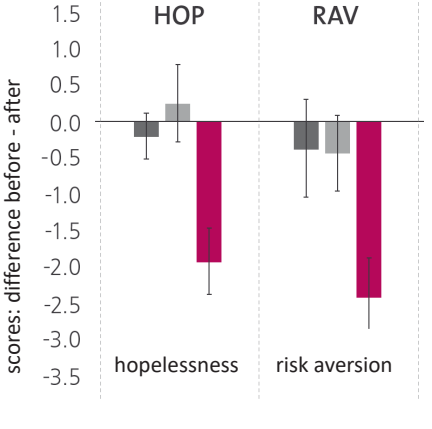
Subjects were significantly more positive

People with high score for positive affect perceive the environment differently. They are **kinder, more energetic** and less prone to illness.

Δ **probiotic**
Δ placebo
Δ **no intervention**

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

OMNi-BiOTiC® STRESS reduces hopelessness and risk aversion



after 4 weeks of probiotics:

subjects felt **more hopeful** and **confident**

Δ **probiotic**
Δ placebo
Δ **no intervention**

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

Part 2: brain signatures

Does OMNi-BiOTiC® STRESS affect memory and emotional decision making?

→ recognition task of neutral and unpleasant images during functional MRI measurement

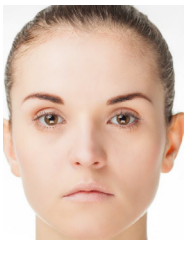
Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

Task: emotional decision making and memory

Which is the most neutral picture?

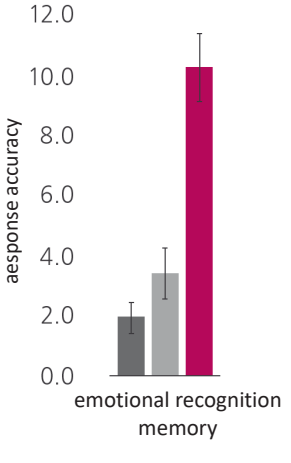
1 hour later:
Have you seen this picture before?

} fMRI-Scans of the brain while performing these tasks



Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

OMNi-BiOTiC® STRESS improves brain function

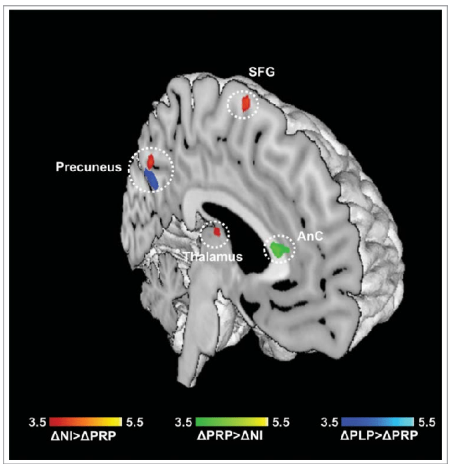


improvement of emotional memory ($p < 0{,}001$)

PLUS: subjects were more certain in their decisions and less prone to change them

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

Neural activity: results of the fMRI-scans



Increased brain activity in the probiotics group in:

AnC
(anterior cingulum):
→ mediates emotional processes

Precuneus:
→ plays an important role in decision-making processes

Bagga D, et al. *Probiotics drive gut microbiome triggering emotional brain signatures.* Gut Microbes, 9:6, p.486-496 (2018)

OMNi-BiOTiC® STRESS improves funtional connectivity

European Journal of Nutrition
https://doi.org/10.1007/s00394-018-1732-z

ORIGINAL CONTRIBUTION

CrossMark

Influence of 4-week multi-strain probiotic administration on resting-state functional connectivity in healthy volunteers

Deepika Bagga[1,2] · Christoph Stefan Aigner[3] · Johanna Louise Reichert[1,2] · Cinzia Cecchetto[1,2] · Florian Ph. S. Fischmeister[1] · Peter Holzer[2,4] · Christine Moissl-Eichinger[2,5] · Veronika Schöpf[1,2]

Received: 4 April 2018 / Accepted: 22 May 2018
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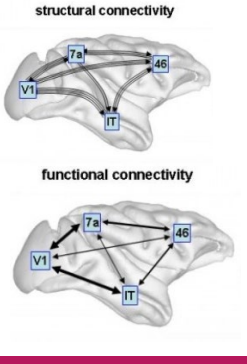
Bagga D, et al. *Influence of 4-week multi-strain probiotic administration on resting-state functional connectivity in healthy volunteers.* Eur J Nutr 58, 1821–1827 (2018)

Part 3: Influence on functional connectivity

Functional connectivity = simultaneous activation of multiple brain regions in complex tasks

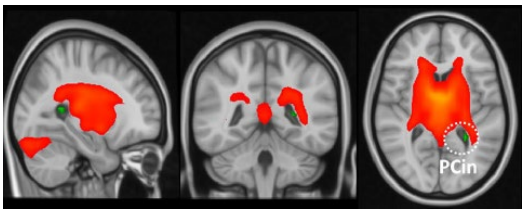
Structural connectivity:
= the presence of axonal connections

Functional connectivity:
observation of activity patterns → inferring possible connections of brain regions



http://www.imsid.uniklinikum-jena.de/imsid_media/Vorlesung_Modellierung_VL8bisVL10.pdf

OMNi-BiOTiC® STRESS increases attention



Salience Network (SN)
identifies the most important of the existing stimuli

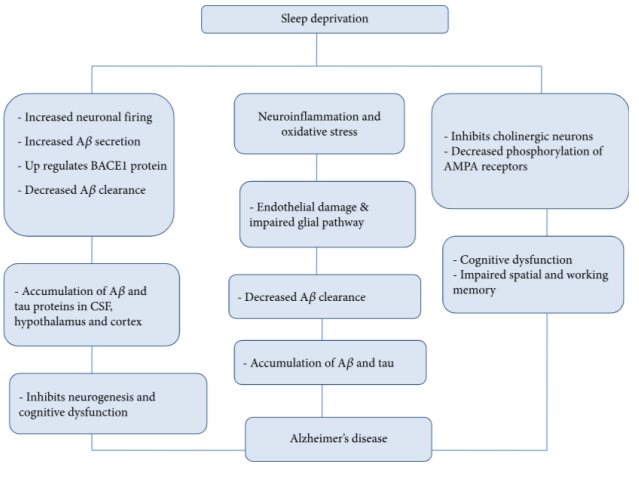
OMNi-BiOTiC® STRESS increases activity in the salience network

→ indicates improved attention

salience … the quality of being particularly noticeable or important; prominence.

Bagga D, et al. *Influence of 4-week multi-strain probiotic administration on resting-state functional connectivity in healthy volunteers.* Eur J Nutr 58, 1821–1827 (2018)

SLEEP AND NEUROLOGICAL DISORDERS



https://doi.org/10.1038/emm.2014.121

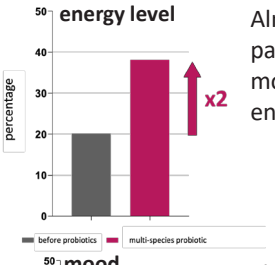
The positive effect of probiotics in stress resistance and quality of sleep

A Probiotic to Improve Sleep Quality during COVID-19 Pandemic: A Questionnaire Study

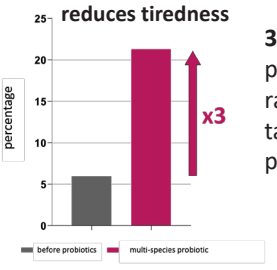
Angela Horvath[1,2], Jolana Wagner-Skacel[3], Verena Stiegelbauer[4], Vanessa Stadlbauer[1,2*]

9897 participants during the COVID-19

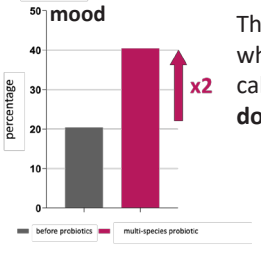
Multispecies probiotic



Almost **twice** as many participants felt mostly/always full of energy!



3 times as many participants felt rarely/never tired after taking the multispecies probiotic



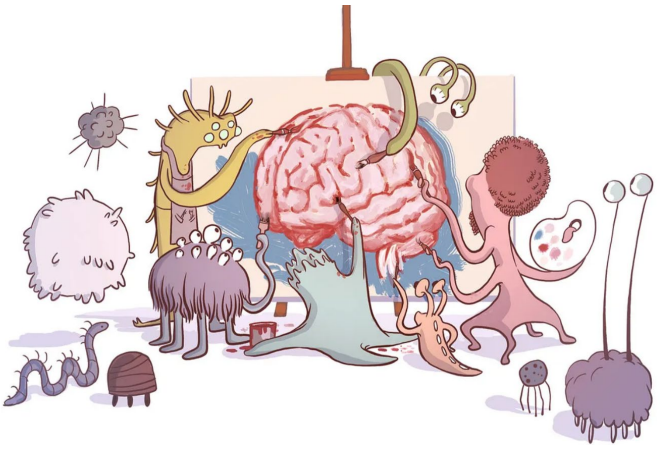
The number of people who mostly/always felt calm and relaxed was **doubled**!

Angela Horvath, Jolana Wagner-Skacel, Verena Stiegelbauer, and Vanessa Stadlbauer. A Probiotic to Improve Sleep Quality during COVID-19 Pandemic: A Questionnaire Study. Journal of Biotechnology and Biomedicine 6 (2023): 80-91.

Summary

- increasing prevalence gut-brain-axis dysfunctions
- microbiome is a potential target to treat
- indication sepcific multispecies probiotics to change the microbiome / microbial genetic pool / metabolome
- clinical trials demonstrate that the probiotic OMNi-BiOTiC® STRESS
  - significantly reduces leaky gut
  - increases *Faecalibacterium prausnitzii* abundance
  - reduces inflammation
  - reduces IBS-SSS
  - improves mood, improves brain function
  - improves sleep quality
  - influences energy level, mood, tiredness positively

Thank you very much for your attention!



https://www.npr.org/sections/health-shots/2013/11/18/244526773/gut-bacteria-might-guide-the-workings-of-our-minds