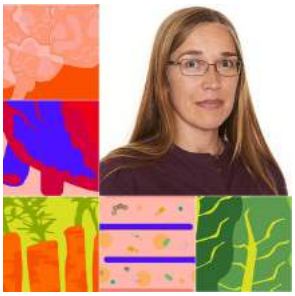
Microbiome, Gut and Systemic Health: New Frontiers in Personalised Nutrition



Justine Bold

Chronic Pain Management, the Microbiome, and Nutritional Interventions

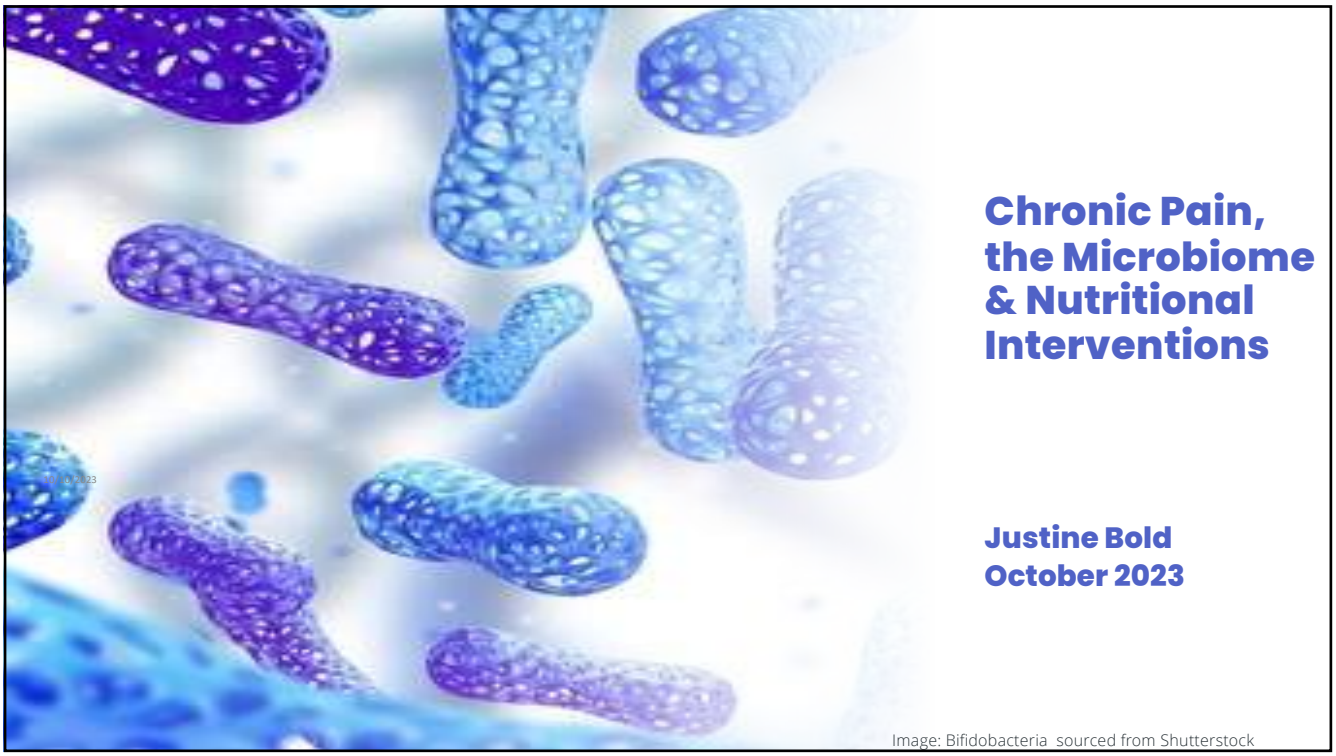
2:00-2:45pm

10/10/2023

An event by: Nutritional Medicine Institute

Platinum sponsors: regenerus labs VIRIDIAN Effective | Ethical | Pure pure encapsulations

1



Chronic Pain, the Microbiome & Nutritional Interventions

10/10/2023

Justine Bold
October 2023

Image: Bifidobacteria sourced from Shutterstock

2

Contents

Image: Sourced from Shutterstock

3

01 Introduction

4

Affiliations

- Cardiff University Medical School since January 2019
  - Programme Director CPD
  - Work on both postgraduate and undergraduate Medical Education
- University of Worcester since January 2008
  - Senior Lecturer on teaching and research contract
  - MSc Nutrition and Lifestyle Medicine
- NMI editorial board member
- Delivered some educational CPD for Lamberts
- University of Northampton, Visiting Lecturer
- Fellow of BANT, PhD Student

10/10/2023

University of Worcester

Nutrition Health Worcester

justineboldfood

justine_bold

Picture Justine Bold
https://www.cardiff.ac.uk/people/view/1584414-bold-justine

5

Microbiome has an important role in all future practice

- Disturbances in microbiome associated with many diseases:-
  - E.g. Periodontitis, inflammatory bowel disease, antibiotic associated diarrhea.
- Important to remember skin, oral, bladder, vaginal microbiota too.
- Modulation identified as therapeutic target in many conditions.
- Hasn't been covered in medical training historically.

10/10/2023

Relman DA. The Human Microbiome and the Future Practice of Medicine. *JAMA*. 2015;314(11):1127-1128.

Opinion

VIEWPOINT

SCIENTIFIC DISCOVERY AND THE FUTURE OF MEDICINE

The Human Microbiome and the Future Practice of Medicine

David A. Relman, MD
Departments of Medicine and of Microbiology and Immunology, Stanford University, Stanford, California; and Veterans Affairs Palo Alto Health Care System, Palo Alto, California.

All animals coexist in intimate, dependent relationships with microbes. Humans are no exception. Host-associated microbes, like nearly all others on this planet, form communities in which the overall composition, structure, and function are explained by ecological processes and environmental factors. Evidence of coadaptation and mutual benefit are key features of these symbioses between hosts and their microbial communities, or microbiotas.[1] The human microbiota is a fundamental component of what it means to be human.

Recent work suggests that the benefits derived by humans from their microbiotas may have profound consequences for health. These benefits include differentiation of host mucosa, food digestion and nutrition, regulation of metabolism, processing and detoxification of environmental chemicals, development and ongoing regulation of the immune system, and prevention of invasion and growth of pathogens. Conversely, disturbance and alterations of the human microbiota and its collective genes and genomes, ie, the microbiome, are associated with a wide variety of human diseases, such as chronic periodontitis, inflammatory bowel disease, and antibiotic-associated diarrhea.

The first direct observations and measurements of the human microbiota were made more than 300 years ago with the advent of the microscope and scrapings from teeth. Since then, the tools for studying the human microbiota have expanded in scope, sophistication, and availability. The current surge of interest in this topic reflects in part recent advances in DNA sequencing technology and its use in characterizing the microbial world directly from environmental samples, as well as a renewed appreciation for ecological principles, including the importance of interactions among organisms; the formation, activities, and stability of communities of microbes; and the relationships between communities and their environment. The study of the human microbiota has substantial potential for improving the management of human health and disease.

Body site is one of the strongest determinants of variation in human microbiota compositional diversity.[2] For example, microbial communities on the exposed tooth surfaces of a healthy individual generally have more similar taxonomic compositions to those on the teeth of another healthy individual than they do to those on the tongue of the same individual. In contrast, when specimens from the same body site are compared among a group of different healthy individuals, individual-specific microbiota features are apparent. Biogeographic patterns in microbiota taxonomic and genomic composition reflect differing selective pressures found at distinct body sites, as well as priority effects on community succession, rates of dispersal, and local microbial diversification. The subtle distinguishing features between microbial communities may teach us about important, underlying variation in both normal and abnormal human physiology and cell biology. For example, distinct types of epithelium, temperatures, and other environmental conditions are associated with differing bacterial communities between the anterior naris of the nose and the middle meatus and sinoethmoidal recess and associated with distinct competitive interactions among community members of interest, like *Staphylococcus aureus*.[3]

From high-diversity oral habitats to low-diversity vaginal habitats, microbial biogeography suggests distinct ecological zones across the human landscape (**Figure**). On the skin, there are 3 types of microbial communities, each characteristic of either dry, moist, or sebaceous environments.[4] *Propionibacterium acnes*, commensal staphylococci, *Corynebacterium* species, and *Propionibacterium* phage explain the greatest amount of variation between these community types; fungi and other eukaryotic microbes are relatively rare. *P acnes* strains tend to be specific to an individual, whereas *Staphylococcus epidermidis* strains tend to be specific to body site. DNA viruses, especially bacterial viruses (phage), vary considerably in number and type between individuals on the skin and elsewhere across human body mucosa and are relatively abundant in the nose and vagina. Because they can kill bacteria or modify them by carrying in new genes, phages help shape the structure and function of the human microbiota.

Site-specific features of microbiota structure and function may serve as early markers of future local disease. Focal processes such as chronic periodontitis, dental caries, atopic dermatitis, and Crohn disease are attractive settings for the identification of such features.

Small molecules mediate a wide variety of interactions among members of microbial communities, and in so doing, promote community stability; with regard to the human microbiota, small molecules also mediate and facilitate host adaptation. With the advent of computational tools for identifying the putative products of complex mixed communities based on their "meta-genomes," a wealth of new molecules have been discovered within the human microbiota. A number of them have demonstrated drug-like activities with significant medical potential.[5]

A recent examination of metagenomic sequence data obtained directly from human microbiota samples at 5 body sites identified more than 3000 biosynthetic gene clusters, each predicted to produce a small mol-

Corresponding Author: David A. Relman, MD, VA Palo Alto Health Care System 154T, 3801 Miranda Ave, Palo Alto, CA 94304 (relman@stanford.edu).

jama.com

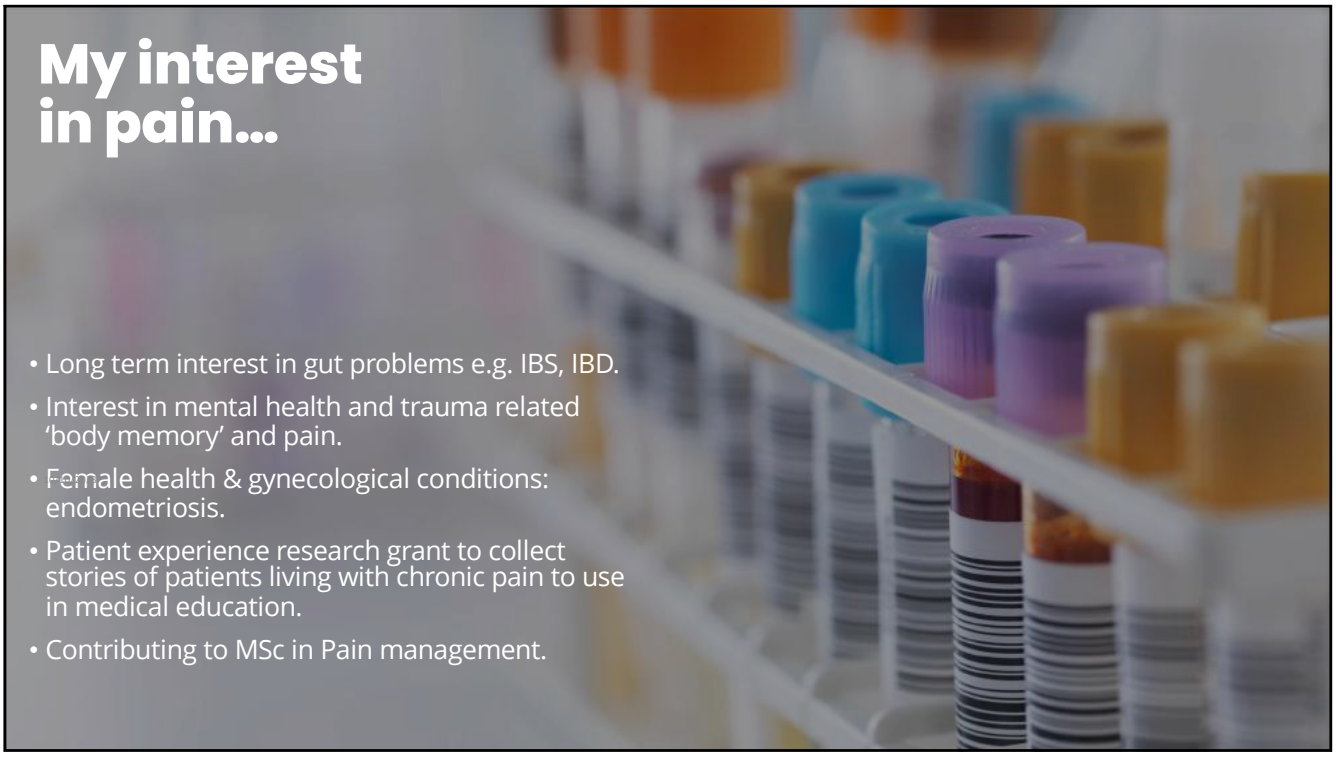
JAMA September 15, 2015 Volume 314, Number 11 112

Copyright 2015 American Medical Association. All rights reserved.

Downloaded From: https://jamanetwork.com/ by a Cardiff University User on 12/16/2021

6

My interest in pain...



- Long term interest in gut problems e.g. IBS, IBD.
- Interest in mental health and trauma related 'body memory' and pain.
- Female health & gynecological conditions: endometriosis.
- Patient experience research grant to collect stories of patients living with chronic pain to use in medical education.
- Contributing to MSc in Pain management.

7

02 Pain

10/10/2023

8

Pain

Acute or Chronic.

Pain receptors are called nociceptors, they send signals to the central nervous system (CNS).

3 main types of pain:

- **Nociceptive** (potentially harmful stimuli being detected by nociceptors around the body usually associated with tissue damage).
- **Neuropathic** (from nerve injury or nerve impairment).
- **Inflammatory**

Nociceptive pain can be categorised into **visceral** or **somatic** pain (which is further classified into two kinds: deep somatic and superficial pain).

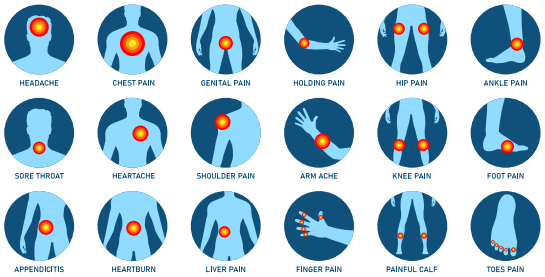


Image: Pain spots sourced from Shutterstock

9

Pain

Physiological processes:-
- Transduction
- Transmission
- Modulation
- Perception

Morreale C, Bresesti I, Bosi A, et al. Microbiota and Pain: Save Your Gut Feeling. *Cells*. 2022;11(6):971. Published 2022 Mar 11. doi:10.3390/cells11060971

10/10/2023

Overall pain is influenced by the balance of the excitatory and inhibitory influences that act on neuron circuits.

Image: Pain sourced from Shutterstock

10

Chronic pain classifications

Revised International Association for the Study of Pain (IASP) classifications for International Classification of Diseases (ICD-11) take account of both the primary and secondary causes of chronic pain.

10/10/2023

Scholz J. Finally, A Systematic Classification of Pain (the ICD-
Pract Pain Manag. 2019;19(3).

Reference 5 from above article
Treede RD, Rief W, Barke A, et al. Chronic pain as a sympton
a disease: the IASP classification of chronic pain for the International Classification of Diseases (ICD-11). *Pain*. 2019;160(1):19–27.

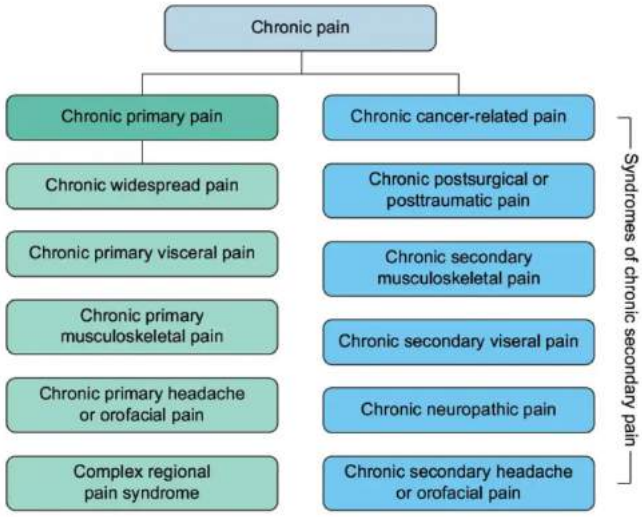


Figure 1. Chronic pain in the ICD-11. The classification distinguishes between conditions of *Chronic primary pain* and syndromes of known etiology or established pathophysiology that are associated with chronic (secondary) pain. Figure created by author, modified from Reference 5.

11

Chronic pain

Affects 20% of the European population (1 in 5 people).

More common in women and older people.

10/10/2023

van Hecke O, Torrance N, Smith BH. Chronic pain epidemiology and its clinical relevance. *Br J Anaesth*. 2013;111(1):13-18. doi:10.1093/bja/aet123

British Journal of Anaesthesia 111 (1): 13–18 (2013)
doi:10.1093/bja/aet123

BJA

REVIEW ARTICLES

Chronic pain epidemiology and its clinical relevance

O. van Hecke, N. Torrance and B. H. Smith*

Population Health Sciences Division, Medical Research Institute, University of Dundee, Dundee, UK

* Corresponding author: Mackenzie Building, Kirsty Semple Way, Dundee DD2 4BF, UK. E-mail: b.h.smith@dundee.ac.uk

Editor's key points

- Identifying risk factors allows development of healthcare strategies to reduce the burden of chronic pain.
- Around 20% of the population may be affected, with a huge impact on the wider society.
- Some risk factors cannot be changed (e.g. gender, age); others can be modified (e.g. pain severity, mood).
- Further epidemiological studies are an essential part of a chronic pain research strategy.

Summary. Chronic pain affects ~20% of the European population and is commoner in women, older people, and with relative deprivation. Its management in the community remains generally unsatisfactory, partly because of lack of evidence for effective interventions. Epidemiological study of chronic pain, through an understanding of its distribution and determinants, can inform the development, targeting, and evaluation of interventions in the general population. This paper reviews current knowledge of risk markers associated with chronic pain and considers how these might inform management and prevention. Risk factors include socio-demographic, clinical, psychological, and biological factors. These are relevant to our understanding of chronic pain mechanisms and the nature of, and responses to, current and future treatments.

Keywords: chronic pain; pain, psychological variables; risk; statistics, epidemiology

12

Chronic pain

Paper states management options are reported as unsatisfactory.

van Hecke O, Torrance N, Smith BH. Chronic pain epidemiology and its clinical relevance. *Br J Anaesth*. 2013;111(1):13-18. doi:10.1093/bja/aet123

BJA

Box 1. Socio-demographic factors associated with chronic pain

- Female gender
- Older age
- Lower socio-economic status
- Geographical and cultural background
- Employment status and occupational factors
- History of abuse or interpersonal violence

I believe this is where Nutrition and Lifestyle medicine have role to play…

13

In the UK

- Prevalence of chronic pain in the UK is 45.6% (disabling pain has a 10 to 14% prevalence).

  Fayaz A, Croft P, Langford RM, Donaldson LJ, Jones GT. Prevalence of chronic pain in the UK: a systematic review and meta-analysis of population studies. *BMJ Open*. 2016;6(6):e010364.

- It is difficult to manage in a primary care setting as pharmacological approaches are not always recommended. People continuing to take opioids have limited benefits and there are addictive side effects and Non-Steroidal Anti-inflammatories (NSAIDs) increase the risk of myocardial infarctions and gastrointestinal bleeding.

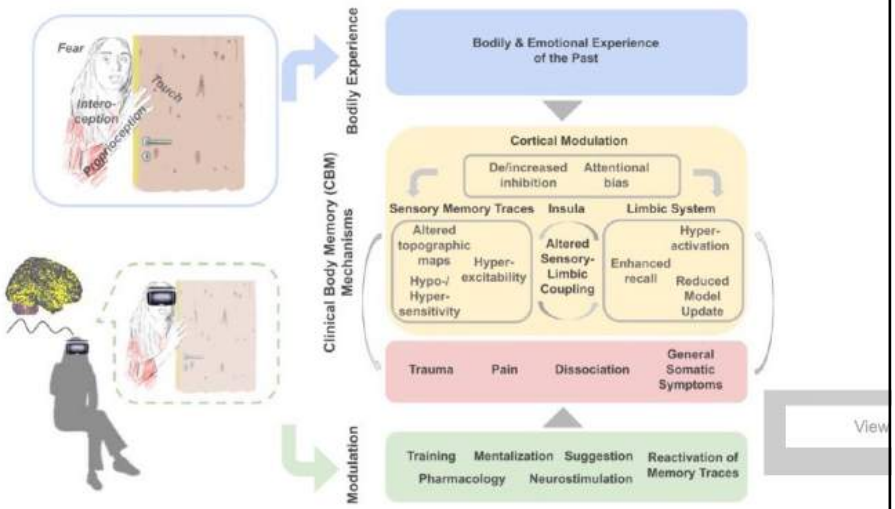
- Topical NSAIDs are recommended for the management of osteoarthritis. (NICE Osteoarthritis Care and Management 2014).

14

Trauma body memory & chronic pain

Proposed mechanism as to why people can experience real physical pain when re-living trauma without any obvious physiological basis.

Gentsch A, Kuehn E. Clinical Manifestations of Body Memories: The Impact of Past Bodily Experiences on Mental Health. *Brain Sci*. 2022;12(5):594. Published 2022 May 3. doi:10.3390/brainsci12050594



Clinical Body Memory (CBM) Mechanisms. The key hypothesis discussed here is that stored bodily experiences of the past and associated emotions (blue boxes) can contribute to the development of Clinical Body Memory (CBM) mechanisms including trauma, pain, dissociation and general somatic symptoms (red box) via neuronal and cognitive mechanisms that mediate their storage and retrieval (yellow box). Experimental investigation may allow empirical access and modulation of CBMs (green box), for example, via using Virtual Reality (VR) paradigms (left bottom).

15

03 Microbiome & Pain

16

Microbiota and pain

- 2022 narrative review.
- Microbiome has many influences on pain.
- Influence is driven via the gut brain axis.

10/10/2023

Morreale C, Bresesti I, Bosi A, et al. Microbiota and Pain: Save Your Gut Feeling. *Cells*. 2022;11(6):971. Published 2022 Mar 11. doi:10.3390/cells11060971

cells

MDPI

Review

Microbiota and Pain: Save Your Gut Feeling

Chiara Morreale [1,†], Ilia Bresesti [1,*,†], Annalisa Bosi [2], Andreina Baj [2], Cristina Giaroni [2], Massimo Agosti [1] and Silvia Salvatore [1]

[1] Department of Woman and Child, ASST-Settelaghi, University of Insubria, 21100 Varese, Italy; cmorreale@studenti.uninsubria.it (C.M.); massimo.agosti@uninsubria.it (M.A.); silvia.salvatore@uninsubria.it (S.S.)
[2] Department of Medicine and Surgery, University of Insubria, 21100 Varese, Italy; a.bosi@uninsubria.it (A.B.); andreina.baj@uninsubria.it (A.B.); cristina.giaroni@uninsubria.it (C.G.)
* Correspondence: ilia.bresesti@uninsubria.it
† These authors contributed equally to the manuscript.

Abstract: Recently, a growing body of evidence has emerged regarding the interplay between microbiota and the nervous system. This relationship has been associated with several pathological conditions and also with the onset and regulation of pain. Dysregulation of the axis leads to a huge variety of diseases such as visceral hypersensitivity, stress-induced hyperalgesia, allodynia, inflammatory pain and functional disorders. In pain management, probiotics have shown promising results. This narrative review describes the peripheral and central mechanisms underlying pain processing and regulation, highlighting the role of the gut-brain axis in the modulation of pain. We summarized the main findings in regard to the stress impact on microbiota's composition and its influence on pain perception. We also focused on the relationship between gut microbiota and both visceral and inflammatory pain and we provided a summary of the main evidence regarding the mechanistic effects and probiotics use.

Keywords: pain; microbiota; probiotics; gut-brain axis

check for updates

Citation: Morreale, C.; Bresesti, I.; Bosi, A.; Baj, A.; Giaroni, C.; Agosti, M.; Salvatore, S. Microbiota and Pain: Save Your Gut Feeling. *Cells* **2022**, *11*, 971. https://doi.org/10.3390/cells11060971

Academic Editor: Alexander E. Kalyuzhny

Received: 3 February 2022
Accepted: 8 March 2022
Published: 11 March 2022

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https://creativecommons.org/licenses/by/4.0/).

1. Introduction

Pain is a natural protective mechanism of the body that arises from nociceptors and involves the interaction of different neuroanatomic and neurochemical systems. According to the International Association for the Study of Pain (IASP): "Pain is an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage" [1]. Experience of pain is the result of the interplay between several compartments: receptors, neurotransmitters involved in the regulation of pain perception, pain-related emotions and memory [2]. Acute pain is an alarm system that protects us from tissue damage. Chronic pain (which has a duration of more than three months) is a persistent pain associated with injury, disorders or diseases (i.e., arthritis, functional gastrointestinal disorders, inflammatory bowel diseases, diabetes or tumor growth). This can be a consequence of damage to nerve fibers which leads to an increased spontaneous firing or alterations in their conduction or neurotransmitter properties [3]. Chronic pain is considered to be a disease state and the Institute of Medicine claims that more than 100 million Americans suffer from chronic pain [4]. Recently, a growing body of evidence has shown that the gut microbiota may play a role in modulating visceral pain and inflammatory and neuropathic pathways [5]. Although still limited, emerging research reports the involvement of the gut microbiota in the release of signal molecules (i.e., metabolites, neurotransmitters, neuromodulators), which are directly involved in pain transmission and modulation [5]. In this narrative review, we aim to provide a comprehensive overview regarding the influence of gut microbiota on pain perception and the potential application of probiotics in pain management.

17

Gut brain axis (GBA)

Bidirectional communication system between the gut and brain.

Regulates homeostasis and physiological functions of the GI tract, immune system and behaviour.

10/10/2023

Main effects via metabolites of gut microorganisms.

Morreale C, Bresesti I, Bosi A, et al. Microbiota and Pain: Save Your Gut Feeling. *Cells*. 2022;11(6):971. Published 2022 Mar 11. doi:10.3390/cells11060971

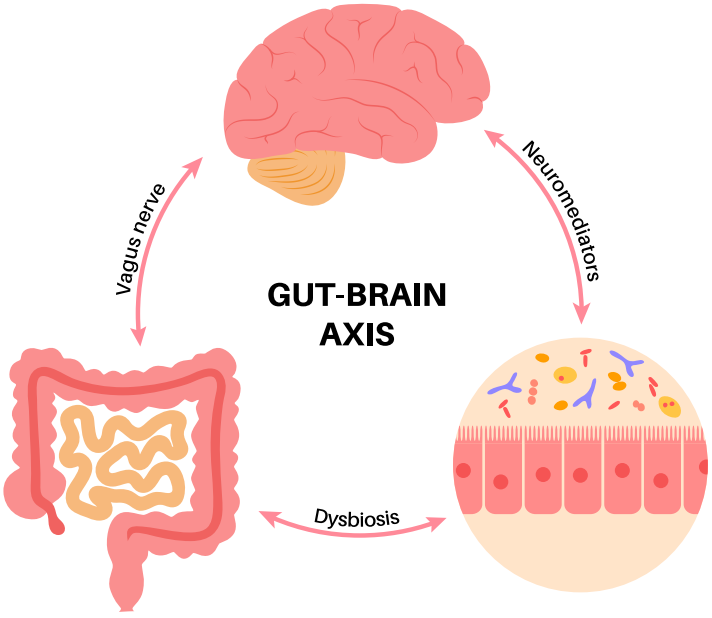
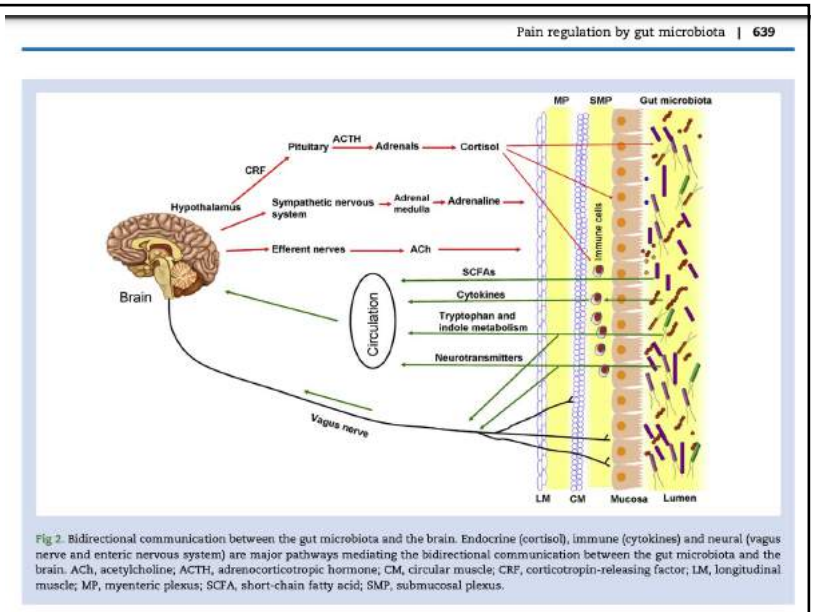


Image: Gut brain axis sourced from Shutterstock

18

Gut brain axis and the vagus nerve

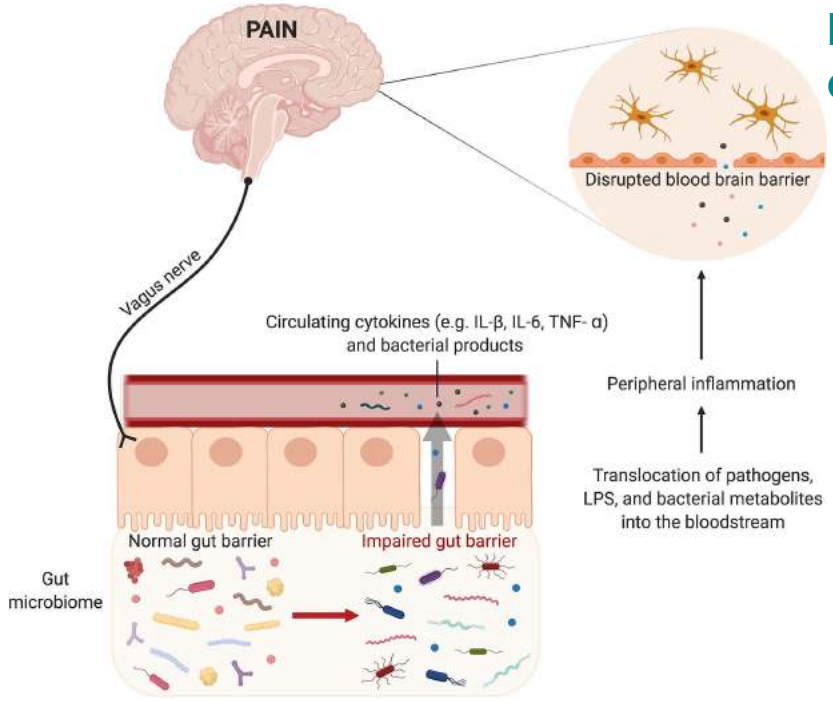
Vagus nerve (VN) is a major pathway in the bidirectional communication, it is the longest cranial nerve – it regulates multiple body systems helping to maintain homeostasis, including respiratory, cardiovascular, immune, endocrine, autonomic systems – and it courses from the medulla to the colon. Microbiota can activate the VN.



Guo R, Chen LH, Xing C, Liu T. Pain regulation by gut microbiota: molecular mechanisms and therapeutic potential. *Br J Anaesth*. 2019;123(5):637-654.

19

Pathophysiology chronic pain



Involves sensory, immune, and inflammatory interactions within both the central and peripheral nervous systems.

Issues with barrier integrity - both gut and blood brain barrier (BBB).

Zoë Dworsky-Fried, Bradley J. Kerr, Anna M.W. Taylor, Microbes, microglia, and pain, Neurobiology of Pain, Volume 7, 2020, 100045, https://doi.org/10.1016/j.ynpai.2020.100045.

20

Image: Blood Brain Barrier sourced from Shutterstock

Blood Brain Barrier

Multi-functional interface made up of cerebral endothelial cells and their linking tight junctions.

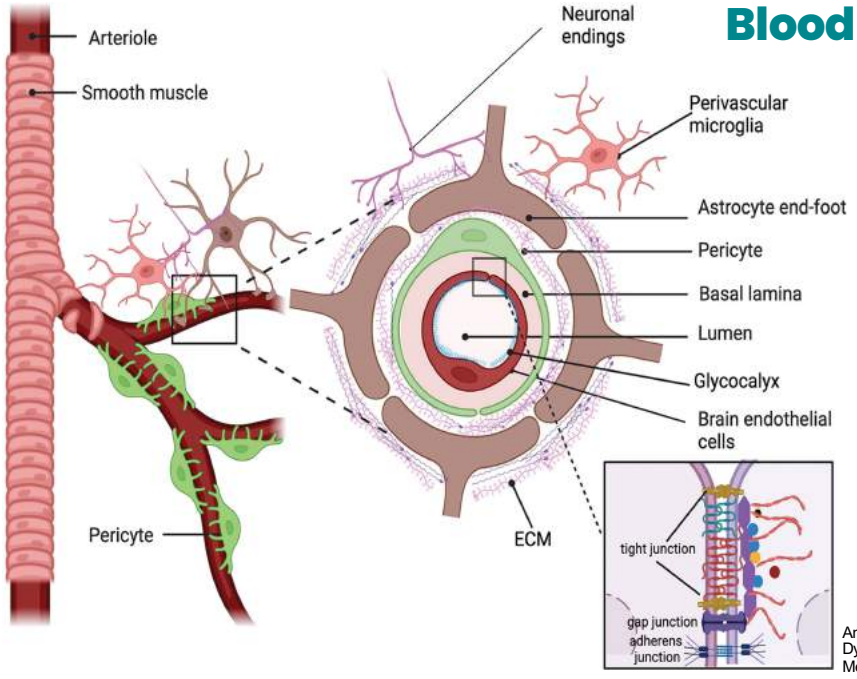
Andjelkovic AV, Situ M, Citalan-Madrid AF, Stamatovic SM, Xiang J, Keep RF. Blood-Brain Barrier Dysfunction in Normal Aging and Neurodegeneration: Mechanisms, Impact, and Treatments. *Stroke*. 2023;54(3):661-672. doi:10.1161/STROKEAHA.122.040578

Together with pericytes, astrocytes, microglia and the surrounding basement membrane, the BBB forms a selective physical barrier that separates the bloodstream from the brain.

Kang R, Gamdzyk M, Lenahan C, Tang J, Tan S, Zhang JH. The Dual Role of Microglia in Blood-Brain Barrier Dysfunction after Stroke. *Curr Neuropharmacol*. 2020;18(12):1237-1249. doi:10.2174/1570159X18666200529150907

21

Blood Brain Barrier



Microglia (macrophages of CNS) play an important role in BBB function.

Anuska V. Andjelkovic. Stroke. Blood-Brain Barrier Dysfunction in Normal Aging and Neurodegeneration: Mechanisms, Impact, and Treatments, Volume: 54, Issue: 3, Pages: 661-672, DOI: (10.1161/STROKEAHA.122.040578)

© 2023 American Heart Association, Inc.

22

2020 Paper Reduction of microglial activation from strategies that restore microbiome

Microglia are involved in the initiation and persistence of pain. They respond to signals from the CNS but are also modulated by signals from the GI tract.

Preclinical and clinical studies suggest that **communication between the gut microbiome and microglia is involved in producing chronic pain**.

Image: Lactobacillus sourced from Shutterstock

Strategies that manipulate or restore the gut microbiome have been shown to reduce microglial activation and alleviate symptoms associated with inflammation. Indicating that manipulations of the gut microbiome in chronic pain patients might be a viable strategy in improving pain outcomes.

Zoë Dworsky-Fried, Bradley J. Kerr, Anna M.W. Taylor, Microbes, microglia, and pain, Neurobiology of Pain, Volume 7, 2020, 100045, https://doi.org/10.1016/j.ynpai.2020.100045.

23

Effects of dysregulation of gut brain axis on pain

- Dysregulation of the gut brain axis can lead to:-
  - Visceral hypersensitivity.
  - Stress-induced hyperalgesia (where nocioceptors become damaged or sensitive).
  - Allodynia which is perception of pain from stimuli that are not normally painful.
  - Inflammation and inflammatory pain.

cells

MDPI

Review

Microbiota and Pain: Save Your Gut Feeling

Chiara Morreale [1,†], Ilia Bresesti [1,*,†], Annalisa Bosi [2], Andreina Bai [2], Cristina Giaroni [2], Massimo Agosti [1] and Silvia Salvatore [1]

Abstract: Recently, a growing body of evidence has emerged regarding the interplay between microbiota and the nervous system. This relationship has been associated with several pathological conditions and also with the onset and regulation of pain. Dysregulation of the axis leads to a huge variety of diseases such as visceral hypersensitivity, stress-induced hyperalgesia, allodynia, inflammatory pain and functional disorders. In pain management, probiotics have shown promising results. This narrative review describes the peripheral and central mechanisms underlying pain processing and regulation, highlighting the role of the gut-brain axis in the modulation of pain. We summarized the main findings in regard to the stress impact on microbiota's composition and its influence on pain perception. We also focused on the relationship between gut microbiota and both visceral and inflammatory pain and we provided a summary of the main evidence regarding the mechanistic effects and probiotics use.

Keywords: pain; microbiota; probiotics; gut-brain axis

1. Introduction

Pain is a natural protective mechanism of the body that arises from nociceptors and involves the interaction of different neuroanatomic and neurochemical systems. According to the International Association for the Study of Pain (IASP): "Pain is an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage" [1]. Experience of pain is the result of the interplay between several compartments: receptors, neurotransmitters involved in the regulation of pain perception, pain-related emotions and memory [2]. Acute pain is an alarm system that protects us from tissue damage. Chronic pain (which has a duration of more than three months) is a persistent pain associated with injury, disorders or diseases (i.e., arthritis, functional gastrointestinal disorders, inflammatory bowel diseases, diabetes or tumor growth). This can be a consequence of damage to nerve fibers which leads to an increased spontaneous firing or alterations in their conduction or neurotransmitter properties [3]. Chronic pain is considered to be a disease state and the Institute of Medicine claims that more than 100 million Americans suffer from chronic pain [4]. Recently, a growing body of evidence has shown that the gut microbiota may play a role in modulating visceral pain and inflammatory and neuropathic pathways [5]. Although still limited, emerging research reports the involvement of the gut microbiota in the release of signal molecules (i.e., metabolites, neurotransmitters, neuromodulators), which are directly involved in pain transmission and modulation [6]. In this narrative review, we aim to provide a comprehensive overview regarding the influence of gut microbiota on pain perception and the potential application of probiotics in pain management.

Cells 2022, 11, 971. https://doi.org/10.3390/cells11060971 https://www.mdpi.com/journal/cells

Morreale C, Bresesti I, Bosi A, et al. Microbiota and Pain: Save Your Gut Feeling. *Cells*. 2022;11(6):971. Published 2022 Mar 11. doi:10.3390/cells11060971

10/10/2023

24

Gut microorganism's metabolites, neurotransmitters and effects on CNS

Examples of gut microorganisms that are able to produce molecules with effects on CNS and behavior (↑ = increase; ↓ = decrease).

Gut Microorganism	Molecules, Metabolites and Neurotransmitters Involved	Effects on Gut-Brain Axis	Notes re study
Bifidobacterium longum NCC3001	↑ BDNF	↓ Anxiety and depressive behavior [30,58] ↑ Neuronal plasticity of ENS [58]	reviews review
Bifidobacterium dentium	↑ GABA	↓ Visceral hypersensitivity [59]	murine
Lacticaseibacillus rhamnosus JB-1	↑ GABA	↓ Anxiety and depressive behavior [59] ↓ Intestinal damage and inflammation [64]	murine rat model
10/10/2023 *Escherichia coli* Nissle 1917	↑ C12AsnGABAOH	↓ Visceral hypersensitivity [61] ↑ Epithelial permeability of GABA [61]	Chromatography/spectrometry to identify metabolite *Escherichia coli* Nissle 1917 in vitro and in viv0
Limosilactobacillus reuteri ATTC-PTA 6475	↑ Oxytocin	↓ Restores social deficits of ASD [24] Promotes DC maturation and immune modulation via IL-10 [65]	murine murine

Morreale C, Bresesti I, Bosi A, et al. Microbiota and Pain: Save Your Gut Feeling. *Cells*. 2022;11(6):971. Published 2022 Mar 11. doi:10.3390/cells11060971

25

Role of microbiota in pain regulation

BJA

British Journal of Anaesthesia, 123 (5): 637–654 (2019)

doi: 10.1016/j.bja.2019.07.026

Advance Access Publication Date: 21 September 2019

Review Article

Pain regulation by gut microbiota: molecular mechanisms and therapeutic potential

Ran Guo[2,†], Li-Hua Chen[2,†], Chungen Xing[4,*] and Tong Liu[3,5,*]

[1]Department of Pain, Zhejiang Provincial People's Hospital, People's Hospital of Hangzhou Medical College, Hangzhou, China, [2]Jiangsu Key Laboratory of Preventive and Translational Medicine for Geriatric Diseases, Department of Nutrition and Food Hygiene, School of Public Health, Suzhou, China, [3]Jiangsu Key Laboratory of Neuropsychiatric Diseases and Institute of Neuroscience, Soochow University, Suzhou, China, [4]Department of General Surgery, The Second Affiliated Hospital of Soochow University, Suzhou, China and [5]College of Life Sciences, Yanan University, Yanan, China

*Corresponding authors. E-mails: xingcg@suda.edu.cn, liutong80@suda.edu.cn

†R. Guo and L.-H. Chen contributed equally to this work.

Summary

The relationship between gut microbiota and neurological diseases, including chronic pain, has received increasing attention. The gut microbiome is a crucial modulator of visceral pain, whereas recent evidence suggests that gut microbiota may also play a critical role in many other types of chronic pain, including inflammatory pain, headache, neuropathic pain, and opioid tolerance. We present a narrative review of the current understanding on the role of gut microbiota in pain regulation and discuss the possibility of targeting gut microbiota for the management of chronic pain. Numerous signalling molecules derived from gut microbiota, such as by-products of microbiota, metabolites, neurotransmitters, and neuromodulators, act on their receptors and remarkably regulate the peripheral and central sensitisation, which in turn mediate the development of chronic pain. Gut microbiota-derived mediators serve as critical modulators for the induction of peripheral sensitisation, directly or indirectly regulating the excitability of primary nociceptive neurones. In the central nervous system, gut microbiota-derived mediators may regulate neuroinflammation, which involves the activation of cells in the blood–brain barrier, microglia, and infiltrating immune cells, to modulate induction and maintenance of central sensitisation. Thus, we propose that gut microbiota regulates pain in the peripheral and central nervous system, and targeting gut microbiota by diet and pharmabiotic intervention may represent a new therapeutic strategy for the management of chronic pain.

Keywords: inflammation; gut–brain axis; microbiome; microbiome–gut–brain axis; pain; pharmabiotic

10/10/2023

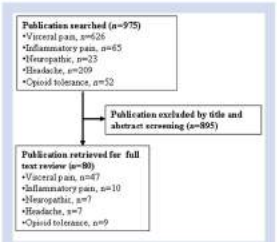
"The gut microbiome is a crucial modulator of visceral pain, whereas recent evidence suggests that gut microbiota may also play a critical role in many other types of chronic pain, including inflammatory pain, headache, neuropathic pain."

"can directly modulate dorsal root ganglia neuronal excitability, and regulate neuroinflammation in the peripheral and central nervous systems under chronic pain conditions."

Guo R, Chen LH, Xing C, Liu T. Pain regulation by gut microbiota: molecular mechanisms and therapeutic potential. *Br J Anaesth*. 2019;123(5):637-654.

26

2019 review – human studies included



Most studies on visceral pain - interventions include antibiotic treatments and administration of probiotics and there's a mix of animal and human studies.

Guo R, Chen LH, Xing C, Liu T. Pain regulation by gut microbiota: molecular mechanisms and therapeutic potential. *Br J Anaesth*. 2019;123(5):637-654.

Table 1 Chronic pain regulated by gut microbiota in preclinical and clinical studies. ABX, antibiotic cocktail; CFA, complete Freund's adjuvant; GF, germ-free; IBS, irritable bowel syndrome; MSU, monosodium urate monohydrate; NMD, neonatal maternal deprivation; PAG, periaqueductal grey; PFC, prefrontal cortex; TLR, Toll-like receptor; PDX/GOS, polydextrose/galacto-oligosaccharide.

Pain condition	Reference	Type of study	Involvement of microbiota
Visceral pain	Pimentel and colleagues[49]	Human clinical trial	Antibiotic rifaximin reduced IBS-related pain.
	Verdu and colleagues[38]	Preclinical animal study	Visceral pain to colorectal distension was increased by antibiotic cocktail and reverses by *Lactobacillus paracasei* in mice.
	Saulnier and colleagues[51]	Human clinical trial	A mixture of *Bifidobacterium infantis* M-63, *breve* M-16V, and *longum* BB536 improved abdominal pain in paediatric IBS patients.
	Ringel-Kulka and colleagues[52]	Human clinical trial	*Lactobacillus acidophilus* NCFM reduced functional abdominal pain in adults.
	Kannampalli and colleagues[45]	Preclinical animal study	Administration of the probiotic *Lactobacillus* GG or the prebiotic combination of PDX/GOS reduced neonatal inflammation-induced visceral hypersensitivity in rats.
	O'Mahony and colleagues[29]	Preclinical animal study	Treatment with antibiotic vancomycin and cocktail in early life lead to an increased visceral pain by colorectal distension in rats.
	Aguilera and colleagues[37]	Preclinical animal study	Antibiotic cocktail reduced visceral pain by acetic acid and intracolonic capsaicin in mice.
	Perez-Burgos and colleagues[108]	Preclinical animal study	Administration of *Lactobacillus reuteri* DSM 17938 reduced jejunal spinal nerve firing evoked by gastric distension or injection of capsaicin in rodents.
	Miquel and colleagues[43]	Preclinical animal study	NMD leads to visceral pain and a reduced diversity of faecal microbiota and administration of *Faecalibacterium prausnitzii* reduces visceral pain after NMD in rats.
	Weizman and colleagues[109]	Human clinical trial	A randomised, double-blind, placebo-controlled trial found administration of *Lactobacillus reuteri* DSM 17938 is beneficial in functional abdominal pain of childhood.
	Spiller and colleagues[110]	Human clinical trial	Randomised, double-blind, placebo-controlled trial found *Saccharomyces cerevisiae* CNCM I-3856 at the dose of 1000 mg day^{-1} does not improve intestinal pain and discomfort in general IBS patients.
	Luczynski and colleagues[46]	Preclinical animal study	GF mice displayed visceral hypersensitivity accompanied by up-regulation of TLRs and cytokines in the spinal cord, which were abolished by postnatal colonisation with microbiota from conventionally colonised.
	Pokusaeva and colleagues[111]	Preclinical animal study	Oral administration of a GABA-producing *Bifidobacterium* strain (*B. dentium* ATCC 27678) reduced visceral hypersensitivity in a rat faecal retention model.
	Zhao and colleagues[112]	Preclinical animal study	Treatment with *Clostridium butyricum* exerted a beneficial effect on visceral hypersensitivity of IBS by inhibiting low

14/10/2023

27

2011 Study in 2019 review

Meta analysis **Lactobacillus rhamnosus GG for abdominal pain** - related functional gastrointestinal disorders in childhood.

Table 1 | Characteristics of included studies

Study	Participants	Diagnostic criteria	LGG (dose)	Comparison	Primary outcome	Secondary outcomes
Bausserman *et al.* 2005[18]	Children & adolescents (mean age: 12 years, range: 6–17 years)	Rome II criteria for IBS	10^{10} CFU, twice daily, for 6 weeks	Placebo (inulin)	Change in the abdominal pain severity score	Number of responders vs. nonresponders in each group and changes in the symptoms on the Gastrointestinal Symptom Rating Scale (15-item GSRS) by syndrome. Responders were classified as patients with a decrease in abdominal pain severity (1 point or more on the 4-point Likert scale)
Francavilla *et al.* 2010[12]	Children (mean age: 6.4 years, range: 5–14 years)	Rome II criteria for IBS or FAP	3×10^9 CFU, twice daily, for 8 weeks	Placebo (inert powder)	Change in abdominal pain (frequency/severity) according to the VAS score from baseline to the end of the treatment period	(i) A decrease of at least 50% in the number of episodes and intensity of pain (treatment success); (ii) A decrease in the perception of children's pain according to their parents; (iii) Modification of intestinal permeability
Gawronska *et al.* 2007[19]	Children (mean age: 11.6 years, range: 6–16 years)	Rome II criteria for IBS, FAP, FD	3×10^9 CFU, twice daily, for 4 weeks	Placebo (maltodextrin)	Treatment success defined as no pain (a relaxed face, score of 0, on the Faces Pain Scale) at the end of the intervention	Improvements defined as a change in: (i) the Faces Pain Scale by at least 2 faces scores; (ii) self-reported severity of pain during the preceding week measured on the Faces Pain Scale; (iii) self-reported frequency of pain during the preceding week; (iv) use of medication for abdominal pain; and (v) school absenteeism because of abdominal pain

CFU, colony forming units; IBS, irritable bowel syndrome; FAP, functional abdominal pain; FD, functional dyspepsia; LGG, *Lactobacillus rhamnosus* GG; VAS, visual analogue scale.

Interventions lasted not less than 4weeks, which is in line with the Rome Foundation document providing guidance for the design of treatment trials in patients with FGD.

Horvath A, Dziechciarz P, Szajewska H. Meta-analysis: Lactobacillus rhamnosus GG for abdominal pain-related functional gastrointestinal disorders in childhood. *Aliment Pharmacol Ther*. 2011;33(12):1302-1310. doi:10.1111/j.1365-2036.2011.04665.x

14/10/2023

28

Findings from 2011 Study in 2019 review

Table 1 | Characteristics of included studies

Study	Participants	Diagnostic criteria	LGG (dose)	Comparison	Primary outcome	Secondary outcomes
Bausserman et al. 2005[18]	Children & adolescents (mean age: 12 years, range: 6–17 years)	Rome II criteria for IBS	10^{10} CFU, twice daily, for 6 weeks	Placebo (inulin)	Change in the abdominal pain severity score	Number of responders vs. nonresponders in each group and changes in the symptoms on the Gastrointestinal Symptom Rating Scale (15-item GSRS) by syndrome. Responders were classified as patients with a decrease in abdominal pain severity (1 point or more on the 4-point Likert scale)
Francavilla et al. 2010[12]	Children (mean age: 6.4 years, range: 5–14 years)	Rome II criteria for IBS or FAP	3×10^9 CFU, twice daily, for 8 weeks	Placebo (inert powder)	Change in abdominal pain (frequency/severity) according to the VAS score from baseline to the end of the treatment period	(i) A decrease of at least 50% in the number of episodes and intensity of pain (treatment success); (ii) A decrease in the perception of children's pain according to their parents; (iii) Modification of intestinal permeability
Gawronska et al. 2007[19]	Children (mean age: 11.6 years, range: 6–16 years)	Rome II criteria for IBS, FAP, FD	3×10^9 CFU, twice daily, for 4 weeks	Placebo (maltodextrin)	Treatment success defined as no pain (a relaxed face, score of 0, on the Faces Pain Scale) at the end of the intervention	Improvements defined as a change in: (i) the Faces Pain Scale by at least 2 faces scores; (ii) self-reported severity of pain during the preceding week measured on the Faces Pain Scale; (iii) self-reported fre quency of pain during the preceding week; (iv) use of medication for abdominal pain; and (v) school absenteeism because of abdominal pain

CFU, colony forming units; IBS, irritable bowel syndrome; FAP, functional abdominal pain; FD, functional dyspepsia; LGG, *Lactobacillus rhamnosus* GG; VAS, visual analogue scale.

Compared with placebo, LGG supplementation was associated with a significantly higher rate of treatment responders (defined as no pain or a decrease in pain intensity) in the overall population with abdominal pain-related functional gastrointestinal disorders.

Horvath A, Dziechciarz P, Szajewska H. Meta-analysis: Lactobacillus rhamnosus GG for abdominal pain-related functional gastrointestinal disorders in childhood. *Aliment Pharmacol Ther*. 2011;33(12):1302-1310. doi:10.1111/j.1365-2036.2011.04665.x

29

From the review – Lactobacillus acidophilus modulates intestinal pain

Oral administration of specific Lactobacillus strains induced the expression of mu-opioid and cannabinoid receptors in intestinal epithelial cells, and mediated analgesic functions in the gut-similar to the effects of morphine.

These results suggest that the microbiology of the intestinal tract influences our visceral perception.

nature medicine BRIEF COMMUNICATIONS

Lactobacillus acidophilus modulates intestinal pain and induces opioid and cannabinoid receptors

Christel Rousseaux[1–3], Xavier Thuru[1–3,10], Agathe Gelot[4–6,10], Nicolas Barnich[7], Christel Neut[7–9], Laurent Dubuquoy[1–3], Caroline Dubuquoy[1–3], Emilie Merour[1–3], Karen Geboes[8], Mathias Chamaillard[1–3], Arthur Ouwehand[9], Greg Leyer[9], Didier Carcano[9], Jean-Frédéric Colombel[1–3], Denis Ardid[4–6] & Pierre Desreumaux[1–3]

Abdominal pain is common in the general population and, in patients with irritable bowel syndrome, is attributed to visceral hypersensitivity. We found that oral administration of specific *Lactobacillus* strains induced the expression of μ-opioid and cannabinoid receptors in intestinal epithelial cells, and mediated analgesic functions in the gut—similar to the effects of morphine. These results suggest that the microbiology of the intestinal tract influences our visceral perception, and suggest new approaches for the treament of abdominal pain and irritable bowel syndrome.

© 2007 Nature Publishing Group http://www.nature.com/naturemedicine

Gut microbiota are composed of different bacterial species, which are involved in the metabolism of nutrients, the maturation of the intestinal epithelium, vasculature and lymphoid tissue, and protection from pathogens[1,2]. The interest in probiotics as a means to restore and maintain health continues to gain momentum. The administration of certain probiotics may prevent abdominal symptoms observed in irritable bowel syndrome—the most common gastrointestinal disease, with unknown etiology and symptoms such as abdominal pain, bloating and altered bowel habits[3–6]. Therefore, we hypothesized that probiotics may induce the expression of receptors on epithelial cells that locally control the transmission of nociceptive information to the intestinal nervous system. Promising candidates include opioid (μ-, δ- and κ-) receptors and cannabinoid receptor 2 (CB2) (refs. 7,8), which have been shown to exert analgesic effects and to have anti-inflammatory functions in several experimental models of colitis[8–11].

We first evaluated the ability of five well known and representative probiotic bacteria belonging to the *Lactobacillus* and *Bifidobacterium* genera (*L. acidophilus* NCFM, *L. salivarius* Ls-33, *L. paracasei* Lpc-37, *B. lactis* Bi-07 and *B. lactis* Bl-04), compared to commensal and adherent-invasive (LF82) *Escherichia coli*, to induce the expression of analgesic receptors. *L. acidophilus* NCFM and *L. salivarius* Ls-33 induced a sustained increase of *OPRM1* mRNA expression in human HT-29 epithelial cells, starting 1 h after bacterial stimulation (**Fig. 1a**). This induction was of the same magnitude as that observed in HT-29 epithelial cells stimulated for 2 h with a positive control (TNF-α; ref. 7). The other probiotics, *L. paracasei* Lpc-37, *B. lactis* Bi-07 and *B. lactis* Bl-04, and the two *E. coli* strains were ineffective (**Fig. 1a**). We found no increase in the expression of δ- and κ-opioid receptors in bacteria-stimulated HT-29 epithelial cells. Concerning cannabinoid receptor expression, only the *L. acidophilus* NCFM strain was able to induce significant *CNR2* mRNA expression compared to that observed in resting epithelial cells ($P < 0.01$, **Fig. 1a**). We observed no induction of *CNR2* mRNA expression in TNF-α–stimulated HT-29 epithelial cells. The inducible effect of NCFM on *OPRM1* and *CNR2* expression in epithelial cells was equally reproduced when we used bacteria killed by 80 °C heat (**Fig. 1b**). Mouse *Rela*$^{-/-}$ embryonic fibroblasts lost their ability to respond to NCFM stimulation, compared to wild-type cells, illustrating the essential role of the NF-κB pathway in the induction of *OPRM1* and *CNR2* by this strain. Next, we conducted a series of *in vivo* experiments to investigate the expression and function of MOR and CB2 in mice and rats, using the live NCFM strain. In an immunohistochemistry analysis using antibodies to opioid receptor μ1 (MOR1) and CB2, we detected the expression of both these receptors in HT-29 epithelial cells incubated with the NCFM strain (**Fig. 1c**). To confirm these results *in vivo*, we orally administered NCFM, at a clinically relevant concentration (10^9 colony-forming units (CFU) per day for 15 consecutive days), to Balb/c mice and Sprague-Dawley rats. We found a rapid and sustained enhancement of NCFM DNA in the feces of mice and rats (**Fig. 1d**). In the colonic section of untreated mice and rats, we detected MOR1 and CB2 expression in approximately 0–20% of epithelial cells; in contrast, the administration of NCFM induced the expression of these proteins in 25–60% of epithelial cells (**Fig. 1e,f**). Over the 15 d during which live NCFM was administered, we observed that the changes in weight and the duration of oro-anal transit were similar in all treated and untreated rodents. We found no macroscopic or histological alteration in NCFM-treated rodents, demonstrating that NCFM does not induce adverse effects in the intestinal tract.

To determine the functional role of NCFM-induced analgesic receptors (**Fig. 2a**), we assessed the visceral perception of rats using

[1]Institut National de la Santé et de la Recherche Médicale (INSERM) U795, Hôpital Swynghedauw, Rue A Verhaeghe, 59037 Lille Cedex, France. [2]University of Lille 2, Hôpital Swynghedauw, Rue A Verhaeghe, 59037 Lille Cedex, France. [3]Digestive Tract Diseases and Nutrition Department, Huriez Hospital, 1 Place de Verdun, 59037 Lille Cedex, France. [4]INSERM U766, 28 Place Henri Dunant, BP 38, 63001 Clermont-Ferrand, France. [5]Laboratoire de Pharmacologie Médicale, University of Clermont 1, Unité de Formation et de Recherche Médecine, 28 Place Henri Dunant, BP 38, 63001 Clermont-Ferrand, France. [6]Institut Universitaire et Technologique de Biologie, Ensemble Universitaire des Cézeaux, BP 86, 63172 Aubière, France. [7]Laboratoire de Pathogénie Bactérienne Intestinale, Centre Biomédical de Recherche et de Valorisation, Facultés de Médecine et de Pharmacie, 28 Place Henri Dunant, 63001 Clermont-Ferrand, France. [8]Department of Pathology and Gastroenterology, University of Leuven, Minderbroedersstraat 12, 3000 Leuven, Belgium. [9]Danisco Cultures Division, Danisco, 20 Rue Brunel, 75017 Paris, France. [10]These authors contributed equally to this work. Correspondence should be addressed to P.D. (pdesreumaux@hotmail.com).

Received 18 July; accepted 13 November; published online 10 December 2006; doi:10.1038/nm1521

Rousseaux, C., Thuru, X., Gelot, A. *et al. Lactobacillus acidophilus* modulates intestinal pain and induces opioid and cannabinoid receptors. *Nat Med* **13**, 35–37 (2007). https://doi.org/10.1038/nm1521

30

From the review feasibility study - probiotics in migraine

29 patients with migraine took 2 g/d of a probiotic food supplement (Ecologic(®)Barrier, 2.5×10(9) cfu/g) for 12 weeks.

The mean±standard deviation (SD) **number of migraine** days/month **decreased significantly** from 6.7±2.4 at baseline to 5.1±2.2 (P=0.008) in week 5-8 and 5.2±2.4 in week 9-12 (P=0.001).

The mean±SD **intensity of migraine decreased significantly** from 6.3±1.5 at baseline to 5.5±1.9 after treatment (P=0.005). **But, the mean Headache Disability Inventory did not change significantly.**

10/10/2023

Probiotics may decrease migraine, supporting a possible role for the intestine in migraine management.

Image: Lactobacillus sourced from Shutterstock

Feasibility and lack of adverse reactions justify further placebo-controlled studies.

de Roos NM, Giezenaar CG, Rovers JM, Witteman BJ, Smits MG, van Hemert S. The effects of the multispecies probiotic mixture Ecologic®Barrier on migraine: results of an open-label pilot study. *Benef Microbes*. 2015;6(5):641-646. doi:10.3920/BM2015.0003

31

Formulation used in the study on probiotics in migraine treatment

Formulation details

Indication	Reducing vulnerability to depression and improving brain functioning under stress.			
Colony forming units (CFU)	2.5 x 10^9 CFU/gram.			
Recommended daily dosage	2-4 grams.			
Bacterial strains	*B. bifidum* W23 *B. lactis* W51 *B. lactis* W52	*L. acidophilus* W37 *L. brevis* W63	*L. casei* W56 *L. salivarius* W24	*Lc. lactis* W19 *Lc. lactis* W58
PROBIOACT® Technology	Protective and nutritional ingredients that improve the stability of the formulation, GI survival and metabolic activity of the bacteria.			
Treatment period	For as long as desired/needed.			
Storage and stability	2 years stable at room temperature, no refrigeration needed.			
Available dosage forms	Dry powder which can be supplied as bulk or sachets and fully packed (with your design).			
Safety and Quality Profile	All probiotic strains have the Qualified Presumption of Safety (QPS) status[4]. Winclove is a NSF International Certified GMP Facility for manufacturing dietary supplements and is ISO 22000:2005 certified for the development and production of pre-and probiotics.			
Marketing	Medically endorsed under private label on a co-branding basis. Co-branding enables our business partners to use the scientific data in their marketing communication.			

10/10/2023

https://www.knowde.com/stores/winclove-probiotics/documents/270103

32

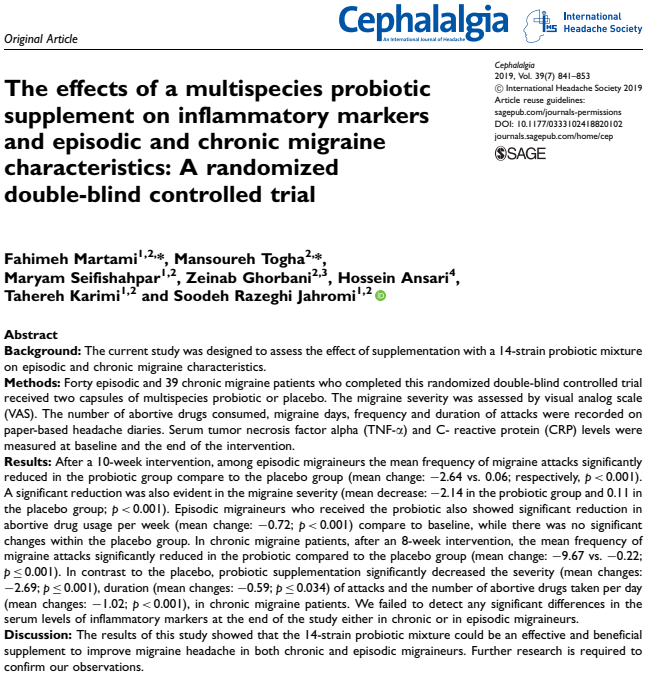
Probiotics in migraine treatment (not from the review)

Controlled RCT, 2 capsules a day
10 week intervention period, inflammatory markers measured at baseline and end of intervention - 14 strain mixture (BioKult)

Mean frequency of migraines significantly reduced and severity of migraines, no significant differences in inflammatory markers

10/11/2023

Martami F, Togha M, Seifishahpar M, et al. The effects of a multispecies probiotic supplement on inflammatory markers and episodic and chronic migraine characteristics: A randomized double-blind controlled trial. *Cephalalgia*. 2019;39(7):841-853. doi:10.1177/0333102418820102



Original Article

Cephalalgia
An International Journal of Headache
International Headache Society

Cephalalgia
2019, Vol. 39(7) 841–853
© International Headache Society 2019
Article reuse guidelines:
sagepub.com/journals-permissions
DOI: 10.1177/0333102418820102
journals.sagepub.com/home/cep
SAGE

The effects of a multispecies probiotic supplement on inflammatory markers and episodic and chronic migraine characteristics: A randomized double-blind controlled trial

Fahimeh Martami[1,2,*], Mansoureh Togha[2,*], Maryam Seifishahpar[1,2], Zeinab Ghorbani[2,3], Hossein Ansari[4], Tahereh Karimi[1,2] and Soodeh Razeghi Jahromi[1,2]

Abstract

Background: The current study was designed to assess the effect of supplementation with a 14-strain probiotic mixture on episodic and chronic migraine characteristics.

Methods: Forty episodic and 39 chronic migraine patients who completed this randomized double-blind controlled trial received two capsules of multispecies probiotic or placebo. The migraine severity was assessed by visual analog scale (VAS). The number of abortive drugs consumed, migraine days, frequency and duration of attacks were recorded on paper-based headache diaries. Serum tumor necrosis factor alpha (TNF-α) and C- reactive protein (CRP) levels were measured at baseline and the end of the intervention.

Results: After a 10-week intervention, among episodic migraineurs the mean frequency of migraine attacks significantly reduced in the probiotic group compare to the placebo group (mean change: −2.64 vs. 0.06; respectively, $p < 0.001$). A significant reduction was also evident in the migraine severity (mean decrease: −2.14 in the probiotic group and 0.11 in the placebo group; $p < 0.001$). Episodic migraineurs who received the probiotic also showed significant reduction in abortive drug usage per week (mean change: −0.72; $p < 0.001$) compare to baseline, while there was no significant changes within the placebo group. In chronic migraine patients, after an 8-week intervention, the mean frequency of migraine attacks significantly reduced in the probiotic compared to the placebo group (mean change: −9.67 vs. −0.22; $p \leq 0.001$). In contrast to the placebo, probiotic supplementation significantly decreased the severity (mean changes: −2.69; $p \leq 0.001$), duration (mean changes: −0.59; $p \leq 0.034$) of attacks and the number of abortive drugs taken per day (mean changes: −1.02; $p < 0.001$), in chronic migraine patients. We failed to detect any significant differences in the serum levels of inflammatory markers at the end of the study either in chronic or in episodic migraineurs.

Discussion: The results of this study showed that the 14-strain probiotic mixture could be an effective and beneficial supplement to improve migraine headache in both chronic and episodic migraineurs. Further research is required to confirm our observations.

33

GABA and visceral hypersensitivity (animal study)

GABA is an inhibitory neurotransmitter and regulator of abdominal/central pain perception from peripheral afferent neurons.

Gut bacteria are reported to produce GABA, it is not known whether the microbial-derived neurotransmitter modulates abdominal pain.

To investigate the potential analgesic effects of microbial GABA, we performed daily oral administration of a specific Bifidobacterium strain (B. dentiumATCC 27678) in a rat fecal retention model of visceral hypersensitivity, and subsequently evaluated pain responses.

10/11/2023

Daily oral administration of this specific Bifidobacterium strain modulated sensory neuron activity in a rat fecal retention model of visceral hypersensitivity.

Pokusaeva K, Johnson C, Luk B, et al. GABA-producing Bifidobacterium dentium modulates visceral sensitivity in the intestine. *Neurogastroenterol Motil*. 2017;29(1):e12904. doi:10.1111/nmo.12904

34

Review on SCFA's and inflammation

The ability of leukocytes to migrate to the foci of inflammation and to destroy microbial pathogens also seems to be affected by the SCFAs.

The effects of these fatty acids on isolated cells (leukocytes, endothelial and intestinal epithelial cells) and, particularly, on the recruitment and activation of leukocytes are discussed.

10/10/2023

Therapeutic application of these fatty acids for the treatment of inflammatory pathologies is also highlighted.

Vinolo MA, Rodrigues HG, Nachbar RT, Curi R. Regulation of inflammation by short chain fatty acids. *Nutrients*. 2011;3(10):858-876. doi:10.3390/nu3100858

35

SCFA's and inflammation

SCFAs acetate, propionate and butyrate are the main metabolic products of anaerobic bacteria fermentation in the gut.

Provide fuel for intestinal epithelial cells and they **modulate processes in the GI tract** such as electrolytes and water absorption.

Image: SCFA test sourced from Shutterstock

SCFAs act on leukocytes and endothelial cells through two mechanisms: activation of G protein coupled receptors GPCRs (GPR41 and GPR43) and inhibition of histone deacetylase (HDAC).

10/10/2023

SCFAs regulate several leukocyte functions including production of cytokines (TNF-α, IL-2, IL-6 and IL-10), eicosanoids and chemokines (e.g., MCP-1 and CINC-2).

Vinolo MA, Rodrigues HG, Nachbar RT, Curi R. Regulation of inflammation by short chain fatty acids. *Nutrients*. 2011;3(10):858-876. doi:10.3390/nu3100858

36

04 Interventions

10/10/2023

37

Nutritional factors

- ✓ **Prebiotics**
  Fibre, soluble fibre.
- ✓ **Live probiotic foods**
  Kefir, yoghurts, kimchi, miso, kraut, aged cheese.
- ✓ **Probiotics Supplements**
  .
- ✓ **Enhancers**
  Omega 3, antioxidants.
- x **Irritants**
  Alcohol, high fat, high sugar, pesticides

10/10/2023

38

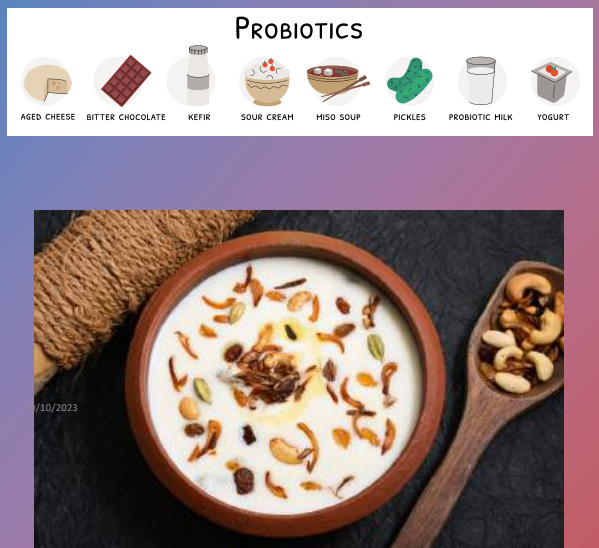
Prebiotics

- Prebiotics are a group of nutrients that are degraded by gut microbiota.
- They can feed the intestinal microbiota, and their degradation products are short-chain fatty acids that are released into blood circulation affecting not only the gastrointestinal (GI) tract but also other organs.

Davani-Davari D, Negahdaripour M, Karimzadeh I, et al. Prebiotics: Definition, Types, Sources, Mechanisms, and Clinical Applications. Foods. 2019;8(3):92. Published 2019 Mar 9.

- Includes fruit, veggies, coffee, also omega 3 has probiotic effects.

39



Probiotic foods

- Foods containing beneficial bacteria.
- Eat a range and some daily.
- Explore supplements of probiotics too and gut mucosa support using functional medicine informed approach.

40

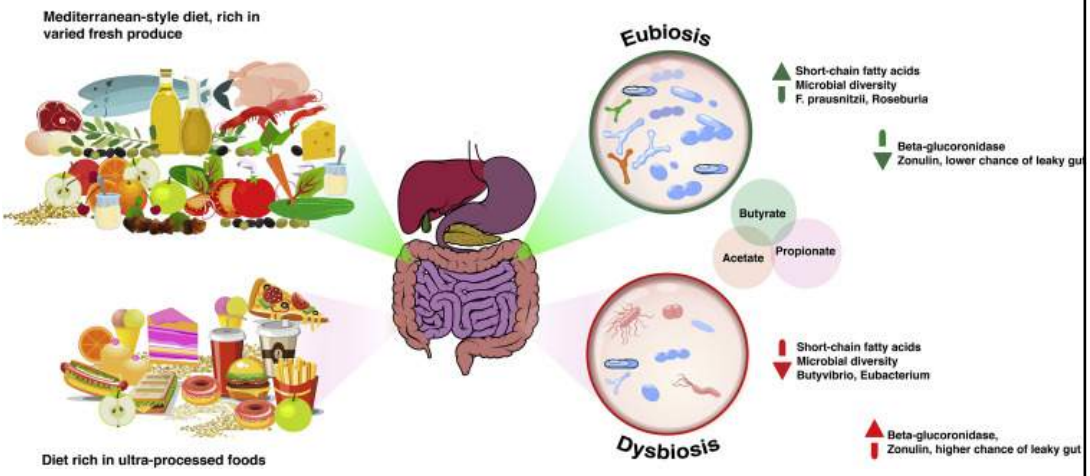
Mediterranean diet

A varied Mediterranean-style diet is associated with more ***diversity***

Miguel Toribio-Mateas, Adri Bester, Chapter 43 - Diet and the microbiome in precision medicine, Editor(s): Joel Faintuch, Salomão Faintuch, Precision Medicine for Investigators, Practitioners and Providers, Academic Press, 2020, Pages 445-452

10/10/2023

Figure adapted from Toribio–Mateas, M. Harnessing the power of microbiome assessment tools as part of neuroprotective nutrition and lifestyle medicine interventions. Microorganisms 2018;6:35.



41

Enhancers – Omega 3 & microbiota 2017 study

International Journal of *Molecular Sciences*

MDPI

Review

Impact of Omega-3 Fatty Acids on the Gut Microbiota

Lara Costantini † , Romina Molinari † , Barbara Farinon and Nicolò Merendino *

Department of Ecological and Biological Sciences (DEB), Tuscia University, Largo dell'Università snc, 01100 Viterbo, Italy; lara.cost@libero.it (L.C.); rominamolinari@libero.it (R.M.); barbara.farinon@gmail.com (B.F.)
* Correspondence: merendin@unitus.it; Tel.: +39-0761-357-133
† These authors contributed equally to this work.

Received: 31 October 2017; Accepted: 1 December 2017; Published: 7 December 2017

Abstract: Long-term dietary habits play a crucial role in creating a host-specific gut microbiota community in humans. Despite the many publications about the effects of carbohydrates (prebiotic fibers), the impact of dietary fats, such as omega-3 polyunsaturated fatty acids (PUFAs), on the gut microbiota is less well defined. The few studies completed in adults showed some common changes in the gut microbiota after omega-3 PUFA supplementation. In particular, a decrease in *Faecalibacterium*, often associated with an increase in the *Bacteroidetes* and butyrate-producing bacteria belonging to the *Lachnospiraceae* family, has been observed. Coincidentally, a dysbiosis of these taxa is found in patients with inflammatory bowel disease. Omega-3 PUFAs can exert a positive action by reverting the microbiota composition in these diseases, and increase the production of anti-inflammatory compounds, like short-chain fatty acids. In addition, accumulating evidence in animal model studies indicates that the interplay between gut microbiota, omega-3 fatty acids, and immunity helps to maintain the intestinal wall integrity and interacts with host immune cells. Finally, human and animal studies have highlighted the ability of omega-3 PUFAs to influence the gut–brain axis, acting through gut microbiota composition. From these findings, the importance of the omega-3 connection to the microbiota emerges, encouraging further studies.

- Increase in Bacteroidetes and butyrate producing bacteria with omega 3 supplementation.
- Decrease in faecalibacterium.
- In combination with gut microbiota, omega 3 help maintain intestinal wall integrity and interact with immune cells and influences the gut brain axis.

42

Prebiotic effect of omega 3 2020 paper

GUT MICROBES
2020, VOL. 13, NO. 1, e1863133 (11 pages)
https://doi.org/10.1080/19490976.2020.1863133

Taylor & Francis
Taylor & Francis Group

RESEARCH PAPER

OPEN ACCESS Check for updates

The prebiotic effects of omega-3 fatty acid supplementation: A six-week randomised intervention trial

Amrita Vijay[a,b], Stuart Astbury[c,d], Caroline Le Roy[a], Tim D Spector[a], and Ana M Valdes[a,b,d]

[a]Department of Twin Research and Genetic Epidemiology, King's College London, London, UK; [b]Division of Rheumatology, Orthopaedics and Dermatology, School of Medicine, University of Nottingham, Nottingham, UK; [c]Nottingham Digestive Diseases Centre, School of Medicine, University of Nottingham, Nottingham, UK; [d]NIHR Nottingham Biomedical Research Centre, Nottingham University Hospitals NHS Trust and the University of Nottingham, Nottingham, UK

ABSTRACT

Prebiotics are compounds in food that benefit health *via* affecting the gut microbiome. Omega-3 fatty acids have been associated with differences in gut microbiome composition and are widely accepted to have health benefits, although recent large trials have been inconclusive. We carried out a 6-week dietary intervention comparing the effects of daily supplementation with 500 mg of omega-3 versus 20 g of a well-characterized prebiotic, inulin. Inulin supplementation resulted in large increases in *Bifidobacterium* and Lachnospiraceae. In contrast, omega-3 supplementation resulted in significant increases in *Coprococcus spp*. and *Bacteroides spp*, and significant decreases in the fatty-liver associated *Collinsella spp*. On the other hand, similar to the results with inulin supplementation which resulted in significant increases in butyrate, iso-valerate, and iso-butyrate ($p < .004$), omega-3 supplementation resulted in significant increases in iso-butyrate and isovalerate ($p < .002$) and nearly significant increases in butyrate ($p < .053$). *Coprococcus*, which was significantly increased post-supplementation with omega-3, was found to be positively associated with iso-butyric acid (Beta (SE) = 0.69 (0.02), $P = 1.4 \times 10^{-3}$) and negatively associated with triglyceride-rich lipoproteins such as VLDL (Beta (SE) = −0.381 (0.01), $P = .001$) and VLDL-TG (Beta (SE) = −0.372 (0.04), $P = .001$) after adjusting for confounders. Dietary omega-3 alters gut microbiome composition and some of its cardiovascular effects appear to be potentially mediated by its effect on gut microbial fermentation products indicating that it may be a prebiotic nutrient.

ARTICLE HISTORY
Received 2 July 2020
Revised 24 November 2020
Accepted 4 December 2020

KEYWORDS
Dietary intervention; Omega 3; Fibre; Prebiotic; Gut microbiota

"Dietary omega 3 alters gut microbiome composition and some of its cardiovascular effects appear to be potentially mediated by its effect on gut microbial fermentation indicating it maybe a pre-biotic nutrient".

43

2021 review paper: high fat & sugar diets associated with microbial changes leading to inflammation or barrier dysfunction

It has been established that long-term dietary habits influence gut bacterial diversity and are capable of inducing changes in hypothalamic energy homeostasis.

The biological effects are mediated by microglial activation, systemic inflammation, and vagal afferent nerve signalling, culminating in neuroinflammation.

10/10/2023

It has been emphasized the need for a further approach regarding the influence of the dietary factors as well as their direct impacts or outcomes on the gut dysbiosis. **This review aimed to understand the role of some dietary triggers of neuroinflammation on changes in the gut microbiota**.

Jamar G, Ribeiro DA, Pisani LP. High-fat or high-sugar diets as trigger inflammation in the microbiota-gut-brain axis. *Crit Rev Food Sci Nutr*. 2021;61(5):836-854.

44

2021 review aper: high fat & sugar diets associated with microbial changes leading to inflammation or barrier dysfunction

Each of the diets significantly altered the microbial composition in distinct ways, leading to neuroadaptations.

Hyperlipidic diets (SFA and MUFA) can stimulate TLR4 inflammatory pathway by increased LPS translocation and LBP activation and modulate brain functions, mainly in the centre of feeding.

10/10/2023

Overconsumption of sucrose seems to be more detrimental for metabolic alterations, whereas fructose has a more pronounced effect on gut barrier dysfunction and subclinical inflammation; nevertheless, sucrose absorption favours fructose bioavailability, contributing to adiposity and sugar addiction.

Jamar G, Ribeiro DA, Pisani LP. High-fat or high-sugar diets as trigger inflammation in the microbiota-gut-brain axis. *Crit Rev Food Sci Nutr*. 2021;61(5):836-854.

45

High fat diets and microbial changes

- Believed high fat diets induce obesity and change gut microbiota.
- BUT inconsistent results on effects of high fat diet on microbiota.

10/10/2023

- This animal study reports high fat diet increased diversity and Short chain fatty acids (SCFAs).

Wang B, Kong Q, Li X, et al. A High-Fat Diet Increases Gut Microbiota Biodiversity and Energy Expenditure Due to Nutrient Difference. *Nutrients*. 2020;12(10):3197.

nutrients MDPI

Article

A High-Fat Diet Increases Gut Microbiota Biodiversity and Energy Expenditure Due to Nutrient Difference

Botao Wang [1,2], Qingmin Kong [1,2], Xiu Li [1,2], Jianxin Zhao [1,2,3,4], Hao Zhang [1,2,4,5,6], Wei Chen [1,2,5,7] and Gang Wang [1,2,3,4,*]

1 State Key Laboratory of Food Science and Technology, Jiangnan University, Wuxi 214122, China; jnwangbotao@foxmail.com (B.W.); konggjingmin2@163.com (Q.K.); liciu@jiangnan.edu.cn (X.L.); zhaojianxin@jiangnan.edu.cn (J.Z.); zhanghao@jiangnan.edu.cn (H.Z.); chenwei668@jiangnan.edu.cn (W.C.)
2 School of Food Science and Technology, Jiangnan University, Wuxi 214122, China
3 International Joint Research Center for Probiotics & Gut Health, Jiangnan University, Wuxi 214122, China
4 (Yangzhou) Institute of Food Biotechnology, Jiangnan University, Yangzhou 225004, China
5 National Engineering Center of Functional Food, Jiangnan University, Wuxi 214122, China
6 Wuxi Translational Medicine Research Center and Jiangsu Translational Medicine Research Institute Wuxi Branch, Wuxi 214122, China
7 Beijing Innovation Centre of Food Nutrition and Human Health, Beijing Technology and Business University (BTBU), Beijing 102488, China
* Correspondence: wanggang@jiangnan.edu.cn; Tel.: +86-510-85912155

Received: 23 September 2020; Accepted: 16 October 2020; Published: 20 October 2020

Abstract: A high-fat diet (HFD) can easily induce obesity and change the gut microbiota and its metabolites. However, studies on the effects of high-fat diets on the host have drawn inconsistent results. In this study, the unexpected results showed that the refined HFD increased gut microbiota diversity and short-chain fatty acids (SCFAs), causing an increase in energy metabolism. Further analysis revealed these changes were caused by the different fiber content in these two diets. Male C57BL/6J mice (4–5 weeks old) were fed either HFD or refined low-fat diet (LFD) for 14 weeks. The metabolic rates, thermogenesis, gut microbiome, and intestinal SCFAs were tested. The HFD triggered obesity and disturbed glucose homeostasis. Mice fed HFD ingested more fiber than mice fed LFD ($p < 0.0001$), causing higher intestinal SCFA concentrations related to the increased abundances of specific bacteria in the HFD group. Also, the HFD increased metabolic heat and up-regulated thermogenesis genes uncoupling protein 1(*Ucp-1*), peroxisome proliferator-activated receptor-γ coactivator-1α (*Pgc-1α*) expression in the brown adipose tissue (BAT). It was revealed by 16S rRNA gene sequencing that the HFD increased gut microbial diversity, which enriched Desulfovibrionaceae, *Rikenellaceae RC9 gut group*, and *Mucispirillum*, meanwhile, reduced the abundance of *Lactobacillus*, *Bifidobacterium*, *Akkermansia*, *Faecalibaculum*, and *Blautia*. The predicted metabolic pathways indicated HFD increased the gene expression of non-absorbed carbohydrate metabolism pathways, as well as the risks of colonization of intestinal pathogens and inflammation. In conclusion, the HFD was obesogenic in male C57BL/6J mice, and increased fiber intake from the HFD drove an increase in gut microbiota diversity, SCFAs, and energy expenditure. Meanwhile, the differences in specific nutrient intake can dissociate broad changes in energy expenditure, gut microbiota, and its metabolites from obesity, raising doubts in the previous studies. Therefore, it is necessary to consider whether differences in specific nutrient intake will interfere with the results of the experiments.

46

High fat diets

- 2015 review paper.
- Outlines complex role of HFD in obesity, but important to note that it is more complex than just energy balance – as HFDs seem to alter microbiota, increasing intestinal permeability and inflammation.

10/10/2023

- HFDs decrease bacteriodetes increase Firmicutes.

Murphy EA, Velazquez KT, Herbert KM. Influence of high-fat diet on gut microbiota: a driving force for chronic disease risk. *Curr Opin Clin Nutr Metab Care*. 2015;18(5):515-520.

Published in final edited form as:
Curr Opin Clin Nutr Metab Care. 2015 September ; 18(5): 515–520. doi:10.1097/MCO.0000000000000209.

Influence of High-Fat-Diet on Gut Microbiota: A Driving Force for Chronic Disease Risk

E. Angela Murphy[1], **Kandy T. Velazquez**[1], and **Kyle M. Herbert**[1]
[1]Department of Pathology, Microbiology, and Immunology, School of Medicine, University of South Carolina, Columbia, SC 29209, USA

X

Abstract

Purpose of review—This review will examine the recent scientific literature surrounding high-fat-diet (HFD)-induced alterations in gut microbiota and subsequent development of obesity and chronic disease risk.

Recent findings—Excessive consumption of HFDs has undoubtedly contributed to the obesity epidemic. However, the mechanisms responsible for this relationship are likely to be more complex than the simple concept of energy balance. In fact, emerging literature has implicated HFD-induced alterations in gut microbiota in the obesity epidemic. HFD consumption generally leads to a decrease in *Bacteroidetes* and an increase in *Firmicutes*, alterations that have been associated with obesity and subsequent development of chronic diseases. Potential mechanisms for this effect include 1) an improved capacity for energy harvest and storage and, 2) enhanced gut permeability and inflammation. We highlight the most important recent advances linking HFD-induced dysbiosis to obesity, explore the possible mechanisms for this effect, examine the implications for disease development, and evaluate the possibility of therapeutic targeting of the gut microbiome to reduce obesity.

Summary—A better understanding of the mechanisms linking HFD to alterations in gut microbiota is necessary to allow for the regulation of dysbiosis and ensuing promotion of anti-obesity effects.

47

Butter and dairy products

- Butter and dairy can still be useful additions to a balanced diet.
- Think about moderation and overall balance with omega 3.
- As SCFA's including butyric acid are important for gut health.
  - Butyrate is a functionally versatile molecule that is produced in the mammalian gut by fermentation of dietary fibre and is enriched in butter and other dairy products.

10/10/2023

Stilling RM, van de Wouw M, Clarke G, Stanton C, Dinan TG, Cryan JF. The neuropharmacology of butyrate: The bread and butter of the microbiota-gut-brain axis?. *Neurochem Int*. 2016;99:110-132

48

Alcohol and microbial changes

- Limited human data, but studies highlight changes in the intestinal microbiota in alcohol-related disorders.
- Alcohol-induced changes in the microbiota composition and metabolic function may contribute to the link between alcohol-induced oxidative stress, intestinal hyperpermeability to luminal bacterial products, and the development of alcoholic liver disease (ALD).

10/10/2023

Engen PA, Green SJ, Voigt RM, Forsyth CB, Keshavarzian A. The Gastrointestinal Microbiome: Alcohol Effects on the Composition of Intestinal Microbiota. *Alcohol Res*. 2015;37(2):223-236.

49

Alcohol and microbial changes

Chronic alcohol intake leads to intestinal inflammation, increased intestinal permeability and alters gut microflora.

Alcohol promotes both dysbiosis and bacterial overgrowth which in turn leads to an increase in the release of endotoxins, produced by gram-negative bacteria - endotoxins activate proteins and immune cells that promote inflammation.

10/10/2023

Bishehsari F, Magno E, Swanson G, et al. Alcohol and Gut-Derived Inflammation. *Alcohol Res*. 2017;38(2):163-171.

50

Are pesticides a factor in Gut brain axis dysfunction?

- 2023 paper.
- Further research is needed to establish causation.
- Proposed mechanisms of pesticides potential effects on human behaviour via gut brain axis.

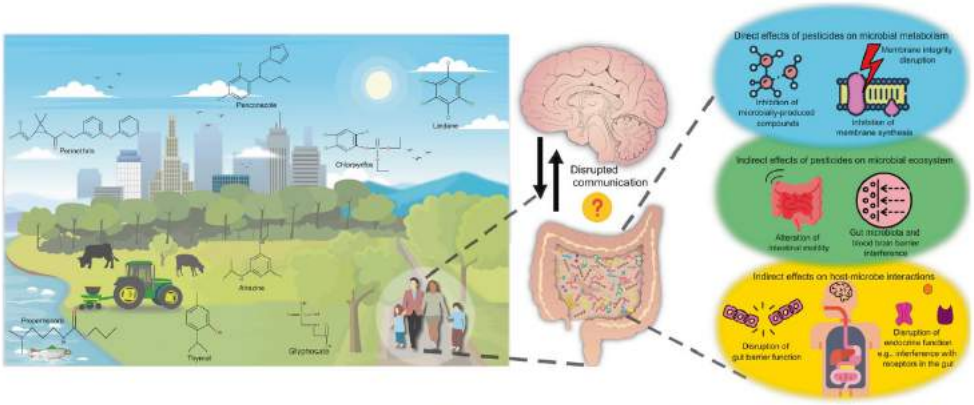


Fig. 1 Proposed mechanisms of environmental pesticides impacting the microbiota-gut-brain axis. The residue of pesticides in the environment (air, soil, water and food) can enter the body of the host leading to disrupted communication between the gut and the brain. While the mechanisms are still being uncovered, some of the potential routes include, solely or in combination of, direct effects on microbial metabolism, indirect effects of pesticides on microbial communities in the gut and indirect effects on host-microbe interactions, which may explain the disrupted communication as seen from behavioural impairments. The icons used in this figure were designed by adriansyah, Flat icons, Freepik and Kalashnyk on https://www.flaticon.com/ and the chemical structure was drawn using BIOVIA, Dassault Systèmes, BIOVIA Draw 2022, San Diego: Dassault Systèmes, 2023.

Matsuzaki, R., Gunnigle, E., Geissen, V. *et al.* Pesticide exposure and the microbiota-gut-brain axis. *ISME J* (2023).

51

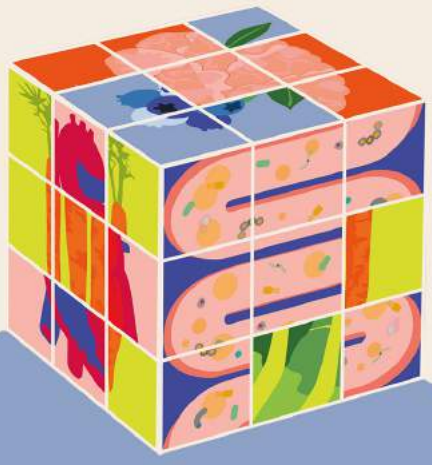
THANK YOU

10/10/2023

52

Microbiome, Gut & Systemic Health:

New Frontiers in Personalised Nutrition



NMI SUMMIT 2023

Saturday 14th October

10/10/2023

Featuring Dr. Gerard Mullin, Professor Glenn Gibson, Dr. Amrita Vijay, Justine Bold, Dr. Jonathan Sutton and Benjamin Brown

An event by: Nutritional Medicine Institute

Platinum sponsors: regenerus labs VIRIDIAN Effective | Ethical | Pure pure encapsulations

53