Insights into the reason of Human-Residential Bifidobacteria (HRB) being the natural inhabitants of the human gut and their potential health-promoting benefits

Jin-zhong Xiao, Ph.D

Next Generation Science Institute, Morinaga Milk Industry Co., Ltd.

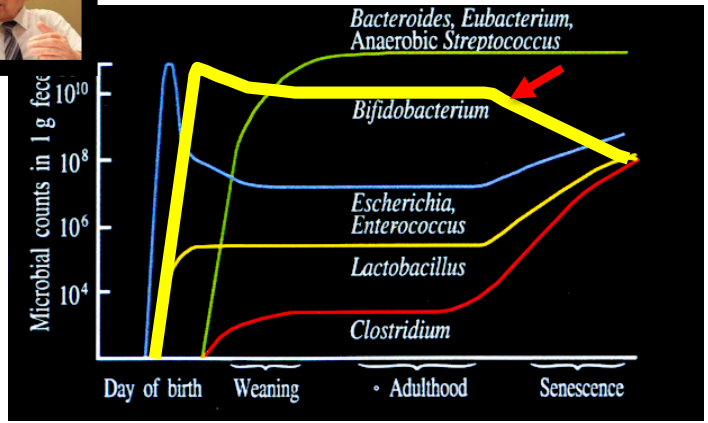
Contents

1. *Bifidobacterium* in human gut, the concept of Human-Residential Bifidobacteria (HRB)
   - Bifidobacterium species and compatibility with human milk
   - The distinguished metabolic profile of HRB

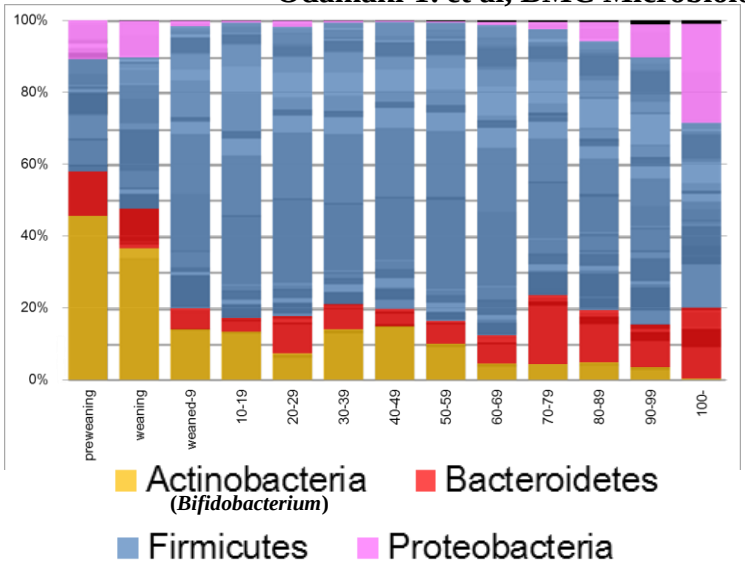
2. Topics of the potential health-promoting effects of HRB
   - Clinical effects to preterm infants
   - Clinical effects in preventing cognitive impairment in elderly.

Bifidobacterium and human health

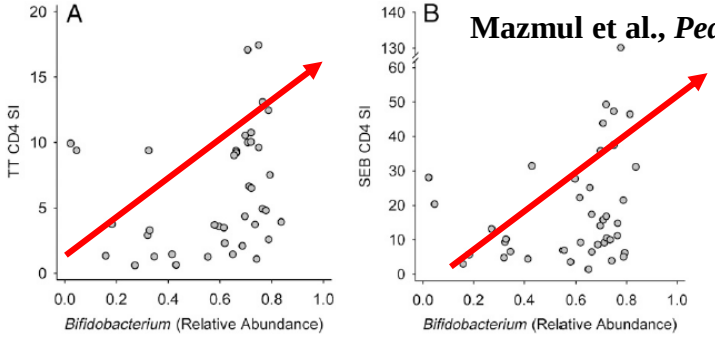
Mitsuoka T, 1978



Odamaki T. et al, BMC Microbiology, 2016



Response to vaccine



Mazmul et al., *Pediatrics* 2014;

Abundance of bifidobacteria

- **Bifidobacteria function in maintaining health and protecting from infection for the infants**

Bifidobacterial species and their residences

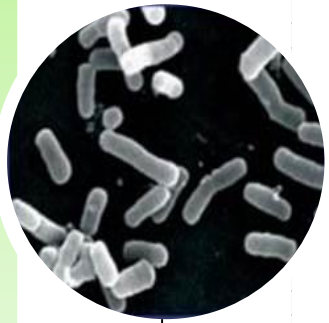
More than 90 species/subspecies of *Bifidobacterium* have been discovered
(http://www.bacterio.net/)

B. longum subsp. infantis
B. longum subsp. longum
B. breve
B. bifidum

B. longum subsp. longum
B. adolescentis
B. pseudocatenulatum
B. Catenulatum
...

HRB

B. animalis subsp. *animalis*
B. animalis subsp. *lactis*
B. thermophilum
B. pseudolongum
...



non−HRB

HRB: Human-residential bifidobacteria
Non-HRB: non-Human-residential bifidobacteria

To answer…

1. What is the mechanism for the characteristic residence of *Bifidobacterium* species?

2. Is there any basic difference between HRB and non-HRB in relation to health benefits?

To understand the difference of HRB and non-HRB

A total of 50 genome sequences from NCBI database or Sequenced by MiSeq

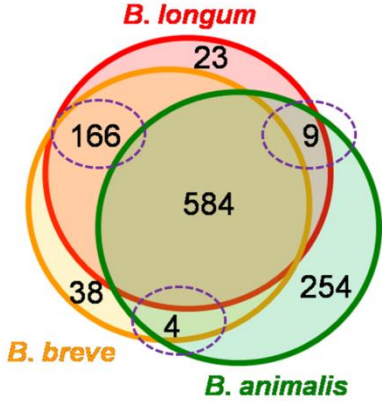
HRB

- **16 strains of *B. longum* (2 ssp. *infantis*, 14 ssp. *longum*)**
- **14 strains of *B. breve***

Non-HRB

- **20 strains of *B. animalis* (5 ssp. *animalis*, 14 ssp. *lactis*)**

Number of common clusters among 3 bifidobacterial species



Common clusters:

***B. longum/B. breve*: 166**

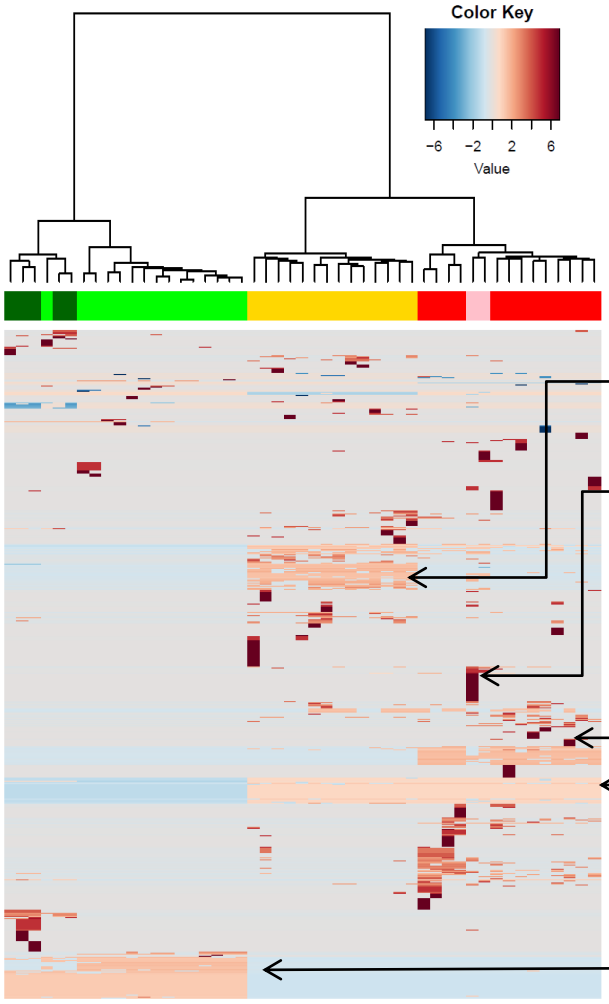
***B. longum/B. animalis*: 9**

***B. breve/B. animalis*: 4**

Odamaki et al., International Journal of Genomics (2015)

Hierarchical clustering based on the genomic information.

Odamaki et al., International Journal of Genomics (2015)



- ***B.longum* subsp. *longum***
- ***B. longum* subsp. *infantis***
- ***B. breve***
- ***B. animalis* subsp. *animalis***
- ***B. animalis* subsp. *lactis***

- ROK family transcriptional regulator
- N-acetylmannosamine-6-phosphate 2-epimerase
- N-acetylneuraminate lyase
- Sialidase A, etc.

Sialidase related cluster

- Exo-alpha-sialidase
- alpha-L-fucosidase
- family 1 extracellular solute-binding protein, etc.

HMO related cluster

- arabinosidase
- alpha-L-arabinofuranosidase A
- xylulose kinase
- extracellular exo-xylanase
- endo-1,4-beta-xylanase D, etc.

Enzymes for plant derived sugar

- GTP cyclohydrolase I
- Dihydroneopterin aldolase
- Dihydropteroate synthase

Folate synthesis

- L-aspartate oxidase
- Quinolinate synthetase A, etc.

Niacin synthesis

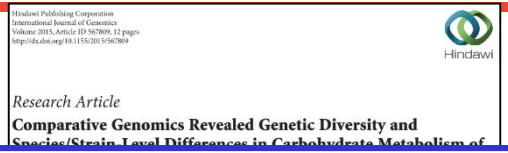
- lacto-N-biose phorylase
- UDP-glucose 4-epimerase

GNB/LNB cluster

- mannan endo-1,4-beta-mannosidase
- beta-mannosidase etc.

Enzymes for plant derived sugar

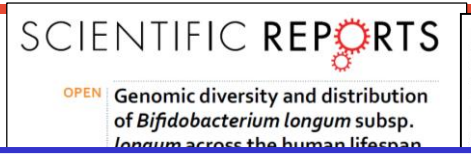
Studies on the difference of HRB and non-HRB


Hindawi Publishing Corporation
International Journal of Genomics
Volume 2015, Article ID 567809, 12 pages
http://dx.doi.org/10.1155/2015/567809

Hindawi

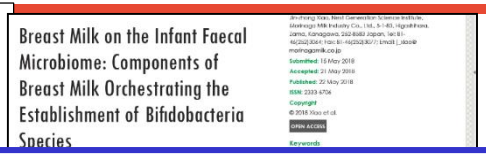
Research Article

Comparative Genomics Revealed Genetic Diversity and


SCIENTIFIC REPORTS

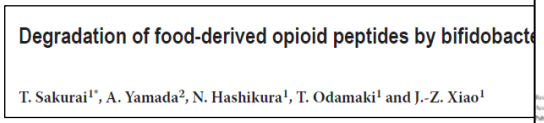
OPEN

Genomic diversity and distribution of *Bifidobacterium longum* subsp. *longum* across the human lifespan

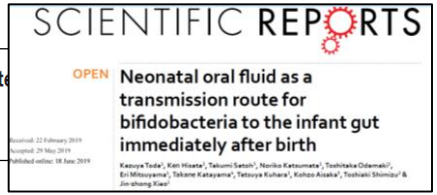

Breast Milk on the Infant Faecal Microbiome: Components of Breast Milk Orchestrating the Establishment of Bifidobacteria Species

Summary	HRB (infant-type)	Non-HRB	
Growth in breast milk	○	×	Minami 2015
Utilization of HMOs	△～○	×	Odamaki 2015, Minami 2015
Tolerance to lysozyme	○	×	Minami 2015, Sakurai 2017
Production of folate	○	×	Odamaki 2015 ,Sugaharai 2015
Degradation of opioid peptides	○	×	Sakurai 2018
Production of tryptophan metabolites（ILA）	○	×	Sakurai, 2019

堀米綾子，小田巻俊孝


Degradation of food-derived opioid peptides by bifidobacte

T. Sakurai[1*], A. Yamada[2], N. Hashikura[1], T. Odamaki[1] and J.-Z. Xiao[1]


SCIENTIFIC REPORTS

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Neonatal oral fluid as a transmission route for bifidobacteria to the infant gut immediately after birth


www.chinadairy.net
zgrpgyzz@163.com
中国乳品工业
dairy INDUSTRY

亲和人体与非亲和人体双歧杆菌之间的生理特征差异

黄勤雯[1]，陈铁涛[1]，肖金忠[1]

FEMS Microbiology Reviews, fuaa010, 1–17

doi: 10.1093/femsre/fuaa010
Advance Access Publication Date: 22 April 2020
Review article

REVIEW ARTICLE

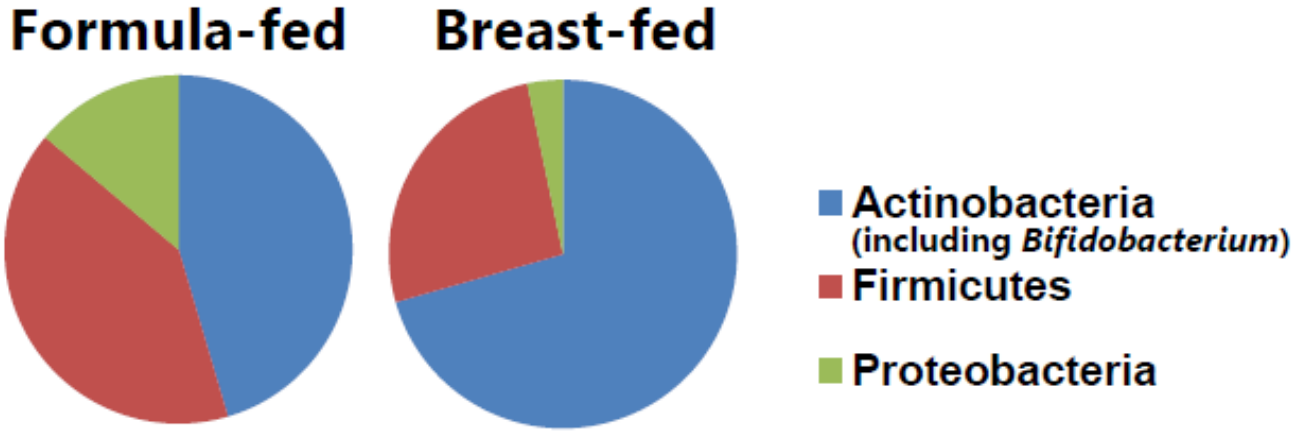
Insights into the reason of Human-Residential Bifidobacteria (HRB) being the natural inhabitants of the human gut and their potential health-promoting benefits

Chyn Boon Wong, Toshitaka Odamaki and Jin-zhong Xiao*

Next Generation Science Institute, Morinaga Milk Industry Co., Ltd., 5-1-83, Higashihara, Zama, Kanagawa,

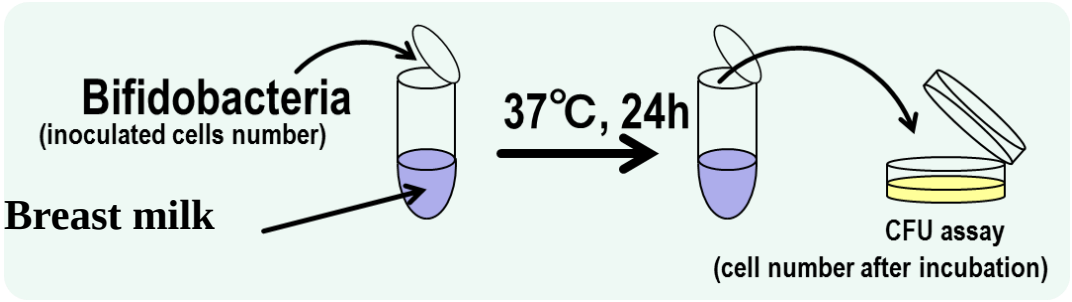
Bifidobacterium and breast milk

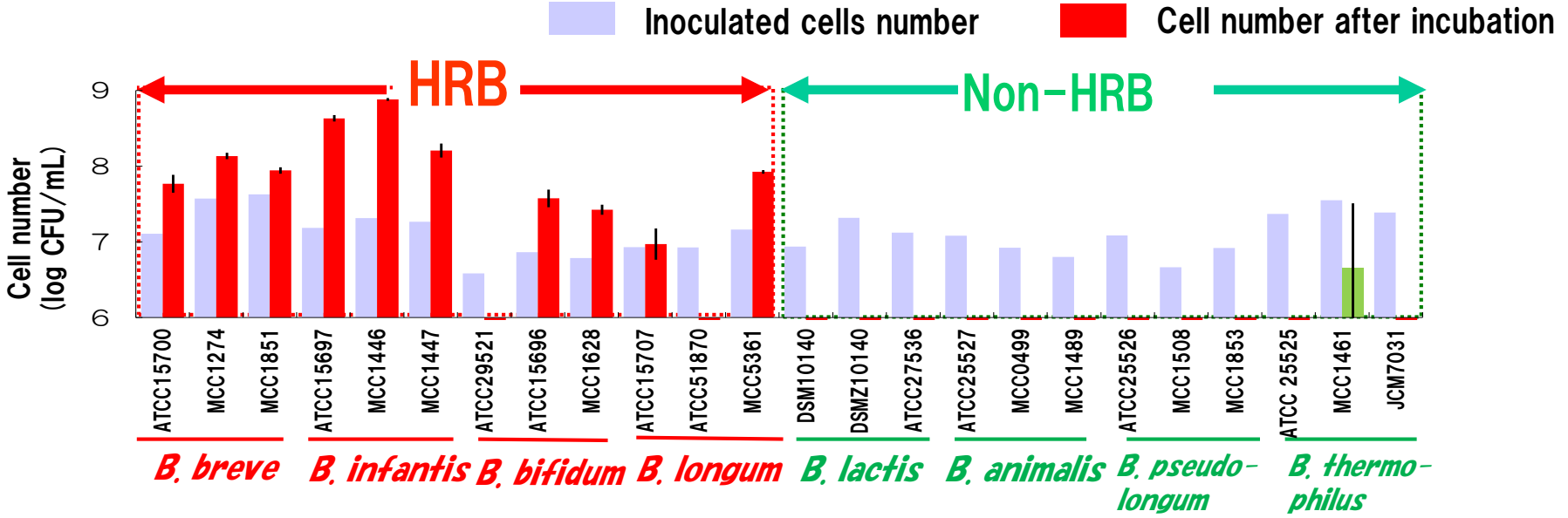
- **The abundance of *Bifidobacterium* is higher in breast-fed infants than formula-fed infants (Vandenplas, 2002)**
- **Milk oligosaccharides (HMOs) in breast milk are the contributing factors for the colonization of *Bifidobacterium*.**



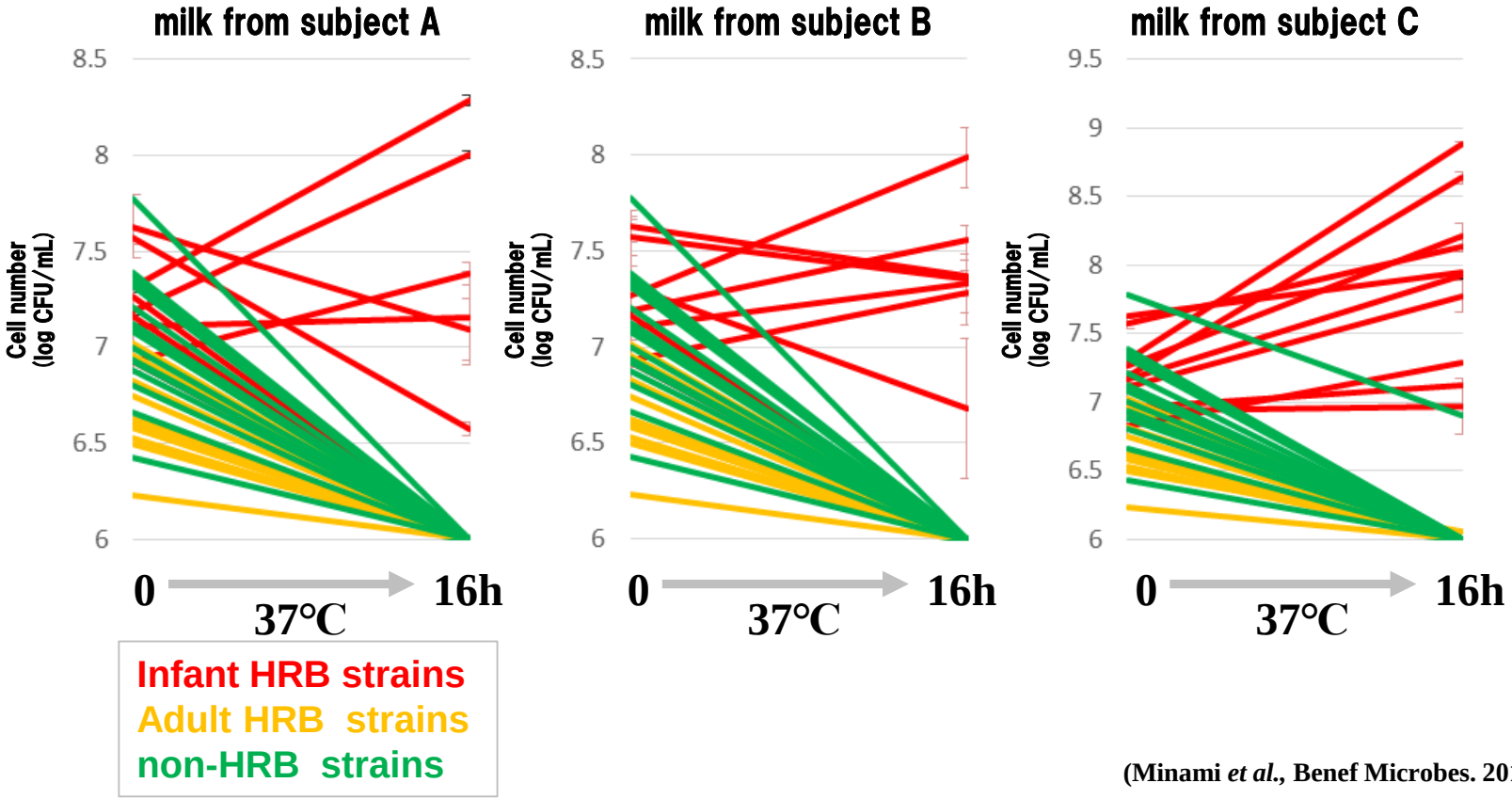
(Modified from Lee et al., Nutrition Research and Practice, 2015)

Growth of *Bifidobacteria* in human breast milk



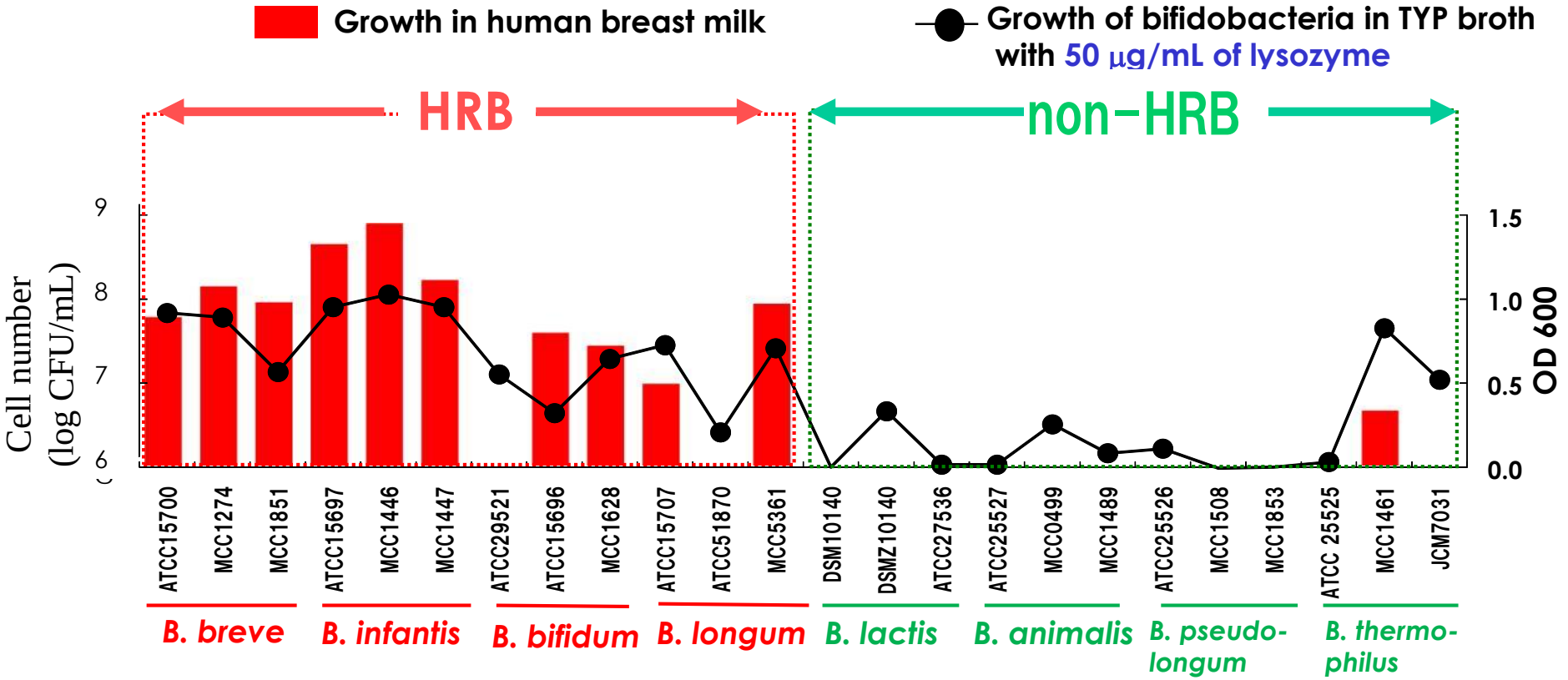


Growth of *Bifidobacterium* in human breast milk



(Minami *et al.*, Benef Microbes. 2015)

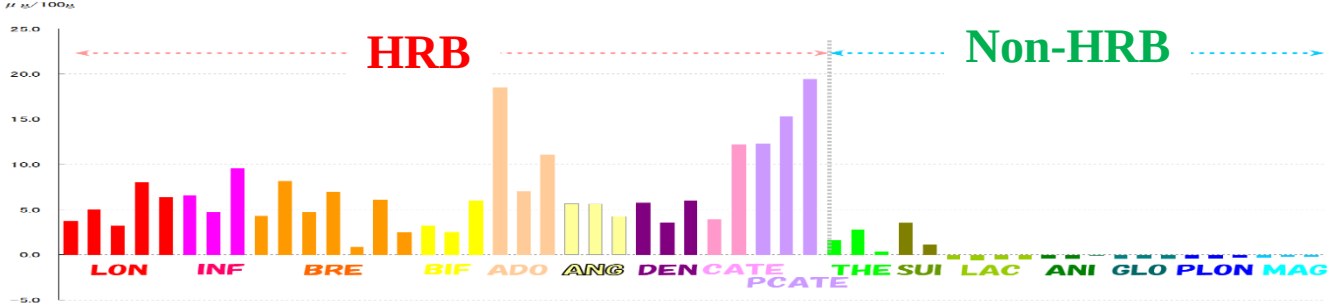
Growth of *Bifidobacterium* in medium containing lysozyme



Minami et al. Ben. Microbes, 2015

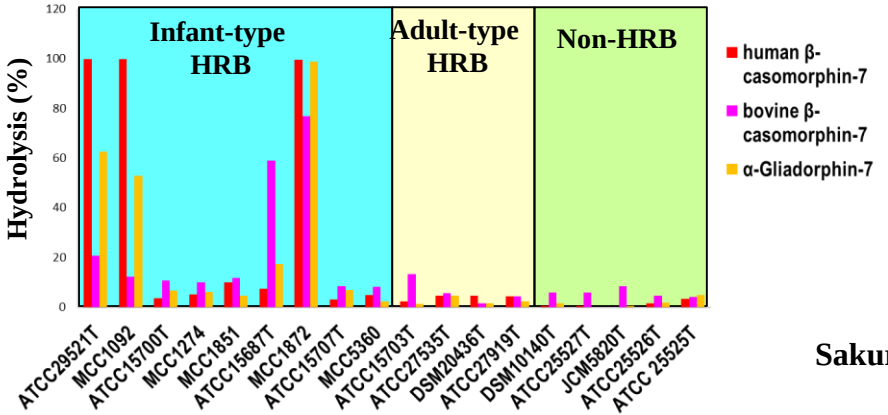
Different metabolisms of HRB and non-HRB

- **Production of folate**



Sugahara et al BMFH, 2015

- **Degradation of food-derived opioid peptides**



Sakurai T et al., Ben. Microbes, 2018

Microbial tryptophan catabolites

nature COMMUNICATIONS

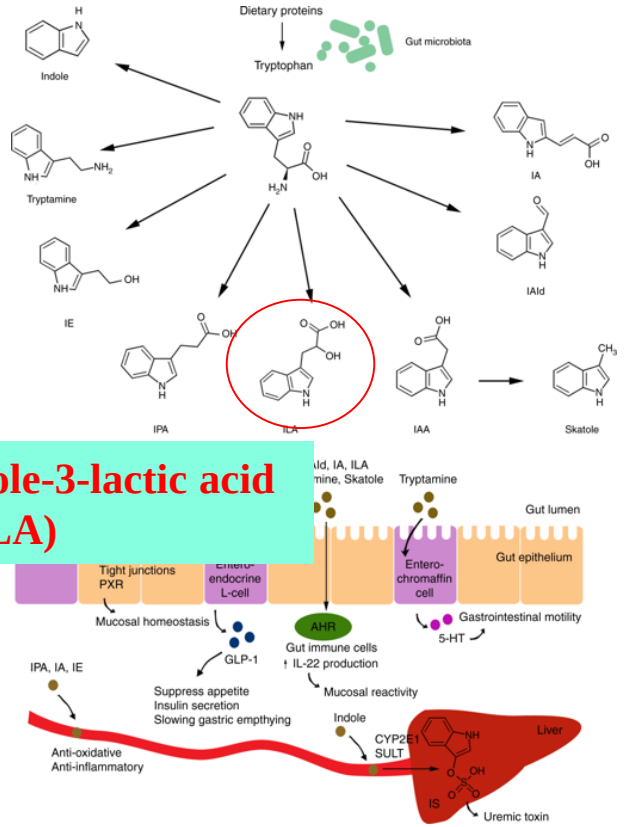
REVIEW ARTICLE

DOI: 10.1038/s41467-018-05470-4 OPEN

Roager et al., 2018

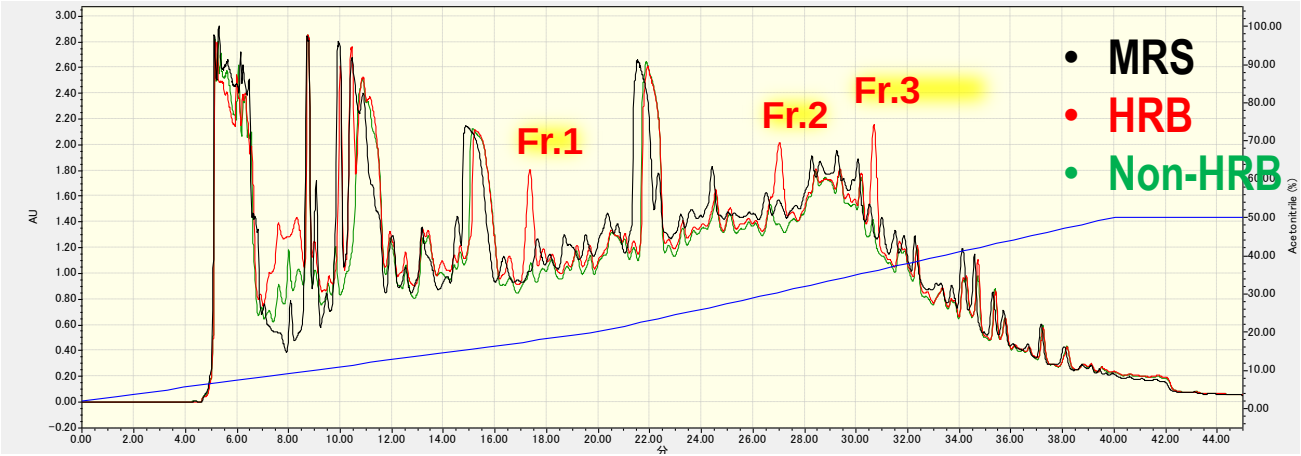
Microbial tryptophan catabolites in health and disease

Microbial tryptophan catabolites in the gut are suggested to activate the immune system through binding to the aryl hydrocarbon receptor (AHR), enhance the intestinal epithelial barrier, stimulate gastrointestinal motility, as well as secretion of gut hormones, exert anti-inflammatory, antioxidative or toxic effects in systemic circulation, and putatively modulate gut microbial composition. Tryptophan catabolites thus affect various physiological processes and may contribute to intestinal and systemic homeostasis in health and disease.



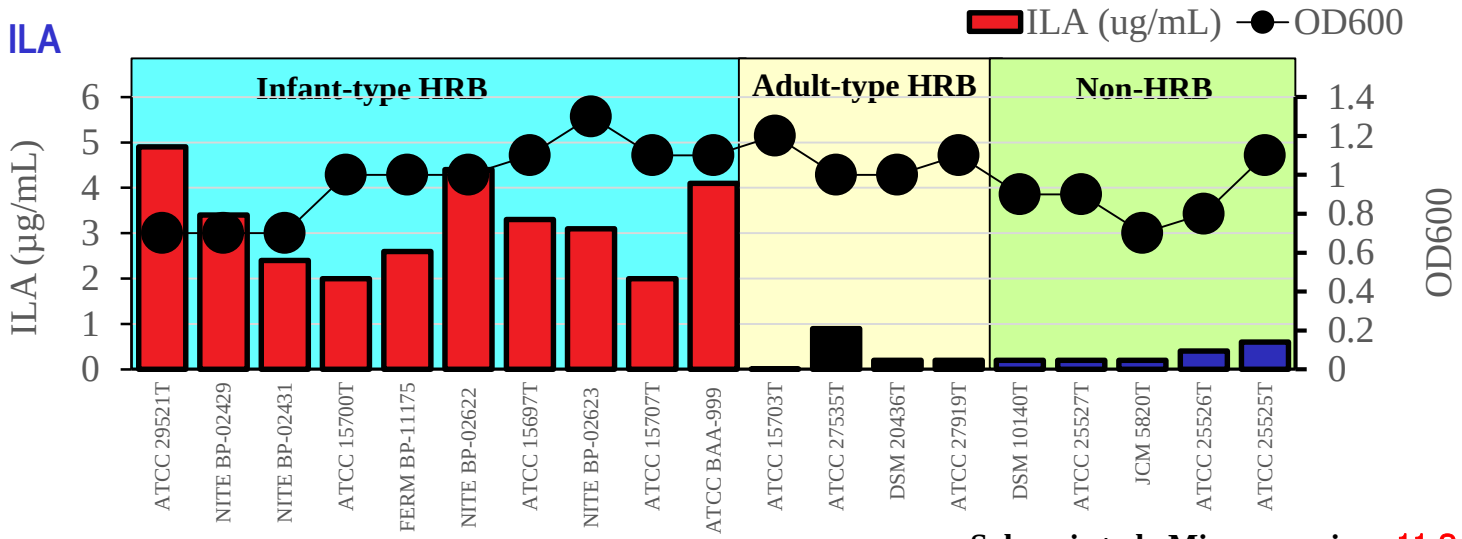
Difference in the metabolite profiles between HRB and non-HRB

HPLC profile of media before and after cultivation with HRB/non-HRB



Aryllactic acids (ALAs) derived from aromatic amino acids

Aryllactic acids (ALAs) by bifidobacterial strains



Sakurai et al., Microorganism, 11 September 2019

Production of ILA by Bifidobacteria

BASIC SCIENCE ARTICLE

Indole-3-lactic acid, a metabolite of tryptophan, secreted by *Bifidobacterium longum* subspecies *infantis* is anti-inflammatory in the immature intestine

Di Meng[1], Eduardo Sommella[2], Emanuela Salviati[2,3], Pietro Campiglia[2,4], Kriston Ganguli[1], Karim Djebali[1], Weishu Zhu[1] and W. Allan Walker[1,4]

Pediatric Res, published online 16 Jan, 2020

Breastmilk-promoted bifidobacteria produce aromatic lactic acids in the infant gut

Martin F. Laursen[1#], Mikiyasu Sakanaka[1,2], Nicole von Burg[3], Daniel Andersen[4], Urs Mörbe[3], Aymeric Rivollier[3], Ceyda T. Pekmez[5], Janne Marie Moll[4], Kim F. Michaelsen[5], Christian Mølgaard[5], Mads Vendelbo Lind[5], Lars O. Dragsted[5], Takane Katayama[2,6], Henrik L. Frandsen[1], Anne Marie Vinggaard[1], Martin I. Bahl[1], Susanne Brix[4], William Agace[3], Tine R. Licht[1*] and Henrik M. Roager[1,5#*]

bioRxiv preprint first posted online Jan. 23, 2020;

Article

Potential Effects of Indole-3-Lactic Acid, a Metabolite of Human Bifidobacteria, on NGF-induced Neurite Outgrowth in PC12 Cells

Chyn Boon Wong, Azusa Tanaka, Tetsuya Kuhara and Jin-zhong Xiao *

Next Generation Science Institute, Morinaga Milk Industry Co., Ltd., Zama 252-8583, Japan;

Microorganisms, published: 12 March 2020

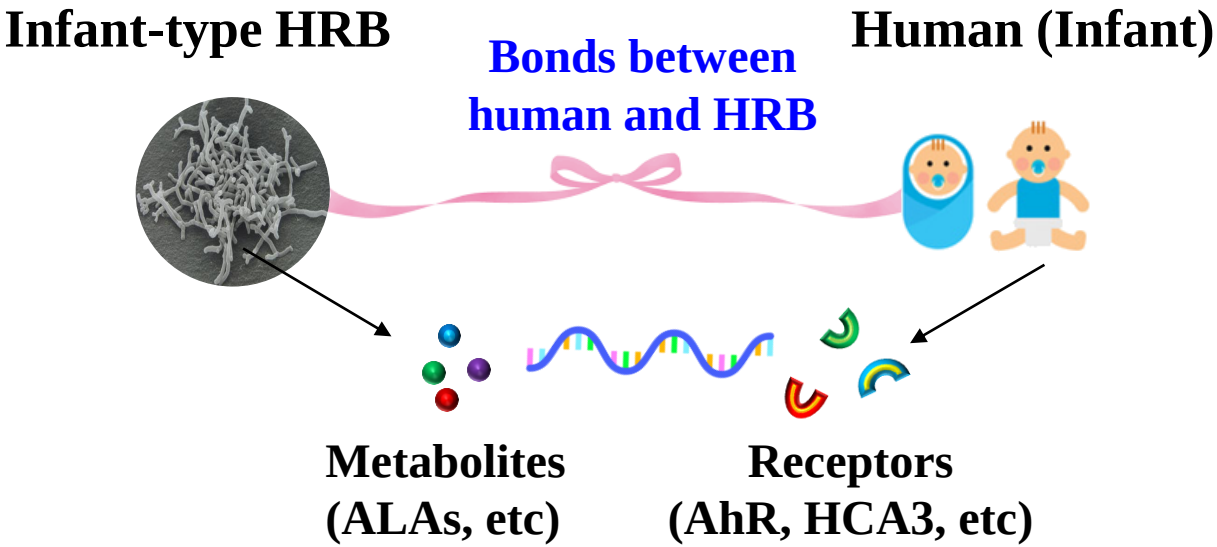
Cell

Article

Bifidobacteria-mediated immune system imprinting early in life

Henrick et al., 2021, Cell 184, 1–15. July 22, 2021

Role of ALAs in infant health



Summary (1) The difference of HRB and non-HRB

Human and animals have different *Bifidobacterium* species (HRB vs non-HRB).

Traits	HRB(infant-type)	Non-HRB
Growth in breast milk	○	×
Utilization of HMOs	△～○	×
Tolerance to lysozyme	○	×
Production of folate	○	×
Degradation of opioid peptides	○	×
Production of tryptophan metabolites (ILA)	○	×

Considering the high prevalence and abundance of *Bifidobacterium* in infant gut, the health benefits of HRB to host may be beyond our expectation!

Which probiotics should we choose for human, in particular for infant ? ? ?

HRB
?

Non-HRB
??

Lactobacilli
Others ??

Whisper of breast milk ……

HRB for infant, the law of nature ！

Contents

1. *Bifidobacterium* in human gut, the concept of Human-Residential Bifidobacteria (HRB)
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   - The distinguished metabolic profile of HRB

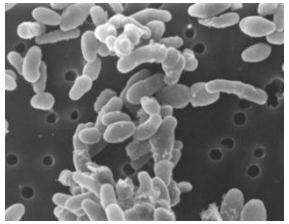
2. **Topics of the potential health-promoting effects of HRB**
   - **Clinical effects on infants**
   - **Clinical effects in the preventing cognitive impairment in elderly.**

Probiotic *Bifidobacterium* strains in Morinaga

B. longum BB536

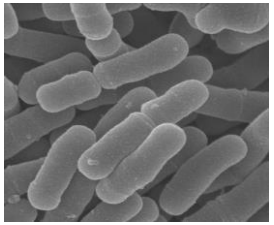
A strain for promoting GIT health, immunity

B. breve M-16V



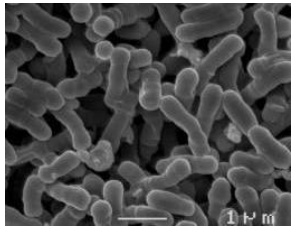
A strain for infant health

B. infantis M-63



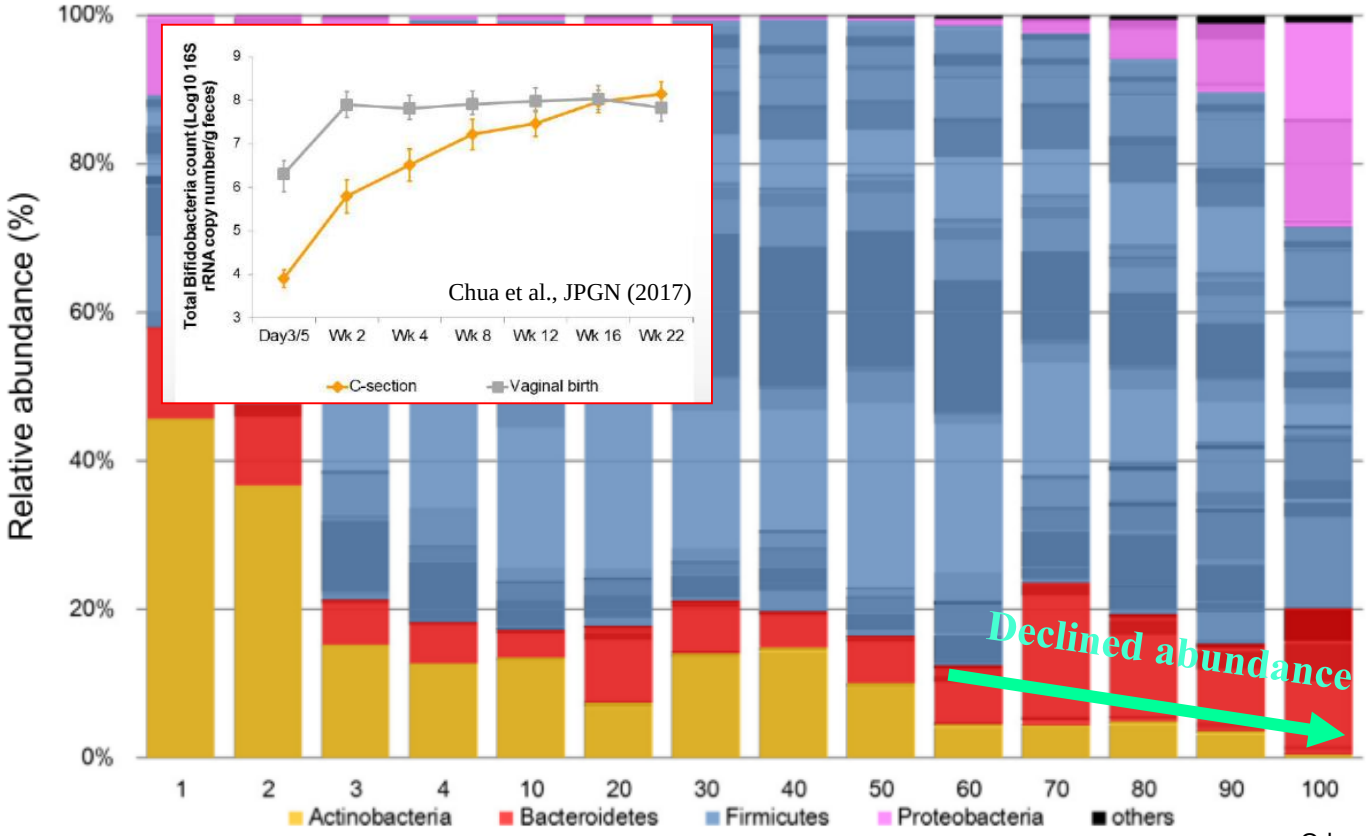
A strain with high ability for utilizing HMOs

B. breve MCC1274



A strain with anti-inflammatory effects (**preventing dementia**)

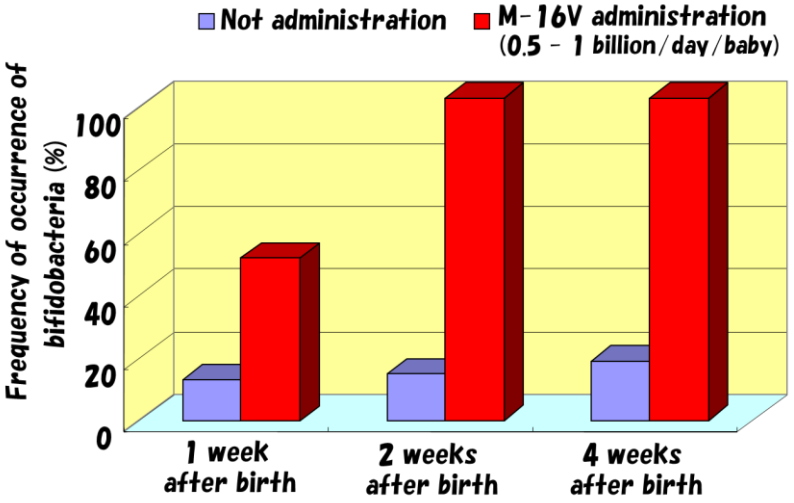
Age-related change of gut microbiota composition



Odamaki *et al.*, *BMC Microbiol.* 2016

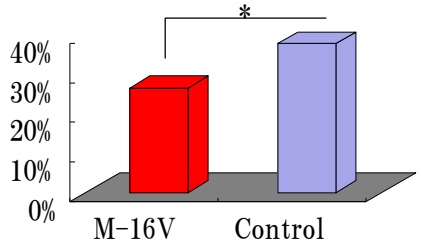
Potentials of improving gut microbiota and health growth

Administration of M-16V to preterm infants

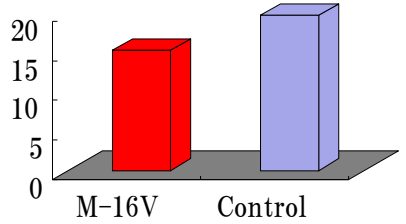


Ishizeki et al., Anaerobe (2013)

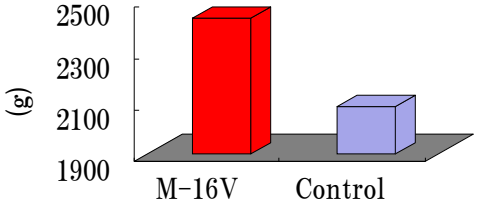
Infection rate



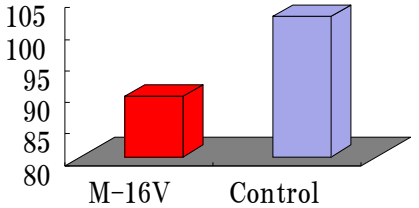
Days to reach 100 ml/d of enteral feedings



Weight at the time of expected delivery



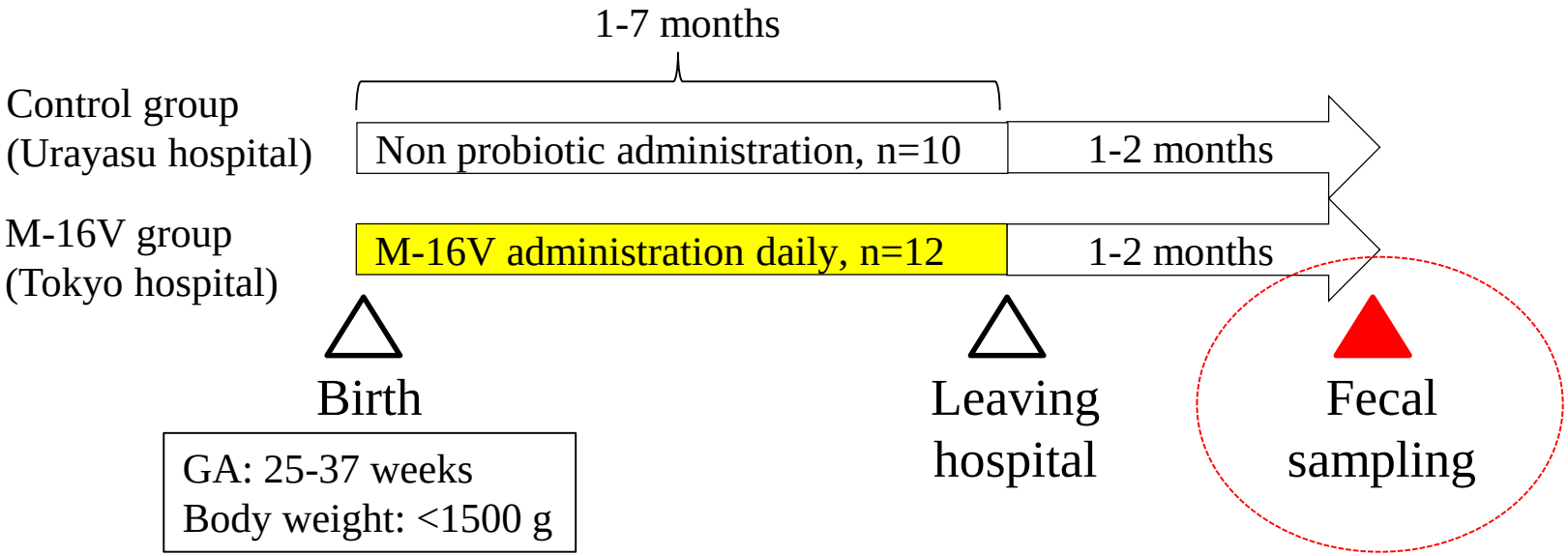
Days of hospital stay



(n = 190)

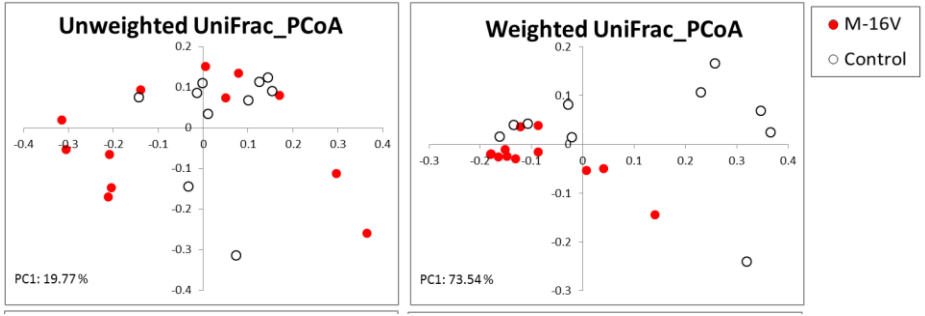
(Satoh, Acta Neotalogica Japonica 2003)

Effects of supplemented *Bifidobacterium breve* M-16V on the gut microbiota and their persistence post administration in preterm infants

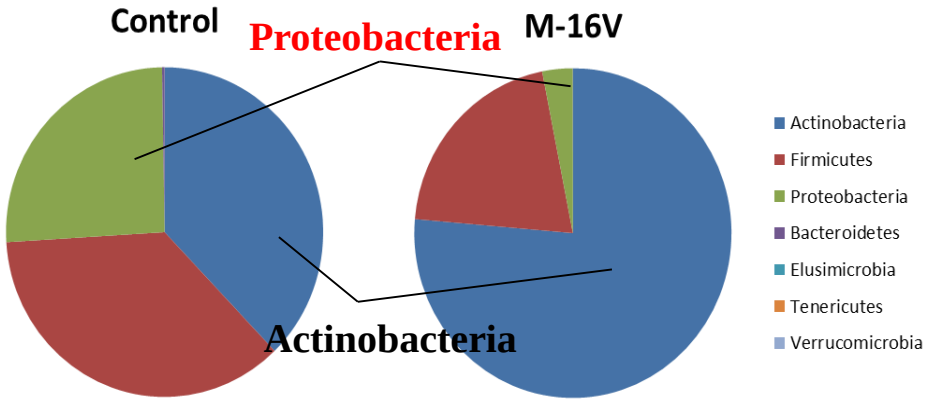


Gut microbiota profile

PCoA



Proportion of bacteria at phylum level



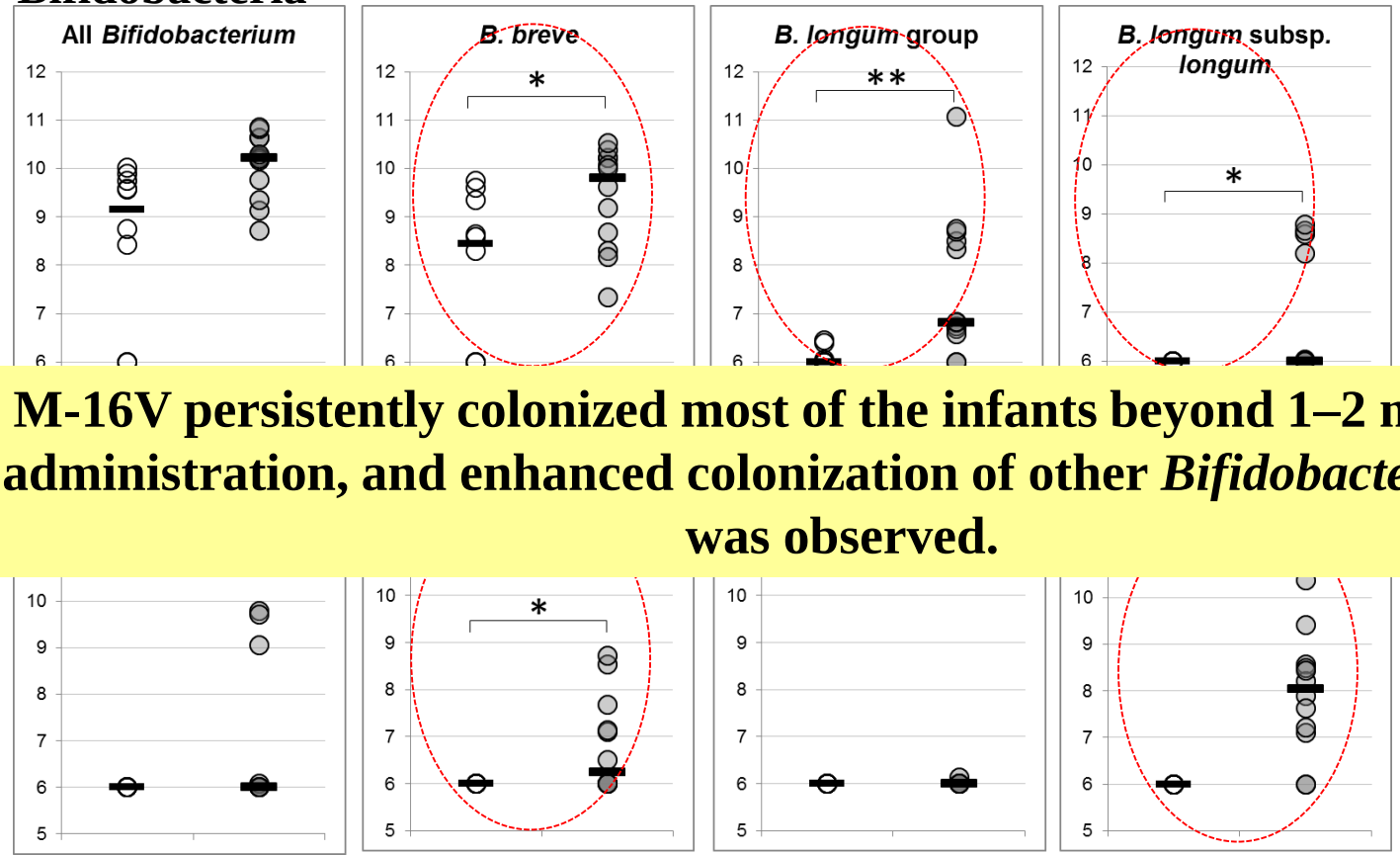
M-16V administration to preterm infants led to the formation of a *Bifidobacterium*-predominant microbiota, beyond 1–2 months post-administration.

Horigome et al. Front. Microbiol., 2021

Group	Median		P value (Mann–Whitney–U)
	M–16V	Control	
k__Bacteria__	0.0%	0.0%	1.000
Actinobacteria	74.0%	33.3%	0.009
Bacteroidetes	0.0%	0.2%	0.134
Elusimicrobia	0.0%	0.0%	1.000
Firmicutes	19.9%	31.9%	0.628
Proteobacteria	3.0%	22.7%	0.000
Tenericutes	0.0%	0.0%	0.481
Verrucomicrobia	0.0%	0.0%	0.455

Cell numbers of *Bifidobacterium* at species level by qPCR

Bifidobacteria

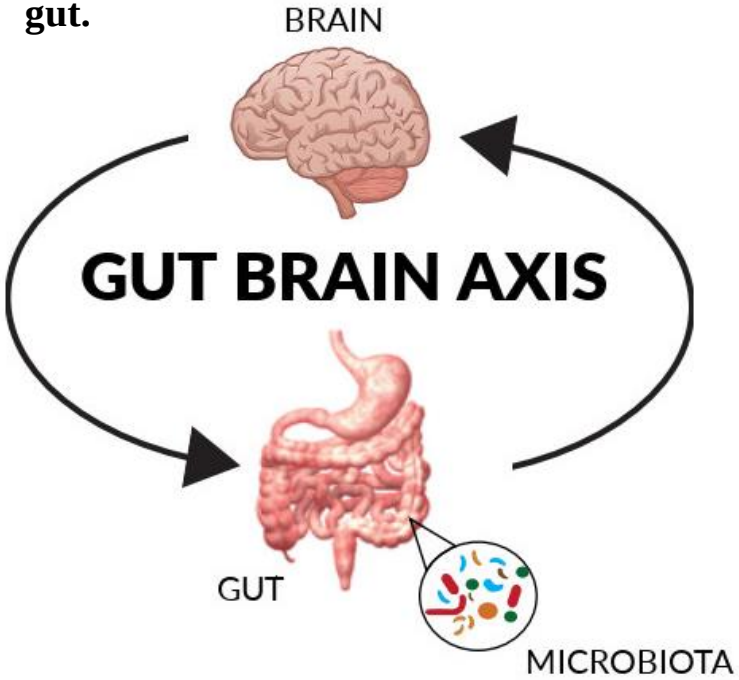


M-16V persistently colonized most of the infants beyond 1–2 months post-administration, and enhanced colonization of other *Bifidobacterium* species was observed.

Horigome et al.
Front. Microbiol., 2021

Brain-Gut Interaction (microbiota-Gut-Brain Axis)

The brain and gut axis is a bilateral interaction between the brain and the gut.



Adapted from Illawarra Athletic Health Hack

Exploring probiotics for preventing cognitive impairment in Alzheimer's disease

↓

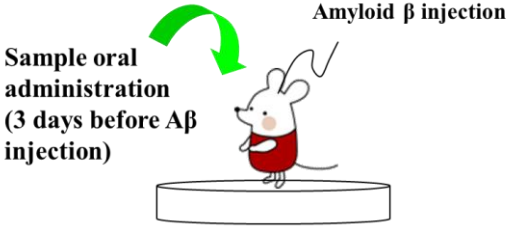
***Bifidobacterium breve* MCC1274**

(synonym, *B. breve* A1)

A strain originated from the gut of an infant

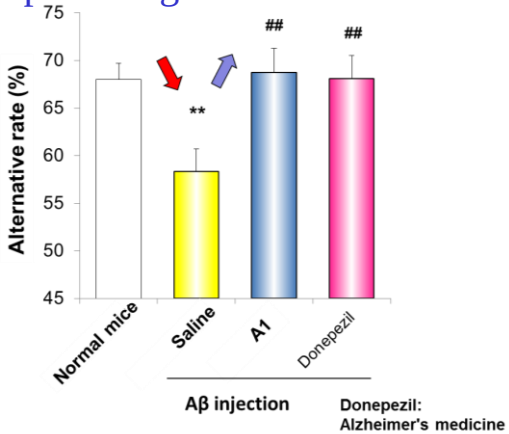
Previous studies

Pre-clinical study

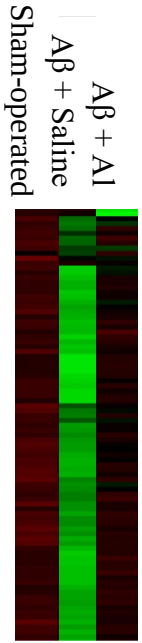


Alzheimer's disease (AD) model mice.

Spatial cognitive function



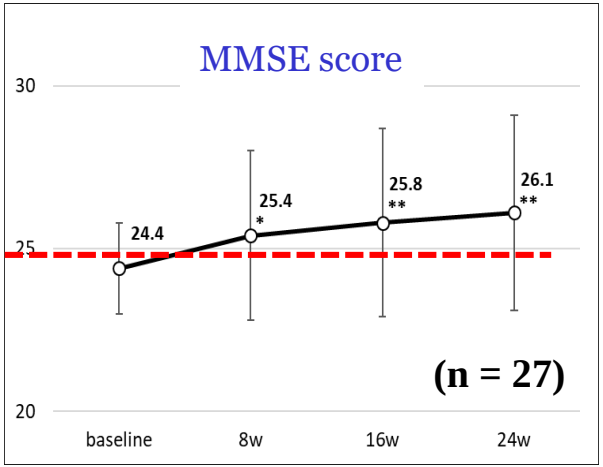
A1 normalized the hippocampal gene expressions related to inflammatory reaction induced by Aβ injection.



Kobayashi et al. Sci. Rep (2017)

Pilot clinical study

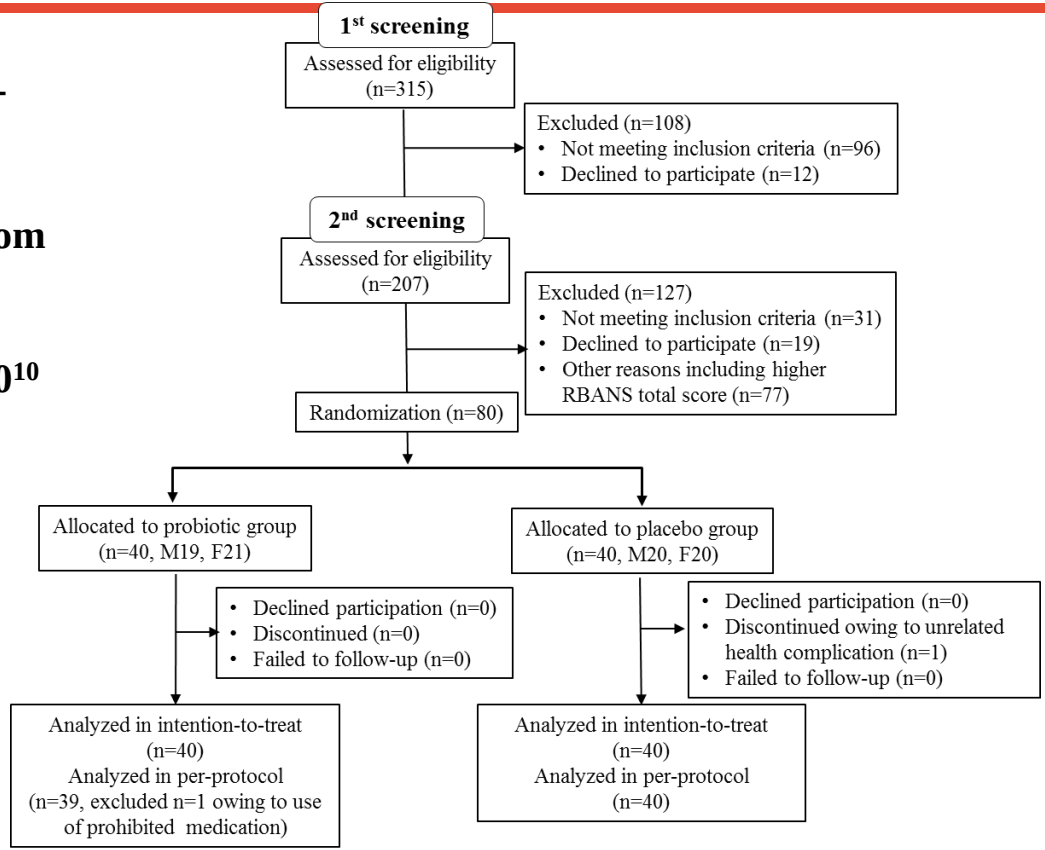
- **An open-label, single-arm study**
- **24-week supplementation of *B. breve* MCC1274 in elderly people with Mild Cognition Impairment (MCI).**



Kobayashi et al., JPAD (2018)

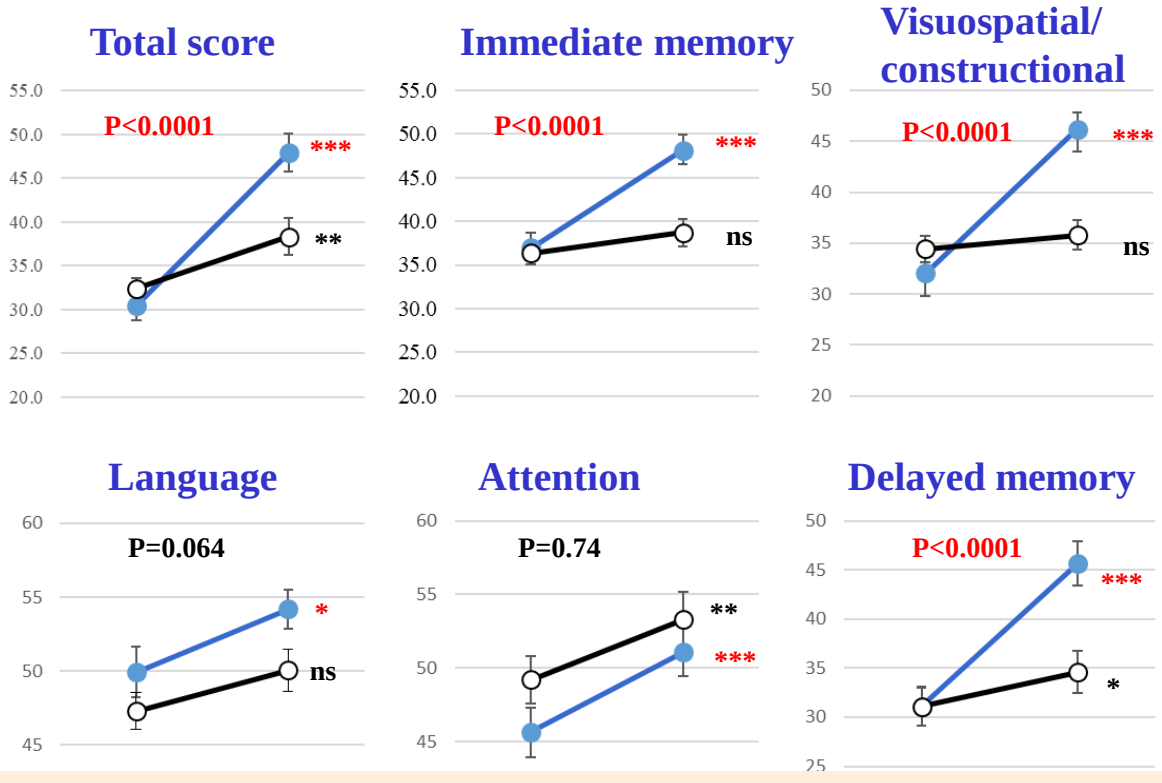
Effect to human, RCT

- **Design:**
  randomized, double-blind, placebo-controlled trial
- **Subjects:**
  80 healthy older adults suffering from MCI
- **Intervention:**
  probiotic (*B. breve* MCC1274, 2×10^{10} CFU) or placebo for 16 weeks.
- **Evaluation:**
  - **Primary outcome:**
    RBANS (the Repeatable Battery for the Assessment of Neuropsychological Status)
  - **Second outcome:**
    the Japanese version of the MCI Screen (JMCIS)



Xiao et al., J. Alzheimer's Disease (2020)

Results: Changes of RBANS Scores（Primary Endpoint）



B. breve MCC1274

Placebo

The Japanese version of RBANS test. It includes 12 standard cognitive subtests grouped in five domains:

- Immediate memory (list learning and story memory)
- Visuospatial/constructional (figure copy and line orientation)
- Language (picture naming and semantic fluency)
- Attention (digit span and digit symbol coding)
- Delayed memory (list recall, list recognition, story recall, and figure recall).

Significant improvement in cognitive function in the probiotic group compared to the placebo group.

P value: Inter-group difference by ANCOVA, *P<0.05, **P<0.01, *** P<0.001, intra-group difference, paired t-test
P values, inter-group difference, ANCOVA

Summary (2) topics of clinical effects

Clinical studies showed the effects of HRB strain in

- **Promoting the formation of health gut microbiota in preterm infants by *B. breve* M-16V.**
- **Improving cognitive function of elderly with MCI by *B. breve* MCC1274.**

These data suggest the potential health-promoting benefits of the specific *Bifidobacterium* strains to host.

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Clinical studies on HRB strains

Thank you for your attention!

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