



April 2022

# Modulation of Gut Microbiome as the Principal Action

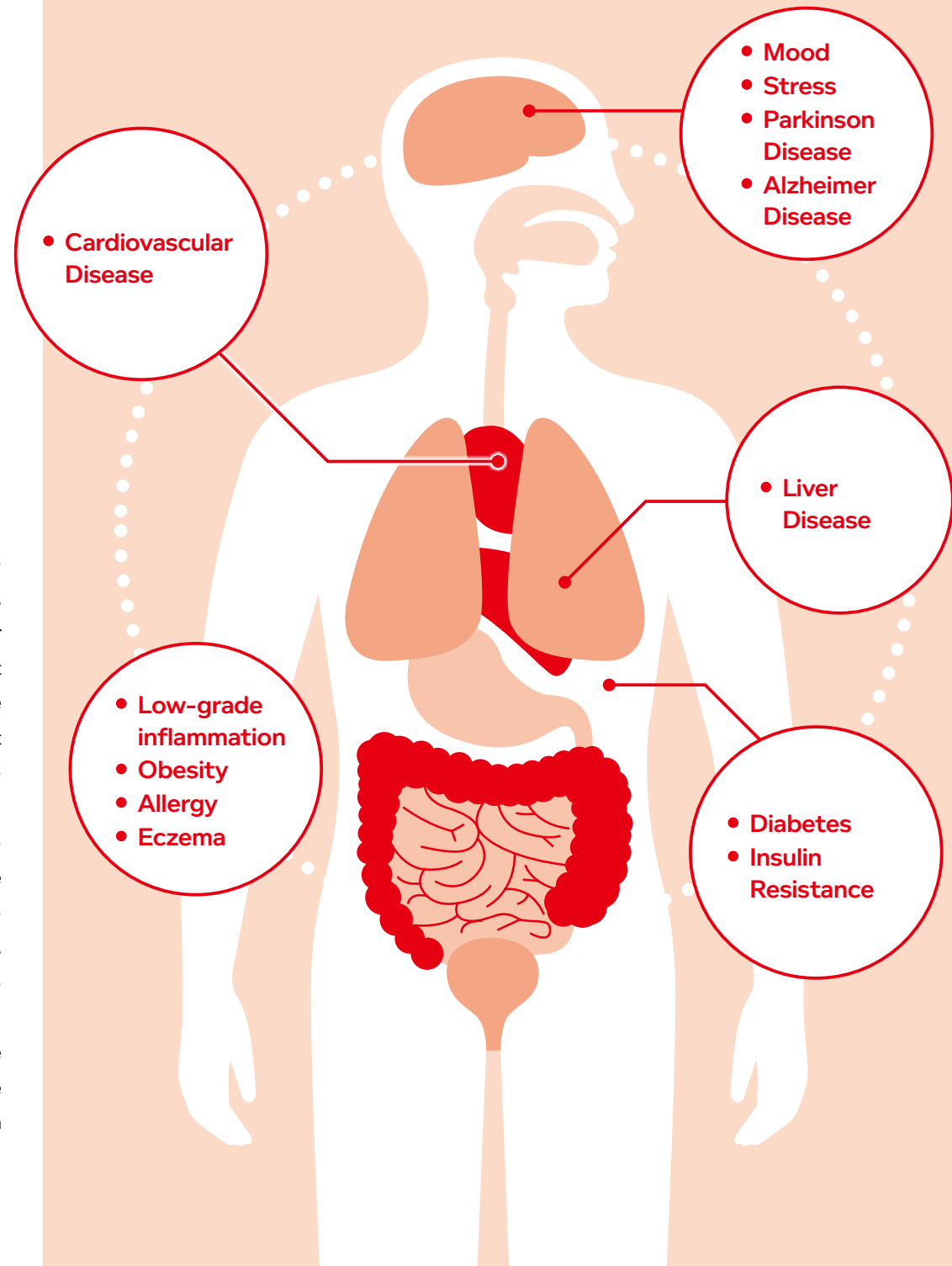


# Gut microbiota is at the intersection of everything

The boom in gut microbiome research in recent years has led to a greater understanding of how we interact with the microbes that live inside our gut. Nowadays, it is accepted that we are living with at least the same number of microorganisms as our human cells <sup>1</sup>. The human gut is home to trillions of microorganisms, including at least 1,000 different species of known bacteria that coexist in a balanced manner. They are collectively known as the gut microbiota. Studies have shown that maintaining the right balance of the gut microbiota, immune responses, and the key metabolites such as short-chain fatty acids, vitamins, etc. are vital for host physiology and immunity.

Nevertheless, the gut microbiota composition, and to a lesser extent their metabolic activity, change throughout the entire life, from birth to old age, and are under the influence of numerous factors (e.g. diet, lifestyle, and host metabolic activities). Accumulating evidence suggests that the gut microbiota are connected with many diseases, including disorders associated with obesity, chronic inflammatory diseases, cardiovascular disease, cancers, stress, and even neurodegenerative disorders <sup>2</sup>.

It has thus brought a multitude of different ways to modulate the gut microbiome to improve its health, including probiotics, prebiotics, and postbiotics. Probiotics are sought after for gut health, as consumers understand that there is a relationship between gut health and overall health.

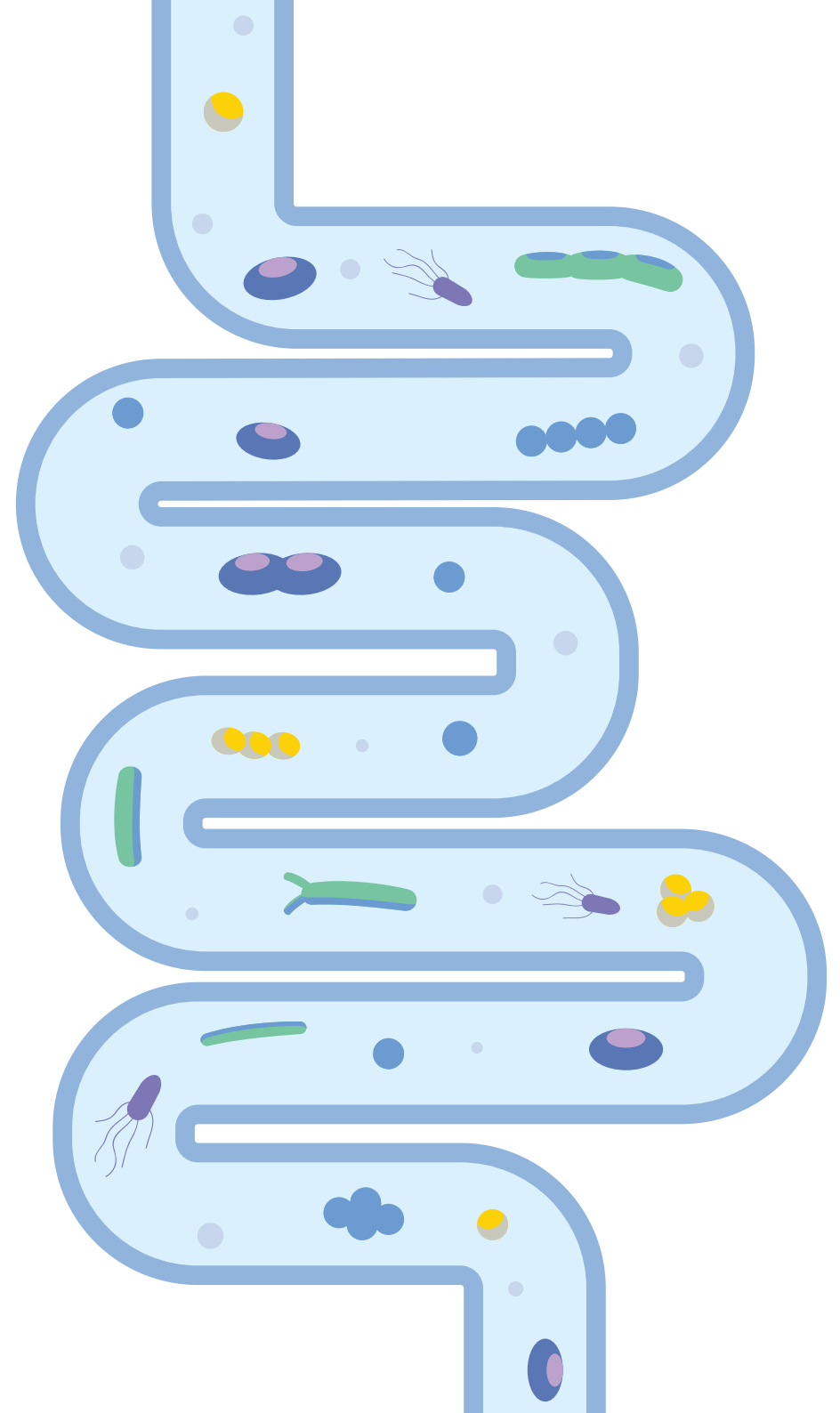




# Gut Health: New Insights into the Old Story

**D**emand for probiotics that deliver gut health isn't a new trend – but it looks set to stick around and potentially accelerate. Gut health goes far beyond solely digestive health, encompassing immunity support and overall wellbeing.

Gut health refers to a well-balanced microbiota in the gastrointestinal tract for which the gut microbiota was found to be affected in vast pathological situations such as diabetes, obesity, or autoimmune disease, and recent research even suggests an impact on mood due to the so-called gut-brain axis.



# At a Glance

## Gut health market insights

1

### A Real Market Opportunity

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The digestive health market alone currently stands at US\$60 billion a year, and it is growing rapidly (Euromonitor International).

2

### A Real Health Concern

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According to Euromonitor International's Health and Nutrition Survey 2020, about one-quartered consumers globally suffer from lower digestive health issues. In half of those cases, consumers claim that this has a moderate or even severe impact on their overall health.

3

### Probiotic in the spotlight

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Thus far, probiotics remain as a key ingredient in the gut health space. The coronavirus outbreak spurred growth in probiotics yogurt in China and Japan as consumers seek products that provide gut and immune support. In Western markets, gut health is gaining popularity among consumers who pursue a holistic approach to healthy living and probiotics continue to be a key ingredient to support a healthy gut microbiome (Euromonitor International).

4

### Post-pandemic impact

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Gut health is expected to gain further traction in 2022 and beyond. Post-Covid-19 consumers are expected to focus much more on preventative health and will pay more attention to healthy nutrition. The link between gut health and immunity support will likely provide a further boost.

5

### See the naturalness of probiotics differently

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Post-pandemic, people are craving connection with nature and there is renewed focus on the natural value of probiotics. With continued efforts in consumer education, consumers see natural probiotics differently. Backed with scientific rigour, most believe that the resident probiotic strains are better.

# Probiotics for gut health, how exactly do they work?

**W**ith growing awareness, probiotics are gaining traction in the gut health space. However, there is still work to be done to reach a wider audience and improve the understanding of gut health and the microbiome.

Although the growing interest has exploded over the past decade, there is still a knowledge gap in the effects of probiotics on the gut microbiome and, by extension, human health. The health benefits of the commercially available probiotics are often captured through only one snapshot. How externally introduced probiotics affect the microbiome, and crucially, how they work to have the desired effects are largely unclear.

**T**o date, there is a pool of commercially available probiotics, each with its origin, safety profile, efficacy, and mechanism of action. Those differences matter, and in the context of gut health, how they modulate the gut microbiome matters the most. Studies show that the efficacy of probiotics is strain-specific. How a probiotic strain work differs even if they are from the same species. Brand manufacturers need to consider this to ensure their probiotic products have the desired impact on human health. Yet, there is a lack of in vitro and human data on many of the available probiotic strains.

**I**n light of the knowledge gap, Morinaga Milk Industry has worked to understand a well-established probiotic strain – *Bifidobacterium longum* BB536. BB536 shows it is possible to build up a detailed understanding of a probiotic strain and its effects on the microbiome. Through a long research history, particularly in the past decade, researchers have sought to shed light on what happens when people consume BB536.

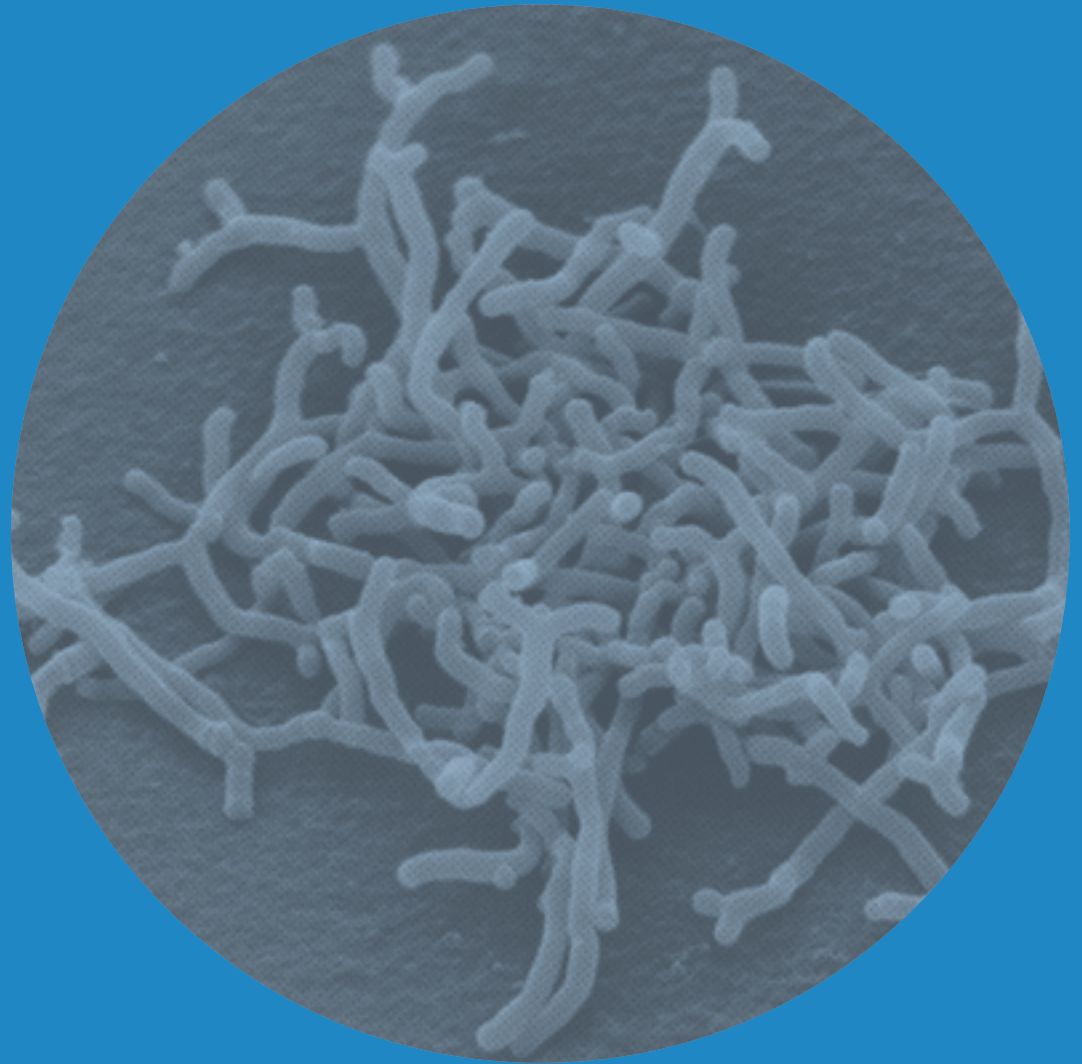


*B. longum*

# BB536: A half-century of discovery

*Bifidobacterium longum* BB536 is a clinically effective, well-established, multifunctional probiotic strain that has a long history of human use. Time proves everything; for more than 50 years, *B. longum* BB536 has been the superior Human-Residential Bifidobacteria (HRB) probiotic strain that provides a consistent beneficial effect in maintaining human health.

BB536 possesses a proven track record of safety and clinical efficacy in improving gastrointestinal, immunological, and infectious conditions, as demonstrated in more than 220 scientific studies (as of March 2022).





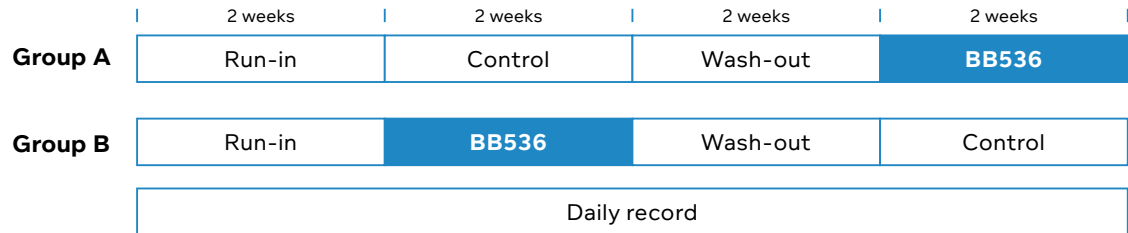
# What the Science Says?

## 1 Improving Gastro-intestinal Conditions

Mounting clinical evidence has shown that intake of BB536 products could improve the frequency of defecation and fecal characteristics in healthy adults and elderly people with irregular bowel movements <sup>3</sup>.

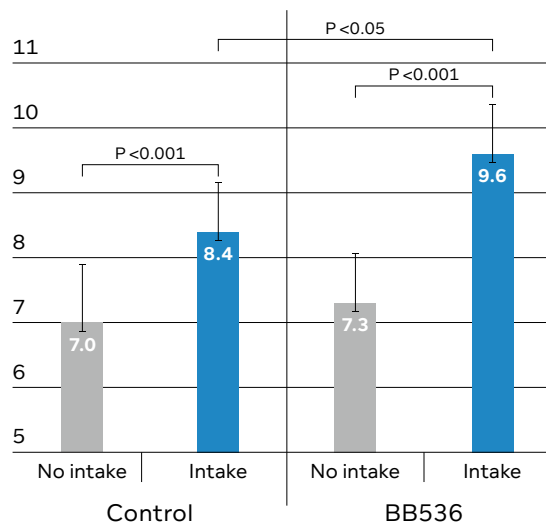
For instance, in a two-way crossover randomized controlled trial involving 55 adults, intake of a drink-type yogurt containing BB536 significantly increased the defecation frequency ( $P < 0.05$ ) and tended to increase the number of days with defecation ( $P = 0.070$ ) as compared to placebo yogurt during the intake period <sup>4</sup>.

Study design: two-way cross-over RCT

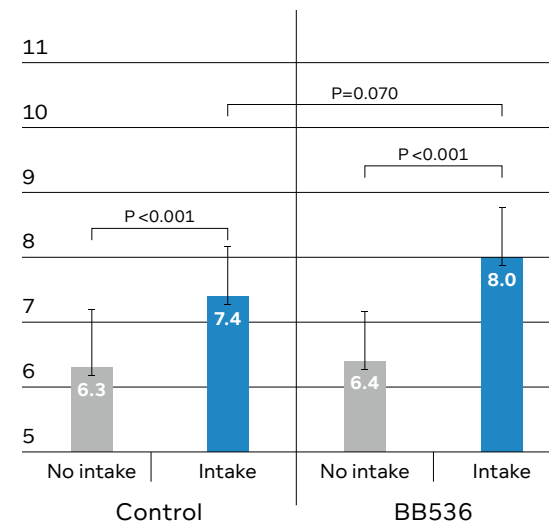


N=55 (aged 31.6+7 y) BB536: 100g of drink-type yogurt (containing BB536 > 210CFU/g)

Defecation frequency / 2 weeks



Number of days with defecation / 2 weeks



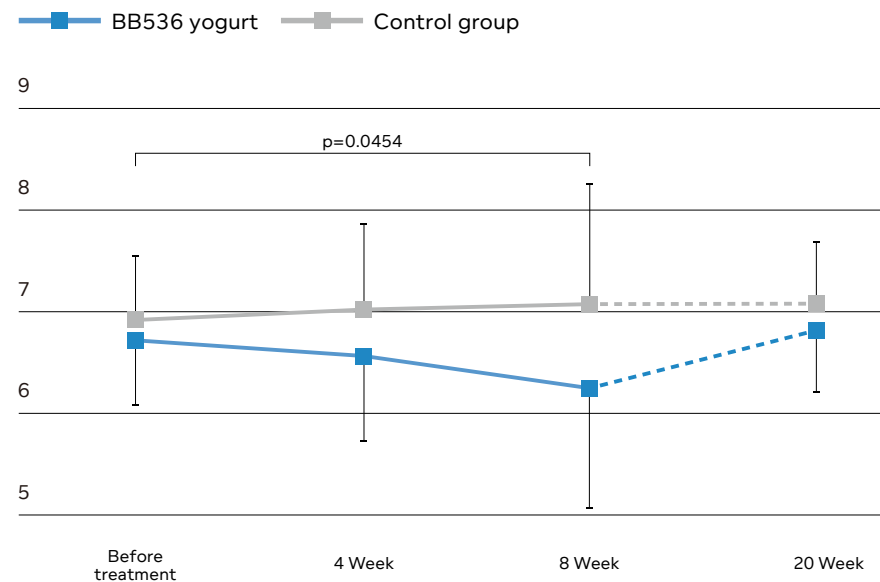
## 2 Improving intestinal microenvironment

Accumulating evidence suggests that gut microbial dysbiosis may represent an etiological factor in the development of gastrointestinal diseases. In particular, it has been reported that certain bacterial species, such as the toxin-producing bacteria, enterotoxigenic *Bacteroides fragilis* (ETBF), are associated with acute and persistent diarrheal disease in patients with inflammatory bowel disease <sup>5</sup> and the development of colorectal cancer <sup>6</sup>.

In an open, randomized, parallel-group trial involving 32 healthy adults who were the carriers of ETBF, ingestion of yogurt containing BB536 for 8 weeks had a discernible effect on the cell numbers of ETBF in the gut microbiota <sup>7</sup>.

As a result, a significant decrease in the cell numbers of ETBF as compared to baseline values was observed in subjects receiving BB536 yogurt but not in the control milk group. However, when the subjects ceased the consumption of BB536 yogurt, the number of ETBF returned to the baseline level. These results suggest that continuous ingestion of BB536 could decrease the opportunistic ETBF pathogens in the gut microbiota and condition the intestinal microenvironment.

Cell number of ETBF (log / g feces)

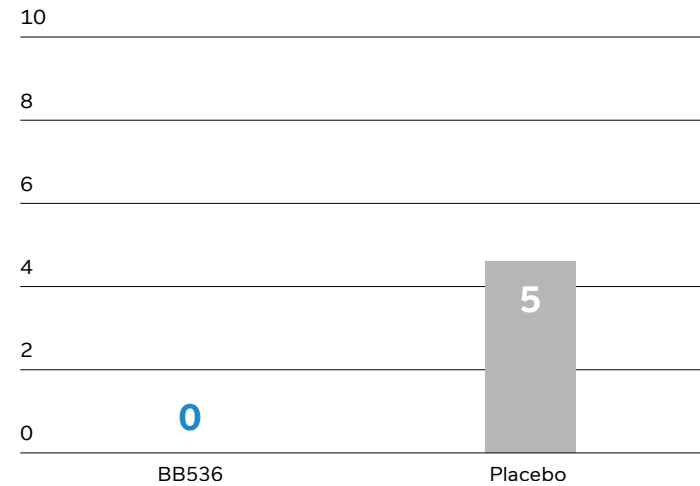


# 3 Immunomodulation

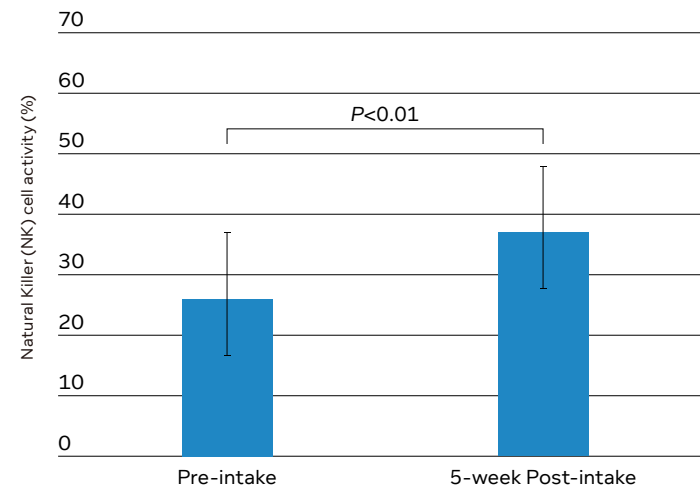
Multiple clinical studies have demonstrated that BB536 could modulate immune responses in human subjects, especially in the elderly <sup>3</sup>. As we age, our cellular immune responses tend to weaken. Studies show that the decline in immune function in the elderly could lead to impaired vaccine efficacy and an increased risk of influenza virus infection. Importantly, BB536 has been found to be able to improve waning immunity in the elderly <sup>8,9</sup>.

In a parallel-group, randomized controlled trial, 27 elderly people aged 65 years and older were pre-administered with BB536 powder (100 billion CFU/day) for 5 weeks, during which they also received influenza vaccination at week 3. The subjects were then randomized into a BB536 group (n=13) and a placebo group (n=14) for 14 weeks. During the study period, the number of subjects with flu symptoms was fewer in those who consumed BB536 than those who did not. Moreover, natural killer cell activity and neutrophil bactericidal activity were increased after BB536 administration, providing evidence of enhanced immunity against the influenza virus in the elderly <sup>8</sup>.

Number of subjects contracted influenza infection



Effect on NK Cell Activity

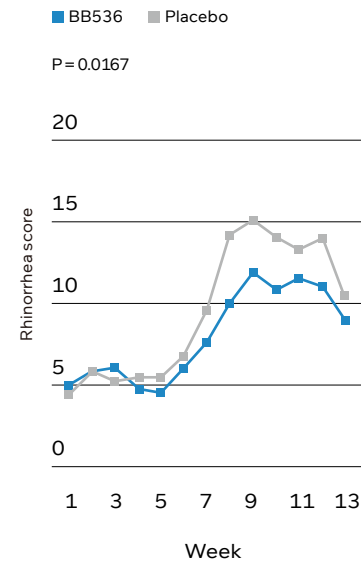


# 4 Anti-allergy

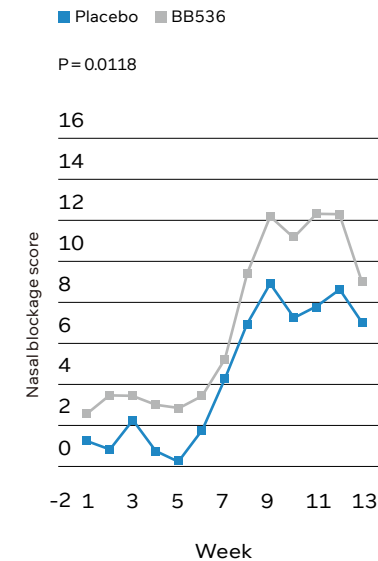
Backed by substantial clinical data and reproducible results, BB536 shows great promise in alleviating symptoms of allergic rhinitis, particularly those induced by the cedar pollens in Japan <sup>3</sup>. Intakes of BB536 yogurt or lyophilized powder have demonstrated a positive effect in alleviating nasal and ocular allergic symptoms and modulating the allergic immune response in subjects sensitive to Japanese cedar pollen.

In a randomized, double-blind, placebo-controlled trial involving 44 allergic rhinitis subjects (aged 26–57 years) during the cedar pollen season in 2005, which was the heaviest season within the past 10 years, intake of BB536 powder at a dose of 50 billion CFU/2 g twice daily for 13 weeks significantly reduced the allergic scores of rhinorrhea and nasal blockage. Intake of BB536 also significantly improved the T-helper 2 (Th-2)-skewed immune response that was occurred along with pollen dispersion. The plasma levels of thymus and activation-regulated chemokine (TARC), a Th-2 type chemokine, were remarkably normalized in subjects receiving BB536 powder than those of the placebo <sup>10</sup>.

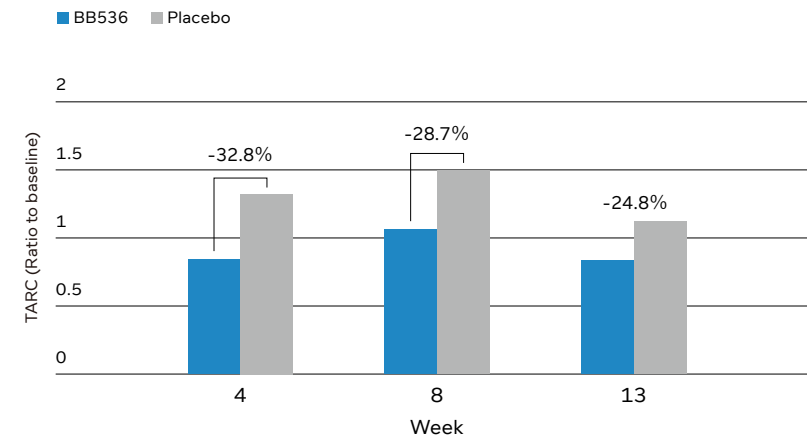
### Rhinorrhea



### Nasal blockage



### Natural killer (NK) cell activity (%)



# How BB536 Modulates the Microbiome?

## 1 Gut homeostasis

Studies suggest BB536 supports gut homeostasis. The increased abundance of bifidobacteria and presumably their associated activity, as stimulated by BB536 intervention, may act as a selective pressure on microbiota composition.

A striking example of the impact of BB536 on microbial homeostasis has been shown in the human trial of animal-based dietary intake <sup>11</sup>. When people consume an animal-based diet, their microbial profile changes, with the proportion of bile-tolerant bacteria such as *Bilophila wadsworthia* rising at the expense of the Firmicutes that metabolize dietary plant polysaccharides. Studies have linked the increased abundance of the bile-tolerant bacteria *Bilophila wadsworthia* to the development of inflammatory bowel disease and colorectal cancer.

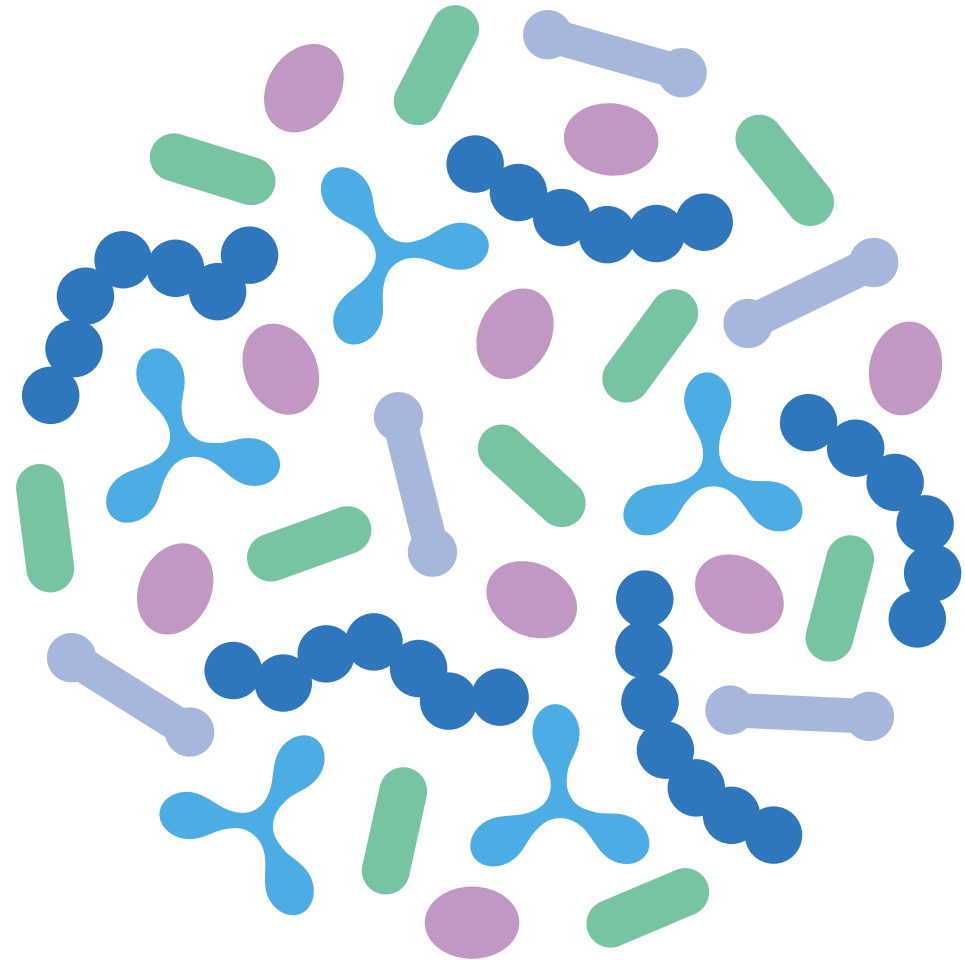
BB536 may be able to counter changes caused by animal-based diets. As demonstrated in the randomized clinical trial, consumption of yogurt supplemented with BB536 maintained a normal microbiota composition when eating an animal-based diet <sup>11</sup>. The finding points to the ability of BB536 to stabilize the gut microbiota, improve the intestinal environment, and enhance gut health.



## 2 Microbial cross-talk

Studies have also shown that BB536 improves gut microbiota homeostasis through its interaction and cross-talk with the endogenous gut microbiota, promoting beneficial metabolite production in the microbiome.

The evidence for cross-feeding of other species of human gut-derived microbiota comes from studies of the levels of intestinal butyrate and the prevalence of butyrate-producing *Eubacterium rectale* [12](#). Studies have shown BB536 makes acetate through the fermentation of carbohydrates, thereby stimulating the growth of butyrate-producing colon bacteria and possibly leads to subsequent. The work is part of a body of evidence suggesting BB536 improves gut homeostasis through cross-talk with other bacteria.



### 3 Immune homeostasis

There is evidence that modulation of the gut microbiome by BB536 could help regulate immune responses associated with allergic reaction. The maintenance of immune homeostasis is crucial to host survival and the gut microbiota is a critical regulator in this process.

A large collection of studies that looked at the effect of BB536 on allergic rhinitis triggered by exposure to certain pollen have demonstrated that BB536 is able to reduce the prevalence of pollen sensitization and alleviate allergic symptoms. It is evident that BB536 improved immune dysfunction by driving a fine-tuned homeostatic balance within the host-microbiome interaction.

Studies show that BB536 modulates immune homeostasis by promoting the stabilization of gut microbiota and consequently restores Th1/Th2 balance to alleviate allergic symptoms <sup>13</sup>. BB536 may also elicit a direct effect on antigen-induced IgE-mediated Th2 skewed immune balance via its bacterial component <sup>14</sup>.



# HRB for differentiation, resident strain matters

**W**ith growing awareness, consumers are seeking for probiotics with a natural value. The Covid-19 pandemic has further accelerated this interest. As such, human-residential bifidobacteria (HRB) are here for differentiation.

Bifidobacteria are commonly used as probiotic agents, with several different species of bifidobacteria have been isolated from the gut of a variety of animals, ranging from insects to mammals. With a few exceptions, bifidobacteria show strong exclusivity and adaptation to their specific hosts. Evolutionary analysis has shown that bifidobacteria have co-speciated with their respective hosts over the last 15 million years <sup>15</sup>, where bifidobacteria can be split into two groups: those that naturally reside in the human intestines and those that are not. The former is called Human-Residential Bifidobacteria (HRB), while

others who are the natural inhabitants of animal intestines, sewage, or foods are called non-HRB.

HRB species are the natural residents of the human gut including *B. infantis*, *B. breve*, *B. longum*, etc. that exhibit superior physiological functions and may promote better human health. Their presence in the human gut has been associated with health-promoting benefits.

Among the pool of commercially available probiotics, only a handful of them is the resident strain that has a natural history with the human host. *B. longum* BB536 is a superior strain of HRB – a natural inhabitant of the human gut that is more adapted to and more functional in the human intestinal environment. BB536 stands out from other probiotics. It offers a true natural value to your products and meets the increasing consumer demand.



# Human-Residential Bifidobacteria (HRB)

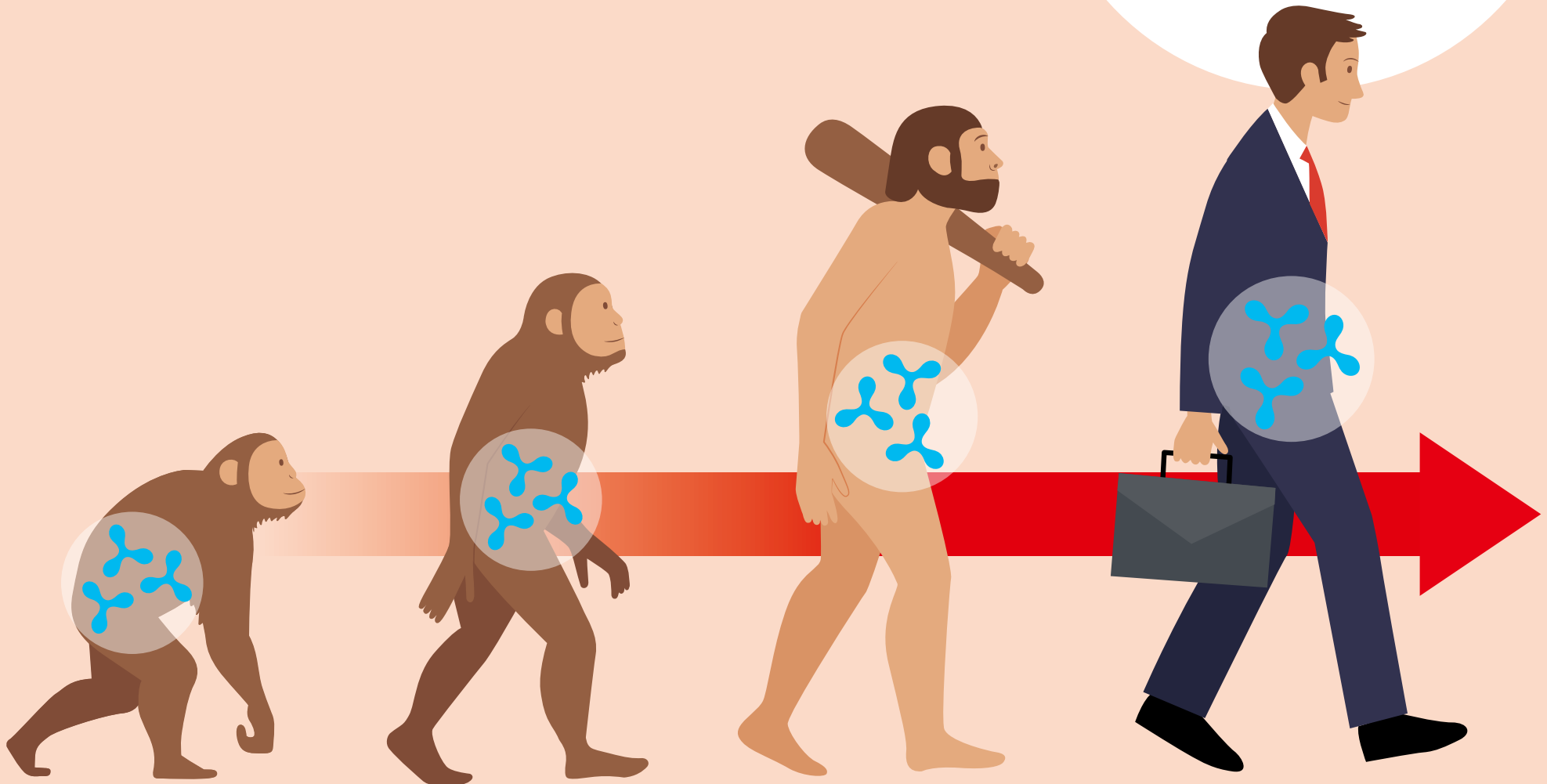
*B. infantis*, *B. breve*, *B. longum*, *B. bifidum*, etc.

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## Non-Human-Residential Bifidobacteria (Non-HRB)

*B. animalis*, *B. lactis*, *B. pseudolongum*,  
*B. thermophilum*, *B. magnum*, etc.

Morinaga Milk focuses its research on Human-Residential Bifidobacteria (HRB) and have developed a premium range of HRB probiotic strains.



# *Bifidobacterium longum* is a key gut determinant

It is estimated that somewhere between 100-200 species of known bacteria live inside our gut. Among these, *Bifidobacterium* spp. is recognized as one of the most critical genera to gut health. The species of bifidobacteria, particularly those of the HRB, predominantly inhabit the infant's gut. However, as we move through different life stages, the level of bifidobacteria in our gut changes. The level of bifidobacteria started to decrease after weaning and continued to decline gradually but remain considerably stable during adulthood. Thereafter, the population of bifidobacteria decreases again in old age <sup>16</sup>.

Notably, *B. longum* subsp. *longum* was widely detected in all segmented age groups ranging from newborns to centenarians <sup>17</sup>, for which some strains were shown to be genetically equipped to utilize both plant-derived and human milk oligosaccharides (HMO)-derived sugars.

In a comparative genome analysis of fecal samples obtained from 453 healthy Japanese subjects aged between 0 and 104 years, based on the obtained species-level gut microbiota composition of these Japanese subjects, only three species (*Blautia wexlerae*, *Streptococcus salivarius*, and *Bifidobacterium longum*) were detected in more than 50% of the subjects across each segmented age groups <sup>18</sup>. This result highlights the broad distribution of *B. longum* across (healthy) human lifespan as a distinctive property among the hundreds of bacterial species that are present in the healthy human gut. It is suggested that *B. longum* – a species that BB536 belongs to – is one of the key determinants in the human gut that plays a crucial role in maintaining human health across the lifespan.



# Innovate with BB536

**A**s explained above, BB536 is a unique HRB probiotic strain genuinely suitable for human consumption. With growing awareness, it will almost definitely become a norm for consumers to choose HRB as probiotics in food and supplement products. The pandemic has certainly accelerated this trend. Therefore, if you have a plan to develop a new product of food/supplement with probiotics right now, you might want to consider incorporating our HRB strain, *B. longum* BB536, to win a stance in the market.

Brands manufacturers can consider launching products with a focus on HRB probiotic strains beneficial to the gut such as BB536 and communicating the importance of a healthy gut microbiome to aid in overall health.



Infant  
formula



Yogurt



Capsule



Oil drop



Powdered  
formula



Chocolate

# What differentiate us?

Our years of research on HRB and BB536 are enabling manufacturers to create probiotic products backed by evidence of strain-specific effects on the gut microbiota and human health.

1

## Excellent stability

BB536 has an exceptionally good stability profile with a long shelf life that made it attractive in accomplishing cost-effectiveness and giving flexibility in formulation.

2

## Versatility with lower overage

BB536 can be incorporated into a wide range of delivery formats with lower overage to meet the needs of consumers for healthy, innovative, and convenient products.

3

## The power of human-residential bifidobacteria (HRB)

Brands manufacturers can take the lead in addressing gut health with human-residential bifidobacteria (HRB) strains such as BB536 that offer a sense of naturalness and familiarity.

4

## Perfect for combination

BB536 shows synergy when combined with other functional ingredients including prebiotics such as lactulose, GOS, FOS, and inulin, postbiotic such as LAC-Shield, and immune-supporting lactoferrin. It expands the possibilities and opportunities.





# Morinaga as your trusted partner

Since the 1960s, Morinaga Milk Industry Co., Ltd. has been researching the safety, functional health benefits, and mechanisms of action of probiotic bifidobacteria to better understand their role in maintaining human health. With its unique HRB probiotic strains and practical knowledge of applications, Morinaga Milk is ready to assist food companies with the new product development of different types of foods, including dietary supplements, functional foods, and infant nutrition. Start your journey today by contacting us to find out how Morinaga Milk's HRB probiotics can help you appeal to smarter consumers.

# Contact us



**Morinaga Milk Homepage**

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**Morinaga Probiotics Center**

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**Morinaga Milk LinkedIn page**



# References

1. Sender R, Fuchs S, Milo R. Revised estimates for the number of human and bacteria cells in the body. *PLoS Biol.* 2016;14(8):e1002533.
2. Cani PD. Gut microbiota—at the intersection of everything? *Nat Rev Gastroenterol Hepatol.* 2017;14(6):321–2.
3. Wong CB, Odamaki T, Xiao J-Z. Beneficial effects of *Bifidobacterium longum* subsp. *longum* BB536 on human health: Modulation of gut microbiome as the principal action. *J Funct Foods.* 2019;54.
4. Xiao J, Kondo S, Odamaki T, Miyagi K, Yaeshima T, Iwatsuki K, et al. Effect of yogurt containing *Bifidobacterium longum* BB536 on the defecation frequency and fecal characteristics of healthy adults: A double-blind cross over study. *Japanese J Lact Acid Bact.* 2007;18(1):31–6.
5. Sears CL. Enterotoxigenic *Bacteroides fragilis*: a rogue among symbiotes. *Clin Microbiol Rev.* 2009;22(2):349–69.
6. Orberg ET, Fan H, Tam AJ, Dejea CM, Shields CED, Wu S, et al. The myeloid immune signature of enterotoxigenic *Bacteroides fragilis*-induced murine colon tumorigenesis. *Mucosal Immunol.* 2017;10(2):421–33.
7. Odamaki T, Sugahara H, Yonezawa S, Yaeshima T, Iwatsuki K, Tanabe S, et al. Effect of the oral intake of yogurt containing *Bifidobacterium longum* BB536 on the cell numbers of enterotoxigenic *Bacteroides fragilis* in microbiota. *Anaerobe.* 2012;18(1):14–8.
8. Namba K, Hatano M, Yaeshima T, Takase M, Suzuki K. Effects of *Bifidobacterium longum* BB536 administration on influenza infection, influenza vaccine antibody titer, and cell-mediated immunity in the elderly. *Biosci Biotechnol Biochem.* 2010;74(5):939–45.
9. Akatsu H, Iwabuchi N, Xiao J, Matsuyama Z, Kurihara R, Okuda K, et al. Clinical effects of probiotic *Bifidobacterium longum* BB536 on immune function and intestinal microbiota in elderly patients receiving enteral tube feeding. *J Parenter Enter Nutr.* 2013;37(5):631–40.
10. Xiao J, Kondo S, Yanagisawa N, Takahashi N, Odamaki T, Iwabuchi N, et al. Probiotics in the treatment of Japanese cedar pollinosis: a double blind placebo controlled trial. *Clin Exp Allergy.* 2006;36(11):1425–35.
11. Odamaki T, Kato K, Sugahara H, Xiao J, Abe F, Benno Y. Effect of probiotic yoghurt on animal-based diet-induced change in gut microbiota: an open, randomised, parallel-group study. *Benef Microbes.* 2016;7(4):473–84.
12. Sugahara H, Odamaki T, Fukuda S, Kato T, Xiao J, Abe F, et al. Probiotic *Bifidobacterium longum* alters gut luminal metabolism through modification of the gut microbial community. *Sci Rep.* 2015;5:13548.
13. Odamaki T, Xiao J, Iwabuchi N, Sakamoto M, Takahashi N, Kondo S, et al. Fluctuation of fecal microbiota in individuals with Japanese cedar pollinosis during the pollen season and influence of probiotic intake. *J Investig Allergol Clin Immunol.* 2007;17(2):92.
14. Takahashi N, Kitazawa H, Shimosato T, Iwabuchi N, Xiao J, Iwatsuki K, et al. An immunostimulatory DNA sequence from a probiotic strain of *Bifidobacterium longum* inhibits IgE production in vitro. *FEMS Immunol Med Microbiol.* 2006;46(3):461–9.
15. Moeller AH, Caro-Quintero A, Mjungu D, Georgiev A V, Lonsdorf E V, Muller MN, et al. Cospeciation of gut microbiota with hominids. *Science (80- ).* 2016;353(6297):380–2.
16. Odamaki T, Kato K, Sugahara H, Hashikura N, Takahashi S, Xiao J, et al. Age-related changes in gut microbiota composition from newborn to centenarian: a cross-sectional study. *BMC Microbiol.* 2016;16(1):90.
17. Kato K, Odamaki T, Mitsuyama E, Sugahara H, Xiao J, Osawa R. Age-related changes in the composition of gut *Bifidobacterium* species. *Curr Microbiol.* 2017;74(8):987–95.
18. Odamaki T, Bottacini F, Kato K, Mitsuyama E, Yoshida K, Horigome A, et al. Genomic diversity and distribution of *Bifidobacterium longum* subsp. *longum* across the human lifespan. *Sci Rep.* 2018;8(1):85.

