Close Size: 8.5” (W) x 11” (H)

Open Size: 17” (W) x 11” (H)

For the use only of a Registered Medical Practitioner or a Hospital or a Laboratory

FLORIMAX/08/01/2022-23

Zuventus Gromaxx

Zuventus House, Plot Y2, CTS No: 358/A2, Near Nahur
Railway Station, Nahur (West), Mumbai - 400 078.

Product Monograph

Florimax™

Lactic Acid Bacteria & Bifidobacteria Capsules

Florimax is Original De Simone Formulation

Florimax Capsules: Each capsule contains at least 112.5 billion (112.5 x 10^9) CFUs

- **Florimax** contains original “De Simone Formulation,” a proprietary blend of live, lyophilized, probiotic bacteria:

1. Lactobacillus acidophilus DSM24735/SD5212
2. Lactobacillus plantarum DSM24730/SD5209
3. Lactobacillus paracasei DSM24733/SD5218
4. Lactobacillus delbrueckii subsp. bulgaricus† DSM24734/SD5210
5. Bifidobacterium longum± DSM24736/SD5220
6. Bifidobacterium breve DSM24732/SD5206
7. Bifidobacterium infantis± DSM24737/SD5219
8. Streptococcus thermophilus DSM24731/SD5207

Close Size: 8.5” (W) x 11” (H)

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Florimax™
Lactic Acid Bacteria & Bifidobacteria Capsules

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CONTENT :

Florimax™
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FLORIMAX : EXECUTIVE SUMMARY

Florimax is a high potency live, lyophilized, Probiotic culture for oral administration. Florimax contains the Original De Simone Formulation, a blend of 8 multispecies, standardised strains of live bacteria, one of the few Probiotic preparations that has undergone rigorous double-blind, placebo-controlled scientific trials.

It is a patented, internationally acclaimed Probiotic formulation (US/EU approved products) combining top-quality Lactobacilli, Bifidobacteria and Streptococcus Thermophilus strains, provides 112.5 billion (112.5 x 10^9) Colony Forming Units (CFUs) per capsule.

The sellers of VSL#3® were ordered by a U.S. court to stop any claims that state or suggest a false continuity between the new formulation sold as VSL#3® and the Original De Simone Formulation.

A state of balance within the microbial population within the gastrointestinal tract can be called “Eubiosis” while an imbalance of the intestinal microbiota, termed “Dysbiosis”.

Gut microbiota Dysbiosis is a condition related with the pathogenesis of intestinal illnesses and extra-intestinal illnesses including cancer, obesity and a variety of bowel disorders.

Many therapeutic strategies have been developed to re-establish intestinal eubiosis. The main and at present best known and most adopted therapeutic strategies include: Probiotics, prebiotics, diet approach to modulate gut microbiota.

Florimax is a medical food as defined by the Orphan Drug Act and additional FDA regulations. **Florimax** is specially formulated and processed to provide a precise mixture of certain bacterial species to the gastrointestinal tract. It is a non-drug therapy that addresses distinct nutritional requirements to promote microbial balance in individuals which cannot be addressed by modification of the diet alone.

Florimax is intended for the clinical dietary management of patients who, (because of therapeutic or chronic medical needs) have special medically-determined nutrient requirements; the dietary management of which cannot be achieved by the modification of the normal diet alone.

Florimax is intended to be used under the supervision of a physician for the dietary management of Dysbiosis associated with Irritable Bowel Syndrome (IBS), Ulcerative Colitis, Antibiotic-Associated Diarrhea, Hepatic Encephalopathy, Pouchitis etc. **The De Simone Formulation in Florimax has been supported with over 75 published clinical trials in human subjects, with extensive clinical research in the dietary management of Dysbiosis associated with IBS, UC, AAD, Pouchitis and HE.**

The Probiotic bacteria in **Florimax** are non-pathogenic, non-toxigenic and Generally Recognized as Safe (GRAS) as food ingredients. **The De Simone Formulation in Florimax has been the subject of over 75 clinical studies involving over 5,000 adults, children and infants - including immuno-compromised individuals.**

Recommended dosage is 2-8 **Florimax** Capsules daily for adults as directed by healthcare provider. **Florimax** Capsules can be consumed directly. Adjustment of the intestinal flora can take a few days or weeks; it may take up to one month for the colonization of the gut to become optimally stable if consumed on a regular basis. Recommended Pediatric daily intake for **Florimax** Capsules, should be as per guidance of healthcare provider. For Pediatric administration **Florimax** Capsules can be pulled apart and sprinkled on apple sauce, yogurt or any cold food.

Florimax™
Lactic Acid Bacteria & Bifidobacteria Capsules

PREFACE

“Death sits in the bowels; a bad digestion is the root of all evil”

- Hippocrates, ca. 400 BC

Nobel laureate Élie Metchnikoff, professor at the Pasteur institute in Paris, in the beginning of the 20th century hypothesized that the reason for the apparent longevity of Bulgarian peasants was that they consumed large quantities of fermented milk products like curd and buttermilk. He believed that the lactic acid bacteria in these products replaced the harmful organisms found in the intestines and thus reduced the production of toxins that lead to disease.

More than 100 trillion microbes inhabit the human gastrointestinal (GI) tract, and the total number of genes derived from these microbes exceeds that of the human genome by at least 100-fold.

Under healthy conditions, the gut microbiota exists in a state of “Normobiosis / Eubiosis” in which microorganisms with beneficial effects on health predominate over harmful species. This situation is crucial for normal gut homeostasis and optimal development of the host.

The gut microbiota exhibits many important physiological functions that include regulation of energy levels and metabolism, neutralization of drugs and carcinogens, modulation of intestinal motility, regulation of immunity, barrier effects, and protection against pathogens. Host behavior and cognitive functions such as learning, memory, and decision-making are also believed to be affected by the gut microbiota. In a broad sense, the gut microbiota appears to be critical to maintain host homeostasis and health.

Probiotics, especially Lactobacillus and Bifidobacterium have been suggested to be associated with alleviation of lactose intolerance; prevention and cure of viral, bacterial and antibiotic or radiotherapy induced diarrhoea; immunomodulation; antimutagenic and anticarcinogenic effects; and even blood cholesterol reduction.

The optimism associated with Probiotics is, however, counter-balanced by skepticism as many “Probiotic” products in the market are unreliable in content and unproven clinically. In India there are no regulatory guidelines for Probiotic foods. In the absence of any such standards and guidelines, there is great scope for spurious products with false claims being marketed. It therefore, becomes imperative that these products fulfil some essential prerequisite conditions before being labelled as a 'Probiotic Product'.

Florimax (De Simone Formulation) is high potency Probiotic live, lyophilized, Probiotic cultures for oral administration. **It is a patented, internationally acclaimed Probiotic formulation (composition similar to Vivomixx, Visbiome US/EU approved products) combining top-quality Lactobacilli, Bifidobacteria and Streptococcus Thermophilus strains & provides 112.5 billion (112.5 x 10^9) Colony Forming Units (CFUs) per capsule.**

Florimax™
Lactic Acid Bacteria & Bifidobacteria Capsules

Florimax contains the original De Simone Formulation of Probiotic. VSL#3® produced before January 31, 2016 contained “De Simone Formulation”. However, after January 2016 the VSL#3® product was changed. The sellers were ordered by a U.S. court to stop any claims that state or suggest a false continuity between the new formulation sold as VSL#3® and the original De Simone Formulation.

This Monograph, dedicated to Florimax (De Simone Formulation), presents an up to date compilation of the scientific evidences for the dietary management of dysbiosis associated with Irritable Bowel Syndrome (IBS), Inflammatory Bowel Diseases (Ulcerative Colitis, Pouchitis), Antibiotic-Associated Diarrhea, Hepatic Encephalopathy.

COMPARISON OF FLORIMAX “DE SIMONE FORMULATION” VS. VSL#3®

FLORIMAX (DSF)	VSL#3
Streptococcus thermophilus DSM 24731	Streptococcus thermophilus BT01
Bifidobacteria B. breve DSM 24732	Bifidobacterium breve BB02
Bifidobacterium longum DSM 24736	Bifidobacterium longum* BL03
Bifidobacterium infantis DSM 24737	Bifidobacterium infantis* BI04
Lactobacillus acidophilus DSM 24735	Lactobacillus acidophilus BA05
Lactobacillus plantarum DSM 24730	Lactobacillus plantarum BP06
Lactobacilus paracasei DSM 24733	Lactobacillus paracasei BP07
Lactobacillus delbrueckii subsp. bulgaricus DSM 24734	Lactobacillus delbrueckii ssp. bulgaricus** BD08
Not less than 112.5 billion CFU / capsule	Not less than 112.5 billion CFU / capsule

HUMAN GUT MICROBIOTA

Bacteria, unicellular eukaryotes, and other organisms inhabit the human body in large numbers. The human gut is dominated by several bacterial phyla including Bacteroidetes, Firmicutes, and Actinobacteria. The term “microbiota,” “microflora,” or “normal flora” is used to designate this vast host of microbes which coexist with the host.

It is estimated that the human microbiota contains as many as 1014 bacterial cells, a number that is 10 times greater than the number of human cells present in our bodies. Virtually every surface of the human body starting from the skin surface to the genitourinary tract, oral cavity, respiratory tract, ear, and the gastrointestinal tract is colonized heavily by various species of bacteria. By far, the most heavily colonized organ is the gastrointestinal tract (GIT) which houses a huge microbial ecosystem; the colon alone is estimated to contain over 70% of all the microbes in the human body.[1]

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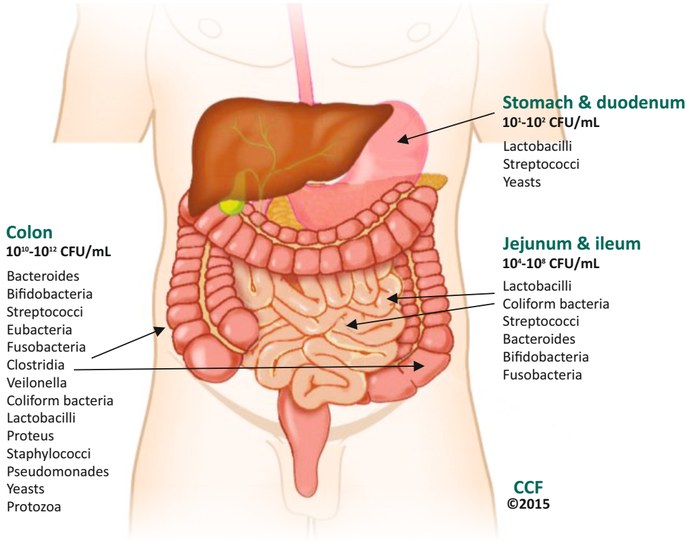

Figure 1 : The human gut microbiota[4]

COMPOSITION OF GUT FLORA

The gastrointestinal tract of an adult human is estimated to harbor about 100 trillion viable bacteria. These live bacteria are known as intestinal or gut flora. Viruses, fungi and protozoa can also be present, but these normally form only a minor component of the total resident population of microorganisms in healthy individuals. The density of microorganisms in the gut flora increases dramatically from 10 - 1,000 CFU/ml in the stomach to 10 - 100 billion CFU/gm in the large intestine and these belong to as many as 400 different species, and anaerobic bacteria outnumber aerobic bacteria by a factor of 1000:1. Anaerobic flora is dominated by bacteroides spp., bifidobacteria, lactobacillus, propionibacteria and clostridia. Among aerobic and anaerobic bacteria enterobacteria, mainly E. coli, and enterococci predominate.[2,3]

The Predominant microflora in the GI tract is as follows (Figure 2) :

- Proximal small intestine : Lactobacilli + Enterococcus faecalis (10 5 - 107 / ml of fluid)
- Distal small intestine : Lactobacilli + Enterococcus faecalis + Coliforms + Bacteroides (108 bacteria / ml of fluid)
- Colon : Bacteroides + Bifidobacteria (1011 bacteria / ml of fluid).[2,3]

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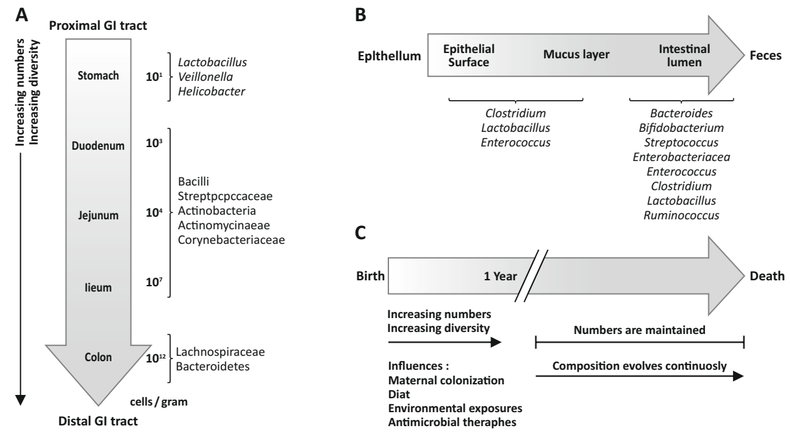
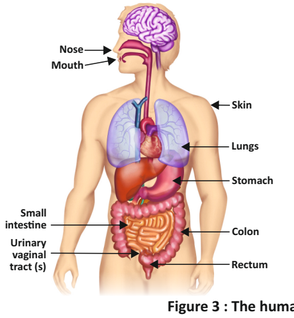


Figure 2 : Variations in microbial numbers & composition across the length of the gastrointestinal tract

A new-born baby has a sterile gut that is colonized by bacteria from the mother and from the baby's surroundings or environment. An adult human has 10 times more bacterial cells on, and in, the entire body as compared to the total human cells (Figure 3). The human microbiome is highly complex and diverse. Its composition and number varies from the nose and mouth to the distal colon and rectum. The composition and complexity of the gut microbiota changes when the baby is weaned to solid foods. Dietary changes in adulthood are also greatly responsible for the composition of gut microbiota. This technique has been used to show that 90% of the bacteria belong to two phyla, namely, the Bacteroidetes and Firmicutes.



Human body 10^{13} calls 23,000 genes ⟷ **Normal flora 10^{14} microbial cells on the human body. 3.3 million genes**

Amount of bacteria per gram of cellular component

- **Stomach - 10^{1} to 10^{3} cells**
- **Duodenum - 10^{3} cells**
- **Jejunum - 10^{4} cells**
- **Ileum 10^{4} to 10^{7} cells**
- **Proximal colon 10^{10} to 10^{12} cells**
- **Transverse colon 10^{11} to 10^{12} cells**
- **Distal colon >10^{12} cells**

“The microbiota can be viewed as a metabolic organ exquisitely tuned to our physiology that performs function we have not had to evolve on our own”

Backhed et al. 2004. PNAS 101:15718-15723

Figure 3 : The human body & number of bacteria present in the total microflora

ROLE OF GUT MICROBIOTA IN HEALTH & DISEASE

Studies have shown that, although there is a heritable component to gut microbiota, environmental factors related to diet, drugs, and anthropometric measures are larger determinants of microbiota composition. Gut microbes are key to many aspects of human health including immune, metabolic and neurobehavioural traits (Fig 3). Different levels of evidence support the role of gut microbiota in human health, from human studies.

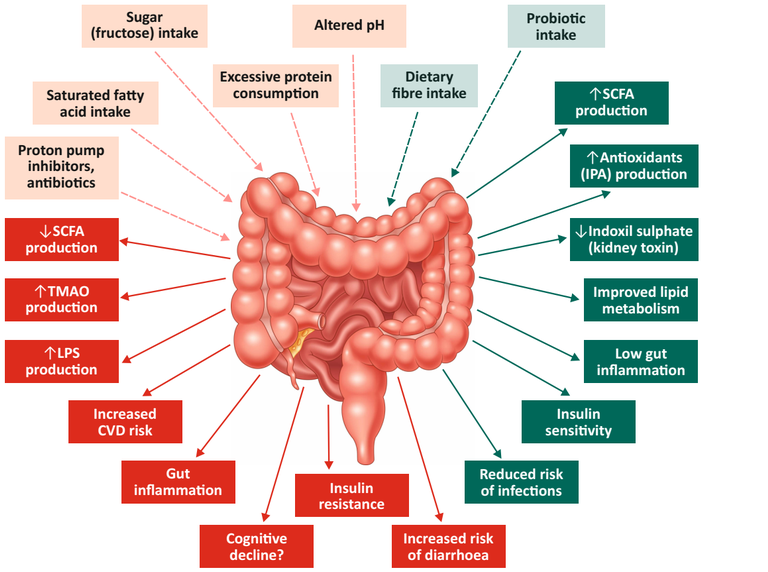


Figure 4 : Schematic representation of the role of the gut microbiota in health & disease giving some examples of inputs & outputs.
CVD: Cardiovascular Disease; IPA: Indolepropionic Acid; LPS: Lipopolysaccharide; SCFA: Short Chain Fatty Acids; TMAO: Trimethylamine N-oxide

The gut microbiota or microflora has a crucial role in human health and disease. The colon or the large intestine is the organ which is the preferred site for bacterial colonization. The GIT is also rich in many molecules which can be used as nutrients by microbes. Hence the GIT has the potential to be heavily colonized by various bacteria both harmful and beneficial. The mucosa of the gastrointestinal tract is

The gut microbiota or microflora has a crucial role in human health and disease. The colon or the large intestine is the organ which is the preferred site for bacterial colonization. The GIT is also rich in many molecules which can be used as nutrients by microbes. Hence the GIT has the potential to be heavily colonized by various bacteria both harmful and beneficial. The mucosa of the gastrointestinal tract is continuously exposed to an environment that is rich in foreign substances, such as food particles and antigens of microbial origin. Particular changes in the intestinal ecosystem might contribute to the development of certain illness. There is therefore a need for an exhaustive review on the functions of the gut microbiota, occurrence of gut dysbiosis (alteration or imbalance of the microflora), how these intestinal bacteria trigger development of disease once the normal flora of a healthy individual is imbalanced, exploiting this intricate and interwoven ecosystem for understanding human health, development of biotherapeutics, and future perspectives.[6]

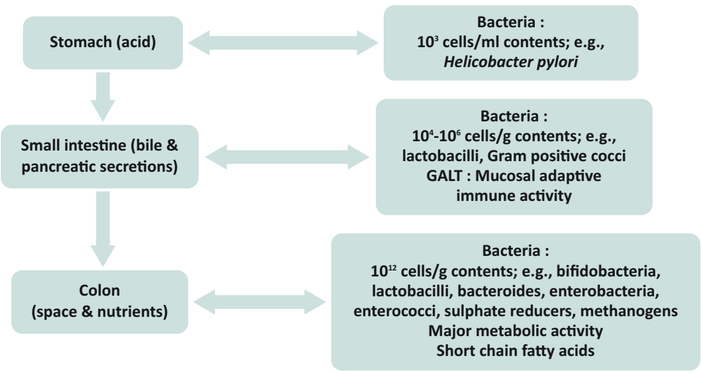


Figure 5 : Reciprocal relationship between human gut bacteria and the host.[5]

Gut bacteria play an important role in human health, including contributing to the host gut defense system and helping the gut to maintain normal function, while its composition can be influenced by the host (Figure 5).[5]

The imbalanced gut bacteria have been studied in diseases such as inflammatory bowel disease, antibiotic-associated diarrhea, colon cancer, hypercholesterolemia and others. Lactic acid bacteria, belonging to the genus Lactobacillus and Bifidobacterium, have been shown to positively influence health. Hence, re-establishing the balance by using these bacteria (termed "probiotics") for disease treatment and prevention should prove advantageous. Probiotics along with prebiotics and synbiotics have been used and studied in various disease areas. **Several studies have indicated that an altered gut microbiota is associated with several diseases that are particularly prevalent in the 21st century.[6]**

WHAT DOES THE GUT MICROBIOTA DO?

The gut microbiota provides essential capacities for the fermentation of nondigestible substrates like dietary fibres and endogenous intestinal mucus. **This fermentation supports the growth of specialist microbes that produce short chain fatty acids (SCFAs) and gases. The major SCFAs produced are acetate, propionate, and butyrate.**[7]

Butyrate

Butyrate is the main energy source for human colonocytes, can induce apoptosis of colon cancer cells, and can activate intestinal gluconeogenesis, having beneficial effects on glucose and energy homeostasis. Butyrate is essential for epithelial cells to consume large amounts of oxygen through β oxidation, generating a state of hypoxia that maintains oxygen balance in the gut, preventing gut microbiota dysbiosis.[7]

Propionate

Propionate is transferred to the liver, where it regulates gluconeogenesis and satiety signalling through interaction with the gut fatty acid receptors.

Butyrate and propionate, but not acetate, seem to control gut hormones and reduce appetite and food intake in mice. Gut microbial enzymes contribute to bile acid metabolism, generating unconjugated and secondary bile acids that act as signalling molecules and metabolic regulators to influence important host pathways.[7]

Acetate

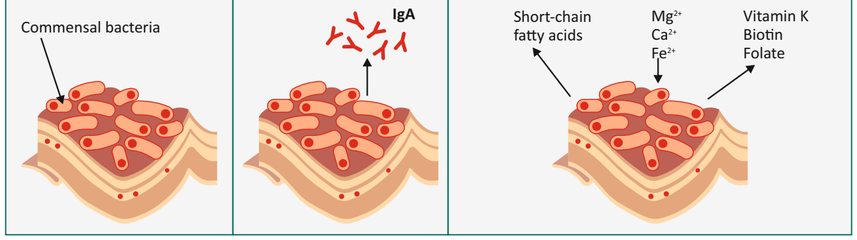
Acetate - the most abundant SCFA and an essential metabolite for the growth of other bacteria - reaches the peripheral tissues where it is used in cholesterol metabolism and lipogenesis, and may play a role in central appetite regulation.[20] Randomised controlled trials have shown that higher production of SCFAs correlates with lower diet-induced obesity[21] and with reduced insulin resistance.[7]

Other specific products of the gut microbiota have been implicated directly in human health outcomes. Examples include trimethylamine and indolepropionic acid. The production of trimethylamine from dietary phosphatidylcholine and carnitine (from meat and dairy) depends on the gut microbiota and thus its amount in blood varies between people. Trimethylamine is oxidised in the liver to trimethylamine N-oxide, which is positively associated with an increased risk of atherosclerosis and major adverse cardiovascular events. Indolepropionic acid is highly correlated with dietary fibre intake[25] and has potent radical scavenging activity in vitro, which seems to reduce the risk of incidence of type 2 diabetes.[7]

FUNCTIONS OF THE INTESTINAL FLORA

Enteric bacteria form a natural defence barrier and exert numerous protective, structural and metabolic effects on the epithelium.

Protective functions	Structural functions	Metabolic functions
Pathogen displacement Nutrient competition Receptor competition Production of anti-microbial factors e.g., bacteriocins, lactic acids	Barrier fortification Induction of IgA Apical tightening of tight junctions Immune system development	Control IEC differentiation and proliferation Metabolize dietary carcinogens Synthesize vitamins e.g., biotin, folate Ferment non-digestible dietary residue and endogenous epithelial-derived mucus Ion absorption Salvage of energy



Commensal bacteria exert a miscellany of protective, structural and metabolic effects on the intestinal mucosa.

ROLE OF THE GI MICROBIOTA IN HEALTH

Relationship between gut flora and humans is not merely commensal (a non-harmful coexistence), but rather a mutualistic relationship. Besides breaking down food compounds and synthesizing vitamins and other nutrients, they play an important role in the development and training of the immune system. They provide colonization resistance, protect against epithelial injury and promote angiogenesis and fat storage. They are also able to modulate bone-mass density, modify the nervous system and metabolize therapeutics into active compounds.

- **Microorganisms perform a host of useful functions:**

a. Fermenting (digestion) unused energy substrates
b. Training the immune system
c. Preventing growth of harmful, pathogenic bacteria
d. Regulating the development of the gut
e. Producing vitamins for the host (e.g. biotin and vitamin k)
f. Producing hormones to direct the host to store fats
g. Defending against some diseases.[8]

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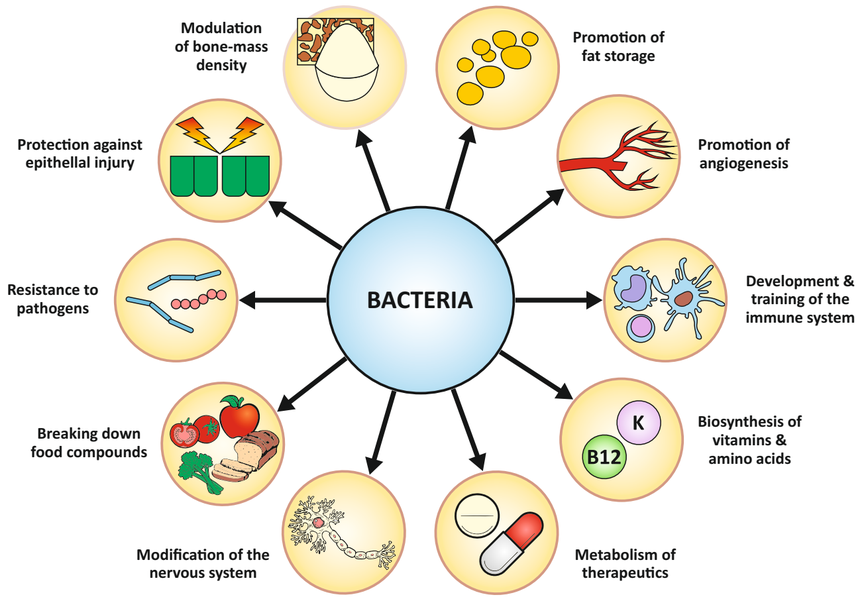

Figure 6 : Main functions of bacteria in the gut. Bacteria benefit the host in many ways.

DYSBIOSIS

A state of balance within the microbial population within the GI tract can be called “eubiosis” while an imbalance is termed “dysbiosis”. An imbalance of the intestinal microbiota, termed “dysbiosis”. Dys means 'faulty' and bios means 'life and growth'. This implies faulty life. [9]

The homeostatic balance of the intestinal microflora is extremely beneficial to the host, however if there is a change in the microbial composition that causes a drastic imbalance between the beneficial and potentially pathogenic bacteria, the gut becomes vulnerable to pathogenic insult with gut microbial alterations. This imbalance in the microbial equilibrium is termed “dysbiosis”, which has been further defined as a disturbance to gut microbiota homeostasis due to an imbalance in the flora itself, changes in their functional composition and metabolic activities, or changes in their local distribution.[9]

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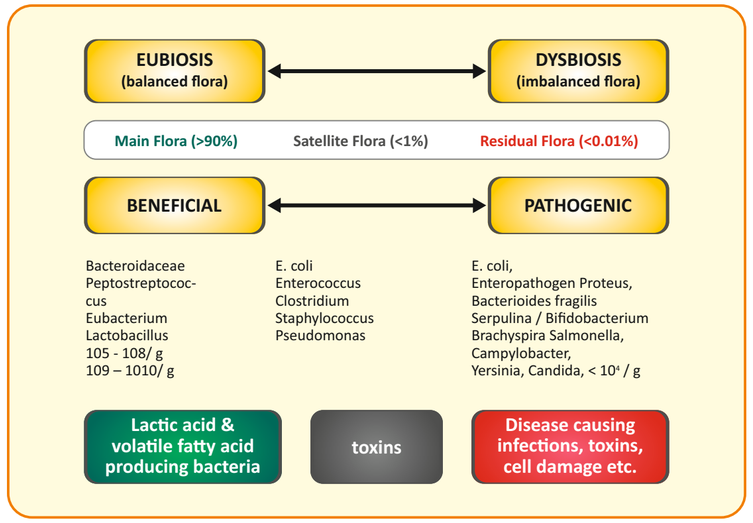

DYSBIOSIS TYPES

In general, dysbiosis can be categorized into three different types :

1. Loss of beneficial organisms
2. Excessive growth of potentially harmful organisms
3. Loss of overall microbial diversity

It has been found that these three types are not mutually exclusive and can occur at the same time, which is most often the case.[9]

THE GUT MICROBIOTA IN HEALTH & INTESTINAL DISEASE

The gastrointestinal microbiota plays a role in host physiology, metabolism and nutrition. An alteration in the gut microbial community is linked to a number of intestinal conditions, including cancer, obesity and a variety of bowel disorders. The contribution of beneficial components of the gut microbiome to host physiology, metabolism and immune function has become the focus of ever more attention, and will undoubtedly lead to new therapeutic approaches.[8]

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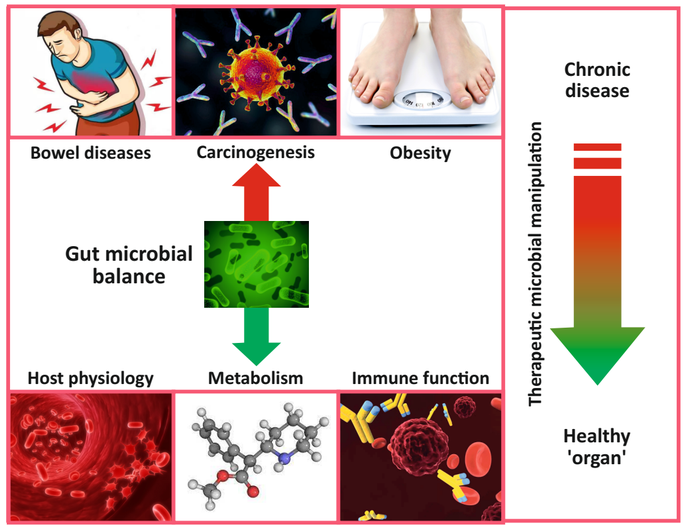

Figure 7 : The gut microbiota in health & intestinal disease.[8]

INFLUENCE OF GUT MICROBIAL COMMUNITIES ON HEALTH

The microbial communities that colonize different regions of the human gut influence many aspects of health.

In the healthy state, they contribute nutrients and energy to the host via the fermentation of nondigestible dietary components in the large intestine, and a balance is maintained with the host's metabolism and immune system.

Negative consequences, however, can include acting as sources of inflammation and infection, involvement in gastrointestinal diseases and possible contributions to diabetes mellitus and obesity. Major progress has been made in defining some of the dominant members of the microbial community in the healthy large intestine and in identifying their roles in gut metabolism.

Furthermore, it has become clear that diet can have a major influence on microbial community composition both in the short and long term, which should open up new possibilities for health manipulation via diet. The extent of inter-individual variation in microbiota composition within the population has also become apparent, and probably influences individual responses to drug administration and dietary manipulation.[12]

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HEALTH	MICROBIAL PRODUCTS OR ACTIVITIES	DISEASE
Supply of nutrients & energy	• SCFA production, vitamin synthesis • Influences on energy supply, gut hormones, satiety, energy expenditure • Lipopolysaccharide, inflammation	Obesity & metabolic syndrome
Cancer prevention	• Butyrate production, phytochemical release • Toxins, carcinogens, inflammation	Cancer promotion
Inhibition of pathogens	• SCFA production, intestinal pH bacteriocins, competition for substrates and/ or binding sites • Toxin production, tissue invasion, inflammation	Source of pathogens
Normal gastrointestinal immune function	• Balance of proinflammatory versus anti-inflammatory signals, development • Inflammation, immune disorders	IBD
Normal gut motility	• Metabolites (SCFA, gases) from nondigestible carbohydrates	IBS (constipation, diarrhoea, bloating)
Cardiovascular health	• Lipid, cholesterol metabolism	Cardiovascular disease

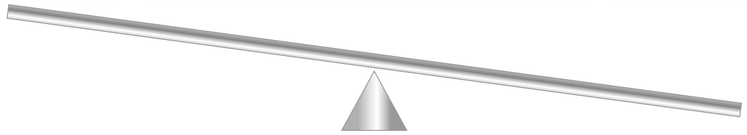

Figure 8 : Influence of gut microbial communities on health. Most of the microbial activities indicated in the centre column are functions of the whole community of gut microbiota rather than being attributable to a single species. The balance of the community & its output determines the net contribution to health or disease. Abbreviation: SCFA, short-chain fatty acid.[12]

DISEASES ASSOCIATED WITH GUT DYSBIOSIS

A microbial ecosystem in which bacteria no longer live in a mutualistic association is called dysbiotis. Gut microbiota dysbiosis is a condition related with the pathogenesis of intestinal illnesses and extra-intestinal illnesses.

Intestinal disorders include :

- Inflammatory bowel disease
- Irritable bowel syndrome
- Coeliac disease
- Colorectal cancer

Extra-intestinal disorders include :

- Allergy, Asthma
- Metabolic syndrome, Obesity, Type 2 diabetes, Cardiovascular disease
- Rheumatoid arthritis
- Parkinson's disease, Alzheimer's disease

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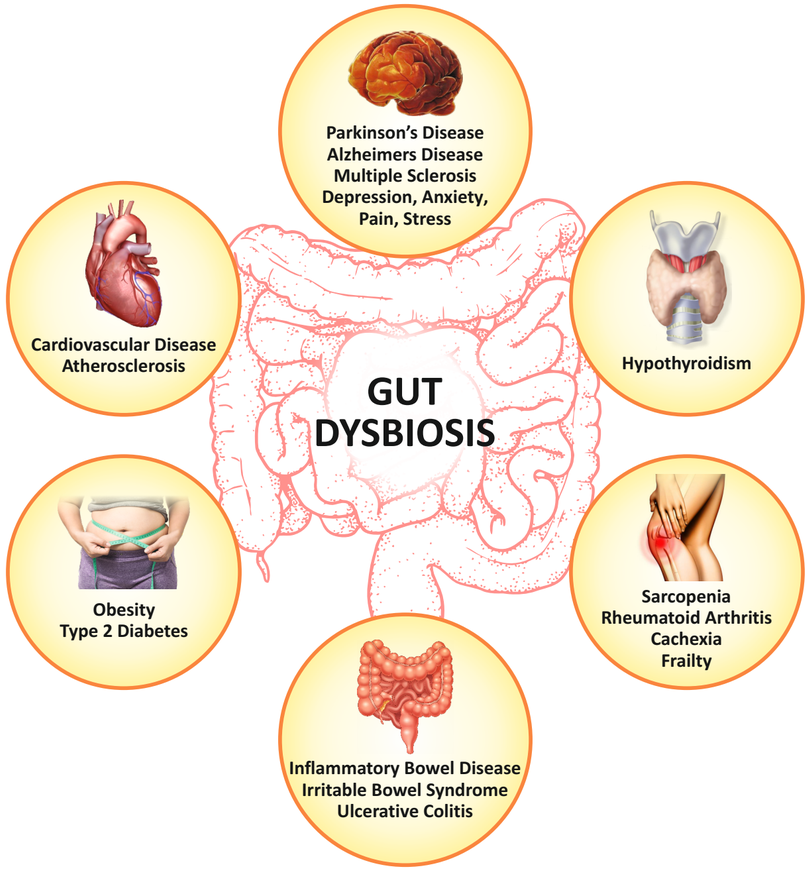

Figure 9 : Representative description of the metabolic diseases, gastrointestinal disorders, neuromusculoskeletal conditions, endocrine pathologies, neurodegenerative, and cardiovascular diseases associated with gut dysbiosis

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MICROBIAL ASSOCIATIONS WITH CHRONIC INTESTINAL DISEASES

As the volume of data relating to the composition and functional potential of the gut microbiota increases, the number of diseases that have been linked with alterations in our gut microbial community has also expanded. Indeed, the many instances of such potential associations are too great to summarize in this review and thus here the focus is on associations that have been the focus of greatest attention, that is, the possibility of a link between the gut microbiota and chronic GI diseases, including irritable bowel syndrome (IBS) and IBD, systemic diseases such as type 2 diabetes (T2D) and obesity, as well as the onset of colorectal cancer (CRC).[8]

Table 1 : Microbial associations with chronic intestinal diseases

Conditions	Microbial association*	
	Increased	Decreased
IBS	*Firmicutes : Bacteroidetes ratio* *- Ruminococcus* *- Dorea* *- Clostridium* *- Gammaproteobacteria (pIBS)* *- Haemophilus influenzae (pIBS)*	*Bifidobacterium* *Faecalibacterium* *Bacteroides*
IBD (incl. CD & UC)	- Increased bacterial numbers in mucosa (CD) - Gamma-proteobacteria - Enterobacteraceae - Adherent invasive escherichia coli (CD) - Clostridium spp.	Bacterial diversity Firmicutes Bacteroidetes Lachnospiracheae Clostridium leptum & coccoides group (Faecalibacterium prausnitzii) Roseburia Phascolarctobacterium
Colorectal Cancer	Increased : *Fusobacterium spp.* *E. coli (pks+)*	
Obesity	Increased : *Firmicutes:Bacteroidetes ratio‡* *Actinobacteria* *Bacteroides ‡* *Prevotellaceae*	Decreased : *bacterial diversity* *C. leptum group* *(Ruminococcus flavefaciens)* *Bifidobacterium* *Methanobrevibacter*
Type 2 Diabetes	Increased : Opportunistic pathogens *(Clostridium spp., E. coli, Eggerthella)*	Decreased: Butyrate-producing organisms

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FACTORS AFFECTING GUT MICROBIOME

The gut microbiome is influenced by multiple factors including mode of infant delivery and feeding, the aging process, diet composition, geography, medications, and stress (Figure 7).

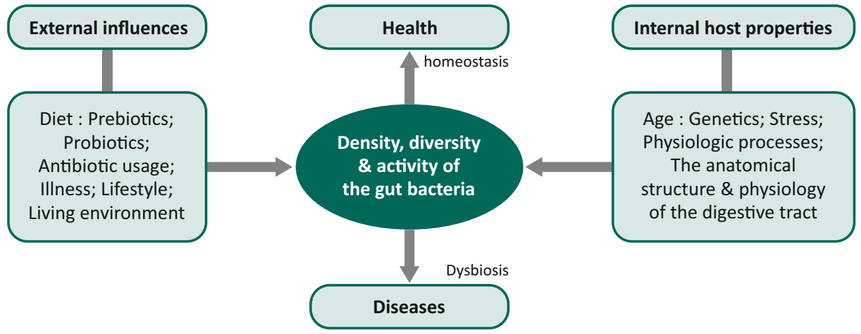

Figure 10 : Several factors influence the density, diversity & activity of the gut bacteria.5

The nature of a steady gut microbiota (including number and variety of microorganisms) is influenced by various factors including[10] :

- Mode of delivery at birth (Normal or Caesarean)
- Gender
- Medications
- Genetics
- Age
- Diet
- Lifestyle
- Supplements like prebiotics and probiotics
- Unwanted impacts like stress
- Geographical location

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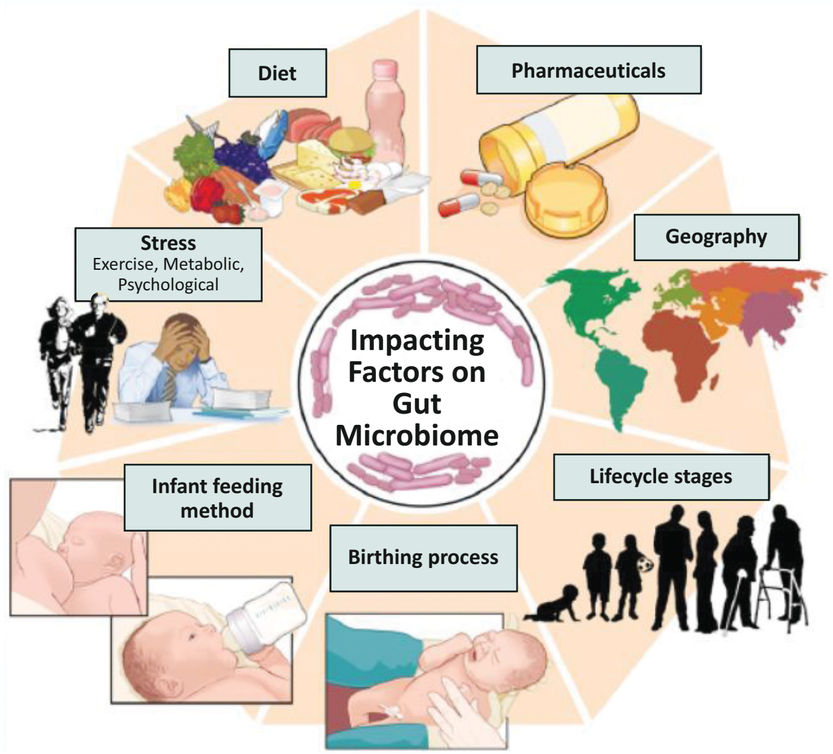

Figure 11 : Factors affecting gut microbiome[4]

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REBALANCE OF THE INTESTINAL ECOSYSTEM

Many therapeutic strategies have been developed to re-establish intestinal eubiosis and new strategies are constantly proposed and investigated. The main and at present best known and most adopted therapeutic strategies include[13] :

1. Probiotics : The administration of probiotic bacteria likely to displace potentially pathogenic bacteria and promote a rebalance of the microbial community.
2. Prebiotics : The administration of prebiotics (i.e., formulations of nutrients being preferentially or exclusively metabolized by probiotic bacteria) to favor the overgrowth of probiotic bacteria.
3. Synbiotics : The administration of probiotics and prebiotics combinations.
4. Diet Approach to Modulate Gut Microbiota (e.g. Mediterranean and Atlantic diets).
5. Other Therapies : More recent therapeutic approaches have been proposed including :
   a. Phage Therapy (viruses that infect bacteria)
   b. Fecal Microbiota Transplantation
   c. Bacterial Consortium Transplantation

All of these strategies share the same goal of replacing harmful microbes with more favorable ones to restore eubiosis.[13]

PROBIOTICS

The word probiotic originates from the Greek word “pro-bios,” which means “for life.”

In 1908, Noble Prize winner Eli Metchnikoff suggested that the long life of Bulgarian peasants resulted from their consumption of fermented milk products. He believed that “lactic acid bacteria” replaced harmful organisms found in intestines and thus reduced production of toxins that lead to diseases. The term “probiotic” was first used in 1965, by Lilly and Stillwell for describing substances secreted by one organism which stimulate the growth of another. Marteau et al, in 2002 defined them as “microbial preparations or components of microbial cells that have a beneficial effect on health and well-being”.[14]

The FAO/WHO definition of a probiotic - “live microorganisms which when administered in adequate amounts confer a health benefit on the host” - was reinforced as relevant and sufficiently accommodating for current and anticipated applications. Since then, this definition has become the most widely adopted and accepted version worldwide.[15]

Lactobacillus, Bifidobacterium, Streptococcus, Saccharomyces, Bacillus and Enterococcus are the most popular probiotics genus sources and their application as probiotics are extensive in fermented food products, non-fermented food items, as well as functional and nutraceutical dietary supplements.[15]

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PROBIOTICS' EFFICIENCY

The probiotics' efficiency may be compromised by several factors : the major challenges

1. An adequate delivery system
2. Food processing and storage conditions (water activity, temperature, pH, oxidation, osmotic pressure, etc.)
3. Survival through the hostile gastrointestinal tract (GIT) environment (acidic pH, bile salts, digestive enzymes, etc.)
4. Ability to colonize the gut

However, microencapsulation seems to be a highly productive and holistic approach to overcome this problem.[16]

PROBIOTICS : RATIONALE OF USE IN MODERN ERA

The use of antibiotics, immunosuppressive therapy and irradiation, amongst other means of treatment, may cause alterations in the composition and have an effect on the GIT flora. Introduction of antibiotic revolutionized field of medicines and increased life expectancy and greatly improved quality of life. However, major drawback with antibiotics was that besides killing bad bacteria it also kills good bacteria and hence it disturbs the ecosystem of the body, causing devastating effects on body like superinfection/drug resistance. Normal microflora is also disturbed in infectious conditions of the gastrointestinal tract and also when there is inflammation of the gastrointestinal tract.[17]

Therefore, the introduction of beneficial bacterial species (Probiotics) to GI tract may be a very attractive option to re-establish the microbial equilibrium and prevent disease. “Probiotic,” selectively removes only the pathogen while leaving the remainder of the ecosystem intact.[17]

CRITERIA OF AN IDEAL MICROORGANISM AS PROBIOTIC

In general, probiotic bacteria need to fulfil several ideal criteria in order to elicit their beneficial effects, summarized is summarised as below [Figure 10] :

- Able to survive the passage through the hostile digestive system (acids and alkali tolerant).
- Able to attach to the intestinal epithelia and colonize.
- Able to maintain good viability.
- Able to utilize the nutrients and substrates in a normal diet.
- Primarily of host (human) origin, non-pathogenic and non-toxic.
- Capable of exerting a beneficial effect on the host.

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- Stability of desired characteristics during processing, storage and transportation.
- Anti-inflammatory, anti-mutagenic, immunostimulatory
- They should be able to interact or to send signals to the immune cells associated with the gut.
- Resistance to processing, transportation
- Short generation time in intestine[18]

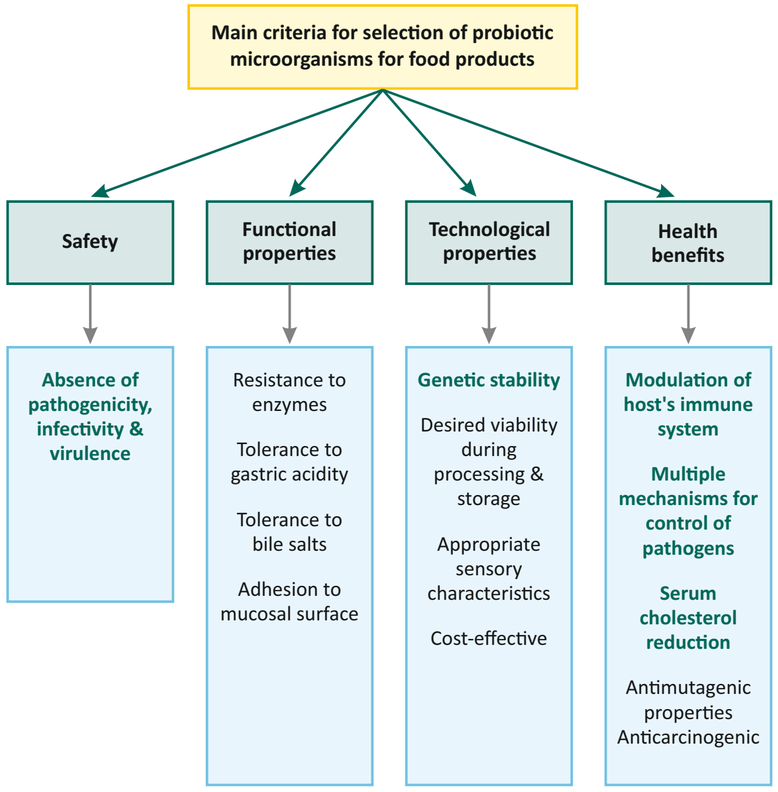

Figure 12 : Desirable criteria for the selection of probiotics in commercial applications[19]

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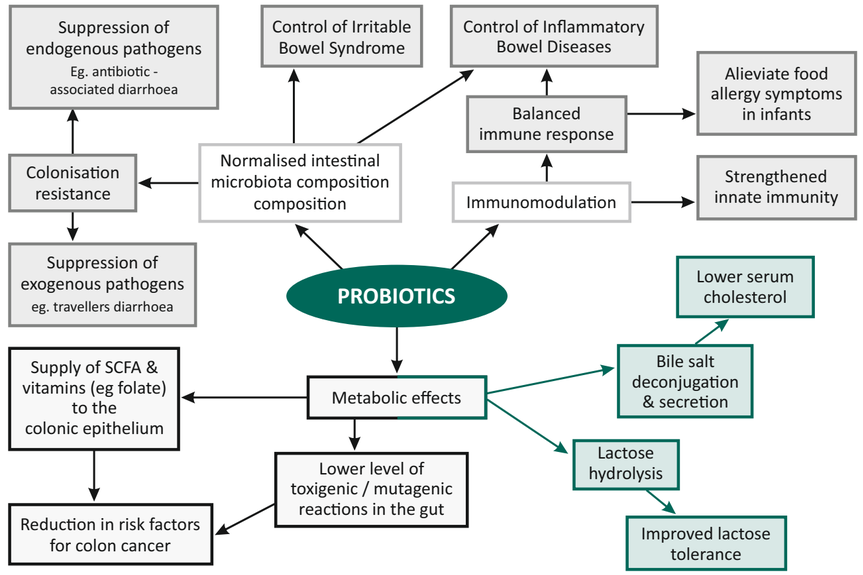
PROPOSED HEALTH BENEFITS



Health Benefits of Probiotic

1. Replenishing the depleted gut microflora, which may have occurred due to use of antibiotics, illness, stress, travel or lifestyle changes.
2. Improving the properties of the indigenous microflora.
3. Offers increased resistance to establishment of infection by potentially pathogenic organisms in the intestine.
4. Decrease the duration of diarrhea (antibiotic associated, travelers', infective).
5. Use in lactose intolerance (promotion of intestinal lactose digestion).
6. Increase nutritional value (better digestibility, increased absorption of vitamins & minerals).
7. Regulation of gut motility (constipation, irritable bowel syndrome).
8. Maintenance of mucosal integrity of the intestine.
9. Reduction in serum cholesterol concentration.
10. Reduction in allergy.
11. Prevention of colon cancer.
12. Reduction in carcinogen /co-carcinogen production.[20]

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PROBIOTIC : MECHANISMS OF ACTION

Major probiotic mechanisms of action include

1. **Enhancement of the epithelial barrier**

   Consumption of non-pathogenic bacteria can contribute to intestinal barrier function, and probiotic bacteria have been extensively studied for their involvement in the maintenance of this barrier. Several studies have indicated that enhancing the expression of genes involved in tight junction signaling is a possible mechanism to reinforce intestinal barrier integrity. **Recent data have indicated that probiotics may initiate repair of the barrier function after damage.**

2. **Increased adhesion to intestinal mucosa**

   The gut microflora competes directly with gut pathogenic organisms for epithelial attachment sites in the gastrointestinal tract, thereby preventing attachment and colonization of the GI tract by the potentially pathogenic organisms.

3. **Concomitant inhibition of pathogen adhesion**

4. **Competitive exclusion of pathogenic microorganisms**

   The gut microflora competes directly with gut pathogenic organisms for the essential nutrients necessary for survival and multiplication, thereby inhibiting the growth and multiplication of potentially pathogenic organisms.

5. **Production of anti-microorganism substances**

   Bacteria of the normal gut flora produce a variety of substances that are inhibitory to both gram - positive and gram - negative bacteria. They produce antimicrobial compounds (bacteriocins), volatile fatty acids, organic acids, and lactic acid, which reduce the intestinal pH. These compounds reduce the number of viable pathogenic organisms in the gastrointestinal tract.

6. **Modulation / stimulation of the immune system**

   The underlying mechanisms of immune stimulation by the gut microflora are not well understood. However, local gut immunity enhancement by the gut microflora may be one possible mechanism of inhibiting growth of potentially pathogenic microorganisms.

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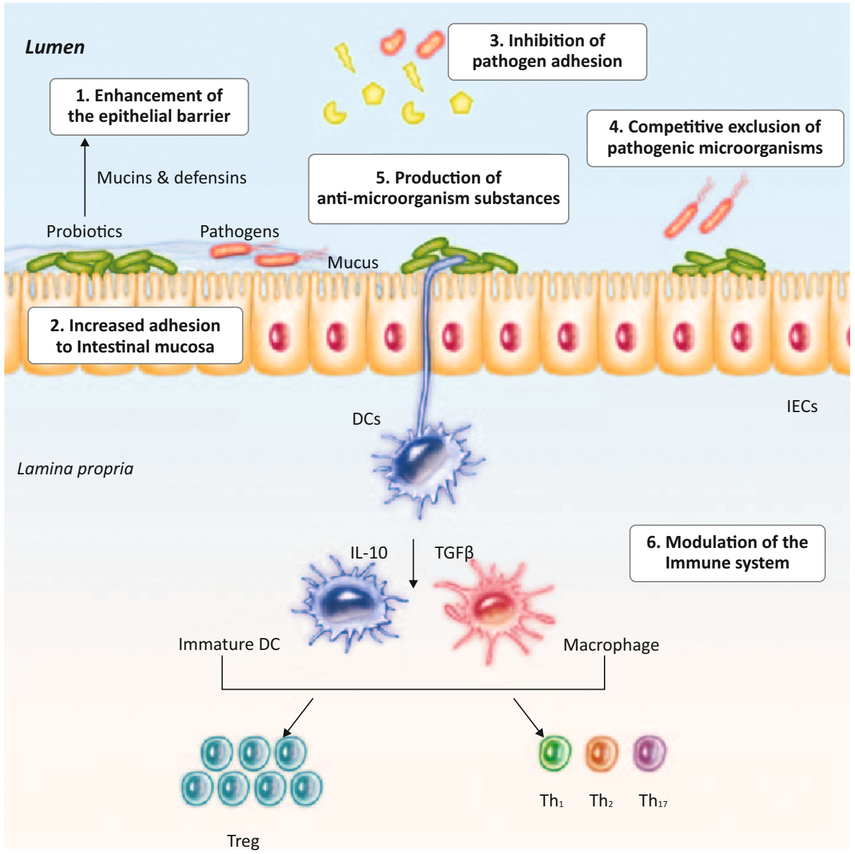

Abbreviations: •DCs - Dendritic Cells •IECs - Intestinal Epithelial Cells •IL-10 - Interleukin 10 •TGF-β - Transforming Growth Factor-β •Treg - T regulatory cells •Th1 - T-helper type 1 •Th2- T-helper type 2 •Th17- T-helper type 17

Figure 13 : Major mechanisms of action of probiotics

Florimax

Florimax is live, lyophilized, Probiotic cultures for oral administration. It is a high potency probiotic combination containing 8 strains of live multispecies and multistrain bacteria.

Florimax contains the Original De Simone Formulation of Probiotic.[21] After January 2016, VSL#3® sellers were ordered by a U.S. court to stop any claims that state or suggest a false continuity between the new formulation sold as VSL#3® and the Original De Simone Formulation.

The De Simone Formulation in Florimax has been supported with over 70 published clinical trials in human subjects, with extensive clinical research in the dietary management of dysbiosis associated with Irritable Bowel Syndrome (IBS), Ulcerative Colitis (UC), Antibiotic Associated Diarrhea (AAD), Pouchitis and Hepatic Encephalopathy (HE).[21]

Florimax is a medical food intended for use under the supervision of a healthcare provider.

COMPOSITION :

Florimax Capsules : Each capsule contains at least 112.5 billion (112.5×10^9) colony forming units (CFUs). **Florimax** contains the original “De Simone Formulation,” a proprietary blend of the following live, lyophilized, probiotic bacteria :

1. Lactobacillus acidophilus DSM24735/SD5212
2. Lactobacillus plantarum DSM24730/SD5209
3. Lactobacillus paracasei DSM24733/SD5218
4. Lactobacillus delbrueckii subsp. bulgaricus† DSM24734/SD5210
5. Bifidobacterium longum± DSM24736/SD5220
6. Bifidobacterium breve DSM24732/SD5206
7. Bifidobacterium infantis± DSM24737/SD5219
8. Streptococcus thermophilus DSM24731/SD5207

Florimax Capsules is a medical food as defined by the Orphan Drug Act and additional FDA regulations.[21]

±Reclassified as Bifidobacterium lactis

†Reclassified as Lactobacillus helveticus

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CHARACTERISTICS OF INDIVIDUAL PROBIOTIC BACTERIA

Probiotic Bacteria (Lactobacillus acidophilus)[62,63,64,65]

1. Lactobacillus acidophilus is a Gram positive, non-spore forming, homo fermentative, catalase-negative rod. It is a common inhabitant of the human intestinal tract, the human mouth and vagina. It is also found in some traditional fermented milks (e.g. curd) and is today widely used in probiotic foods and supplements.
2. **High tolerance to hostile gastrointestinal conditions**

   Excellent Acid and Bile tolerance: extremely resistant to low pH conditions and survives the presence of bile at concentrations present in the duodenum. Excellent Pepsin and Pancreatin resistance.
3. **Strong Adhesion to Intestinal Mucosa :**

   L. acidophilus ability to adhere to different human epithelial cell lines has been confirmed in several studies. It shows very good adherence to cultured intestinal cells (HT-29 and/or Caco-2 cells).

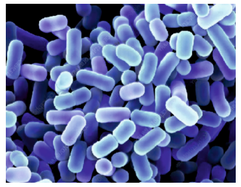
   In addition, the adhesion of lactobacilli to the mucosa can prevent other pathogenic bacteria from adhering, promoting their elimination.
4. **Inhibition of Pathogens :**

   Lactobacillus acidophilus inhibits the growth of several Gram-positive and Gram-negative bacteria by producing lactic acid, acetic acid, hydrogen peroxide, bacteriocins and possibly biosurfactants.

   Antimicrobial substance production: The ability of L. acidophilus to produce antimicrobial substances was confirmed by the genome sequence of the strain. L. acidophilus was found to produce a Bacteriocin, designated lactacin B.
5. **Beneficial modulation of immune functions :**

   May influence immune regulation, as demonstrated by the induction of IL-12 and moderate induction of TNF in vitro.
6. **Long history of safe human consumption**

Lactobacillus plantarum[69,70,71,72,73]



1. Lactobacillus plantarum (L. plantarum) is a rod-shaped, gram-positive lactic acid bacterium. Lactobacillus plantarum is a widely distributed and versatile lactic acid bacterium. It represents part of the microbiota of many foods and feeds, including dairy, meat, fish, vegetable fermented products (e.g., pickled vegetables, sourdoughs), and silage; it is also a natural inhabitant of the human and animal mucosa (oral cavity, gastrointestinal tract, vagina, etc.).
2. L. plantarum is one of the most 'hardy' probiotic bacteria, thanks to its ability to withstand a huge variety of different temperatures. In fact, it can survive in any environment between 1-60 degrees Celsius. It can also adapt to a wide scale of atmospheric pressures. It can grow at temperatures between 15-45^{0}C and at pH levels as low as 3.2.
3. **Survival and adhesion in hostile GI tract**

   The scientific validity of Lb. plantarum strain as probiotics was first evaluated by characterizing bile and acid resistance (safeguard) in the intestinal tracts of animal and human hosts.

   Moreover, Lb. plantarum helped reduce overall symptoms of burden of infection of GI tracts.

   It is believed that adherent probiotic (Lb. plantarum) has beneficial health effects, especially connected to the inhibition of pathogen adhesion to intestinal cell lines.
4. **Health benefits**

   In addition, Lb. plantarum strain is recognized as natural probiotic of the human GI tract and
   • Can decrease intestinal heavy metal absorption • Reduce metal accumulation in tissues
   • Alleviate hepatic oxidative stress
5. **Antifungal Effects**

   Antifungal activity of Lb. plantarum strains has been exhibited to be due to presence of phenyllactic acid (PLA), cyclic dipeptides, fatty acids, and organic acids.
6. **Immune Response**

   Several reports suggested that the supplementation of probiotics (Lb. plantarum strain) can improve the growth, disease resistance, and immune response. Considering the tolerance-inducing immunomodulatory effects of Lb. plantarum, it is of interest to search the opportunity of using allergen expressing Lactobacillus as delivery vehicle in immunotherapy or an allergy vaccine.
7. **Repair GIT**

   Another major benefit of L. plantarum is its ability to help repair the intestinal lining. It does this by using mannose-specific adhesions, which allow it to compete with both gram- positive and gram-negative pathogenic strains for receptor sites.

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It also provides valuable nutrients in the mucosal membrane. This means that L. plantarum is able to secrete anti-microbial substances that help to stop pathogenic colonies from forming in the gut.

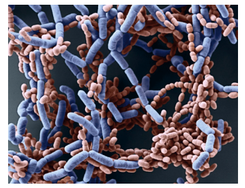
8. **Antimicrobial activity**

   • Lb. plantarum which is a probiotic strain from the traditional fermented dairy products and identified to produce bacteriocin opposing to Gram-negative and Gram-positive bacteria.

   • Lb. plantarum strains are chief factors/components in a variety of fermentation processes whereby their fructification of organic acids, hydrogen peroxide (H2O2), diacetyl, and other antimicrobial components increased the safety and quality of fermented foods.

   • Lactic Acid is the major organic acid produced by Lb. plantarum strain. Other organic acids produced are acetic acid, propionic acid, phenyllactic acid (PLA), formic acid, and succinic acid. The approach of action of organic acids is the reduction of pH in the environment, causing inhibition of several pathogenic microorganisms.

Lactobacillus paracasei[66,67,68]



1. Lactobacillus paracasei is a Gram-positive, non-spore forming, homo fermentative rod that is a common inhabitant of the human intestinal tract. It is found in the human intestinal tract and mouth, but also in foods such as yogurt and naturally fermented vegetables and milk.
2. **Established Safety :**

   L. paracasei is listed in the Inventory of Microorganisms with Documented History of Use in Human Food. The European Food Safety Authority has also included the species on its Qualified Presumption of Safety list. In addition to a long history of safe human consumption of the species, no acquired antibiotic resistance was detected in L. paracasei during screening by the EU-funded PROSAFE project.
3. **Resistance to acid and bile :**

   High tolerance to gastrointestinal conditions (acid and bile)

   In vitro studies have shown that L. paracasei is very resistant to low pH conditions (acidic) and shows moderate resistance to bile at the concentrations present in the duodenum.
4. **High survival rate during manufacture and storage**
5. **Restore the intestinal epithelial barrier function :**

   Lactobacillus paracasei is able to reverse dysfunctions of intestinal epithelial barrier through upregulation of anti-inflammatory cytokines, including interleukin (IL)-10 and tumor growth factor (TGF)-β.

6. **Adhesion to intestinal mucosa :**

   L. paracasei has demonstrated excellent adhesion to human epithelial cell lines (Caco-2) applied in in vitro studies.

7. **Lactic Acid Production :**

   Lactic acid is the most important metabolic end product of fermentation processes by lactic acid bacteria and other microorganisms. L. paracasei only produces L (+) lactic acid.

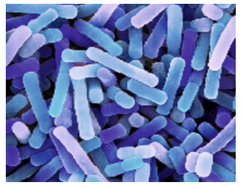
8. **Beneficial modulation of immune functions :**

   L. paracasei may have an influence on immune regulation, as demonstrated through induction of IL-12 in vitro.

9. **Health benefits :** Potential mechanisms include

   • Production of antimicrobial substances such as bacteriocins
   • Enhancing the epithelial barrier through attachment
   • Competition for pathogenic binding sites
   • Modulation of the immune system

Lactobacillus delbrueckii subsp. Bulgaricus[74,75]



1. Lactobacillus delbrueckii subsp. bulgaricus, a Gram-positive, rod-shaped, non-motile and non-spore-forming lactic acid bacterium (LAB), is one of the most common starter bacterial species in industrial fermentation of dairy products. Globally, almost all types of yogurt and a considerable proportion of other types of dairy products contain this bacterium.

2. **Hydrogen peroxide Production :**

   Hydrogen peroxide is another antagonistic metabolite produced by L. bulgaricus in the presence of air. The antimicrobial action of hydrogen peroxide has been attributed to its ability to produce toxic compounds, such as the superoxide anion and other free radicals.

3. **Antimicrobial Substances** (Bacteriocins, Bulgaricins, Bulgarican)

   Bacteriocins produced by L. bulgaricus have antimicrobial activity. Inhibitory activity against Candida albicans, Clostridium difficile, Helicobacter pylori, L. monocytogenes, Salmonella spp., Staphylococcus aureus, and Streptococcus agalactiae has been attributed to some bulgaricins (bacteriocins produced by L. bulgaricus). Bulgarican (not bacteriocin in nature), which showed maximum activity and stability at pH 2.2 and was not affected by autoclaving at 1200C for 1 h, was inhibitory toward both Gram-positive and Gram-negative bacteria, but it had no apparent antifungal activity. Inhibitory compounds against Staphylococcus and Clostridium have been

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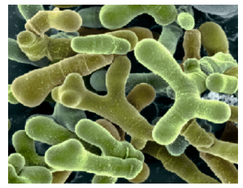
found, which were insensitive to proteolytic enzymes, resistant to heat and active over a wide range of pH.

4. **Lactic acid production :** Lactic acid production during fermentation lowers the pH and creates an environment that is unfavorable to pathogens and spoilage organisms.

In addition, the low pH potentiates the antimicrobial effects of organic acids, which show greater lethality to bacteria than the inorganic acids

Bifidobacterium Lactis - Bifidobacterium longum[76]



1. Bifidobacterium sp. comprises Gram positive, non-spore forming, anaerobic, pleomorphic bacilli that are dominant microbial residents of the colonic microbiota. B. lactis is of human origin and has been shown to grow well in milk.

2. Long history of safe human consumption: Bifidobacterium sp. has long been considered safe and suitable for human consumption with several published studies addressing its safety. Furthermore, Bifidobacterium lactis is listed in the Inventory of Microorganisms with Documented History of Use in Human Food. The European Food Safety Authority has also added the species to the Qualified Presumption of Safety list. No harmful metabolic or toxigenic activities are associated with B. lactis. In addition to its long history of safe human consumption of the species, no acquired antibiotic resistance was detected in B. lactis during screening by the EU-funded PROSAFE project.

3. Resistance to acid and bile

   In vitro studies have shown that B. lactis is extremely resistant to low pH conditions and survive the presence of bile at concentrations present in the duodenum.

4. Adhesion to intestinal mucosa

   B.lactis has demonstrated very good adhesion to human epithelial cell lines (Caco-2) applied in vitro studies

5. Beneficial modulation of immune functions

   May improve specific immune response, as demonstrated in a human clinical study. May influence immune regulation as demonstrated by induction of IL-10 in vitro. Proven in an animal model to protect against inflammation and balance the intestinal mucosal immune response.

6. Lactic acid production

   Lactic acid is the most important metabolic end product of fermentation processes by lactic acid bacteria and other microorganisms. B. lactis only produces L (+) lactic acid.

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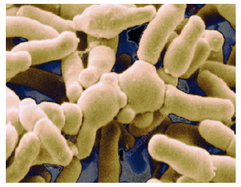
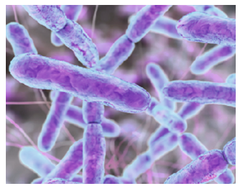
Bifidobacterium breve[77,78]



1. Bifidobacterium breve are anaerobic, rod-shaped, gram-positive bacterium that lack cell motility, sporulation, and a cell capsule. Although B. breve have been found within the gut flora of fully grown adult humans, they are found in much higher quantities within the infant gut and were first isolated from breast-fed infant feces in 1990. B. breve have unique metabolic capabilities that underlie their importance as a dominant commensal bacterium within the gut. B. breve, in particular, plays a key role in human infant metabolism of breast-milk, which contains Human Milk Oligosaccharides (HMO) indigestible by the human host.
2. Strong antimicrobial activity against pathogens
3. Stimulation of mitochondrial dehydrogenase activity of macrophages
4. Inhibition of the growth of E. coli biotypes in in vitro study
5. Immunomodulation

   Reduction of pro-inflammatory TNF-α in blood samples of celiac children Immunomodulation activity by increasing TGF-β1 in preterm infants B. breve possessed a cell surface exopolysaccharide (EPS) able to play an important role in immunomodulation in B cell response.
6. Improvement of glucose metabolism and weight management in obese children
7. Safety : B. breve is included in the list of Qualified Presumption of Safety (QPS) biological agents

Bifidobacterium infantis[76]



1. Bifidobacterium infantis a Gram-positive, anaerobic, branched rod-shaped bacterium. In the intestines, they ferment sugars to produce lactic acid. Bifidobacterium infantis, a gut bacterium uniquely adapted to metabolizing breastmilk carbohydrates. Bifidobacterium longum subspecies infantis is a prominent early colonizer of the infant gut that consumes human milk oligosaccharides (HMOs).
2. Safety

   In 2007, the European Food Safety Authority (EFSA) assigned qualified presumption of safety (QPS) status to the bacterial species B. longum, which includes subspecies infantis, indicating that this taxonomic group does not raise any safety concerns.

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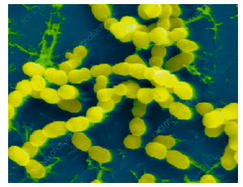
3. **Proposed Mechanisms of Action of B. infantis Based Studies :**

   - B. infantis is highly specialized for the consumption of human milk oligosaccharides (HMO) and has a competitive advantage against other bacteria, allowing increased colonization and resulting in fewer luminal pathogens.
   - **Immune function :**

     B. infantis produces exogenous substances that promote maturation of the immature innate immune response.
   - **Reduces inflammation :**

     HMO “turn on” the repertoire of genes in B. infantis that are important in controlling inflammation within the infant gut. B. infantis improves the intestinal barrier integrity through the production of tryptophan metabolite, indole-3-lactic acid.
   - B. infantis becomes dominant in the gut and reduces pH by its unique ability to metabolize all HMO into acidic end products, lactate, and acetate.

Streptococcus thermophilus[79,80,81]



1. S. thermophilus is a Gram-positive bacterium. S. thermophilus is the only Streptococcus species used in food industry. Because it has been consumed by humans for centuries without giving any disease, it is also the only Streptococcus species to be recognized as a Generally Recognized as Safe bacterium by FDA.
2. **GRAS status :**

   Due to its safe use in food production over the years, S. thermophilus was granted 'Generally Recognized as Safe' (GRAS) status in the USA and the 'Qualified Presumption of Safety' (QPS) status in the European Union
3. **Survival in the digestive environment :**

   Studies showed that S. thermophilus can survive the passage through the human GI tract and be found alive in the feces of people after consumption.

   Resistance Mechanisms of S. thermophilus to acid and bile are composed of a large combination of numerous different mechanisms.
4. **Adhesion to intestinal epithelial cells :**

   S. thermophilus have the ability to adhere to intestinal epithelial cells, which is an important criterion for probiotic strain selection. This biological mechanism promoting interactions with the host leads to gut protection and enhances colonization.

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5. **Beneficial health effects of S. thermophilus**
   • Alleviation of lactose intolerance • Prevention of chronic gastritis • Prevention of diarrhea
6. **Antioxidant activity :**
   These studies indicate that some S. thermophilus strains may have a beneficial antioxidant effect both in vitro and in vivo in animals.
   • Inhibition of lipid peroxidation in liposomes treated by ferrous iron
   • Scavenger of reactive oxygen species or free radicals or may increase antioxidant capacities of intestinal content
7. **Immunomodulation**
   A number of in vitro studies have investigated the ability of S. thermophilus strains to modulate the immune response of various human cell lines, such as intestinal HT-29 cells, Peripheral Blood Mononuclear Cells (PBMCs), monocyte-derived Dendritic Cells (moDCs) or human primary macrophages.
8. **Enhances the intestinal barrier function**
   • S. thermophilus prevents decrease in trans-epithelial resistance induced by entero-invasive Escherichia coli (EIEC).
   • The effect of these bacteria on resistance was found to be accompanied by maintenance (actin) or enhancement (actinin, occluding) of cytoskeletal and tight junctional protein phosphorylation. S. thermophilus also enhanced the barrier function of naive epithelial cells not exposed to any pathogen.
   • Pretreatment with the two strains limited the number of adhered and invasive EIEC. The ability of S. thermophilus to improve intestinal barrier function was confirmed by an in vivo study. Supplementation with S. thermophilus significantly decrease intestinal permeability, both in the small bowel and in the colon of the volunteers.
9. **Possible mechanisms of action of S. thermophilus on the host**
   Health
   • Antimicrobial activity : S. thermophilus is able to synthesize thermophilins which are bacteriocins, small peptides able to inhibit the growth or kill closely related bacteria.
   • Thermophilins have been shown to have in vitro inhibitory activities against LAB but also against Gram positive pathogenic strains such as E. faecalis, Clostridium ©.) botulinum, Staphylococcus aureus and Listeria monocytogenes
   • Thermophilin 1277, which is produced by S. thermophilus SBT1277, shows an antimicrobial activity against several LAB and food spoilage bacteria including C. butyricum, C. sporogenes and Bacillus cereus.
   • Also produces lactic acid, another antimicrobial compound, but this trait, shared by all LAB, is not specific to this bacterium.

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FLORIMAX : MEDICAL FOOD

The Orphan Drug Act of 1988 defines “medical food” as “a food which is formulated to be consumed or administered enterally under the supervision of a physician and which is intended for the specific dietary management of a disease or condition for which distinctive nutritional requirements, based on recognized scientific principles, are established by medical evaluation” 21 U.S.C. 360ee(b)[3]. FDA regulations 21 C.F.R. 101.9(j)[8] set forth additional criteria for makers of medical food products.[21]

Florimax is a medical food as defined by the Orphan Drug Act and additional FDA regulations. **Florimax is specially formulated and processed to provide a precise mixture of certain bacterial species to the gastrointestinal tract.** The gastrointestinal microbiota, or “microbiome”, is important for the normal functioning of the human gastrointestinal tract. Healthy gut microbes compete with pathogens for nutrients and adhesion sites on the gut mucosa. **The gut microbiome also plays a critical role in human digestion with key probiotic bacteria being essential for the fermentation of non-digestible fibers. Fermentation of non-digestible fibers results in the production of short chain fatty acids such as butyrate which is a critical energy source for human colonic cells.[21]**

Additionally, the gut microbiome plays a role in modulating intestinal inflammation, improving gut barrier function. Patients who experience irritable bowel syndrome (IBS), ulcerative colitis (UC), and pouchitis have documented dysbiosis associated with limited bacterial diversity and deficiencies in luminal concentrations of lactobacilli and bifidobacteria compared with healthy individuals. Likewise, the consumption of antibiotics can have a dramatic short- and long-term impact on the healthy gut microbiome with reductions in bacterial evenness and diversity at the both the phylum and species levels. And finally, the gut microbiome of patients with hepatic encephalopathy (HE) has also been shown to be significantly altered compared to healthy controls with significant increase in ammonia producing bacterium present and other differences in bacterial species compared to controls.[21]

Patients with dysbiosis associated with these gastrointestinal and liver disorders have distinct nutritional requirements that differ from the general population and which requires the consumption of high levels of probiotic bacteria to maintain an adequate and balanced microbiota. While alterations in the normal diet can impact the microbiome, **there is insufficient evidence to suggest that, in these patient groups, adjustment of the microbiota can be achieved through modification of the normal diet alone. Florimax** is intended for those who are receiving active and ongoing medical supervision with regular instruction on the use of medical foods.[21]

INTENDED USE :

Florimax is a probiotic medical food intended for the dietary management of dysbiosis associated with :

• Irritable Bowel Syndrome (IBS) • Ulcerative Colitis • Antibiotic-Associated Diarrhea
• Hepatic Encephalopathy • Pouchitis

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Florimax is a non-drug therapy that addresses distinct nutritional requirements to promote microbial balance in individuals which cannot be addressed by modification of the diet alone. **Florimax** is intended to be used under the supervision of a physician.

Florimax is intended for the clinical dietary management of patients who, because of therapeutic or chronic medical needs, have special medically-determined nutrient requirements; the dietary management of which cannot be achieved by the modification of the normal diet alone.

Florimax, as a medical food, must be used under physician supervision.

DOSAGE & ADMINISTRATION

For oral administration

Consume 2- 8 **Florimax Capsules** daily (As directed by Physician)

Florimax Capsules can be consumed directly. Adjustment of the intestinal flora can take a few days or weeks; it may take up to one month for the colonization of the gut to become optimally stable if consumed on a regular basis.

Adult Administration

For the Dietary Management of :	Capsules Per Day*
Irritable Bowel Syndrome	2-4 capsules
Antibiotic-Associated Diarrhea	4-8 capsules during the course of antibiotic treatment and 1-2 capsules for 10 days following antibiotic usage
Hepatic Encephalopathy	4-8 capsules
Ulcerative Colitis (maintenance)	4-8 capsules

*Each Capsule Contains 112.5 Billion Live Bacteria (Colony Forming Units-CFUs

Pediatric Administration

Recommended Pediatric daily intake for **Florimax Capsules.**
Florimax Capsules can be pulled apart and sprinkled on apple sauce, yogurt or any cold food.

For the Dietary Management of :	Less than 2 Years	2-5 Years	6-11 Years	12-17 Years
Irritable Bowel Syndrome (IBS)	1 capsule	2 capsules	2 capsules	2-4 capsules
Ulcerative Colitis (maintenance)	1 capsule	2 capsules	2-4 capsules	4-8 capsules

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CONTRAINDICATIONS

Florimax Capsules should not be used in premature infants in the Neonatal Intensive Care Unit (NICU) setting.

WARNINGS AND PRECAUTIONS

Florimax may contain trace amounts of lactose (less than 0.1 g per 100 g) and dehydrated skim milk or milk protein (casein and beta-lacto globulin of less than 2 mg/kg). The bacteria may be fermented using some dairy ingredients to ensure their health and vitality.

ADVERSE REACTIONS

Mild abdominal bloating has been reported occasionally during the first few days of consuming **Florimax.**

This is generally a readjustment of the microbiota, which usually diminishes within 3 - 4 days. If bloating persists, the patient should reduce their intake for a few days and consult with their healthcare provider.

DRUG INTERACTIONS

There are no known adverse drug interactions associated with consumption of **Florimax.** Some strains of bacteria in **Florimax** may be inactivated by certain antibiotics. Do not consume **Florimax** within four (4) hours before or after taking antibiotics.

USE IN SPECIFIC POPULATIONS

The probiotic formulation in **Florimax** has been the subject of studies in adults, children, and infants. If you are pregnant or nursing, please consult with your healthcare provider before consuming **Florimax.**

SAFETY AND OVERDOSAGE

- Probiotics have a long history of safe use, having been consumed for health benefit and as part of fermented foods for millennia. Many bifidobacteria and lactobacilli species are normal, non-pathogenic inhabitants of the human gastrointestinal tract, oral cavity, skin, and vagina.
- Documented cases of infection attributable to probiotic intake are limited to individual case reports, primarily associated with the use of probiotics in severely immunocompromised patients. **Florimax** has been the subject of clinical trials in ART treated HIV-1 positive patients.
- The probiotic bacteria in **Florimax** are non-pathogenic, non-toxigenic and Generally Recognized as Safe (GRAS) as food ingredients.
- The De Simone Formulation in **Florimax** has been the subject of over 75 clinical studies involving over 5,000 adults, children, and infants - including immunocompromised individuals.
- The most common reported adverse events are abdominal bloating and/or gas, generally reported within the first few days of probiotic consumption.
- **Florimax** has been administered in clinical evaluation in daily dosages of up to 3,600 billion ($3,600 \times 10^9$) CFUs per day for 12 weeks.

GUT MICROBIOTA FOR MANAGEMENT OF CHRONIC LIVER DISEASE (CLD)

The common end stage of liver disease progression regardless of etiology is cirrhosis, which can result in decompensation and development of hepatocellular cancer. Changes in the gut microbiota composition and function have a critical relationship with liver health from precirrhotic stages to cirrhosis, decompensation, and requirement for liver transplantation. Alterations in the gut-liver axis have far-reaching consequences pertaining to the occurrence, progression, prognostication, and treatment of the major liver diseases.

Pre-Cirrhotic Changes in Microbiota and Relationship to Etiology

There is usually a prolong pre-cirrhotic period during which liver inflammation and fibrosis progress with alteration in the gut-liver axis with continued exposure to the etiology. In general, progression in pre-cirrhosis stages involves a reduction in overall diversity, reduction in phyla with predominantly beneficial bacteria (Firmicutes), and increase in phyla Bacteroidetes and Proteobacteria that tend to contain pathobionts.[60]

- In viral hepatitis (hepatitis B and hepatitis C), the stool microbiome shows a loss of diversity and increase in potential pathobionts, such as Enterobacteriaceae and others, such as Bacteroides, well before cirrhosis.
- In alcoholic hepatitis (AH), that is, before CLD onset, use of alcohol results in significant dysbiosis with increased intestinal permeability that is variably reversed after successful alcohol cessation. With continued alcohol misuse, there is further reduction in diversity and an increase in relative abundances of pathogenic bacteria, such as Enterobacteriaceae and Enterococcaceae.
- Finally, in cholestatic liver diseases, studies have also consistently shown an overall lower diversity in microbiota.
- Primary biliary cholangitis showed a distinct pattern with 8 genera (Haemophilus, Veillonella, Clostridium, Lactobacillus, Streptococcus, Pseudomonas, Klebsiella, and an unknown genus in the family of Enterobacteriaceae) being significantly increased in primary biliary cholangitis, while primary sclerosing cholangitis patients tended to have increased pathobionts (Veillonella).[60]

Role of Microbiota in Progression of Liver Diseases

The role of liver disease etiology vs microbiota in causation and progression of disease is important to consider. The preponderance of evidence points towards a noncausal but contributory role of the microbiome and the current accepted hypothesis is that the human intestinal microbiome, when stressed by various disease processes, undergoes dysbiosis, which accelerates liver fibrosis/cirrhosis development through up-regulation of inflammation and subsequently advanced fibrosis/cirrhosis that contributes to its complications, such as hepatic encephalopathy (HE). The data to support this narrative are relatively robust in ALD and NAFLD. For example, alcohol consumption results in direct toxicity to the liver via dysbiosis mediated through small intestinal bacterial overgrowth/large intestinal bacterial overgrowth or direct microbial toxicity, and by direct local injury to the intestinal barrier with resultant increased bacterial translocation and increased inflammation.[60]

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As liver fibrosis progresses, there are simultaneous changes in the metabolic function of the liver that influence the microbiome. With progression of liver fibrosis, there is a reduction in bile acid (BA) production, with lower BAs noted in the intestine, which link directly to bacterial dysbiosis. This is because secondary BAs (transformation of which can only be done by selected colonic microbes, such as Clostridium cluster XVIa via 7α dehydroxylation) are the most potent stimulators of the Farnesoid X Receptor (FXR) in the ileum.[60]

FLORIMAX : PLACE IN THERAPY OF GASTROINTESTINAL DISORDERS

The World Gastroenterology Organization developed global guidelines for the use of probiotics and prebiotics in adults and children in 2011. They determined that there was good evidence from randomized controlled trials (evidence level 1b) or from meta-analyses of randomized trials (level 1a) to support the use of probiotics in a number of adult indications (as given below).[61]

1. Treatment of acute diarrhea
2. Prevention of antibiotic-associated diarrhea
3. Prevention of Clostridium difficile diarrhea
4. Reduction of symptoms associated with lactose maldigestion
5. Alleviation of symptoms of irritable bowel syndrome
6. Maintenance of remission in ulcerative colitis
7. Treatment of mildly active ulcerative colitis or pouchitis
8. Prevention and maintenance of remission in pouchitis

Antibiotic-Associated Diarrhea

Otherwise, unexplained diarrhea that occurs in association with antibiotic treatment is classified as AAD. The use of probiotics, which are thought to ameliorate the disruptive effects of antibiotics on the microbiome, may provide a method for prevention and treatment of AAD and C. difficile-associated diarrhea. A Cochrane systematic review found that probiotics significantly reduced the risk of C. difficile-associated diarrhea in adults and children and may have reduced the risk of adverse events during treatment and reduced the duration of hospital stay.[61]

Travelers' Diarrhea

Probiotics may also be used in the prevention of Travelers' diarrhea. In a meta-analysis by McFarland and colleagues, several probiotics (including Lactobacillus strains) were associated with significant improvements in the prevention of Travelers' diarrhea compared with placebo, and there were no reports of serious adverse events in any trials.[61]

Helicobacter pylori infection

According to the fourth edition of the Maastricht consensus report on the management of H. pylori infection, adjuvant treatment with certain probiotics may reduce the incidence of adverse effects during treatment.

Two meta-analyses found that probiotics generally, and Lactobacillus-containing probiotics specifically, significantly enhance H. pylori eradication rates when administered with eradication therapy. Lactobacillus-containing probiotics were effective in adults, as well as children. The improved eradication rates could be due to a better tolerability of the antibiotic combination therapies and, therefore, better compliance. In the meta-analysis of Zhang and colleagues, the use of adjunctive probiotics also reduced the incidence of adverse events associated with eradication therapy by 41%.[61]

Irritable Bowel Syndrome

There is some recent level 1a evidence to support the use of probiotics for patients with IBS. Ford and colleagues conducted a meta-analysis of 35 placebo-controlled trials of probiotics in adults with IBS involving 3452 patients. They found that probiotics had a significantly favorable effect on a range of outcomes, including persistent symptom relief, abdominal pain or global symptoms, bloating, and flatulence.

More studies were available for combination probiotic formulations than for individual strains, and these combinations significantly reduced all outcomes. Of the individual probiotics assessed, Lactobacillus strains significantly improved flatulence, Bifidobacterium improved abdominal or global symptom scores, and Streptococcus strains both provided persistent symptom relief. Finally, several studies conducted across the world found that probiotics altered the microbiota and exerted therapeutic benefits for patients with IBS.[61]

Inflammatory Bowel Disease

One meta-analysis found that De Simone Formulation (a mixture of S. thermophilus, Lactobacillus, and Bifidobacterium strains) was effective for inducing remission in patients with active UC. This analysis included data from a double-blind, placebo-controlled study conducted in India, in which De Simone Formulation proved to be more effective than placebo for relieving symptoms and inducing remission in adults with mild-to-moderate UC. De Simone Formulation was also significantly more effective than placebo for preventing relapses in patients with UC or ileo-anal pouch anastomosis.[61]

Other indications

There are several other potential indications for probiotic use and include lactose intolerance, post-infectious IBS, tropical sprue, small intestinal bacterial overgrowth (e.g., after prolonged use of

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proton pump inhibitors), non-alcoholic fatty liver disease, complications of liver disease, constipation, and enteropathy associated with non-steroidal anti-inflammatory drugs. Probiotics are also being actively investigated in a range of indications aside from those affecting the gastrointestinal tract, including respiratory tract infections, urinary tract infections, bacterial vaginosis, pollenosis, allergic rhinitis and asthma, diabetes mellitus, hyperlipidemia, obesity/metabolic syndrome, and minimal hepatic encephalopathy. However, there is currently limited evidence supporting a recommendation for probiotics in these indications.[61]

CLINICAL EVIDENCE

In the 1990's, Professor Claudio De Simone invented and patented a number of multi-strain probiotic formulations with specific biochemical and immunologic profiles. One of these formulations - the De Simone Formulation - was licensed to the VSL Pharmaceuticals, Inc. (“VSL Inc”) and was subsequently produced under the brand name, “VSL#3” from 2002 to January 31, 2016. When De Simone terminated his relationship with VSL Inc. he partnered with ExeGi Pharma, LLC to market his De Simone Formulation under the brand, VISBIOME (**Florimax** in India). Therefore, clinical trials performed with the De Simone Formulation and are applicable to the evaluation of **Florimax** as a medical food.

Florimax contains the original De Simone Formulation of Probiotic. VSL#3® produced before January 31, 2016 contained “De Simone Formulation”. However, after January 2016 the VSL#3® product was changed. The sellers were ordered by a U.S. court to stop any claims that state or suggest a false continuity between the new formulation sold as VSL#3® and the original De Simone Formulation.

CLINICAL EXPERIENCE - IBS DIETARY MANAGEMENT

The De Simone Formulation has been the subject of over 75 published clinical trials in human subjects, with extensive clinical research in the dietary management of dysbiosis associated with irritable bowel syndrome (IBS), ulcerative colitis (UC), pouchitis, hepatic encephalopathy (HE), and antibiotic-associated diarrhea (AAD). **Florimax** is a medical food. The De Simone Formulation has been the subject of ten clinical trials involving over 550 adult and pediatric patients in the dietary management of dysbiosis associated with IBS.[22,23,24,25,26,27,28,29,30,31]

In one study, 25 patients with diarrhea-predominant IBS received placebo or the De Simone Formulation for eight weeks. Patients receiving the De Simone Formulation as a medical food experienced a statistically significant reduction in abdominal bloating.[23] In a second study, 48 patients with Rome II IBS were randomized in a double-blind design to the probiotic or placebo. Patients receiving the De Simone Formulation medical food experienced a statistically significant reduction in flatulence (p=0.01).[22]

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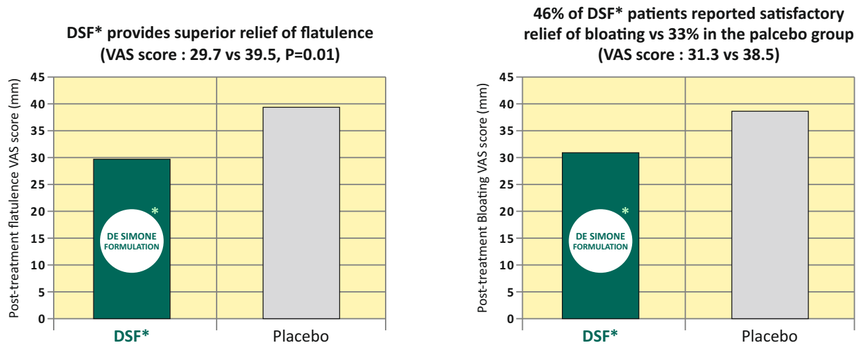
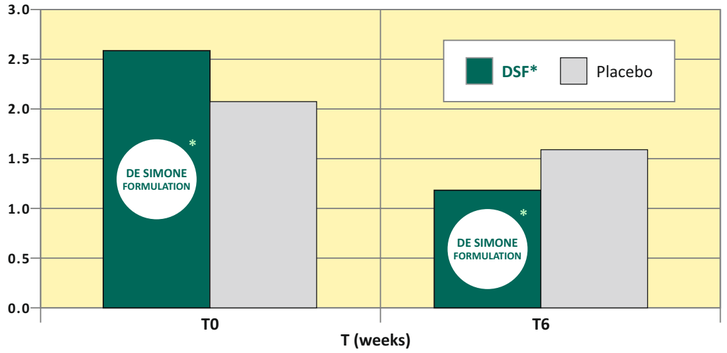


*Prof. Claudio De Simone’s Original Formulation

Figure : [Kim et al 2005] flatulence & bloating scores in the probiotic & in the placebo groups after 8 weeks of supplementation



*Prof. Claudio De Simone’s Original Formulation

Figure : [Guandalini S et al] Abdominal bloating score rated on a scale of 0 to 4

The De Simone Formulation was well tolerated with no adverse events reported in either IBS studies.

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CLINICAL EXPERIENCE - ULCERATIVE COLITIS (UC) MANAGEMENT

The De Simone Formulation has been the subject of published clinical studies in ulcerative colitis involving nearly 500 adults and 47 Pediatric patients.[32,33,34,35,36,37,38] In these studies, daily consumption of the De Simone Formulation was associated with effective dietary management of ulcerative colitis.

In one study involving 90 adult patients, the De Simone Formulation plus low-dose balsalazide was compared to balsalazide, or mesalamine alone in the dietary management of acute ulcerative colitis. The De Simone Formulation plus low-dose balsalazide was superior to balsalazide or mesalamine alone in achieving dietary management of remission (85.7% vs. 80.8% vs. 72.7%; p <0.02) with improved time to remission (4 days vs. 7.5 vs. 13; p <0.001). In a second study involving 34 adult patients with acute UC, dietary management with the De Simone Formulation resulted in a combined 77% remission/response rate with no adverse effects, as measured by UCDAI score (53% remission, 24% response).[37]

In the dietary management of UC, the De Simone Formulation was also shown to help achieve remission when added to standard therapies (mesalazine, azathioprine, or 6-mercaptopurine). In a multicenter, randomized, double blind, placebo-controlled trial (n=147) patients consuming the De Simone Formulation had significantly higher remission rates vs. placebo (43% vs. 16%; p< 0.001). In the same study, UCDAI scores for patients consuming the De Simone Formulation showed a significant decrease by 50% from baseline (p<0.001)[35]

The European Society for Clinical Nutrition and Metabolism (ESPEN) recognizes the De Simone Formulation as one of two probiotics which should be considered as a dietary aid in the maintenance of remission and induction of remission in patients with ulcerative colitis.[39,40]

CLINICAL EXPERIENCE - ANTIBIOTIC-ASSOCIATED DIARRHEA (AAD)

In a double-blind, randomized, controlled clinical trial, 229 hospitalized patients on systemic antibiotics were administered the De Simone Formulation probiotic or placebo. Patients were administered a variety of antibiotics in intravenous and oral formulations, including penicillins, broad spectrum penicillins, cephalosporins, quinolones, macrolides, aminonglycosides, imidazoles, and others. The probiotic or placebo was administered within 48 hrs of first hospital antibiotic treatment. The rate of AAD was significantly lower in the active group vs placebo on a per protocol analysis (0% active vs 11.4% placebo; P=0.006).[41]

CLINICAL EXPERIENCE - POUCHITIS DIETARY MANAGEMENT

In three double-blind, placebo-controlled trials and one open trial, the De Simone Formulation has been shown to aid in the dietary management of pouchitis.[42,43,44]

In Gionchetti et al. (2000), 40 patients with chronic relapsing pouchitis were randomized to the probiotic or placebo group after one month of antibiotic treatment. In the dietary management of

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remission, 20 patients consuming the probiotic were still in remission after nine months compared to zero in the placebo group (p <0.0001).[42] **The formulation found in Florimax is recognized as a tool for the dietary management of pouchitis by the American College of Gastroenterology,[45] the European Society for Clinical Nutrition and Metabolism[46] the British Society of Gastroenterology,[46] the Asian Working Group[48] and The Cochrane Collaboration.[47]**

CLINICAL EXPERIENCE - HEPATIC ENCEPHALOPATHY (HE) MANAGEMENT

In the dietary management of dysbiosis associated with hepatic encephalopathy (HE), the De Simone Formulation has been the subject of multiple controlled clinical studies involving over 800 patients.[54,55,56,57,58] In one placebo controlled trial involving 160 cirrhotic patients, those consuming the De Simone Formulation for dysbiosis experienced a reduced incidence of HE, reduced ammonia levels, and improvements in psychometric tests compared to controls. Seven patients in the probiotic group experienced overt HE vs. 14 in the control group (p<0.05)[54]

In a second study, 235 cirrhotic patients who had prior episodes of HE were evaluated after consuming the De Simone Formulation, lactulose or no therapy. There was a significant difference in the development HE in the probiotic vs. no treatment groups (p=0.02) and in the lactulose vs. no treatment group (p=0.001), but no difference between the probiotic group vs. lactulose (p=0.134).[56]

CLINICAL EXPERIENCE - PEDIATRIC

The De Simone Formulation was the subject of two trials involving patients between the ages of 1.7 and 17 years of age with active ulcerative colitis (UC). In one trial, 29 patients were randomized to receive dietary management with the De Simone Formulation or placebo concomitantly with standard UC treatment (steroids, 5-ASA). Thirteen patients (92.8%) of those supplemented with the De Simone Formulation and standard therapy achieved remission vs. four patients (36.4%) in the placebo arm (p<0.001).[59]

In addition, 21.4% of patients consuming the De Simone Formulation and standard UC therapy and 73.3 % patients consuming placebo and standard therapy relapsed within 1 year of follow-up (p= 0.014). At six months, 12 months, or at time of relapse, endoscopic and histological scores were lower in the probiotic group than in placebo group. 66 In a second study, the De Simone Formulation was administered open-label for eight weeks in pediatric patients with mild to moderate acute UC. Ten patients (56%) achieved remission and the combined remission/response rate was 61%.[34]

The De Simone Formulation was studied in 59 pediatric IBS patients, aged 4 to 18, diagnosed with IBS using the Rome II criteria. The group who was administered the De Simone Formulation as a medical food had statistically significant improvements in the primary endpoint of subjective assessment of relief of symptoms (P <0.05%) abdominal bloating/gassiness (P<0.05%) and family assessment of life disruption. However, there were no significant changes in stool pattern.[26]

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OBESITY AND NON-ALCOHOLIC FATTY LIVER DISEASE (NAFLD)

In a non-controlled study, Loguercio et al. analyzed the effect of the probiotic mix for 3 months in several groups of patients with various liver diseases, including NAFLD. Patients with NAFLD showed a decrease in serum aminotransferases, oxidative damage and nitric oxide production.

Recently, Alisi et al. reported a double-blind placebo-controlled randomized trial in which they studied 44 obese children with NAFLD to evaluate the effect of the probiotic mix or placebo for 4 months. Although liver biopsy was not performed at the end of the study period, in children receiving the probiotic, the authors observed a statistically significant decrease in body mass index and an improvement in the severity of NAFLD evaluated by ultrasonography.

Obesity often leads to serious cardiovascular diseases and diabetes, and represents a heavy economic burden.

Rajkumar et al. conducted a randomized placebo-controlled study in overweight adults in 4 arms receiving the probiotic mix with or without Omega-3, Omega-3 alone or placebo and observed that the patients receiving the probiotics had significant reduction in total cholesterol, triglyceride, LDL (low density lipoprotein) and VLDL (very low density lipoprotein) and increase in HDL (high density lipoprotein) cholesterol. The combination with Omega-3 had a more pronounced effect on HDL, insulin sensitivy and amelioration of inflammation (hsCrP).

FLORIMAX : SUMMARY OF THE CLINICAL OBSERVATIONS IN PATIENTS WITH CIRRHOSIS

- Prevention of the first episode of hepatic encephalopathy
- Prevention of recurrence of hepatic encephalopathy
- Improvement in psychometric tests in patients with minimal hepatic encaphalopathy
- Decrease in the need for hospitalization due to hepatic encephalopathy
- Improvement in liver function tests
- Improvement in health-related quality of life
- Decrease in ammonemia
- Modulation of inflammatory response
- Decrease in portal pressure

The specific mix of bacteria contained in the Florimax (De Simone Formulation- DSF) has been mentioned for the first time in the Recommendations for Probiotics 2015 from the proceedings of a workshop organized by Yale and Harvard Universities for the managament of liver conditions, in particular Non-Alcoholic Steato-Hepatitis and Hepatic Encephalopathy. In particular level A evidence was recognized in Hepatic Encephalopathy.

Indeed, several randomized clinical trials including a large number of cirrhosis patients have demonstrated the possible application of the specific probiotic combination contained in the Florimax (De Simone Formulation) in primary and secondary prevention of HE and in minimal HE. Other effects observed in these trials included an improvement in liver function tests and a decrease in the need for hospitalization. There is a potential role for DSF to decrease portal pressure and to prevent related complications in patients with cirrhosis. One main positive aspect represented by the use of DSF in these indications is that it improves the intestinal permeability, which prevents or reduces bacterial translocation thus reducing the inflammation of the liver. Other settings in which DSF could be useful include NAFLD and Alcoholic Liver Disease. In particular, one important target may be Obesity in children, which has become the most common cause of Chronic Liver Disease in children and is a significant burden on healthcare systems worldwide. The severity of hepatic steatosis is affected by intestinal permeability and Intestinal Bacterial Overgrowth. There is a difference in the distinct composition of the gut microbiome among children and adolescents with Non-Alcoholic Steato-Hepatitis (NASH), Obese children without NASH, and healthy individuals, and modulation of the intestinal microbiota may offer an important therapeutic target for NAFLD as suggested by Miloh. Obesity in adults leads to high risks of metabolic syndrome and the use of this specific combination of probiotic strains to improve lipid profile, insulin sensitivity, and inflammatory responses may help reduce the risks of heart disease, diabetes, and stroke, in a healthy overweight population. The prevention of hepatocellular carcinoma, and the prophylaxis of bacterial infections avoiding the development of bacterial resistances observed with antibiotic prophylaxis are future targets for research.

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FLORIMAX (DE SIMONE FORMULATION) LIVER DISEASE STUDIES

Sr. no.	Title of the study	Probiotic Intervention	Study population & Duration	Results & Main outcome	Conclusions
1	Secondary Prophylaxis of Hepatic Encephalopathy in Cirrhosis : An Open-Label, Randomized Controlled Trial of Lactulose, Probiotics & No Therapy Agrawal A, et al. Am J Gastroenterol. 2012 Jul;107(7):1043-50.	**Florimax (De Simone Formulation)**	360 patients with Consecutive cirrhotic patients who had recovered from hepatic encephalopathy. One group received lactulose (Gp-L, 30 ml three times per day), three capsules of probiotics (Gp-P) per day containing 112.5 billion viable lyophilized bacteria per capsule, other group have no therapy (Gp-N). Duration of the follow-up is 12 months dosage 112.5 billion per capsule for 3 times per day.	This study showed that the use of probiotics as compared with no therapy is more effective in secondary prophylaxis of HE. Of 197 patients, 77 (39.1%) developed an episode of overt HE over a follow-up period of 12 months. In all, 18 (26.2%) in Gp-L, 22 (34.4%) in Gp-P, and 37 (56.9 %) in Gp-N developed HE (P = 0.001). In our previous study, 12 (19.6%) of 61 patients in the HE-L group & 30 (46.8%) of 64 in the HE-NL group (P = 0.001) developed HE over a median follow-up of 14 months (range 1-20 months)	Lactulose and Probiotics are effective for secondary prophylaxis of HE in patients with cirrhosis. Overall, study findings propose De Simone Formulation probiotics are equally effective in secondary prophylaxis of hepatic encephalopathy.
2	Randomised clinical trial : the beneficial effects of De Simone Formulation VSL#3 in obese children with non-alcoholic steatohepatitis Alisi A, et al. Aliment Pharmacol Ther. 2014 Jun;39(11):1276-85.	**Florimax (De Simone Formulation)**	Of 48 randomised children, 44 (22 De Simone Formulation and 22 placebo) completed the study. The main outcome was the change in fatty liver severity at 4 months as detected by ultrasonography. Secondary outcomes were the changes in triglycerides, insulin resistance as detected by the homoeostasis model assessment (HOMA), alanine transaminase (ALT), body mass index (BMI), glucagon-like peptide 1 (GLP-1) and activated GLP-1 (aGLP-1). Ordinal and linear models with cluster confidence intervals were used to evaluate the efficacy of De Simone Formulation vs. placebo at 4 months. Duration 4 months. Dosage : 112.5 billion per day.	At baseline, moderate and severe NAFLD were present in 64% and 36% of PLA children and in 55% and 45% of De Simone Formulation children. The probability that children supplemented with De Simone Formulation had none, light, moderate or severe FL at the end of the study was 21%, 70%, 9% and 0% respectively with corresponding values of 0%, 7%, 76% and 17% for the placebo group (P<0.001). No between-group differences were detected in triglycerides, HOMA and ALT while BMI decreased and GLP-1 and aGLP1 increased in the De Simone Formulation group (P < 0.001 for all comparisons).	Overall, study findings confirm 4-month supplement of De Simone Formulation significantly improves NAFLD in children. The De Simone Formulation -dependent GLP-1 increase could be responsible for these beneficial effects.
3	Probiotic VSL#3 De Simone Formulation Reduces Liver Disease Severity and Hospitalization in Patients with Cirrhosis: A Randomized, Controlled Trial Dhiman RK, et al. Gastroenterology. 2014 Dec;147(6):1327-37.e3.	**Florimax (De Simone Formulation)**	Authors performed a double-blind trial at a tertiary care hospital in India. Patients with cirrhosis who had recovered from an episode of HE during the previous month were assigned randomly (using computer-generated allocation) to groups given a probiotic preparation (De Simone Formulation, daily for 6 months. Dosage 112.5 billion one sachet per day.	There was a trend toward a reduction in the development of breakthrough HE among patients receiving the probiotic (34.8% in the probiotic group vs 51.6% in the placebo group; hazard ratio [HR], 0.65; 95% confidence interval [CI], 0.38-1.11; P ¼ .12). Fewer patients in the probiotic group were hospitalized for HE (19.7% vs 42.2%, respectively; HR, 0.45; 95% CI, 0.23-0.87; P ¼ .02) or for complications of cirrhosis (24.2% than in the placebo group (45.3%) (HR, 0.52; 95% CI, 0.28-0.95; P ¼ .034). Child-Turcotte-Pugh and model for end-stage liver disease scores improved significantly from baseline to 6 months in the probiotic group, but not in the placebo group. There were no adverse events related to De Simone Formulation.	Study showed over a 6-month period, daily intake of De Simone Formulation significantly reduced the risk of hospitalization for HE, as well as Child-Turcotte-Pugh and model for end-stage liver disease scores, in patients with cirrhosis.

Sr. no.	Title of the study	Probiotic Intervention	Study population & Duration	Results & Main outcome	Conclusions
4	Shigh potency multistrain probiotic improves liver histology in nonalcoholic fatty liver disease (NAFLD) : a randomised, double-blind, proof of concept study. Duseja A, et al. BMJ Open Gastroenterol. 2019 Aug 7;6(1):e000315.	**Florimax (De Simone Formulation)**	Thirty-nine liver biopsy-proven patients with NAFLD were randomised in a double-blind fashion to either lifestyle modifications plus an oral multistrain probiotic (675 billion bacteria daily, n=19) or identical placebo (n=20) for 1 year. Lifestyle modifications included regular exercise for all and control of overweight/obesity (with additional dietary restrictions), hypertension and yperlipidaemia in those with these risk factors. Primary objective of the study was the histological improvement in NAFLD activity score (NAS) and its components and secondary objectives were improvement in alanine transaminase (ALT) and cytokine profile. Duration 1 year. (675 billion bacteria daily.	A significant improvement in levels of ALT (p=0.046), leptin (p=0.006), tumour necrosis factor-α (p=0.016) and endotoxins (p=0.017) was observed in probiotic De Simone Formulation group in comparison to placebo at 1 year.	Study showed that patients with NAFLD managed with lifestyle modifications and multistrain probiotic. De Simone Formulation showed significant improvement in liver histology, ALT and cytokines.
5	Effects of the adjunctive probiotic VSL#3 De Simone Formulation on portal haemodynamics in patients with cirrhosis and large varices : a randomized trial. Gupta N, et al. Liver Int. 2013 Sep;33(8):1148-57.	**Florimax (De Simone Formulation)**	Randomized double-blind placebo-controlled trial conducted in G.B. Pant Hospital, New Delhi. A total of 94 cirrhotic patients having large oesophageal varices without history of variceal bleeding were randomized to three treatment groups and given 2 months' treatment with propranolol plus placebo, propranolol plus antibiotics (norfloxacin 400 mg BD) or propranolol plus probiotic (De Simone Formulation, 900 billion/day) randomly assigned in 1:1:1 ratio. Outcome measures were change in Hepatic venous pressure gradient (HVPG): Response rate (Percentage of patients having a decrease from baseline of _20% or to _12 mm Hg) and changes from baseline; biochemical markers of inflammation: changes from baseline. Duration 2 months. Dosage 900 billion/day.	Adjunctive probiotics De Simone Formulation increased the response rate compared with propranolol alone (58% vs. 31%, P = 0.046).	Probiotic De Simone Formulation improved the response rate to propranolol therapy and was safe and well tolerated in patients with cirrhosis. Adjunctive probiotic therapy merits further study for reduction in portal pressure.
6	VSL#3_De Simone Formulation probiotic therapy does not reduce portal pressures in patients with decompensated cirrhosis. Jayakumar S, et al. Liver Int. 2013 Nov;33(10):1470-7.	**Florimax (De Simone Formulation)**	Seventeen patients with decompensated cirrhosis and an HVPG of ≥10 mmHg was randomized to receive 2 months of De Simone Formulation _ or an identical placebo. HVPG, endotoxin, interleukin (IL)-6, IL-8, IL-10, renin, aldosterone, nitric oxide and stool microbiota were measured at baseline and study end. Duration 8 weeks. Dosage 112.5 billion one sachet per day.	There was a significant reduction in the plasma aldosterone level in the De Simone Formulation group.	De Simone Formulation has a positive impact on portal pressure reduction in patients with decompensated cirrhosis.

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7	Beneficial Effects of a Probiotic VSL#3 De Simone Formulation on Parameters of Liver Dysfunction in Chronic Liver Diseases. Loguercio C. J Clin Gastroenterol. 2005 Jul;39(6):540-3.	**Florimax (De Simone Formulation)**	A total of 22 nonalcoholic fatty liver disease (NAFLD) and 20 alcoholic liver cirrhosis (AC) patients were enrolled in the study and compared with 36 HCV- positive patients with chronic hepatitis without (20, CH) or with (16, CC) liver cirrhosis. All patients were treated with the probiotic De Simone Formulation. Routine liver tests, plasma levels of tumor necrosis factor alpha (TNF-a), interleukin (IL)-6 and -10, malondialdehyde (MDA), and 4-hydroxynonenal (4-HNE), S-nitrosothiols (S-NO), were evaluated on days 0,30, 90, and 120. Duration of the study 4 months . Dosage 450 billion per day.	Treatment with VSL#3 exerted different effects in the various groups of patients : in NAFLD and AC groups, it significantly improved plasma levels of MDA and 4-HNE, whereas cytokines (TNF-a, IL-6, and IL-10) improved only in AC patients.	Results of the study suggest that manipulation of intestinal flora with De Simone Formulation should be taken into consideration as possible adjunctive therapy in chronic liver disease.
8	Probiotics Prevent Hepatic Encephalopathy in Patients with Cirrhosis : A Randomized Controlled Trial. Lunia MK, et al. Clin Gastroenterol Hepatol. 2014 Jun;12(6):1003-8.e1.	**Florimax (De Simone Formulation)**	Study was conducted a prospective trial at a tertiary care referral institute in New Delhi, India, from January 2012 through March 2013, of patients with cirrhosis without overt HE (age, 48.6 - 11.1 y; 96 men and 64 women); 25 were Child-Turcotte-Pugh (CTP) class A, 51 were CTP class B, and 84 where CTP class C. Subjects were assigned randomly to groups given probiotics colony-forming units, 3 times daily. Duration is 3 months. Dosage 112.5 billion three sachet per day.	Three months of probiotic administration significantly reduced levels of arterial ammonia, SIBO, and OCTT; increased psychometric hepatic encephalopathy scores; and increased CFF thresholds, compared with baseline. Seven subjects in the probiotic group and 14 controls developed overt HE (P < .05; hazard ratio for controls vs probiotic group, 2.1; 95% confidence interval, 1.31-6.53). Psychometric hepatic encephalopathy scores, CTP scores, and SIBO correlated with the development of overt HE.	In this prospective, randomized controlled trial, De Simone Formulation probiotics were found to be effective in preventing HE in patients with cirrhosis.
9	The effect of short-term treatment with probiotic vsl#3 De Simone Formulation on various Clinical and biochemical parameters in patients with liver cirrhosis. Marlicz W, et al. J Physiol Pharmacol. 2016 Dec;67(6):867-877.	**Florimax (De Simone Formulation)**	In our study twenty patients with compensated and decompensated liver cirrhosis and ten healthy controls received probiotic De Simone Formulation daily for 28 days. Plasma levels of interleukin 6 (IL-6), vascular endothelial growth factor (VEGF), plasminogen activator inhibitor (PAI), macrophage inflammatory protein 3a (MIP-3 a/CCL20), monocyte chemotactic protein-1a (MCP-1/CCL2), human myeloperoxidase (MPO), nitric oxide (NO), prostaglandins, thromboxane (TXB2) and big-endothelin were measured at baseline, day 14 and 28 of probiotic administration. Duration 28 days. Dosage 112.5 billion three sachet per day.	Probiotic administration in patients with liver cirrhosis led to modulation of plasma levels of several molecules and compounds measured (MIP-3a/CCL20.	De Simone Formulation manipulates selected plasma molecules and compounds involved in hyperdynamic circulatory dysfunction. Short term De Simone Formulation administration affects several clinical and biochemical parameters commonly altered in liver cirrhosis.

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10	Urinary 1H-NMR-based metabolic profiling of children with NAFLD undergoing VSL#3 De Simone Formulation treatment. Miccheli A, et al. Int J Obes (Lond). 2015 Jul;39(7):1118-25.	**Florimax (De Simone Formulation)**	Urine samples from a group of 31 pediatric NAFLD patients, enrolled in a De Simone Formulation clinical trial, were analyzed by high resolution proton nuclear magnetic resonance spectroscopy in combination with analysis of variance-Simultaneous Component. Analysis model and multivariate data analyses. Urinary metabolic profiles were interpreted in terms of clinical patient feature, treatment and chronology pattern correlations. Duration 4 months. Dosage 112.5 billion three sachet per day.	De Simone Formulation treatment induced changes in NAFLD urinary metabolic phenotype mainly at level of host amino-acid metabolism (That is, valine, tyrosine, 3-amino-isobutyrate or β-aminoisobutyric acid (BAIBA)), nucleic acid degradation (pseudouridine), creatinine metabolism (methylguanidine) and secondarily at the level of gut microbial amino-acid metabolism (that is, 2-hydroxyisobutyrate from valine degradation). Furthermore, some of these metabolites correlated with clinical primary and secondary trial end points after De Simone Formulation treatment : tyrosine and the organic acid U4 positively with alanine aminotransferase (R = 0.399, P = 0.026) and BMI (R = 0.36, P = 0.045); BAIBA and tyrosine negatively with active glucagon-like-peptide 1 (R = – 0.51, P = 0.003; R = -0.41, P = 0.021, respectively).	De Simone Formulation treatment-dependent urinary metabotypes of children with NAFLD may be considered as non-invasive effective biomarkers to evaluate the response to treatment.
11	A randomized controlled trial comparing lactulose, probiotics, and L-ornithine L-aspartate in treatment of minimal hepatic encephalopathy. Mittal VV, et al. Eur J Gastroenterol Hepatol. 2011 Aug;23(8):725-32.	**Florimax (De Simone Formulation)**	Consecutive patients with cirrhosis were screened for MHE. MHE was diagnosed by two or more abnormal psychometric tests (number/figure connection tests A and B, block design test, picture completion test). Patients were randomized to no treatment (GpA), lactulose 30-60 ml/twice per day (GpB), probiotics 110 billion colony forming units twice in a day (GpC), LOLA 6 g three times per day (GpD) for 3 months. Arterial ammonia and HRQoL assessment using SIP questionnaire was done at baseline and end point. Duration of the treatment is 3 months. dosage 112.5 billion three sachet per day.		De Simone Formulation probiotics, and LOLA significantly improve MHE and HRQoL in patients with chronic liver disease.
12	Effect of probiotic De Simone Formulation VSL#3 in the treatment of minimal hepatic encephalopathy : A non-inferiority randomized controlled trial. Pratap Mouli V et al. Hepatol Res. 2015 Aug;45(8):880-9.	**Florimax (De Simone Formulation)**	Patients with CLD (n = 227) were screened for MHE using neuropsychometric tests (number connection tests A and B [or figure connection tests A and B]) and/or neurophysiological test (P-300 auditory event-related potential), and 120 (53%) were diagnosed with MHE by abnormal tests. MHE patients were randomized to lactulose (30-60 mL/ day) or probiotic (four capsules of De Simone Formulation; total of 450 billion CFU/day) for 2 months. Dosage 112.5 billion 4capsule per day.	Of 120 patients randomized, 40 in the lactulose arm and 33 in the probiotic arm completed 2 months of intervention. MHE improved in 25 (62.5%) patients taking lactulose and 23 (69.7%) taking probiotics. The effect size of difference of improvement in MHE between lactulose and probiotic was 0.072 per per-protocol analysis and 0.040 as per intention to treat analysis (within −20% of non-inferiority margin). Serum ammonia was comparable between groups at baseline and 2 months; it decreased in patients in whom MHE improved, while increased in patients with no improvement in MHE.	The probiotic De Simone Formulation was non-inferior to the standard therapy, lactulose in the treatment of MHE.

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13	Effect of Probiotic (VSL#3) De Simone Formulation and Omega-3 on Lipid Profile, Insulin Sensitivity, Inflammatory Markers, and Gut Colonization in Overweight Adults : A Randomized, Controlled Trial. Rajkumar H, et al. Mediators Inflamm. 2014;2014:348959.	**Florimax (De Simone Formulation)**	N=60 After initial screening the subjects were randomized into four groups with 15 per group. The four groups received, respectively, placebo, omega-3 fatty acid, probiotic De Simone Formulation, or both omega-3 and probiotic, for 6 weeks. Blood and fecal samples were collected at baseline and after 6 weeks. dosage 112.5 billion 1 capsule per day.	The probiotic (VSL#3) supplemented group had significant reduction in total cholesterol, triglyceride, LDL, and VLDL and had increased HDL ($p < 0.05$) value. VSL#3 improved insulin sensitivity ($p < 0.01$), decreased hsCRP, and favorably affected the composition of gut microbiota.	De Simone Formulation had more pronounced effect on HDL, insulin sensitivity and hsCRP. Subjects with low HDL, insulin resistance, and high hsCRP.
14	Oral probiotic VSL#3 De Simone Formulation attenuates the circulatory disturbances of patients with cirrhosis and ascites. Rincón D, et al. Liver Int. 2014 Nov;34(10):1504-12.	**Florimax (De Simone Formulation)**	Seventeen patients with cirrhosis and ascites were prospectively included, five of whom abandoned this study prematurely. Hepatic and systemic haemodynamic evaluations were performed at baseline and after 6 weeks of receiving an oral De Simone Formulation probiotic preparation. Two sachets per day of a high potency probiotic preparation containing 450 billion colony forming units.	Administration of the probiotic mixture De Simone Formulation improved the hepatic and systemic haemodynamics and serum sodium levels in patients with cirrhosis.	Administration of the probiotic mixture Florimax improved the hepatic and systemic haemodynamics and serum sodium levels in patients with cirrhosis. Our results identify major effects of De Simone Formulation probiotics in liver disease and provide the rationale for assessing their therapeutic potential against the progression of portal hypertension and its complications in future clinical trials.
15	Effect of a Multistrain Probiotic on Cognitive Function and Risk of Falls in Patients With Cirrhosis : A Randomized Trial. Román E. Hepatol Commun. 2019 Mar 12;3(5):632-645.	**Florimax (De Simone Formulation)**	The aim of this study was to evaluate the effect of a multistrain probiotic on cognitive function, risk of falls, and inflammatory response in patients with cirrhosis. Consecutive outpatients with cirrhosis and cognitive dysfunction (defined by a Psychometric Hepatic Encephalopathy Score [PHES] < −4) and/or falls in the previous year were randomized to receive either a sachet of a high-concentration multistrain probiotic containing 450 billion bacteria twice daily for 12 weeks or placebo. Two sachets per day of a high potency probiotic preparation containing 450 billion colony forming units.	The main finding of the present study is that treatment with a multi strain De Simone Formulation probiotic improved cognitive function and decreased the risk of falling in patients with cirrhosis who had cognitive dysfunction and/or previous falls.	The study concluded that this multi strain De Simone Formulation probiotic improved cognitive function, decreased the risk of falls, and modulated the systemic inflammatory response in patients with cirrhosis and cognitive dysfunction and/ or previous falls. This probiotic mixture could be useful in the prevention of falls and complications, especially hepatic encephalopathy and bacterial infections, in patients with cirrhosis.
16	Effects of probiotic therapy on portal pressure in patients with cirrhosis : a pilot study Tandon P, et al. Liver Int. 2009 Aug;29(7):1110-5.	**Florimax (De Simone Formulation)**	Eight patients with compensated or very early decompensated cirrhosis and hepatic venous pressure gradient (HVPG) 410mmHg, received 2 months of De Simone Formulation (3600 billion bacteria daily). The HVPG, intestinal permeability, endotoxin, tumour necrosis factor (TNF)-a, interleukin (IL)-6, IL-8, renin and aldosterone were measured at baseline and study end Each sachet of probiotics (De Simone Formulation contained 900 billion per day for 2 months.	De Simone Formulation a trend to reduction in plasma endotoxin (P = 0.09), a mild but significant increase in serum TNF-α (P = 0.02) and a significant reduction in plasma aldosterone (P = 0.03).	The study concluded that reductions in endotoxin and aldosterone suggest possible beneficial effects of probiotics De Simone Formulation for this patient population.

INFLAMMATORY BOWEL DISEASE (IBD)

Sr. no.	Title of the study	Probiotic Intervention	Study population & Duration	Results & Main outcome	Conclusions
1	Effect of a probiotic preparation (De Simone Formulation) in patients with mild to moderate ulcerative colitis. Lee J. Korean J Gastroeterol 2012 Aug;60(2):94-101.	**"De Simone Formulation" 4 sachets daily in 2 divided doses for 8 weeks.**	Open-label Study 24 eligible patients with mild to moderate Ulcerative Colitis 8 weeks study	• Study demonstrated remission in 45.8% of subjects (n=11) • Partial response in 20.8% (n=5) • No change or worse in 25.0% (n=6) of subjects • The mean ulcerative colitis disease activity index (UCDAI) scores decreased from 7.09±1.81 to 1.45±1.29 in patients with a remission (p<0.001) • The mean endoscopic scores had also significantly decreased from 1.91±0.54 to 0.63±0.50 in patients with a remission (p<0.001)	Study demonstrated that DSF is effective in achieving clinical responses and remissions in patients with mild-to moderately active UC, further supporting the potential role in UC therapy.
2	Effects of intervention with sulindac & inulin/ De Simone Formulation on mucosal & luminal factors in the pouch of patients with familial adenomatous polyposis. Friederich P. Int J Colorectal Dis. 2011 May;26(5):575-82	**Three intervention with** **1. Sulindac,** **2. Inulin / De Simone Formulation** **3. Sulindac / inulin / De Simone Formulation**	17 patients with familial adenomatous polyposis 4-week intervention period	• Cell proliferation was lower after sulindac or De Simone Formulation / inulin, the combination treatment with sulindac / inulin / De Simone Formulation showed the opposite. • Glutathione S-transferase enzyme activity was increased after sulindac or De Simone Formulation / inulin, the combination treatment showed the opposite effect.	Our study revealed non-significant decreased cell proliferation and increased detoxification capacity after treatment with sulindac or De Simone Formulation /inulin.
3	Treatment of relapsing mild-to-moderate ulcerative colitis with the probiotic De Simone Formulation as adjunctive to a standard pharmaceutical treatment : a double-blind, randomized, placebo-controlled study. Tursi A. Am J Gastroenterol. 2010 Oct;105(10):2218-27.	**De Simone Formulation at a dose of 3,600 billion CFU/day**	144 patients were randomly treated for 8 weeks with De Simone Formulation at a dose of 3,600 billion CFU/day (71 patients) or with placebo (73 patients).	• The decrease in ulcerative colitis disease activity index (UCDAI) scores of 50% or more was higher in the De Simone Formulation group than in the placebo group (63.1 vs. 40.8). • Remission was higher in the De Simone Formulation group than in the placebo group (47.7% vs. 32.4%). Eight patients on De Simone Formulation (11.2%) and nine patients on placebo (12.3%) reported mild side effects.	De Simone Formulation supplementation is safe and able to reduce UCDAI scores in patients affected by relapsing mild-to-moderate UC who are under treatment with 5-ASA and/or immunosuppressants. Moreover, De Simone Formulation improves rectal bleeding and seems to reinduce remission in relapsing UC patients after 8 weeks of treatment.

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4	The probiotic preparation, De Simone Formulation induces remission in patients with mild-to-moderately active ulcerative colitis. Sood A. Clin Gastroenterol Hepatol. 2009 Nov;7(11):1202-9, 1209.e1.	**De Simone Formulation 3.6 x 10^{12} CFU or Placebo**	Adult patients with mild-to-moderate UC were assigned randomly to groups that were given 3.6 x 1012 CFU De Simone Formulation (n = 77) or placebo (n = 70), twice daily for 12 weeks.	• At week 6, the percentage of patients with an improvement in Ulcerative Colitis Disease Activity Index (UCDAI) score that was greater than 50% was significantly higher in the group given De Simone Formulation (25; 32.5%) than the group given placebo (7; 10%) (P = .001). • At week 12, there were 33 patients given De Simone Formulation (42.9%) who achieved remission, compared with 11 patients given placebo (15.7%) (P < .001). • Furthermore, significantly more patients given De Simone Formulation (40; 51.9%) achieved a decrease in their UCDAI that was greater than 3 points, compared with those given placebo (13; 18.6%) (P < .001). • The De Simone Formulation group had significantly greater decreases in UCDAI scores and individual symptoms at weeks 6 and 12, compared with the placebo group.	De Simone Formulation is safe and effective in achieving clinical responses and remissions in patients with mild-to-moderately active UC.
5	Probiotic preparation De Simone Formulation induces remission in children with mild to moderate acute ulcerative colitis: a pilot study. Huynh HQ. Inflamm Bowel Dis. 2009 May;15(5):760-8.	**De Simone Formulation daily in 2 divided doses**	18 eligible patients between the ages of 3-17 with mild to moderate acute UC received open-label De Simone Formulation daily in 2 divided doses for 8 weeks.	• 13 patients completed 8 weeks of De Simone Formulation treatment and 5 patients were withdrawn due to lack of improvement. • Remission (defined as SCCAI <or=3) was achieved in 56% of children (n = 10); response (decrease in SCCAI >or=2, but final score <or=5) in 6% (n = 1); and no change or worsening in 39% (n = 7). • Post- De Simone Formulation treatments demonstrated a bacterial taxonomy change in rectal biopsy. • The De Simone Formulation was well tolerated in clinical trials and no biochemical and clinical adverse effects attributed to De Simone Formulation were identified.	Treatment of pediatric patients diagnosed with mild to moderate UC with De Simone Formulation resulted in a remission rate of 56% and a combined remission/response rate of 61%.

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6	Effect of a probiotic preparation (De Simone Formulation) on induction & maintenance of remission in children with ulcerative colitis. Miele E. Am J Gastroenterol. 2009 Feb;104(2):437-43.	**De Simone Formulation 450-1,800 billion bacteria/day**	29 patients (mean age: 9.8 yrs) with newly diagnosed UC were randomized to receive either De Simone Formulation (450-1,800 billion bacteria/day; n=14) or an identical placebo (n=15) in conjunction with concomitant steroid induction and mesalamine maintenance treatment.	• All 29 patients responded to the inflammatory bowel disease (IBD) induction therapy. • Remission was achieved in 13 patients (92.8%) treated with De Simone Formulation and IBD therapy and in 4 patients (36.4%) treated with placebo and IBD therapy (P<0.001). • Overall, 3 of 14 (21.4%) patients treated with De Simone Formulation and IBD therapy and 11 of 15 (73.3%) patients treated with placebo and IBD therapy relapsed within 1 year of follow-up (P=0.014; RR=0.32; CI=0.025-0.773; NNT=2). All 3 patients treated with De Simone Formulation and 6 of 11 (54.5%) patients treated with placebo relapsed within 6 months of diagnosis. • At 6 months, 12 months, or at time of relapse, endoscopic and histological scores were significantly lower in the De Simone Formulation group than in the placebo group (P<0.05). There were no biochemical or clinical adverse events related to De Simone Formulation.	Trial that suggests the efficacy and safety of a highly concentrated mixture of probiotic bacterial strains (De Simone Formulation) in active UC and demonstrates its role in maintenance of remission.
7	Probiotic administration in patients with ileal pouch-anal anastomosis for ulcerative colitis is associated with expansion of mucosal regulatory cells. Pronio A. Inflamm Bowel Dis. 2008 May;14(5):662-8.	**De Simone Formulation**	31 patients at different periods from surgery without signs and symptoms of pouchitis were randomized to 2 sachets of De Simone Formulation once daily or no treatment for 12 months.	• During the study period, De Simone Formulation -treated patients showed a significant reduction in PDAI score and a significant increase in the percentage of mucosal CD4+CD25 (high) and CD4+ LAP-positive cells compared with baseline values. • Tissue samples at different points showed a significant reduction in IL-1beta mRNA expression, and a significant increase in Foxp3 mRNA expression.	De Simone Formulation administration in patients with IPAA modulates the PDAI and expands the number of mucosal regulatory T cells.
8	Balsalazide and/or high-potency probiotic mixture (De Simone Formulation) in maintaining remission after attack of acute, uncomplicated diverticulitis of the colon. Tursi A. Int J Colorectal Dis. 2007 Sep;22(9):1103-8.	**Group A : balsalazide 2.25 g daily** **Group B : De Simone Formulation**	Group A: Balsalazide 2.25 g daily for 10 days every month plus De Simone Formulation 450 billions/day for 15 days every month and Group B: De Simone Formulation alone 450 billion/day for 15 days every month.	• At the end of follow-up, 11 patients were completely symptom-free (73.33%), whilst 2 patients complained of only mild, recurrent symptoms (13%). • Two group B patients (13.33%) showed relapse of the disease at the 5th and 8th month of follow-up, respectively. • At the end of follow-up, 8 patients were completely symptom-free 60%), 2 patients complained of mild, recurrent symptoms (13.33%), 1 patient (6.66%) complained of mild but continuous symptoms.	Combination probiotic / anti-inflammatory drug was found better than probiotic treatment in preventing relapse of uncomplicated diverticulitis of the colon, even if without statistical significance. No side effects were recorded throughout the follow-up in both groups.

Sr. no.	Title of the study	Probiotic Intervention	Study population & Duration	Results & Main outcome	Conclusions
9	High-dose probiotics for the treatment of active Pouchitis. Gionchetti P. Dis Colon Rectum. 2007 Dec;50(12):2075-82.	**De Simone Formulation, 2 sachets B.I.D. (3,600 billion bacteria/day)**	23 patients with mild pouchitis, treated with De Simone Formulation, 2 sachets B.I.D. (3,600 billion bacteria/day) for four weeks.	• Sixteen of 23 patients (69 percent) were in remission after treatment. • The median total Pouchitis Disease Activity Index scores before and after therapy were 10 (range, 9-12) and 4 (range, 2-11), respectively (P < 0.01). • The median Inflammatory Bowel Disease Questionnaire score also significantly improved from 110 (range, 90-140) to 200 (range, 95-220; P < 0.001). All 16 patients who went into remission maintained remission during maintenance treatment. • Only one patient experienced a transient bloating at the beginning of treatment. • Patients who developed pouchitis while treated with placebo had low bacterial and high fungal diversity. Bacterial diversity was increased and fungal diversity was reduced in patients in remission maintained with De Simone Formulation (p = 0.001). • Real time PCR experiments demonstrated that De Simone Formulation increased the total number of bacterial cells (p = 0.002) and modified the spectrum of bacteria towards anaerobic species. • Taxa specific clone libraries for Lactobacilli and Bifidobacteria showed that the richness and spectrum of these bacteria were altered under probiotic therapy.	Probiotic therapy with De Simone Formulation increases the total number of intestinal bacterial cells as well as the richness and diversity of the bacterial microbiota, especially the anaerobic flora. The diversity of the fungal flora is repressed. Restoration of the integrity of a "protective" intestinal mucosa related microbiota could therefore be a potential mechanism of probiotic bacteria in inflammatory barrier diseases of the lower gastrointestinal tract.
11	Probiotics (DSF) in arthralgia in patients with ulcerative colitis & Crohn's disease: A pilot study. Karimi O, Pena AS, van Bodegraven AA. Drugs Today (Barc). 2005 Jul;41(7):453-9.	**An open-label trial using De Simone Formulation**	Sixteen of 29 patients completed the trial; in 29 patients of inflammatory bowel disease (IBD)	• 10 of the 16 patients a statistically significant improvement was documented by the Ritchie Articular Index. • No one of the patients had a relapse of intestinal disease while on probiotics. • These preliminary results suggest that the probiotic mixture De Simone Formulation may be an alternative treatment for arthralgia in patients with IBD without inducing exacerbation of the disease.	Because probiotics may be effective in the treatment of IBD as well, our results suggest that patients with active disease and arthralgia may also derive benefit from this treatment. Proper randomized controlled studies are indicated.

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12	Beclometasone Diproprionate (BDP) Plus De Simone Formulation for the Treatment of Mild to Moderate Diverticular Colitis : An Open Pilot Study. Tursi A. J Clin Gastroenterol. 2005 Aug;39(7):644-5.	**De Simone Formulation 450 billion viable lyophilized bacteria once daily**	12 patients were treated with BDP 10 mg/day for 4 weeks plus De Simone Formulation 2.5 g/day for 15 consecutive days containing 450 billion viable lyophilized bacteria of followed by BDP 5 mg/day for further 4 weeks plus De Simone Formulation De Simone Formulation was administered for only 15 days/month	• Ten patients (per-protocol, 90.90%; on intention-to-treat, 83.33%) were asymptomatic already at the 4th week and at the 8th week of treatment (overall score, 0), while only 1 patient (per-protocol, 9.10%; on intention-to-treat, 8.33%) experienced still slight symptoms. • About the time to obtain remission, it was obtained within 4 days in 7 of 11 patients (63.64%) and within 6 days in the other 4 patients (36.36%).	Study shows that the association BDP/ De Simone Formulation may be effective in treating acute mild to moderate diverticular colitis.
13	De Simone Formulation Probiotic-Mixture Induces Remission in Patients with Active Ulcerative Colitis. Bibiloni R. Am J Gastroenterol. 2005 Jul;100(7):1539-46.	**De Simone Formulation, 3,600 billion bacteria daily**	34 ambulatory patients with active UC received open label De Simone Formulation, 3,600 billion bacteria daily in two divided doses for 6 wk.	Intent to treat analysis demonstrated remission (UCDAI < or = 2) in 53% (n = 18); response (decrease in UCDAI > or = 3, but final score > or =3) in 24% (n = 8); no response in 9% (n = 3); worsening in 9% (n = 3); and failure to complete the final sigmoidoscopy assessment in 5% (n = 2). There were no biochemical or clinical adverse events related to DSF.	Treatment of patients with mild to moderate UC, not responding to conventional therapy, with De Simone Formulation resulted in a combined induction of remission/ response rate of 77% with no adverse events. At least some of the bacterial species incorporated in the probiotic product reached the target site in amounts that could be detected.
14	Once Daily High Dose Probiotic Therapy (De Simone Formulation) for Maintaining Remission in Recurrent or Refractory Pouchitis. Mimura T. Gut. 2004 Jan;53(1):108-14.	**De Simone Formulation 6 g once daily or identical placebo sachets.** **300 billion bacteria/g**	36 patients with Pouchitis • 20 patients to De Simone formulation • 16 to placebo	Remission was maintained at one year in 17 patients (85%) on De Simone Formulation and in one patient (6%) on placebo ($p<0.0001$). The IBDQ score remained high in the De Simone Formulation group ($p = 0.3$) but deteriorated in the placebo group ($p = 0.0005$).	The once daily high dose probiotic De Simone Formulation is effective in maintaining antibiotic introduced remission for at least a year in patients with recurrent or refractory pouchitis. This is associated with a high level of quality of life.

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15	Low dose balsalazide plus a high potency probiotic preparation is more effective than balsalazide alone or mesalazine in the treatment of acute mild to moderate ulcerative colitis. Tursi A. Med Sci Monit. 2004 Nov;10(11):PI126-31.	**Group A : 2.25 g Balsalazide 750 mg daily, plus 3 g De Simone Formulation, daily 300 billion viable lyophilized bacteria per gram Group B : 4.50 g Balsalazide daily Group C : 2.4 g Mesalazine daily**	Ninety patients (30 per group) were randomly enrolled, with a treatment duration of 8 weeks.	• Balsalazide/ De Simone Formulation was significantly superior to balsalazide alone and to mesalazine in obtaining remission: 24 patients of group A were in remission [per-protocol: 85.71%, on intention-to-treat: 80%], while 21 group B [per-protocol: 80.77% on intention-to-treat: 77%] and 16 group C patients [per-protocol: 72.73% on intention-to-treat: 53.33%] were in remission ($p<0.02$). • Balsalazide with or without De Simone Formulation was better tolerated than mesalazine: two group C patients were withdrawn from the study because of severe side-effects; 1 group A (3.33%), 3 group B (10%) and 4 group C (13.33%) patients experienced slight side-effects. • The balsalazide/ De Simone ormulation combination was faster in obtaining remission than balsalazide alone or mesalazine (4, 7.5, and 13 days in groups A, B and C, respectively) and also better in improving all parameters evaluated.	Balsalazide/De Simone Formulation may be a very good choice in the treatment of active mild-to-moderate active ulcerative colitis instead of balsalazide alone or mesalazine.
16	Prophylaxis of pouchitis onset with probiotic therapy : a double-blind, placebo-controlled trial. Gionchetti P. Gastroenterology. 2003 May;124(5):1202-9.	**De Simone Formulation (900 billion bacteria/day)** **versus** **placebo**	40 consecutive patients who underwent ileal pouch-anal anastomosis for ulcerative colitis were randomized to receive either De Simone Formulation (n = 20) or an identical placebo (n = 20) immediately after ileostomy closure for 1 year.	Two of the 20 patients (10%) treated with De Simone Formulation had an episode of acute pouchitis compared with 8 of the 20 patients (40%) treated with placebo (log-rank test, $z = 2.273$; $P < 0.05$). Treatment with De Simone Formulation determined a significant improvement in Inflammatory Bowel Disease Questionnaire score, whereas this was not the case with placebo.	Treatment with De Simone Formulation is effective in the prevention of the onset of acute pouchitis and improves quality of life of patients with ileal pouch-anal anastomosis.
17	Oral bacteriotherapy as maintenance treatment in patients with chronic pouchitis : a double-blind, placebo-controlled trial. Gionchetti P. Gastroenterology. 2000 Aug;119(2):305-9.	**De Simone Formulation (5×10^{11} per gram of viable lyophilized bacteria)** **Versus** **Placebo**	Forty patients in clinical and endoscopic remission were randomized to receive either De Simone Formulation, 6 g/day, or an identical placebo for 9 months.	• Three patients (15%) in the De Simone Formulation group had relapses within the 9-month follow-up period, compared with 20 (100%) in the placebo group ($P < 0.001$). • Fecal concentration of lactobacilli, bifidobacteria, and S. thermophilus increased significantly from baseline levels only in the De Simone Formulation -treated group ($P < 0.01$).	These results suggest that oral administration of this new probiotic preparation is effective in preventing flare-ups of chronic pouchitis.

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18	Expression of cytokines, inducible nitric oxide synthase, & matrix metalloproteinases in pouchitis: effects of probiotic treatment. Ulisse S. Am J Gastroenterol. 2001 Sep;96(9):2691-9.	**De Simone Formulation containing 300 billion viable lyophilized bacteria/g given p.o. b.i.d. (2 x 3 g/day) for 9 months**	Pouch biopsy samples were obtained from seven patients with Pouchitis before and after antibiotic and probiotic treatment.	• Tissue levels of tumor necrosis factor a increased ($p < 0.01$) in Pouchitis relative to uninflamed pouches and reduced after antibiotic and probiotic treatment. Also, interferon y and interleukin 1alpha (IL-1alpha) augmented in Pouchitis, but their increase did not reach statistical significance. The latter, however, were lower ($p < 0.05$) after treatment with the antibiotics and probiotics. • Tissue levels of IL-4 and IL-10 were unchanged in inflamed pouches and unaffected by antibiotic treatment. However, IL-10 increased ($p < 0.05$) after probiotic treatment. • Moreover, inflamed pouches had higher levels of inducible nitric oxide synthase and gelatinase activities, which decreased after treatment.	The ability of antibiotic and probiotic treatments to increase tissue levels of IL-10, at a higher level than those observed in control pouches, and to decrease, to levels present in control pouches, pro-inflammatory cytokine, inducible nitric oxide synthase, and matrix metalloproteinase activity may suggest a mechanism of action to explain the efficacy of this therapeutic regime in Pouchitis.
19	Impact on the composition of the faecal flora by a new probiotic preparation: preliminary data on maintenance treatment of patients with ulcerative colitis. Venturi A. Aliment Pharmacol Ther. 1999 Aug;13(8):1103-8.	**De Simone Formulation**	20 patients with ulcerative colitis, intolerant or allergic to 5-ASA, have been treated with a new probiotic preparation De Simone Formulation containing 5x10(11) cells/g. Two doses of 3 g were administered o.d. for 12 months.	• Faecal concentrations of treptococcus salivarius ssp. thermophilus, lactobacilli and bifidobacteria increased significantly in all patients, compared to their basal level, from the 20th day of treatment ($P<0.05$) and remained stable throughout the study. • Concentrations of Bacteroides, clostridia, coliforms, total aerobic and anaerobic bacteria did not change significantly during treatment (P = N.S.). • Fifteen of 20 treated patients remained in remission during the study, one patient was lost to follow up, while the remaining relapsed.	Results show that this probiotic preparation is able to colonize the intestine, and suggest that it may be useful in maintaining the remission in ulcerative colitis patients intolerant or allergic to 5-ASA. No significant side-effects have been reported

IRRITABLE BOWEL SYNDROME

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1	Effectiveness and Safety of a Probiotic-Mixture for the Treatment of Infantile Colic : A Double-Blind, Randomized, Placebo-Controlled Clinical Trial with Fecal Real-Time PCR and NMR-Based Metabolomics Analysis. Baldassarre ME. Nutrients. 2018 Feb 10;10(2).	**effectiveness and the safety of a probiotic-mixture DeSimone Formulation®; Danisco-DuPont, Madison, WI, USA) for the treatment of infantile colic in breastfed infants, compared with a placebo**	A randomized, double-blind, placebo-controlled trial was conducted in 53 exclusively breastfed infants with colic, randomly assigned to receive a probiotic-mixture or a placebo for 21 days.	• Infants receiving the probiotic-mixture had less minutes of crying per day throughout the study by the end of treatment period (68.4 min/day vs. 98.7 min/day; p = 0.001). • A higher rate of infants from the probiotic-mixture group responded to treatment (defined by reduction of crying times of ≥50% from baseline), on day 14, 12 vs. 5 (p = 0.04) and on day 21, 26 vs. 17 (p = 0.001). • A higher quality of life, assessed by a 10-cm VAS, was reported by parents of the probiotic-mixture group on day 14, 7.1 ± 1.2 vs. 7.7 ± 0.9 (p = 0.02); and on day 21, 6.7 ± 1.6 vs. 5.9 ± 1.0 (p = 0.001).	Administration of a probiotic-mixture appears safe and reduces inconsolable crying in exclusively breastfed infants. No adverse events were reported.
2	Symptom Severity Following Rifaximin and the Probiotic DeSimone Formulation® in Patients with Chronic Pelvic Pain Syndrome (Due to Inflammatory Prostatitis) Plus Irritable Bowel Syndrome. Vicari E. Nutrients. 2017 Nov 3;9(11):1208.	**Rifaximin** **vs.** **DeSimone Formulation®**	75 patients and control groups were prescribed treatment with Rifaximin, (200 mg, 2 tablets bid) for seven days per month for three months followed by a probiotic combination DeSimone Formulation® (450 × 109 CFU/day)	• In IIIa prostatitis patients, the total NIH-CPSI scores significantly ($p < 0.05$) decreased from a baseline mean value of 21.2 to 14.5 at Visit-3, as did all subscales, and in the IIIb the total NIH-CPSI score also significantly decreased (from 17.4 to 15.1). • Patients with IBS alone showed no significant differences in NIH-CPSI score. • At V3, significantly greater improvement in the IBS-SSS and responder rate were found in IIIa patients.	Our study revealed non-significant decreased cell proliferation and increased detoxification capacity after treatment with sulindac or De Simone Formulation /inulin.

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3	Diet Low in FODMAPs Reduces Symptoms in Patients with Irritable Bowel Syndrome and Probiotic Restores Bifidobacterium Species : A Randomized Controlled Trial. Staudacher HM. Gastroenterology. 2017;153:936-947.	**4 groups** **1. 27 receiving sham-diet/ placebo,** **2. 26 receiving sham diet/ probiotic,** **3. 24 receiving low FODMAP diet /placebo and** **4. 27 receiving low FODMAP diet/probiotic** **DeSimone Formulation® 2 sachets per day (11.95 log 10 bacteria) to be taken in the morning with cold food or fluid.**	104 patients with IBS Patients were randomly assigned to groups given counselling to follow a sham diet or diet low in FODMAPs for 4 weeks, along with a placebo or multistrain probiotic formulation.	• In the intention-to-treat analysis, a higher proportion of patients in the low FODMAP diet had adequate symptom relief (57%) than in the sham diet group (38%), although the difference was not statistically significant (P = .051). • In the per-protocol analysis, a significantly higher proportion of patients on the low FODMAP diet had adequate symptom relief (61%) than in the sham diet group (39%) (P = .042). • Total mean IBS-Severity Scoring System score was significantly lower for patients on the low FODMAP diet (173 ± 95) than the sham diet (224 ± 89) (P = .001), but not different between those given probiotic (207 ± 98) or placebo (192 ± 93) (P = .721) • Abundance of Bifidobacterium species was lower in fecal samples from patients on the low FODMAP diet (8.8 rRNA genes/g) than patients on the sham diet (9.2 rRNA genes/g) (P = .008), but higher in patients given probiotic (9.1 rRNA genes/g) than patients given placebo (8.8 rRNA genes/g) (P = .019).	In a placebo-controlled study of patients with IBS, a low FODMAP diet associates with adequate symptom relief and significantly reduced symptom scores compared with placebo. It is not clear whether changes resulted from collective FODMAP restriction or removal of a single component, such as lactose. Co-administration of the multistrain probiotic increased numbers of Bifidobacterium species, compared with placebo, and might be given to restore these bacteria to patients on a low FODMAP diet.
4	Change of Fecal Flora and Effectiveness of the Short-term DeSimone Formulation® Probiotic Treatment in Patients with Functional Constipation (FC). Kim S. J Neurogastroenterol Motil, 2015 Jan 1;21(1):111-20.	**Formulation® sachet contained 450 billion lyophilized bacteria**	30 patients Functional Constipation and 30 controls were enrolled. Fecal samples were obtained before and after DeSimone Formulation® intake (one sachet twice daily for 2 weeks) and flora were examined by RT-PCR.	• After taking DeSimone Formulation®, the fold differences in Lactobacillus, Bifidobacterium and Bacteroides species increased in controls (P = 0.022, P = 0.018, and P = 0.076), but not in FC. Mean Bristol scores and complete spontaneous bowel movements (CSBMs)/week increased significantly in FC after ingesting DeSimone Formulation® (both P < 0.001). • Relief of subjective CSBM frequency, stool consistency and abdominal bloating were reported in 70%, 60%, and 47% of patients. • After DeSimone Formulation® cessation, 44.4% of patients with symptom improvement experienced constipation recurrence mostly within one month.	Bifidobacterium and Bacteroides species might be quantitatively altered in Functional Constipation. A short-term DeSimone Formulation® treatment can improve clinical symptoms of Functional Constipation.

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5	Melatonin Regulation as a Possible Mechanism for Probiotic (DeSimone Formulation®) in Irritable Bowel Syndrome: A Randomized Double-Blinded Placebo Study. Wong R. Dig Dis Sci. 2015 Jan; 60(1):186-94.	**DeSimone Formulation®** **or** **Placebo**	42 IBS patients were randomly assigned to receive DeSimone Formulation® or Placebo for 6 weeks.	• Abdominal pain duration and distension intensity decreased significantly in the probiotic group, long with an increase in rectal distension pain thresholds. • A correlation between increase in pain tolerance and improvement in abdominal pain scores (r = 0.51, p = 0.02) was seen with probiotic. There was an increase in salivary morning melatonin levels in males treated with VSL#3, which correlated (r = 0.61) with improved satisfaction in bowel habits. • When grouped based on baseline diurnal melatonin levels, patients with normal diurnal fluctuations showed an increase in morning melatonin levels with DeSimone Formulation® treatment, which significantly correlated with improved satisfaction in bowel habits (r = 0.68). • They also had reduced symptom severity scores and abdominal pain duration when treated with DSF, as well as satisfaction with bowel movements and quality-of-life.	DeSimone Formulation® improved symptoms and increased rectal pain thresholds. Symptom improvement correlated with a rise in morning melatonin, significant in males and subjects with normal circadian rhythm. This suggests that probiotics may act by influencing melatonin production, hence modulating IBS symptoms, in individuals with a normal circadian rhythm.
6	Chronic bacterial prostatitis & irritable bowel syndrome : effectiveness of treatment with rifaximin followed by the probiotic DeSimone Formulation®. Vicari E. Asian J Androl 2014 Sep-Oct;16(5):735-9.	**Rifaximin Vs. DeSimone Formulation®** **Group A = "6Tx/6-": treatment for the initial 6 & no treatment for the following 6 months (n = 26); Group B = "12Tx": 12 months of treatment (n = 22); Group C = "6-/6Tx": no treatment for the initial 6 months and treatment in the last 6 months (n = 23); Group D = "12-": no treatment (n = 24).**	A total of 106 selected infertile male patients with bacteriologically cured chronic bacterial prostatitis (CBP) and irritable bowel syndrome (IBS) were randomly prescribed Rifaximin (200 mg, 2 tablets bid, for 7 days monthly for 12 months) and probiotic containing multiple strains DeSimone Formulation®. (450×10^9 FU per day) or no treatment.	• The patients of Groups A = "6Tx/6-" and B = "12Tx" had the highest frequency of chronic prostatitis (88.5% and 86.4%, respectively). In contrast, group "12-": patients had the lowest frequency of prostatitis (33.4%). • The progression of prostatitis into PV in groups "6Tx/6-" (15.5%) and "6-/6Tx" (13.6%) was lower than that found in the patients of group "12-" (45.8%). • Finally, no patient of groups "6Tx/6-" and "6-/6Tx" had PVE, whereas it was diagnosed in 20.8% of group "12-" patients.	Long-term treatment with Rifaximin and the probiotic DeSimone Formulation® is effective in lowering the progression of prostatitis into more complicated forms of male accessory gland infections in infertile patients with bacteriologically cured CBP plus IBS.

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7	Effect of probiotic bacteria on the intestinal microbiota in irritable bowel syndrome. Ng S. Journal of Gastroenterology and Hepatology 2013 Oct;28(10):1624-31.	**DeSimone Formulation® sachets contained 900 billion viable lyophilized bacteria**	10 IBS patients received open-label treatment with probiotic mix DeSimone Formulation® twice daily (total 1800 billion bacteria) for 4 Week.	• At week 4 of probiotic therapy, six patients showed symptom improvement on global symptom assessment compared with baseline (P = 0.031). • Before therapy, intestinal microbiota of IBS subjects differed significantly from that of healthy controls, with less diversity and evenness than controls (n = 9; P < 0.05), increased abundance of Bacteroidetes (P = 0.014) and Synegitestes (P = 0.017), and reduced abundance of Actinobacteria (P = 0.004). • The classes Flavobacteria (P = 0.028) and Epsilonproteobacteria (P = 0.017) were less enriched in IBS. • Abundance differences were largely consistent from the phylum to genus level. • Probiotic treatment in IBS patients was associated with a significant reduction of the genus Bacteroides (all taxonomy levels; P < 0.05) to levels similar to that of controls.	Global and deep molecular analysis demonstrates an altered mucosal microbiota composition in IBS. Probiotic leads to detectable changes in the microbiota. These effects of probiotic bacteria may contribute to their therapeutic benefit.
8	Gut Microbiota is Not Modified by Randomized, Double-Blind, Placebo-Controlled Trial of DeSimone Formulation® in Diarrhea-Predominant Irritable Bowel Syndrome. Michail S. Probiotics Antimicrob Proteins. 2011 Mar;3(1):1-7.	**DeSimone Formulation®** **or** **placebo**	Twenty four subjects were randomized to receive DeSimone Formulation® or placebo for 8 weeks.	• A favorable change in Satiety subscale was noted in the DeSimone Formulation® groups. • However, the consumption of the probiotic did not change the gut microbiota. • There were no adverse events or any safety concerns encountered during this study.	To summarize, the use of DeSimone Formulation® in this pilot study was safe and showed improvement in specific GSRS-IBS scores in diarrhea-predominant IBS subjects. The gut microbiota was not affected by DeSimone Formulation® consumption suggesting that the mechanism of action is not directly linked to the microbiota.

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9	DeSimone Formulation® Improves Symptoms in Children with Irritable Bowel Syndrome: A Multicenter, Randomized, Placebo-Controlled, Double-Blind, Crossover Study. Guandalini S. J Pediatr Gastroenterol Nutr. 2010 Jul;51(1):24-30.	**DeSimone Formulation® or Placebo Patients were randomized to receive either 1 sachet of DeSimone Formulation® (once per day for children 4–11 years of age; twice per day for those 12–18 years old) or an identical looking and tasting placebo for 6 weeks**	59 children 4 to 18 years of age, were enrolled and then randomized to receive either DeSimone Formulation® or a placebo for 6 weeks, with controls every 2 weeks.	• DeSimone Formulation® was significantly superior to it (P < 0.05) in the primary endpoint, the subjective assessment of relief of symptoms; as well as in 3 of 4 secondary endpoints: abdominal pain/discomfort (P < 0.05), abdominal bloating/gassiness (P < 0.05), and family assessment of life disruption (P < 0.01). • No significant difference was found (P = 0.06) in the stool pattern. • No untoward adverse effect was recorded in any of the patients.	DeSimone Formulation® is safe and more effective than placebo in ameliorating symptoms and improving the quality of life in children affected by IBS.
10	A randomized controlled trial of probiotic combination DeSimone Formulation® and placebo in irritable bowel syndrome with bloating. Kim HJ. Neurogastroenterol Motil. 2005 Oct;17(5):687-96.	**DeSimone Formulation®** **or** **placebo**	48 patients with Rome II IBS were randomized in a parallel group, double-blind design to placebo or DeSimone Formulation® twice daily (31 patients received 4 weeks and 17 patients 8 weeks of treatment).	• Treatment with DeSimone Formulation® was associated with reduced flatulence over the entire treatment period (placebo 39.5 +/- 2.6 vs DeSimone Formulation® 29.7 +/- 2.6, P = 0.011); similarly, during the first 4 weeks of treatment, flatulence scores were reduced (placebo 40.1 +/- 2.5 vs DeSimone Formulation® 30.8 +/- 2.5, P = 0.014). • Proportions of responders for satisfactory relief of bloating, stool-related symptoms, abdominal pain and bloating scores were not ifferent. • Colonic transit was retarded with DeSimone Formulation® relative to placebo (colon geometric center 2.27 +/- 0.20 vs 2.83 +/- 0.19, P = 0.05 respectively).	DeSimone Formulation® reduces flatulence scores and retards colonic transit without altering bowel function in patients with IBS and bloating.
11	A randomized controlled trial of a probiotic, VSL#3, on gut transit and symptoms in diarrhoea-predominant irritable bowel syndrome. Kim HJ. Aliment Pharmacol Ther. 2003 Apr 1;17(7):895-904.		25 patients with diarrhoea-predominant irritable bowel syndrome were randomly assigned to receive DeSimone Formulation® powder (450 billion lyophilized bacteria/day) or matching placebo twice daily for 8 weeks after a 2-week run-in period.	• Abdominal bloating was reduced (P = 0.046) with DSF [mean post-minus pre-treatment score, - 13.7; 95% confidence interval (CI), - 2.5 to - 24.9], but not with placebo (P = 0.54) (mean post- minus pre-treatment score, - 1.7; 95% CI, 7.1 to - 10.4). • With the exception of changes in abdominal bloating, DSF had no effect on other individual symptoms: abdominal pain, gas and urgency. • All patients tolerated DSF well.	DeSimone Formulation® appears to be promising in the relief of abdominal bloating in patients with diarrhoea-predominant irritable bowel syndrome. This is unrelated to an alteration in gastrointestinal or colonic transit.

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References :

1. Thursby E, Juge N. Biochem J. 2017;474(11):1823-1836. Published 2017 May 16. doi:10.1042/BCJ20160510
2. Holzapfel WH, Haberer P et al. Int J of Food Microbiol 1998; 41:85-101.
3. Orrhage K., Nord C.E. Drugs Exptl. Clin. Res. 2000; XXVI (3):95 111.
4. Cresci GA, Bawden E. Nutr Clin Pract. 2015 Dec;30(6):734-46. doi: 10.1177/0884533615609899.
5. Zhang YJ, Li S, Gan RY, Zhou T, Xu DP, Li HB. Int J Mol Sci. 2015;16(4):7493-7519.
6. Vyas U, Ranganathan N. Gastroenterol Res Pract. 2012;2012:872716. doi: 10.1155/2012/872716. Epub 2012 Sep 19. PMID: 23049548; PMCID: PMC3459241.
7. Valdes A M, Walter J, Segal E, Spector T D. BMJ 2018; 361 :k2179 doi:10.1136/bmj.k2179
8. Guinane CM, Cotter PD. Therap Adv Gastroenterol. 2013;6(4):295-308. doi:10.1177/1756283X13482996
9. DeGruttola AK, Low D, Mizoguchi A, Mizoguchi E. Inflamm Bowel Dis. 2016;22(5):1137-1150. doi:10.1097/MIB.0000000000000750
10. Salami M. Front Neurosci. 2021 Feb 11;15:613120. doi: 10.3389/fnins.2021.613120. PMID: 33642976; PMCID: PMC7904897.
11. Baptista LC, Sun Y, Carter CS, Buford TW. Front Nutr. 2020 Mar 11;7:17. doi: 10.3389/fnut.2020.00017. PMID: 32219095; PMCID: PMC7078157.
12. Flint HJ, Scott KP, Louis P, Duncan SH. Nat Rev Gastroenterol Hepatol. 2012 Sep 4;9(10):577-89. doi: 10.1038/nrgastro.2012.156. PMID: 22945443.
13. Gagliardi A, Totino V, Cacciotti F, et al. Int J Environ Res Public Health. 2018;15(8):1679. Published 2018 Aug 7. doi:10.3390/ijerph15081679
14. Gupta V, Garg R. Indian J Med Microbiol. 2009 Jul-Sep;27(3):202-9. doi: 10.4103/0255-0857.53201. PMID: 19584499.
15. Hill, C., Guarner, F., Reid, G. et al. Nat Rev Gastroenterol Hepatol 11, 506–514 (2014). https://doi.org/10.1038/nrgastro.2014.66.
16. Aponte M, Murru N, Shoukat M. Front Microbiol. 2020;11:562048. Published 2020 Sep 11. doi:10.3389/fmicb.2020.562048.
17. Gupta V, Garg R. Indian J Med Microbiol. 2009 Jul-Sep;27(3):202-9. doi: 10.4103/0255-0857.53201. PMID: 19584499.
18. Kuljit Singh, Tejinder Kaur, Alka Rao. Editor(s): P.V. Mohanan, In Woodhead Publishing Series in Biomaterials, Biomedical Product and Materials Evaluation, Woodhead Publishing, 2022, Pages 635-665, ISBN 9780128239667, https://doi.org/10.1016/B978-0-12-823966-7.00027-X.
19. Kareb O, Aïder M. Probiotics Antimicrob Proteins. 2019 Jun;11(2):348-369. doi: 10.1007/s12602-018-9427-6. PMID: 29732479.
20. Kechagia M, Basoulis D, Konstantopoulou S, et al. ISRN Nutr. 2013;2013:481651. Published 2013 Jan 2. doi:10.5402/2013/481651
21. Product Inserts. https://cdn.shopify.com/s/files/1/0736/3645/files/Visbiome_Cap_Unf_EX_Package_Insert_02_2022.pdf?v=1644249516
22. Kim et al. Neurogastroenterol Motil (2005) 17, 1-10.
23. Kim et al. Aliment Pharmacol Ther 2003; 17:895-904.
24. Wong et al. Dig Dis Sci. 2014.
25. Bazzocchi, G., Gionchetti, P., Almerigi, P.F., Amadini, C. Campieri, M. Digestive and Liver Diseases 2002; 34(2):S48-53.
26. Guandalini et al. JPGN. 2010;51:24-30.
27. Staudacher, H, et al. Gastroenterology 2017;153:936-947.
28. Ng S.C., Lam E.F., Lam T.T., Chan Y., Law W., Tse P., Kamm M.A., Sung J.J., Chan F.K., Wu J. Journal of Gastroenterology and Hepatology 2013; 28(10):1624-31. doi: 10.1111 jgh. 12306.
29. Brigidi P, et al. Res. Microbiol. 152 (2001) 735-741.
30. Kim SE, et al. J. Neurogastroenterol Motil. 2015 Jan 1, 21 (1) 111-20.
31. Michail S, Kenche H. Probiotics Antimicro. Prot. 2011 3:1-7.

Florimax™
Lactic Acid Bacteria & Bifidobacteria Capsules

32. Tursi et al. Med Sci Monit 2004; 10(11). Med Sci Monitor 2004;10(11).
33. Tursi, et al. American Journal Of Gastro. 2010.
34. Huynh et al. Inflam Bowel Dis. 2008.
35. Sood et al. Clinical Gastroenterology and Hepatology 2009;7:1202-1209.
36. Ng S.C., et al. Inflamm Bowel Dis. 2010.
37. Bibiloni et al. Am J Gastroenterology 2005;100:1539-1546.
38. Miele E, et al. Am J Gastroenterol. 2009;104:437-443.
39. Forbes A. et al. ESPEN guideline: Clinical Nutrition. (2017) 321-347.
40. Erratum: Jia et al. Medicine. 97; 51:e13792.
41. Selinger et al. J. of Hospital Infection. 84 (2013 159-165.
42. Gionchetti P, et al. Gastroenterology. 2000;119(2):305-309.
43. Gionchetti P, et al. Gastroenterology.2003b;124(5):1202-1209.
44. Mimura et al. Gut. 2004; 53:108-114.
45. Su et al. Gastroenterology. 2020;159 697-705.
46. Bischoff S.C. et al. ESPEN practical guideline: Clinical Nutrition. 39 (2020) 632-653.
47. Lamb et al. Gut. 2019,0:1-106.
48. Sood et al. Indian Journal of Gastroenterology 2019, 38(3)220-246.
49. Dhiman et al. Gastroenterology. 2014;147:1327-1337.
50. Agrawal A. et al., American Journal of Gastroenterology. 2012.
51. Mittal, V.V. et al, European Journal of Gastroenterology and Hepatology. 2011, 23:725-732.
52. Mouli, V. et al. Hepatology Research. 2014.
53. Lunia MK, et al. Clin Gastroenterol Hepatol. 2014 Jun;12(6):1003-8.
54. Dhiman et al. Gastroenterology. 2014;147:1327-1337.
55. Agrawal A. et al., American Journal of Gastroenterology. 2012.
56. Mittal, V.V. et al, European Journal of Gastroenterology and Hepatology. 2011, 23:725-732.
57. Mouli, V. et al. Hepatology Research. 2014.
58. Lunia MK, et al. Clin Gastroenterol Hepatol. 2014 Jun;12(6):1003-8.
59. Miele E, et al. Am J Gastroenterol. 2009;104:437-443.
60. Acharya C, Bajaj JS. Chronic Liver Diseases and the Microbiome-Translating Our Knowledge of Gut Microbiota to Management of Chronic Liver Disease. Gastroenterology. 2021 Jan;160(2):556-572.
61. Ghoshal, U. C., Gwee, K.-A., Holtmann, G., Li, Y., Park, S. J., Simadibrata, M., … Cohen, H. (2017). The role of the microbiome and the use of probiotics in gastrointestinal disorders in adults in the Asia-Pacific region - background and recommendations of a regional consensus meeting. Journal of Gastroenterology and Hepatology, 33(1), 57-69. doi:10.1111/jgh.13840.
62. Anjum, N., Maqsood, S., Masud, T., Ahmad, A., Sohail, A., & Momin, A. (2014). Lactobacillus acidophilus: Characterization of the Species and Application in Food Production. Critical Reviews in Food Science and Nutrition, 54(9), 1241-1251. doi:10.1080/10408398.2011.621169.
63. Selle, K.M. (2014). Encyclopedia of Food Microbiology | | LACTOBACILLUS | Lactobacillus acidophilus. , (), 412-417. doi:10.1016/B978-0-12-384730-0.00179-8
64. Matthew Bull, Sue Plummer, Julian Marchesi, Eshwar Mahenthiralingam, The life history of Lactobacillus acidophilus as a probiotic: a tale of revisionary taxonomy, misidentification and commercial success, FEMS Microbiology Letters, Volume 349, Issue 2, December 2013, Pages 77-87, https://doi.org/10.1111/1574-6968.12293.

Florimax™
Lactic Acid Bacteria & Bifidobacteria Capsules

65. Gomes, A. M. P., & Malcata, F. X. (1999). Bifidobacterium spp. and Lactobacillus acidophilus: biological, biochemical, technological and therapeutical properties relevant for use as probiotics. Trends in Food Science & Technology, 10(4-5), 139-157. doi:10.1016/s0924-2244(99)00033-3.
66. Hill D, Sugrue I, Tobin C, Hill C, Stanton C, Ross RP. The Lactobacillus casei Group: History and Health Related Applications. Front Microbiol. 2018 Sep 10;9:2107.
67. Jones, R.M. (2017). The Microbiota in Gastrointestinal Pathophysiology || The Use of Lactobacillus casei and Lactobacillus paracasei in Clinical Trials for the Improvement of Human Health. , (), 99-108. doi:10.1016/B978-0-12-804024-9.00009-4
68. Gobbetti, M., & Minervini, F. (2014). LACTOBACILLUS | Lactobacillus casei. Encyclopedia of Food Microbiology, 432-438. doi:10.1016/b978-0-12-384730-0.00180-4.
69. Behera SS, Ray RC, Zdolec N. Lactobacillus plantarum with Functional Properties: An Approach to Increase Safety and Shelf-Life of Fermented Foods. Biomed Res Int. 2018 May 28;2018:9361614. doi: 10.1155/2018/9361614. PMID: 29998137; PMCID: PMC5994577.
70. Corsetti, A. (2011). Encyclopedia of Dairy Sciences || Lactic Acid Bacteria | Lactobacillus spp.: Lactobacillus plantarum. , (), 111–118. doi:10.1016/B978-0-12-374407-4.00263-6.
71. Kleerebezem, M et al. "Complete genome sequence of Lactobacillus plantarum WCFS1" (2002).
72. Flórez AB et al. "Susceptibility of Lactobacillus plantarum strains to six antibiotics and definition of new susceptibility-resistance cutoff values“ (2006).
73. Mangell, P. "Adhesive capability of Lactobacillus plantarum 299v is important for preventing bacterial translocation in endotoxemic rats" (2006).
74. Teixeira, P. (2014). LACTOBACILLUS | Lactobacillus delbrueckii ssp. bulgaricus. Encyclopedia of Food Microbiology, 425-431. doi:10.1016/b978-0-12-384730-0.00177-4.
75. El Kafsi, H., Binesse, J., Loux, V. et al. Lactobacillus delbrueckii ssp. lactis and ssp. bulgaricus: a chronicle of evolution in action. BMC Genomics 15, 407 (2014). https://doi.org/10.1186/1471-2164-15-407.
76. Chichlowski M, Shah N, Wampler JL, Wu SS, Vanderhoof JA. Bifidobacterium longum Subspecies infantis (B. infantis) in Pediatric Nutrition: Current State of Knowledge. Nutrients. 2020 May 28;12(6):1581. doi: 10.3390/nu12061581. PMID: 32481558; PMCID: PMC7352178.
77. Bozzi Cionci N, Baffoni L, Gaggìa F, Di Gioia D. Therapeutic Microbiology: The Role of Bifidobacterium breve as Food Supplement for the Prevention/Treatment of Paediatric Diseases. Nutrients. 2018 Nov 10;10(11):1723. doi: 10.3390/nu10111723. PMID: 30423810; PMCID: PMC6265827.
78. Butta, Hena; Sardana, Raman; Vaishya, Raju; Singh, Kailash N; Mendiratta, Leena (2017). "Bifidobacterium: An Emerging Clinically Significant Metronidazole-resistant Anaerobe of Mixed Pyogenic Infections". Cureus. 9 (4): e1134. doi:10.7759/cureus.1134. ISSN 2168-8184. PMC 5418030. PMID 28480152.
79. Martinović A, Cocuzzi R, Arioli S, Mora D. Streptococcus thermophilus: To Survive, or Not to Survive the Gastrointestinal Tract, That Is the Question! Nutrients. 2020 Jul 22;12(8):2175.
80. Uriot, Ophélie; Denis, Sylvain; Junjua, Maira; Roussel, Yvonne; Dary-Mourot, Annie; Blanquet-Diot, Stéphanie (2017). Streptococcus thermophilus : From yogurt starter to a new promising probiotic candidate?. Journal of Functional Foods, 37(), 74-89. doi:10.1016/j.jff.2017.07.038.
81. Burton, J.P. (2017). The Microbiota in Gastrointestinal Pathophysiology || Common Organisms and Probiotics: Streptococcus thermophilus (Streptococcus salivarius subsp. thermophilus)., 165-169. doi:10.1016/B978-0-12-804024-9.00019-7.

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