Intestinal Flora

What Are Probiotics?

Probiotics are dietary supplements containing friendly bacteria. While the word "bacteria" usually has a negative association, certain species of intestinal microflora are considered beneficial. While we usually start life with a relatively healthy intestinal tract, the effects of infections, antibiotics, alcohol, stress and poor diet very often devastate healthy bacteria leading to constipation or diarrhea and a number of life threatening diseases. Supplementing our diets with an effective probiotics can reverse this trend, improve intestinal and overall health and help guard against disease. The intestinal microflora are important for maturation of the immune system, the development of normal intestinal morphology and in order to maintain a chronic and immunologically balanced inflammatory response. The microflora reinforce the barrier function of the intestinal mucosa, helping in the prevention of the attachment of pathogenic microorganisms and the entry of allergens.

Probiotics means "for life" and this name is now mostly used to refer to concentrated supplements of beneficial or good bacteria taken by humans and animals. A "probiotic", by the generally accepted definition, is a "live microbial feed supplement which beneficially affects the host animal by improving its intestinal microbial balance." Human beings, as all animals, play host to many types and high numbers of microbes. Microbes live on our skin, in our mouths, in women's vaginal tracts, and all the way through our gastrointestinal tract. In fact, in reference to the human body, it has been estimated that there are more microbes associated with it (about 10^{14}, or 100,000,000,000,000 bacterial cells) than there are human cells in it (about 10^{13}). In addition to this very large number of bacteria, there also is a very large diversity of bacteria. It has been estimated that more than 400 different species, or types, of bacteria make their homes on humans.

Friendly and *pathogenic* bacteria form a delicate and constantly changing balance as they compete to take hold and remain. Both friendly and pathogenic bacteria *can be* and *are* affected by changes in the intestinal environment. Bacterial infections, stress, traveling, antibiotic treatment, alcohol, poor diet and a number of other factors can and do disturb the delicate balance in our intestinal tract, often decreasing the number of beneficial bacteria while allowing an increase in pathogenic bacteria. Some common symptoms of this imbalance include flatulence, constipation and/or diarrhea. If this imbalance is left untreated the symptoms can become chronic, as in *Irritable Bowel Syndrome* (IBS), and can compromise the immune system and lead to other serious illnesses.

Lactobacillus (one of many friendly species of intestinal microflora) is considered an especially beneficial bacteria in its ability to aid in the breakdown of proteins, carbohydrates and fats in food, and help absorption of necessary elements and nutrients such as minerals, amino acids and vitamins required for humans and other animals to survive. Probiotic supplements contain the beneficial bacteria like Lactobacillus help to adjust this balance. Prevention of illness by a well-maintained microflora balance is accomplished by a method referred to as "competitive exclusion." That is, the friendly bacteria take up positions known as *enteric sites* therefore preventing the pathogenic bacteria from establishing itself and proliferating to become the predominant bacteria.

Surprisingly, despite this impressive list of *therapeutic* and *prophylactic* attributes, probiotics are not commonly part of the medical practitioner's arsenal of prescription drugs. Instead, probiotics are available from retail outlets, usually supermarkets, grocery and health food stores. The probiotics are available to the consumer as powders or tablets, but most commonly as milk-based products. The growth in the production of probiotics by the dairy industry means that it is now increasingly difficult to purchase yogurts that do not contain "probiotic" bacteria such as *Lactobacillus acidophilus*. There are consumers who hold a firm belief that their health is improved by regular consumption of a probiotic. It becomes almost an article of faith. Others are suspicious of the probiotic industry, and question the validity of the claims made in relation to health benefits.

History of Probiotics

Probiotics in the form of substances containing *lactobacillus, bifidobacterium* and *acidophilus* cultures have been used for centuries as food preservatives and natural sources to promote good human health without specific knowledge of their active ingredients or how they work. Lactobacillus was first identified by Pasteur (1845–1895) in France, the pioneer of modern microbiology. A real understanding of how probiotics function began when the Nobel Prize winning Russian physiologist, Metchnikoff (1845–1916), introduced his *intoxication theory*. He stated that the main cause of aging is "toxicants" formed by intestinal putrefaction and fermentation and suggested drinking beverages such as yogurt containing lactic acid bacteria would prevent aging. *Lactobacilli* suddenly attracted world attention. Since Metchnikoff, a lot of research and clinical trials have been made but his "Eternal Youth Theory" has repeated a cycle of disappearing for a time and then returning again to public notice with the issue never really being resolved in either a scientific or practical sense.

Recently, thanks to remarkable advances in microbiology and intestinal bacteriology, it has been made clear that certain lactobacilli, especially a *Lactobacillus* genus and a *Bifidobacterium* genus, have high mucus membrane chemical affinity, and play important roles in human health. Metchnikoff's assumption has been substantiated and even become common knowledge in the health sciences.

There are hundreds of papers published on many health benefits of probiotic cultures. Described in some of these publications are clinical studies designed to determine how probiotic cultures may influence a variety of health conditions. These are very complicated questions, and research is still actively being conducted to clarify the role of probiotics in human health.

What Do They Do?

Probiotic bacteria favorably alter the intestinal microflora balance, inhibit the growth of harmful bacteria, promote good digestion, boost immune function, and increase resistance to infection. People with flourishing intestinal colonies of beneficial bacteria are better equipped to fight the growth of disease-causing bacteria. Lactobacilli and bifidobacteria maintain a healthy balance of intestinal flora by producing organic compounds—such as *lactic acid, hydrogen peroxide*, and *acetic acid*—that increase the acidity of the intestine and inhibit the reproduction of many harmful bacteria. Probiotic bacteria also produce substances called bacteriocins, which act as natural antibiotics to kill undesirable microorganisms.

Immune function tends to decline with age. Twice daily supplementation with *Bifidobacterium lactis* (a particular strain of bifidobacteria) in milk was found in a double-blind trial to significantly enhance various aspects of immune function in a group of healthy elderly people. Benefits were apparent after only six weeks of supplementation. Yogurt has been purported to support immune function, due to its inclusion of lactic-acid bacteria. While *B. lactis* is a different organism than that found in yogurt, effects on immunity may be similar.

Regular ingestion of probiotic bacteria may help prevent vaginal yeast infection. A review of the research concluded that both topical and oral use of acidophilus can prevent yeast infection caused by candida overgrowth.

Diarrhea flushes intestinal microorganisms out of the gastrointestinal tract, leaving the body vulnerable to opportunistic infections. Replenishing the beneficial bacteria with probiotic supplements can help prevent new infections. The incidence of "traveler's diarrhea," caused by pathogenic bacteria in drinking water or undercooked foods, can be reduced by the preventive use of probiotics.

Most people associate lactobacilli with *L. acidophilus*, the most popular species in this group of probiotic bacteria. However, research shows that other Lactobacillus species may be beneficial as well. For example, *L. rhamnosus* and *L. plantarum* appear to be protective intestinal bacteria. They are involved in the production of several "gut nutrients," such as short-chain fatty acids, and the amino acids, *arginine, cysteine*, and *glutamine*. These beneficial bacteria may also help remove toxins from the gut and exert a beneficial effect on cholesterol levels. In a double-blind trial, administration of a preparation containing *L. plantarum* to people with acute pancreatitis reduced the number of complications severe enough to require surgery.

One probiotic, *Saccharomyces boulardii*, has prevented diarrhea in several human trials. Double-blind research studying critically ill patients found this strain of yeast to prevent diarrhea when 500 mg is taken four times per day. Probiotics are important in recolonizing the intestine during and after antibiotic use. Probiotic supplements replenish the beneficial bacteria, preventing up to 50% of infections occurring after antibiotic use. Probiotics also promote healthy digestion. Enzymes secreted by probiotic bacteria aid digestion. Acidophilus is a source of *lactase*, the enzyme needed to digest milk sugar, which is lacking in lactose-intolerant people.

Fructo-oligosaccharides (FOS) are naturally occurring carbohydrates that cannot be digested or absorbed by humans. They support the growth of bifidobacteria, one of the beneficial bacterial strains. Due to this effect, some doctors recommend that patients taking bifidobacteria also supplement with FOS. Several trials have used 8 grams per day. However, a review of the research has suggested that 4 grams per day appears to be enough to significantly increase the amount of bifidobacteria in the gut.

Some Bacteria and Yeasts Used as Probiotics

Bifidobacterium

Bifidobacteria are normal inhabitants of the human and animal colon. Newborns, especially those that are breast-fed, are colonized with bifidobacteria within days after birth. Bifidobacteria were

first isolated from the feces of breast-fed infants. The population of these bacteria in the colon appears to be relatively stable until advanced age when it appears to decline. The bifidobacteria population is influenced by a number of factors, including diet, antibiotics and stress. Bifidobacteria are gram-positive anaerobes. They are non-motile, non-spore forming and catalase-negative. They have various shapes, including short, curved rods, club-shaped rods and bifurcated Y-shaped rods. Their name is derived from the observation that they often exist in a Y-shaped or bifid form. The *guanine* and *cytosine* content of their DNA is between 54 mol% and 67mol%. They are *saccharolytic* organisms that produce *acetic* and *lactic acids* without generation of CO_2, except during degradation of *gluconate*. They are also classified as *lactic acid bacteria* (LAB). To date, 30 species of bifidobacteria have been isolated. Bifidobacteria used as probiotics include *Bifidobacterium adolescentis, Bifidobacterium bifidum, Bifidobacterium animalis, Bifidobacterium thermophilum, Bifidobacterium breve, Bifidobacterium longum, Bifidobacterium infantis* and *Bifidobacterium lactis*. Specific strains of bifidobacteria used as probiotics include *Bifidobacterium breve strain Yakult, Bifidobacterium breve RO7O, Bifidobacterium lactis Bb12, Bifidobacterium longum RO23, Bifidobacterium bifidum RO71, Bifidobacterium infantis RO33, Bifidobacterium longum BB536* and *Bifidobacterium longum SBT-2928.*

Lactobacillus

Lactobacilli are normal inhabitants of the human intestine and vagina. Lactobacilli are gram-positive facultative anaerobes. They are non-spore forming and non-flagellated rod or *coccobacilli*. The *guanine* and *cytosine* content of their DNA is between 32 mol% and 51 mol%. They are either *aerotolerant* or *anaerobic* and strictly fermentative. In the *homofermentative* case, glucose is fermented predominantly to lactic acid. Lactobacilli are also classified as *lactic acid bacteria* (LAB).

To date, 56 species of the genus *Lactobacillus* have been identified. Lactobacilli used as probiotics include *Lactobacillus acidophilus, Lactobacillus brevis, Lactobacillus bulgaricus, Lactobacillus casei, Lactobacillus cellobiosus, Lactobacillus crispatus, Lactobacillus curvatus, Lactobacillus fermentum, Lactobacillus GG (Lactobacillus rhamnosus or Lactobacillus casei* subspecies *rhamnosus), Lactobacillus gasseri, Lactobacillus johnsonii, Lactobacillus plantarum and Lactobacillus salivarus. Lactobacillus plantarum 299v* strain originates from sour dough. *Lactobacillus plantarum* itself is of human origin. Other probiotic strains of Lactobacillus are *Lactobacillus acidophilus BG2FO4, Lactobacillus acidophilus INT-9, Lactobacillus plantarum ST31, Lactobacillus reuteri, Lactobacillus johnsonii LA1, Lactobacillus acidophilus NCFB 1748, Lactobacillus casei Shirota, Lactobacillus acidophilus NCFM, Lactobacillus acidophilus DDS-1, Lactobacillus delbrueckii* subspecies *delbrueckii, Lactobacillus delbrueckii* subspecies *bulgaricus type 2038, Lactobacillus acidophilus SBT-2062, Lactobacillus brevis, Lactobacillus salivarius UCC 118* and *Lactobacillus paracasei subsp paracasei F19*.

Lactococcus

Lactococci are gram-positive facultative anaerobes. They are also classified as *lactic acid bacteria* (LAB). *Lactococcus lactis* (formerly known as *Streptococcus lactis*) is found in dairy products and is commonly responsible for the souring of milk. Lactococci that are used or are being developed as probiotics include *Lactococcus lactis, Lactococcus lactis* subspecies *cremoris* (*Streptococcus*

cremoris), *Lactococcus lactis* subspecies *lactis NCDO 712*, *Lactococcus lactis* subspecies *lactis NIAI 527*, *Lactococcus lactis* subspecies *lactis NIAI 1061*, *Lactococcus lactis* subspecies *lactis biovar diacetylactis NIAI 8 W* and *Lactococcus lactis* subspecies *lactis biovar diacetylactis ATCC 13675.*

Saccharomyces

Saccharomyces belongs to the yeast family. The principal probiotic yeast is *Saccharomyces boulardii*. *Saccharomyces boulardi*i is also known as *Saccharomyces cerevisiae Hansen CBS 5296* and *S. boulardii*. *S. boulardi*i is normally a nonpathogenic yeast. *S. boulardii* has been used to treat diarrhea associated with antibiotic use.

Streptococcus Thermophilus

Streptococcus thermophilus is a gram-positive facultative anaerobe. It is a cytochrome-, oxidase- and catalase-negative organism that is nonmotile, non-spore forming and homofermentative. *Streptococcus thermophilus* is an *alpha-hemolytic* species of the viridans group. It is also classified as a *lactic acid bacteria* (LAB). *Steptococcus thermophilus* is found in milk and milk products. It is a probiotic and used in the production of yogurt. *Streptococcus salivarus* subspecies *thermophilus type 1131* is another probiotic strain.

Enterococcus

Enterococci are gram-positive, facultative anaerobic cocci of the *Streptococcaceae* family. They are spherical to ovoid and occur in pairs or short chains. Enterococci are catalase-negative, non-spore forming and usually nonmotile. Enterococci are part of the intestinal microflora of humans and animals. *Enterococcus faecium SF68* is a probiotic strain that has been used in the management of diarrheal illnesses.

Where are they found?

Beneficial bacteria present in fermented dairy foods—namely live culture yogurt—have been used as a folk remedy for hundreds, if not thousands, of years. Yogurt is the traditional source of beneficial bacteria. However, different brands of yogurt can vary greatly in their bacteria strain and potency. Some (particularly frozen) yogurts do not contain any live bacteria. Supplements in powder, liquid extract, capsule, or tablet form containing beneficial bacteria are other sources of probiotics.

Probiotics have been used in connection with the following conditions: Diarrhea; Tooth decay (*Lactobacillus GG*); Vaginitis; Yeast infection; Canker sores; Crohn's disease (*Saccharomyces boulardii*); Eczema; Food allergies; HIV support (Saccharomyces boulardii); Immune function; Infection; Pancreatitis (acute) (*Lactobacillus plantarum*); Ulcerative colitis; Chronic candidiasis.

Reliable and relatively consistent scientific data showing a substantial health benefit. Contradictory, insufficient, or preliminary studies suggesting a health benefit or minimal health benefit. An herb is primarily supported by traditional use, or the herb or supplement has little

scientific support and/or minimal health benefit. People using antibiotics, eating a poor diet, or suffering from diarrhea are more likely to have depleted colonies of friendly bacteria.

Mechanism Of Action

Lactobacillus plantarum 299v, which is derived from sour dough and which is used to ferment sauerkraut and salami, has been demonstrated to improve the recovery of patients with enteric bacterial infections. This bacterium adheres to reinforce the barrier function of the intestinal mucosa, thus preventing the attachment of the pathogenic bacteria to the intestinal wall. *Bifidobacterium breve* was found to eradicate *Campylobacter jejuni* from the stools of children with enteritis, although less rapidly than in those treated with erythromycin. *Lactobacillus GG* was found to eradicate *Clostridium difficile* in patients with relapsing colitis, and supplementation of infant formula milk with *Bifidobacterium bifidum* and *Streptococcus thermophilus* reduced rotavirus shedding and episodes of diarrhea in hospitalized children.

The antimicrobial activity of probiotics is thought to be accounted for, in large part, by their ability to colonize the colon and reinforce the barrier function of the intestinal mucosa. Probiotics, such as *Lactobacillus bulgaricus*, which do not adhere as well to the intestinal mucosa, are much less effective against enteric pathogens. In addition, some probiotics have been found to secrete antimicrobial substances. These substances are known as *bacteriocins*. Such a bacteriocin has been isolated from *Lactobacillus plantarum ST31*, a probiotic derived from sour dough. The substance was found to be a 20 amino acid peptide. A different bacteriocin was isolated from another strain of *Lactobacillus plantarum*. The bacteriocin has 27 amino acids and contains *lanthionine* residues. This type of bacteriocin is classified as a *lantibiotic*.

Lactobacillus casei has been demonstrated to increase levels of circulating *immunoglobulin A* (IgA) in infants infected with *rotavirus*. This has been found to be correlated with shortened duration of rotavirus-induced diarrhea. *Lactobacillus GG* has also been shown to potentiate intestinal immune response to rotavirus infection in children. *Lactobacillus acidophilus* and *Bifidobacterium bifidum* appear to enhance the nonspecific immune phagocytic activity of circulating blood granulocytes. This effect may account, in part, for the stimulation of IgA responses in infants infected with rotavirus. In healthy individuals, *Lactobacillus salivarius UCC118* and *Lactobacillus johnsonii LA1* were demonstrated to produce an increase in the phagocytic activity of peripheral blood monocytes and granulocytes. Also, *Lactobacillus johnsonii LA1*, but not *Lactobacillus salivarius UCC118*, was found to increase the frequency of interferon-gamma-producing peripheral blood monocytes. *Lactobacillus GG* has been shown to inhibit chemically induced intestinal tumors in rats. The probiotic appears to alter the initiation and/or promotional events of the chemically-induced tumors. *Lactobacillus GG* also binds to some chemical carcinogens.

Saccharomyces boulardii has been shown to prevent antibiotic-associated diarrhea and also to prevent diarrhea in critically ill tube-fed patients. The mechanism of this antidiarrheal effect is not well understood. *S. boulardii* has been found to secrete a protease which digests two protein exotoxins, *toxin A* and *toxin B,* which appear to mediate diarrhea and colitis caused by *Clostridium difficile*. The protective effects of *S. boulardii* on *C. difficile*-induced inflammatory diarrhea may, in part, be due to proteolytic digestion of toxin A and toxin B by a secreted protease.

Dietary antigens may induce an immunoinflammatory response that impairs the barrier function of the intestine, resulting in aberrant absorption of intralumenal antigens. This may account, in part, for food allergies. Probiotics that colonize the colon may be helpful in the management of some with food allergies by reinforcing the barrier function of the intestinal mucosa. *Lactobacillus rhamnosus GG* and *Bifidobacterium lactis Bb12* were found to produce significant improvement of atopic eczema in children with food allergies. The decrease in the signs and symptoms of atopic eczema occurred in parallel with a reduction in the concentration of circulating *CD4+ T lymphocytes* and an increase in *transforming growth factor beta1* (TGF-beta1), indicating suppressive effects on T cell functions in this disorder. These probiotics may help restore the *Th1/Th2* balance in atopic eczema.

Lactobacillus GG was found to scavenge *superoxide anion radicals*, inhibit lipid peroxidation and chelate iron in vitro. The iron chelating activity of *Lactobacillus GG* may account, in part, for its antioxidant activity. Other lactic acid bacteria, including strains of *Lactobacillus acidophilus, Lactobacillus bulgaricus, Bifidobaterium longum* and *Streptococcus thermophilus,* have also demonstrated antioxidative ability. Mechanisms include chelation of metal ions (iron, copper), scavenging of reactive oxygen species and reducing activity.