Proven Therapeutic Benefits of High Quality Probiotics

BY ROBERT ROUNTREE, M.D.

ABSTRACT: Probiotics such as select Lactobacillus and Bifidobacterium species are microorganisms that improve intestinal microflora balance and confer health benefits. Probiotics have been used for many years to aid in restoring and maintaining a healthy intestinal balance in favor of healthful bacteria, which is essential in maintaining good health. A major consideration in the choice of probiotics is to choose a strain that possesses essential characteristics that enable it to survive and establish itself under the conditions encountered in the intestinal environment. Perhaps of even greater importance is the scientific evidence supporting its clinical efficacy. The L. acidophilus NCFM® strain has been the subject of over 50 research studies and is recognized for its ability to survive in the presence of bile or stomach acid and adhere to the intestinal mucosa. In addition, many health benefits of the L. acidophilus NCFM strain have been demonstrated, such as stimulation of immune function, cholesterol assimilation, and reduction of procarcinogenic fecal bacterial enzyme activity.

INTESTINAL MICROFLORA

The human host and its microflora constitute a complex ecosystem whose equilibrium serves as a remarkable example of reciprocal adaptation. Establishment of the intestinal microflora begins at birth and progresses throughout life. The gastrointestinal (GI) tract is the most densely colonized region of the human body, with approximately 10¹² bacteria per gram of contents in the large intestine.¹ This complex ecosystem of intestinal microflora is estimated to contain approximately 500 different bacterial species of anaerobes as well as facultative and aerobic organisms, constituting a highly active society of organisms that can have both beneficial and detrimental influences on the host.²

Some bacterial species that may colonize the gut are potentially harmful as a result of toxin production, mucosal invasion, or activation of carcinogens and inflammatory responses.² These pathogens can modify the function of the intestinal epithelium to enhance their penetration across this barrier, serving as a link between inflammatory diseases of the gut and systemic inflammatory disorders.^{2,3} Conversely, the indigenous, healthy intestinal flora provides resistance to colonization by these pathogenic microorganisms through several mechanisms:

- Strengthening the epithelial barrier, thereby preventing translocation of pathogenic bacteria
- Competing with pathogenic bacteria for attachment to epithelial cells
- Producing antimicrobial compounds
- Creating a local environment that is unfavorable for the growth of many pathogens
- · Enhancing the mucosal immune response to pathogens

In return for providing this defense, the indigenous microflora gain access to a nutrient-enriched, stable environment and thus maintains a symbiotic relationship with the host.

ASSAULTS ON HEALTHY MICROFLORA

There is a progressive increase in immune-mediated, gut-related health problems such as allergies and autoimmune and inflammatory diseases in Western society.² Many factors are theorized to contribute to a disruption of healthy bacterial balance and a decline in host defense mechanisms in the gut. These include medications such as gastric acid inhibitors or neutralizers, high fat/low fiber diets, and contaminated food and water. But perhaps the most prevalent cause of indigenous microflora disruption is the widespread and chronic use of antibiotics.^{4,5}

Consequences of Antibiotic Use

Because antibiotics do not distinguish between pathogenic and healthful bacteria, they can severely disrupt intestinal and urogenital microbial balance. Suppression of the indigenous microflora may lead to reduced colonization resistance and overgrowth of pre-existing, naturally resistant microorganisms, such as *Clostridium difficile*, which may cause diarrhea and/or colitis.⁴ For example, a recent study found that after a course of antibiotics, patients were 3 times as likely to report functional bowel complaints 4 months after treatment compared with patients who did not receive antibiotics.⁶ Additionally, by altering the microflora of the vagina, antibiotics can increase a woman's propensity for urinary tract infections (UTIs) and vaginal candidiasis.⁷ New colonization by resistant pathogens may also occur, thus promoting the emergence and spread of antibiotic-resistant strains.⁵

The dramatic overuse and misuse of antibiotics and other antimicrobials has led to a global emergence of antibiotic resistance. To illustrate, 70% of bacteria that cause hospital-acquired infections are resistant to at least one commonly used antibiotic, and some organisms are resistant to all approved antibiotics and must be treated with experimental drugs with potential toxicity.⁸ More than 90% of *Staphylococcus aureus* are resistant to penicillin and related antibiotics, and the incidence of vancomycin-resistant enterococci in the U.S. increased 20-fold from January 1989 to March 1993.⁹ Community-acquired antibiotic resistance is also a growing problem, as evidenced by the fact that in some communities up to 40% of *Streptococcus pneumoniae* strains are now resistant to penicillin.⁹ Additionally, there is an increasing prevalence of resistance to several antibiotics used to treat UTIs.¹⁰

A well-balanced intestinal microflora reduces the risk of infections in the gut and urogenital tract, and thus is an important factor in reducing the need for antibiotics. Furthermore, a healthy microflora can prevent establishment of resistant strains. In this context, it is easy to see that if the microflora balance and colonization resistance of the host could be maintained at a healthy, normal level, the risk of emergence and spread of resistant strains could be mitigated. This may be accomplished through the therapeutic application of beneficial microorganisms, or *probiotics*.

THE USE OF PROBIOTICS TO PROMOTE INTESTINAL MICROFLORA BALANCE

Probiotics are viable microorganisms that alter the microflora of the host and exert beneficial health effects. The concept of using beneficial flora for disease treatment and prevention as well as health restoration and maintenance is not new: fermented dairy products such as yogurt and milk have been used for hundreds of years to promote health.¹¹ Today, the numerous health effects attributed to the use of probiotics are well documented and supported by modern science.^{23,11,29} These benefits include:

- Protection against infection by pathogenic bacteria
- Lower frequency and duration of diarrhea associated with antibiotic use
- Improvement of lactose intolerance
- Stimulation of immune function
- Decreased fecal concentrations of enzymes, mutagens, and secondary bile salts that may be involved in colon carcinogenesis

Other benefits of probiotics currently under study include reduction of allergic symptoms and atopic dermatitis, relief from constipation, irritable bowel syndrome (IBS), and inflammatory bowel disease, improvement in mineral absorption, and lowering of cholesterol.^{11,30-32}

THE IMPORTANCE OF PROBIOTIC SELECTION

The criteria for a microorganism to be defined as a probiotic include that the strain be of human origin, be safe for human use, be stable in acid and bile, and adhere to the intestinal mucosa.^{21,3,34} The genera most frequently used as probiotics are *Lactobacillus* and *Bifidobacterium*. While many lactobacilli strains are available for human use, relatively few have substantial scientific evidence that supports their effectiveness.^{21,34,35} Furthermore, many questions have been raised about reliability, viable content, and the general quality of many probiotic products.

The NCFM[®] Strain of L. acidophilus

The unique *L. acidophilus* NCFM strain, developed at North Carolina State University, was isolated from a human source in the early 1970s. It is perhaps the most extensively tested and proven probiotic strain available today, with over 50 research studies confirming its many health benefits.^{1215,19-29,31,35-44} Furthermore, DNA studies have revealed great genetic variability within strains that are designated as *L. acidophilus*. NCFM has been verified as a type A1 *L. acidophilus* strain, the only type of strain that should be classified as true *L. acidophilus*.¹⁷ Additional properties that are important in selecting a strain of

lactobacilli include the ability to adhere to the intestinal mucosa, resist acid and bile, and produce antimicrobial substances. The NCFM strain has been shown to not only meet these strict criteria, but also to survive and implant in the gut and produce beneficial effects.

• Intestinal Adherence

The ability to adhere to the intestinal mucosa is a critical selection criteria because adhesion is considered to be a prerequisite for colonization.³³ Adhesion is also considered important for stimulation of the immune system. The stomach and upper intestinal tract represent a hostile environment that can easily discourage growth or survival of microorganisms. Although many lactobacilli survive selective pressures of this environment, flow rates of digesta through the small intestine would wash out any organism unable either to multiply rapidly enough to avoid dilution or to maintain their residence by physical attachment to the intestinal epithelium.³⁶

Unfortunately, many lactobacilli products do not appear to live up to claims regarding human GI adherence.²¹ In fact, NCFM is one of only a limited number of strains that have actually been shown in vitro to adhere. It adheres to human intestinal cells and Caco-2 and mucus-secreting HT-29 cell culture systems.³⁶⁻³⁸ Compared with other lactobacilli assayed, the adherence of NCFM was high and stable at physiologically relevant pH values between 6 and 8.³⁸ Furthermore, an in vivo study showed that the NCFM strain, when consumed in milk, is capable of surviving and implanting in the human intestinal tract.³⁹ Highly significant increases in bile-resistant lactobacilli were seen in the group consuming *L. acidophilus* NCFM, indicating successful implantation.

• Acid and Bile Tolerance

To survive passage through the stomach and upper small intestine, probiotic strains must be able to tolerate the acidic and protease-rich environment of the stomach, and survive in the presence of bile acids.³³ Bile tolerance is considered to be an important characteristic of *L. acidophilus* that enables it to survive, grow, and exert its action in the lower small intestine and colon.^{31,33,40} Although the degree of bile tolerance required for maximum growth of the organism in the intestinal tract is not known, it is important to select an organism having a high degree of bile resistance.³⁵ There is a wide range in the ability of cultures to grow in the presence of bile.³⁵ Studies have shown that the NCFM strain is capable of growing in bile concentrations of up to 3%^{22,31,35,40,41} and an in vivo study demonstrated that the NCFM strain survives in the presence of human gastric juice.³⁷

• Antimicrobial Activity

The ability to competitively exclude or inhibit pathogenic intestinal bacteria through antimicrobial action is a major benefit attributed to *L. acidophilus*, and is an important consideration when selecting strains.⁴² These substances include growth-inhibiting metabolites such as organic acids and hydrogen peroxide, as well as bacteriocins, adhesion inhibitors, and a range of small antimicrobial substances.³³ For instance, the production of organic acids (lactic and acetic acid) reduces intestinal pH and thereby inhibits the growth of acid-sensitive bacteria, including many pathogenic species. The NCFM strain has demonstrated antagonistic activity against foodborne disease agents, *Staphylococcus aureus, Salmonella typhimurium*, enteropathogenic *E.coli*, and *Clostridium perfringens*, among others.^{20,21,23}

In addition to lactic and other acids, lactobacilli have the capacity to secrete numerous metabolites that kill pathogenic bacteria.^{20,45-48} A variety of antibacterial/anti-yeast substances have been isolated such as lactocidin, lactobicillin, lactobreven, and acidolin.^{20,45,49}

Bactericidal proteins with a more specific spectrum of antagonistic activity, termed bacteriocins, are produced by many strains of L. acidophilus and usually prove inhibitory to closely related species that may compete for the same ecological niche.^{17,24,35} It is important to select a culture of L. acidophilus that will be able to compete and grow well in the presence of similar bacteria.³⁵ The NCFM strain has been found to produce a bacteriocin, designated lactacin B, that has a range of activity against closely related lactobacilli such as L. bulgaricus and L. helveticus.²³ This provides NCFM an advantage in being able to establish and grow in the intestinal tract.^{23,35} Because many bacteriocins have been identified for lactobacilli, the practice of mixing L. acidophilus products together with other species groups (i.e., L. bulgaricus, L. leichmannii) may hinder therapeutic efforts due to beneficial strains being rendered innocuous.21,24

CLINICAL EVIDENCE OF HEALTH BENEFITS

Numerous studies have confirmed the many wide-ranging beneficial effects of probiotics, such as improving intestinal tract health, enhancing the immune system, and reducing symptoms of lactose intolerance. In addition, evidence is emerging that potential benefits of probiotics may include treatment and prevention of allergies and inflammatory diseases, cancer prevention, and a reduction in respiratory disease. Several of these clinical benefits are discussed below, with particular reference to the well-studied NCFM strain of *L. acidophilus*.

• Anti-Carcinogenicity

There is considerable interest in the role of the intestinal microflora in colon carcinogenesis and other forms of cancer.^{12,16,19,25,43} Bacterial metabolism of various dietary constituents results in the production of many compounds, some of which may be carcinogenic. Altering the composition of the intestinal flora with probiotics may have a major effect on the production rate of these compounds. Indeed, epidemiological studies indicate a correlation between regular consumption of fermented dairy products and low incidence of colon cancer.^{12,19,25,43}

Several studies have measured the activities of microbial enzymes that play a role in the conversion of procarcinogens to carcinogens, including β -glucuronidase, nitroreductase, and azoreductase.^{19,25,26,43} In a study with 7 human subjects, it was found that supplementing the diet with *L. acidophilus* NCFM for 1 month significantly reduced fecal β -glucuronidase and nitroreductase activities.¹⁹ In a similar study with 21 human subjects, reductions of 2- to 4-fold in the activities of the 3 fecal enzymes were observed during a 4-week period of NCFM supplementation.²⁵

Animal studies indicate that NCFM can delay the initiation of chemically induced colon cancer.^{12,16,27} For example, a recent controlled study evaluated the potential anti-cancer properties of NCFM with respect to preneoplastic lesions (aberrant crypt foci, or ACF) in the colon of mutagenized rats.¹² NCFM produced a dose-dependent inhibition (up to 39%) of induced colonic ACF formation and a significant reduction in β -glucuronidase activity, suggesting its potential use for lowering colon cancer risk.

• Small Bowel Bacterial Overgrowth

Patients with chronic kidney disease often suffer from small bowel bacterial overgrowth (SBBO), which is accompanied by production of toxic amines such as dimethylamine (DMA).^{17,28} These toxic amines cause general chronic renal failure symptoms as well as target organ dysfunction, especially in the brain.⁵⁰ In several controlled studies, *L. acidophilus* NCFM was administered to hemodialysis patients to determine if supplementation could modify SBBO, thereby reducing the toxic levels of amines in the blood.^{13,28} The patients treated with NCFM for at least 1 month showed significant reductions in serum DMA levels as well as levels of nitrosodimethylamine, a potent carcinogen. The researchers concluded that NCFM changed small bowel pathobiology by modifying metabolic actions of SBBO, reducing generation of toxins and carcinogens with no adverse side effects.

• Immune Function

Approximately 70% of the immune system is localized in the GI tract and the intestinal mucosa provides a protective host defense against the constant presence of antigens from food and microorganisms.^{3,51} The intestinal microflora provides important support for this defense barrier, as shown by increased antigen transport across the gut mucosa in its absence.⁵¹ In addition to strengthening the intestine's permeability barrier, probiotics have been shown to enhance immune responses, particularly immunoglobulin A, and thereby promote the intestine's immunologic barrier. For instance, B. animalis, L. acidophilus NCFM, and 2 other lactobacilli species were shown to protect immunodeficient mice colonized with Candida albicans from mucosal and systemic candidiasis and prolong survival.¹⁴ The numbers of C. albicans were significantly reduced in the alimentary tract and the probiotics modulated antibody- and cell-mediated responses to C. albicans, suggesting that they may have potential for the prevention and treatment of this fungal disease.

Probiotics have also been shown to stimulate non-specific host resistance to microbial pathogens and to control the balance of proinflammatory and antiinflammatory cytokines.⁵¹ Therefore, in addition to strengthening the intestinal barrier, probiotics may down-regulate hypersensitivity reactions and hold promise in the prevention and treatment of conditions associated with gut barrier dysfunction and sustained inflammatory responses, such as inflammatory bowel disease. For a complete discussion of gut restoration, refer to the Applied Nutritional Science Report entitled, *Gut Dysfunction and Chronic Disease: The Benefits of Applying the 4R GI Restoration Program.*

Assimilation of Cholesterol

It has been reported that *L. acidophilus* may play a role in reduced serum cholesterol levels in humans and animals.^{31,32} Certain strains of *L. acidophilus*, including NCFM, have the ability in vitro to assimilate cholesterol from a laboratory growth medium, although with wide variation.^{32,35} Among 13 isolates of *L. acidophilus*, the NCFM strain ranked first in cholesterol assimilation over 16 hours of growth. Furthermore, NCFM exhibited cholesterol uptake in the presence of bile and in the absence of oxygen, conditions that exist in the intestinal tract. Such assimilation of cholesterol in the small intestine may be important in reducing the absorption of dietary cholesterol from the digestive system into the blood.^{31,32}

THE HEALTH BENEFITS OF BIFIDOBACTERIA

Bifidobacteria is another probiotic naturally occurring in humans, with *Bifidobacterium infantis* being the dominant

microorganism to colonize the intestines of breast-fed newborns. Research studies have documented several beneficial effects of bifidobacteria when given to infants, such as its effectiveness against a specific strain of enteropathogenic *E. coli*, in prevention of enteric infections, and in decreasing the growth of *C. albicans*.^{14,18,52}

Recent research into the immune-enhancing effectiveness of bifidobacteria in the elderly also shows great promise. For instance, in a study of 30 healthy elderly volunteers consuming either 5 billion or 50 billion *B. lactis* organisms daily for 3 weeks, an increase in total, helper, and activated T lymphocytes and natural killer cells was observed.⁵³ Subjects with poor pre-treatment immune responses showed the greatest positive change in immunity. In general, the 2 doses had similar effectiveness. The researchers suggest that supplementation of bifidobacteria in the elderly may be a natural, non-invasive way to resist the decline in cellular immunity associated with the aging process.

Acids produced by bifidobacteria include short-chain fatty acids (SCFAs) such as acetic, propionic, and butyric acids, as well as lactic and formic acids.^{52,54} These acids have an antimicrobial effect by lowering the pH of the colon, in addition to other inhibitory influences on bacterial cell growth. The most plentiful SCFA produced by bifidobacteria is acetic acid, which exerts a wide range of antimicrobial activity against yeasts and molds as well as bacteria.

PROBIOTICS: THE IMPORTANCE OF VIABILITY

For L. acidophilus and other probiotics to establish and exert benefits within the intestinal environment, the bacteria must be in a viable condition. The viability of the selected strain can be determined through acid and bile tolerance analyses. These tests determine the percentage of colony forming units per unit weight recovered following exposure to 0.3% oxgall bile and modified gastric juice for each batch produced. A high percentage, above 80%, indicates high viability of the organism in the GI tract.²¹ The importance of these analyses cannot be understated, as a study conducted in 1990 suggests that there are serious problems associated with some commercial probiotic preparations.5 Researchers analyzed 11 products claiming to contain L. acidophilus in powder, capsule, and tablet form for number and type of bacteria present. Only 2 of the products were found to contain L. acidophilus, while the remainder contained other Lactobacillus species. Problems with culture viability and contamination with Enterococcus and Clostridium were also found.

In addition to selecting viable strains, the method of packaging and storing of the product is important in maintaining viability.²¹ Temperature, moisture, light, and air can all adversely impact viability. These variables can be controlled through the use of containers that prevent entry of oxygen, moisture, and light. Most importantly, refrigeration of the product from the time of manufacture through delivery and storage is critical in ensuring the potency of the bacterial strains.

THE BENEFICIAL ROLE OF PREBIOTICS

A prebiotic is a non-digestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of desirable bacteria.^{11,54} Intake of prebiotics can significantly modulate the intestinal flora by increasing the number of specific bacteria. Prebiotics identified thus far include lactulose, inulin, and a range of oligosaccharides, which act as a food source for the indigenous healthful microflora.

One specific group of oligosaccharides that has attracted much interest is the fructooligosaccharides (FOS), which occur naturally in a variety of plants such as onion, asparagus, chicory, banana and artichoke. FOS are predominantly fermented by bifidobacteria, but can also be utilized by lactobacilli,⁴⁴ yielding SCFAs as fermentation products, which lower the pH of the colon and inhibit the growth of pathogenic bacteria.^{16,54} FOS have also been shown to improve absorption of essential minerals such as calcium, magnesium, zinc, and iron. In animal studies, FOS-feeding has been shown to prevent the development of anemia and osteopenia following gastrectomy.⁵⁶

Because of their influence on microflora growth, it has been suggested that the ability of probiotic bacteria to ferment oligosaccharides may be an especially important characteristic. A recent study analyzed multiple *Lactobacillus* and *Bifidobacterium* species for their ability to ferment FOS.⁴⁴ *B. infantis* and *L. acidophilus* NCFM proved to be strong fermenters, with final cell densities twice that of the non-fermenting *Lactobacillus* GG strain. Due to the potential synergy between pre- and probiotics, combinations of these ingredients are often referred to as *synbiotics*.

BIOACTIVE PROTEINS

Bioactive proteins are a class of supportive substances that, like prebiotics, may beneficially affect intestinal microflora balance through a variety of mechanisms.

• Immunoglobulins

Immunoglobulins (Ig) are protein molecules that function as antibodies against many pathogenic as well as potentially pathogenic microorganisms.⁵⁷ Secretory IgA is produced in abundance at mucosal surfaces and plays a major role in the immune defense of the intestine by preventing the absorption of, and/or by disposing of, microbial antigens. It also neutralizes viruses and, in the case of bacterial infections, blocks the attachment of pathogens to mucosal tissues and cells. Other immune proteins, such as IgM and especially IgG, may also be helpful because they are known to have remarkably similar specificities.

In human studies it has been shown that a concentrate of bioactive Ig from the milk of cows immunized with human rotavirus could provide passive immunity and prevent rotavirus gastroenteritis when added to an infant's diet.^{58,59} Similar results have been achieved in infants and adults against enteropathogenic *E. coli.*^{60,61} Bovine milk is a rich source of IgG and other immune proteins. Because the functionality of certain types of IgA and IgG are similar, IgG from milk can be considered a valuable nutritional contribution to the health of the intestinal tract.

Lactoferrin and Lactoperoxidase

Lactoferrin is an iron-binding glycoprotein produced during lactation and by epithelial cells at mucosal surfaces, where it functions as a prominent component of the first line of host defense against infection and inflammation.⁶²⁻⁶⁷ It possesses antimicrobial properties against bacteria, fungi, and several viruses including *Helicobacter pylori*, hepatitis C virus, herpes viruses, *E. coli*, *C. albicans, Streptococcus* species, *Staphylococcus epidermidis*, and *Pseudomonas aeruginosa*. Lactoferrin's antimicrobial activity was originally believed to be due to its ability to sequester iron and cause nutritional deprivation in susceptible organisms. However, other mechanisms are now known to contribute its host defense, such as the ability to directly damage bacterial membranes.^{62,63} Lactoferrin may also act as a potent antiinflammatory protein at local sites of inflammation including the respiratory and intestinal tracts.⁶⁴ It modulates the inflammatory process mainly by preventing the release of cytokines and regulating the proliferation of immune cells.

Lactoperoxidase (LP) is one of the most prominent enzymes in bovine milk and also occurs in human secretions such as saliva, tears, and milk.⁵⁷ It has no antibacterial activity itself but forms, with hydrogen peroxide and thiocyanate, a potent natural antibacterial system known as the LP-system. This system catalyzes the inactivation of a wide range of microorganisms such as *Staphylococcus aureus*, *Campylobacter jejuni*, which is recognized as a cause of acute enteritis, *E. coli*, *H. pylori*, and *Streptococcus*, *Salmonella*, and *Pseudomonas* species.⁶⁸⁻⁷¹ Some lactic acid bacteria are unaffected because they contain a "reversal enzyme," which prevents the antimicrobial activity of the LP-system.

CONCLUSION

Old age, antibiotics and other medications, and immunocompromised states can all contribute to a disruption of the indigenous microflora population, putting patients at increased risk for infection and disease. The use of probiotics offers an intervention with essentially no risk that may provide significant health benefits by stabilizing the intestinal microflora. When choosing a probiotic, it is important to select strains that fulfill the criteria that are required for establishment in the intestinal tract and have demonstrated clinical effectiveness. Prebiotics such as FOS and other supportive substances such as lactoferrin are also useful in supporting a healthy bacterial balance.

References

- 1. Klaenhammer TR. Probiotic bacteria: today and tomorrow. J Nutr 2000;130(2S Suppl):415S-16S
- 2. Isolauri E. Probiotics in human disease. Am J Clin Nutr 2001;73(supp1):114S-16S.
- Lu L, Walker WA. Pathologic and physiologic interactions of bacteria with the gastrointestinal epithelium. *Am J Clin Nutr* 2001;73(suppl):1124S-30S.
 Edlund C, Nord CE. Effect on human normal microflora of oral antibiotics for treatment of urinary tract infections.
- Jamin C, Nord CL. Encode on mann formation recorded of our antibiotodes to treatment or annualy uncer mechanis. JAminicol Chemother 2000;46(Suppl 1):41-48.
 Sullivan A, Edlund C, Nord CE. Effect of antimicrobial agents on the ecological balance of human microflora. Lancet
- Infect Dis 2001;1(2):101-14.
- Maxwell PR, Rink E, Kumar D, et al. Antibiotics increase functional abdominal symptoms. Am J Gastroenterol 2002;97(1):104-08.
- 7. Kurowski K, Ghosh R, Singh SK, et al. Clarithromycin-induced alterations in vaginal flora. Am J Ther 2000;7(5):291-95.
- U.S. Food and Drug Administration: Antibiotic resistance. (n.d.). Retrieved March 27, 2002, from http://www.fda.gov/oc/opacom/hottopics/anti_resist.html
 Cassell GH. Mekalanos J. Antibiotic resistance. JAMA 2001:285:601-05.
- Gupta K, Scholes D, Stamm WE. Increasing prevalence of antimicrobial resistance among uropathogens causing acute uncomplicated cystitis in women. JAMA 1999;281(8):736-38.
- Schrezenmeir J, de Vrese M. Probiotics, prebiotics, and synbiotics—approaching a definition. Am J Clin Nutr 2001;73 (suppl):361S-64S.
- Rao CV, Sanders ME, Indranie C, et al. Prevention of colonic preneoplastic lesions by the probiotic Lactobacillus acidophilus NCFM™ in F344 rats. Int J Oncol 1999;14(5):939-44.
- Dunn SR, Simenhoff ML, Ahmed KE, et al. Effect of oral administration of freeze-dried Lactobacillus acidophilus on small bowel bacterial overgrowth in patients with end stage kidney disease: reducing uremic toxins and improving nutrition. Int Dary J 1998;8:545-53.
- Wagner RD, Pierson C, Warner T, et al. Biotherapeutic effects of probiotic bacteria on candidiasis in immunodeficient mice. Infect Immun 1997;65(10):4165-72.
- Reid G. In vitro testing of Lactobacillus acidophilus NCFM as a possible probiotic for the urogenital tract. Int Dairy J 2000;10:415-19.
- Wollowski I, Rechkemmer G, Pool-Zobel BL. Protective role of probiotics and prebiotics in colon cancer. Am J Clin Nutr 2001;73(suppl):4518-55S.
- Sanders ME, Klaenhammer TR. The scientific basis of Lactobacillus acidophilus NCFM functionality as a probiotic. J Dairy Sci 84:319-31.
- Hoyos AB. Reduced incidence of necrotizing enterocolitis associated with enteral administration of Lactobacillus acidophilus and Bifidobacterium infantis to neonates in an intensive care unit. Int J Infect Dis 1999;3(4):197-202.
- Goldin BR, Swenson L, Dwyer J, et al. Effect of diet and Lactobacillus acidophilus supplements on human fecal bacterial enzymes. J Natl Cancer Instit 1980;64:255-61.
- Gilliland SE, Speck ML. Antagonistic action of *Lactobacillus acidophilus* toward intestinal and food borne pathogens in associative cultures. J Food Protection 1977;40(12):820-23.
- Schauss AG. Lactobacillus acidophilus: method of action, clinical application, and toxicity data. J Adv Med 1990;3(3): 163-78.
- Lin MY, Savaiano D, Harlander S. Influence of nonfermented dairy products containing bacterial starter cultures on lactose maldigestion in humans. J Dairy Sci 1991;74:87-95.
- Barefoot SF, Klaenhammer TR. Detection and activity of lactacin B, a bacteriocin produced by Lactobacillus acidophilus. Appl Environ Microbiol 1983;45:1808-15.

- Barefoot SF, Chen YR, Hughes TA, et al. Identification and purification of a protein that induces production of the Lactobacillus acidophilus bacteriocin lactacin B. Appl Environ Microbiol 1994;60:3522-28.
- Goldin BR, Gorbach SL. The effect of milk and Lactobacillus feeding on human intestinal bacterial enzyme activity. *Amer J Clin Nutr* 1984;39:756-61.
- Goldin BR, Gorbach SL. Alterations of the intestinal microflora by diet, oral antibiotics, and Lactobacillus: decreased production of free amines from aromatic nitro compounds, azo dyes, and glucuronides. J Natl Cancer Instit 1984;73:689-95.
- Goldin BR, Gorbach SL. Effect of Lactobacillus acidophilus dietary supplements on 1,2-dimethylhydrazine dihydrochloride-induced intestinal cancer in rats. J Natl Cancer Instit 1980;64:263-65.
- Simenhoff ML, Dunn SR, Zollner GP, et al. Biomodulation of the toxic and nutritional effects of small bowel bacterial overgrowth in end-stage kidney disease using freeze-dried *Lactobacillus acidophilus*. *Miner Electrolyte Metab* 1996;2:29:29.66.
- Kim GS, Gilliland SE. Lactobacillus acidophilus as a dietary adjunct for milk to aid lactose digestion in humans. J Dairy Sci 1983;66:959-66.
 Marteau PR, de Vrese M, Cellier CJ, et al. Protection from gastrointestinal diseases with the use of probiotics. Am J Clin
- Nutr 2001;73(suppl):330S-36S. 31. Walker DK, Gilliland SE. Relationships among bile tolerance, bile salt deconjugation, and assimilation of cholesterol by
- Charles Lactobacillus acidophilus J. Dairy Sci 1993;76:956-61.
 Gilliland SE, Nelson CR, Maxwell C. Assimilation of cholesterol by Lactobacillus acidophilus. Appl Environ Microbio.
- Sz. Olimand SE, Netson CK, Maxwen C. Assimination of chorestero by Lactobactitus actuophilus. Appl Environ Microbiol 1985;49:377-81.
- Tuomola E, Crittenden R, Playne M, et al. Quality assurance criteria for probiotic bacteria. Am J Clin Nutr 2001;73 (suppl):393S-98S.
- 34. Reid G. The scientific basis for probiotic strains of Lactobacillus. Appl Environ Microbiol 1999;65(9):3763-66
- Gilliland SE, Walker DK. Factors to consider when selecting a culture of Lactobacillus acidophilus as a dietary adjunct to produce a hypocholesterolemic effect in humans. J Dairy Sci 1990;73:905-11.
- 36. Kleeman EG, Klaenhammer TR. Adherence of Lactobacillus species to human fetal intestinal cells. J Dairy Sci 1982;65:2063-69.
- 37. Conway PL, Gorbach SL, Goldin BR. Survival of lactic acid bacteria in the human stomach and adhesion to intestinal cells. *J Dairy Sci* 1987;70:1-12.
- Greene JD, Klaenhammer TR. Factors involved in adherence of lactobacilli to human caco-2 cells. *Appl Environ Microbiol* 1994;60:4487-94.
 Gilliland SW, Speck ML, Nauyok DF, et al. Influence of consuming nonfermented milk containing *Lactobacillus*
- Andrew M., Spear M., Walker V. & Start Science of Commercial Control of Commercial Commercia Commercial Comm
- 40. Sanders ME, watter DC, watter DC, watter KM, et al. Performance of commercial cumules in huid mitk applications. *J Dury Sci* 1996;79:943-55.
- Klaenhammer TR, Kleeman EG. Growth characteristics, bile sensitivity, and freeze damage in colonial variants of Lactobacillus acidophilus. Appl Environ Microbiol 1981;41:1461-67.
- Klaenhammer TR. Microbiological considerations in selection and preparation of *Lactobacillus* strains for use as dietary adjuncts. J Dairy Sci 1982;65:1339-49.
- Goldin B, Gorbach SL. Alterations in fecal microflora enzymes related to diet, age, *Lactobacillus* supplements, and dimethylhydrazine. *Cancer* 1977;40:2421-26.
- Kaplan H, Hutkins RW. Fermentation of fructooligosaccharides by lactic acid bacteria and bifidobacteria. Appl Environ Microbiol 2000;66(6):2682-84.
- Shahani KM, Vakil JR, Kilara A. Natural antibiotic activity of Lactobacillus acidophilus and Bulgaricus II. Isolation of acidophilin from L. acidophilus. Cult Dairy Prod J 1977;12(2):8.
 Vincent JG. Veomett R. Rilev R. Antibacterial activity associated with Lactobacillus acidophilus. J Bact 1959;78:447-84.
- to: vincent 50, vooinet R, Ritoj R. Antioacertai activity associated with Eactobacturas actuophilas. 5 Daer
- 47. Sabine D. An antibiotic-like effect of Lactobacillus acidophilus. Nature 1963;199(5895):811.
- Dahiya RS, Speck ML. Hydrogen peroxide formation by lactobacilli and its effect on *Staphylococcus aureus*. J Dairy Sci 1968;51:1568-72.
 Shahani KM, Ayebo AD. Role of dietary lactobacilli in gastrointestinal microecology. Am J Clin Nutr 1980;33:2448-57.
- Simenhoff ML, Dunn SR. Altered gut flora in uremia. J Renal Nutr 1996;6:68-74.
- 51. Isolauri E, Sütas Y, Kankaanpää P, et al. Probiotics: effects on immunity. Am J Clin Nutr 2001;73(suppl):444S-50S
- 52. Rasic JLj, Kurmann JA. Bifidobacteria and Their Role. Boston: Kirkhauser Verlag; 1983.
- Gill HS, Rutherfurd KJ, Cross ML, et al. Enhancement of immunity in the elderly by dietary supplementation with the probiotic *Bifidobacterium lactis* HN019. *Am J Clin Nutr* 2001;74(6):833-39.
- Gibson GR, Roberfroid MB. Dietary modulation of the human colonic microbiota: introducing the concept of prebiotics J Nutr 1995;125:1401-12.
- Hughes VL, Hillier SL. Microbiologic characteristics of *Lactobacillus* products used for colonization of the vagina. Obstet Gynecol 1990;75:244-48.
- Ohta A, Ohtsuki M, Uehara M, et al. Dietary fructooligosaccharides prevent postgastrectomy anemia and osteopenia in rats. J Nutr 1998;128:485-90.
- 57. Guyton AC. Protein Metabolism. In: Textbook of Medical Physiology. 8th ed. Philadelphia: W.B. Saunders; 1991
- Brussow H, Hilpert H, Walther I, et al. Bovine milk immunoglobulins for passive immunity to infantile rotavirus gastroenteritis. J Clin Micro 1987;25(6):982-86.
- Hilpert H, Brussow H, Mietens C, et al. Use of bovine milk concentrate containing antibody to rotavirus to treat rotavirus gastroenteritis in infants. J Infect Dis 1987;156(1):158.
- Mietens C, Keinhorst H, Hilpert H, et al. Treatment of infantile E. coli gastroenteritis with specific bovine anti-E. coli milk immunoglobulins. Eur J Pedeatr 1979;132:239-52.
- Tacket CO, Losonsky G, Link H, et al. Protection by milk immunoglobulin concentrate against oral challenge with enterotoxigenic *Escherichia coli*. *NEJM* 1988;318 (19):1240-43.
- 62. Ward PP, Uribe-Luna S, Conneely OM. Lactoferrin and host defense. Biochem Cell Biol 2002;80(1):95-102.
- 63. Ellison RT 3rd. The effects of lactoferrin on gram-negative bacteria. Adv Exp Med Biol 1994;357:71-90
- 64. Conneely OM. Antiinflammatory activities of lactoferrin. J Am Coll Nutr 2001;20(5 Suppl):389S-97S.
- 65. van der Strate BW, Beljaars L, Molema G, et al. Antiviral activities of lactoferrin. Antiviral Res 2001;52(3):225-39.
- Ajello M, Greco R, Giansanti F, et al. Anti-invasive activity of bovine lactoferrin towards group A streptococci. Biochem Cell Biol 2002;80(1):119-24.
- Arnold RR, Brewer M, Gauthier JJ. Bactericidal activity of human lactoferrin: sensitivity of a variety of microorganisms. Infect Imm 1980;28(3):893-96.
- Shin K, Hayasawa H, Lonnerdal B. Inhibition of *Escherichia coli* respiratory enzymes by the lactoperoxidase-hydrogen peroxide-thiocyanate antimicrobial system. *J Appl Microbiol* 2001;90(4):489-93.
- Shin K, Yamauchi K, Teraguchi S, et al. Susceptibility of *Helicobacter pylori* and its urease activity to the peroxidasehydrogen peroxide-thiocyanate antimicrobial system. *J Med Microbiol* 2002;51(3):231-37.
 Borch E, Rosen C, Björck L. Antibacterial effect of the lactoperoxidase/thiocyanate/hydrogen peroxide system against
- Borch E, Rosen C, Bjorck E. Antibacterial effect of the actoperoxidase-inforvatiate hydrogen peroxide system against strains of campylobacter isolated from poultry. J Food Protect 1989;42(9):639-41.
- Thomas EL, Milligan TW, Joyner RE, et al. Antibacterial activity of hydrogen peroxide and the lactoperoxidase-hydrogen peroxide-thiocyanate system against oral streptococci. *Infect Imm* 1994;6(2):529-35.

Proven Therapeutic Benefits of High Quality Probiotics: A Summary

BY ROBERT ROUNTREE, M.D.

Your intestinal tract plays a vital role in your overall health, not only allowing life-supporting nutrients to be absorbed, but also providing the first line of defense as a physical and immune barrier to food antigens or microorganisms that you may ingest. Within this environment is a highly active society of approximately 500 different species of bacteria that can have both harmful and beneficial effects on your health.² While it is imperative for your overall health that the beneficial bacteria dominate, many factors can lead to an imbalance in favor of harmful bacteria, such as a poor diet, antibiotics, and contaminated food and water.⁵

A proliferation of unhealthy bacteria can damage your intestinal lining and lead to the production of carcinogenic compounds and intestinal symptoms such as bloating, gas, diarrhea, and abdominal pain.²³A damaged intestinal lining allows infectious agents, toxic compounds, and macromolecules to pass through to the bloodstream. Symptoms of this increased intestinal permeability, or "leaky gut syndrome," can include fatigue, diarrhea, and skin rashes. Ultimately, it can lead to many digestive disorders as well as seemingly unrelated illness, including chronic fatigue syndrome, eczema, migraine headaches, rheumatoid arthritis, and cancer.

Conversely, the healthy, or "friendly," microflora provide protection against these harmful bacteria by strengthening the intestinal lining, competing with harmful bacteria for attachment to epithelial cells, producing antimicrobial compounds, and enhancing the intestinal immune system. Thus, maintaining a well-balanced intestinal microflora is important for reducing the risk of infections and supporting overall health. This may be accomplished through the therapeutic use of beneficial microorganisms, or *probiotics*.

Probiotics: "Friendly" Bacteria That Promote Health

Probiotics are live, beneficial bacteria that help to reestablish a healthy microbial balance and exert health benefits. They can be supplied in supplement form (powder or tablets) and in foods such as yogurt and milk. The numerous health effects attributed to probiotics are well documented and supported by modern science:^{2,3,11-32}

- Control harmful bacteria and stimulate immune function
- Decrease side effects of antibiotic therapy, such as diarrhea
- Help control uro-genital infections
- Improve the digestion of lactose for those with lactose intolerance
- Decrease harmful activities of intestinal bacteria that may lead to cancer of the colon or other organs
- Assimilate cholesterol, thereby helping to lower blood cholesterol levels
- Improve the integrity of the intestinal barrier

Other benefits of probiotics currently under study include reduction of allergic symptoms and atopic dermatitis and relief from constipation, irritable bowel syndrome, and inflammatory bowel disease.^{11,30}

How to Choose a Probiotic Supplement

Selecting a high quality probiotic supplement is essential because it has been found that probiotic strains vary greatly in quality.⁹ A high quality probiotic should be a strain of human origin that is safe for human use, able to resist acid and bile, and capable of adhereing to the intestinal lining.^{21,3,34} The two most common health-promoting groups of bacteria are *Lactobacillus* and *Bifidobacterium* species, which includes the popular *L. acidophilus* species. Relatively few strains of *L. acidophilus* have substantial scientific evidence that supports their quality and effectiveness.^{21,34,35} One strain that does is the *L. acidophilus* NCFM* strain, which is perhaps the most extensively tested and proven probiotic strains available today. Over 50 research studies have confirmed the many beneficial properties of NCFM:^{12-15,19-29,35-44}

- Isolated from human flora
- Acid and bile tolerance—enables it to survive in the harsh intestinal environment
- Ability to adhere to the intestinal wall—enables it to establish and flourish
- · Bacteriocin production-helps it compete with other bacteria
- · Produces lactase enzymes, helping those with lactose intolerance
- Decreases the production of intestinal carcinogens that can lead to cancer of the colon or other organs
- Assimilates cholesterol in the small intestine

The Health Benefits of Bifidobacteria

Bifidobacteria is another probiotic naturally occurring in humans. Research studies have documented several beneficial effects of bifidobacteria when given to infants, such as prevention of intestinal infections.¹⁸ Recent research into the immune-enhancing effectiveness of bifidobacteria in the elderly also shows great promise and suggests that supplementation with bifidobacteria may be a natural, non-invasive way to resist the decline in cellular immunity associated with the aging process.⁵³ Bifidobacteria also produce short-chain fatty acids (SCFAs) that have an antimicrobial effect by lowering the pH of the colon, in addition to other inhibitory influences on bacterial cell growth.^{52,54}

The Importance of Viability

For probiotics to be effective, they must be viable, or live, organisms. This can be determined through laboratory analysis of acid and bile tolerance. In addition, the method of culturing, packaging, and handling of the product can make a huge difference in maintaining viability.²¹ Temperature, moisture, light, and air can all adversely impact the stability and potency of the bacterial strains. These variables can be controlled through the use of properly sealed containers and refrigeration from the time of manufacture through delivery and storage (both in the store and at home). Finally, the label on the product should identify exactly what bacteria are in the product and what level of live bacteria are guaranteed through the expiration date.

The Beneficial Role of Prebiotics and Bioactive Proteins

Prebiotics are substances that beneficially and selectively promote the growth and activity of desirable bacteria.^{11,54} As non-digestible carbohydrates, these substances act as a food source for "friendly" bacteria and include fructooligosaccharides (FOS) and inulin. Their breakdown also produces compounds that lower the pH of the colon and inhibit the growth of harmful bacteria.^{16,54}

Bioactive proteins are a class of supportive substances that, like prebiotics, may beneficially affect intestinal microflora balance through a variety of mechanisms. Some examples of bioactive proteins include: 1.) immunoglobulins, which function as antibodies that inactivate foreign bacteria, yeast, and molecules and prevent them from being absorbed⁵⁷ 2.) lactoferrin, an iron-binding protein that is speculated to play a role in the primary defense system against a wide variety of harmful organisms;⁶²⁻⁶⁶ and 3.) lactoperoxidase, an enzyme that combines with hydrogen peroxide and thiocyanate to suppress a wide range of harmful microorganisms.⁶⁸⁻⁷¹

Conclusion

Many factors can contribute to a disruption of the healthy microflora population, putting you at increased risk for infection and disease. The use of probiotics offers an intervention with essentially no risk that may provide significant health benefits by stabilizing the intestinal microflora. When choosing a probiotic, it is important to select strains that fulfill the criteria for establishment in the intestinal tract and have demonstrated clinical effectiveness. Prebiotics and bioactive proteins are also useful in supporting a healthy bacterial balance.