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Probiotic Bacteria
Capitalizing on probiotics and their potential as healthful food ingredients

By Mary Ellen Sanders

Bacteria in food? Many people are certain that this is something to avoid, and for good reason. The popular press coverage of deaths resulting from E. coli and the potential for food poisoning when we cross-contaminate our Thanksgiving gravy with raw turkey juices is quite convincing. However, better behaved bacteria may provide a very capable defense against troublesome bacteria.

Our bodies are colonized by upwards of 10 trillion microbes. Without them, evidence from germ-free animal studies suggests, our defense against pathogens would be paltry. Exposure to the fewest of pathogens could be lethal. These microbes help to activate our immune system and occupy niches in and around our body, making it more difficult for pathogens to take hold. In short, without these microbes, we would be vulnerable to microbial infection.

But the picture is not so simple. The estimated 400 species of microbes in our gastrointestinal tract are capable of an array of enzymatic activities, some of which might not be so good for us. Scientific evidence suggests that some microbes can intensify the carcinogenic or toxic potential of some intestinal compounds. So desirable bacteria are those with positive metabolic tendencies. Can we positively influence the bacteria that are present in our GI tract, or at least what they're doing? This brings us to the concept of probiotic bacteria, and their potential as healthful food ingredients.

The idea of "good" bacteria in foods is not new in the dairy industry. For thousands of years, the chance association of lactic acid bacteria with milk and the lactic acid (and other metabolites) they produced, provided a way to preserve milk and transform it into a plethora of nutritious, tasty cheeses and fermented milks. Some (but certainly not all) of these lactic acid bacteria are also normal residents of the human GI tract. In particular, some members of the genus Lactobacillus and Bifidobacterium are routinely isolated from intestinal contents. These bacteria are not majority components in adult human intestines, but constitute a stable, resident population. Interestingly, these bacteria are known for their low pathogenic potential and for the ability to produce antimicrobial products, such as lactic and acetic acids and bacteriocins that are thought to play a role in keeping harmful intestinal bacteria and their activities in check. In fact, these friendly lactic acid bacteria have been associated with many positive health effects (Table 2), and although more research is needed, early indications suggest a favorable role of these bacteria in intestinal health.

Worldwide, more and more products marketed for their healthful appeal include probiotic bacteria. In the United States, probiotic bacteria are often added to yogurts, but little effort is made to communicate a benefit or appropriate level of probiotic consumption to consumers. Concerns about limited research substantiating benefits and regulator obstacles likely contribute to this product positioning. Fresh, fluid milk products containing Lactobacillus acidophilus with or without Bifidobacterium strains have been sold in the United States since the mid 1970s, but sales are lackluster, comprising just 0.6 percent of the fluid milk market. More recently, regional products such as Land O' Lakes Plus 3, Stonyfield Farms yogurts, Lifeway Food's BasicsPlus, and Bona drinkable yogurt, are examples of more aggressively positioned products, formulated with a diverse array of probiotic species at high doses. But the United States still lags behind

Europe and Japan in probiotic-containing product development and promotion. Yakult, a Japanese company that produces a milk-based, sweetened, probiotic-containing beverage, sells more than 16 million units worldwide each day in dozens of countries. The United States is not one of them. Is there opportunity for U.S. dairy products manufacturers to capitalize on probiotics as ingredients for dairy products? To accomplish this goal, companies must develop a healthful product concept with consumer appeal. They must determine whether or not consumers respond better to messages about intestinal health or immune system modulation without sacrificing taste or convenience. Once this is achieved, careful attention must be paid to which strain or combination of strains of probiotic bacteria best accomplish this goal. Microbiological issues surrounding probiotic product development are many: strain selection, product compatibility, quality control verification and stability are among the most important. After a product concept is developed, product labeling issues must be resolved for compliance with regulatory requirements (e.g., Can a dairy product be sold as a dietary supplement as Lifeway Food's BasicsPlus is?). Product release must be accompanied by a consumer communications program so that consumers can stretch beyond the many negative perceptions of bacteria in foods and recognize the positive ones. And finally, directed research must be funded to address some of the fundamental questions still unanswered, such as:

* What level of what bacteria will cause what health benefits in which consumers?
* Can probiotic function in dairy products be improved?
* What are the mechanisms of health effects?
* What are the best strains for specific applications?
* What is the best means of selecting the right probiotic?
* What are the shelf-life limitations of probiotics in dairy foods?
* What are the best methodologies for monitoring probiotic levels in dairy products?
* What dairy product formulations and processes are compatible with probiotics?

In conclusion, a scientific basis for probiotic function exists, although basic and dairy product-focused research is still needed. Probiotic-containing dairy products are available worldwide, suggesting opportunity for the United States to expand this market for healthy dairy products.

Mary Ellen Sanders is an adjunct research professor at Cal Poly San Luis Obispo, where she conducts research on in vitro characterization of probiotic bacteria. She spends most of her time as a consultant for her own company, Dairy and Food Culture Technologies, in Littleton, CO.

Table 1
Glossary of Terms Related to Probiotics

* **Probiotic**

Oral probiotics are living microorganisms that exert health benefits beyond inherent basic nutrition. Common probiotic bacteria include Lactobacillus and "bifido" Bifidobacterium species.

* **Prebiotic**

Syn: colonic food. A carbohydrate not digested by humans that reaches the large intestine and serves as a substrate for growth for certain beneficial intestinal microbes. Examples include fructooligosaccharides and inulin.

* **Synbiotic**
A combination of probiotic bacteria and prebiotics that will encourage their growth once consumed.

* **Lactobacillus**
Commonly used genera for probiotic applications. Species used include Lactobacillus acidophilus, L. casei, L. reuteri, L. rhamnosus Bifidobacterium Commonly used genera for probiotic applications, especially favored in Japan. Previously classified as a Lactobacillus, but now recognized as its own genus. This bacterium is the dominant microbe in the intestine of breast-fed infants, although relative proportions drop with age.

* **Lactic Acid Bacteria**
A functionally related group of non-pathogenic, phylogenetically diverse bacteria that produce lactic acid as a primary metabolic endproduct from glucose and are often associated with food fermentations. Contains the genera Lactobacillus, Lactococcus, Streptococcus, some Enterococcus. Bifidobacterium is not always included in this group.

Table 2
Glossary of Terms Related to Probiotics

Activities of Probiotic Bacteria

* Intestinal health
* Anti-diarrheal effects
* Anti colon cancer effects
* Suppression of harmful intestinal microbe activities
* Immune system modulation
* Suppression of pathogen translocation
* Improved tolerance to milk
* Lactose intolerance
* Milk allergy
* Vaginal/urinary tract health
* Hypertension
* Cholesterol lowering

Producer Conference
Dairy Research investments explored

As in any business, dairy producers must continually update techniques and technologies to remain competitive. In the early 1980s, the California dairy industry, recognizing the need for research and development, began a tradition of investing in dairy research.

The results of this continued commitment to dairy research provided the focus of a recent one-day conference for the state's dairy producers. Titled the "California Milk Producer Research Conference: Report on Producers' Investments," the October 28 conference held in Visalia, Calif., was the second of its kind to bring producers and researchers together to discuss

developments in dairy research and how they will assist the future growth of California's dairy industry. A similar conference was held in 1993.

"Through their research investments California's dairy producers have enabled significant breakthroughs in the development of new technologies," said Joseph O'Donnell, executive director of the California Dairy Research Foundation (CDRF), sponsor of the conference. "These technologies will significantly help expand the use of dairy products in the food ingredients market."

Dairy producers heard presentations by leading dairy research scientists from the University of California, Davis, and California Polytechnic University, San Luis Obispo.

Speakers included Dr. Jim Cullor of UC Davis, who described Dairy Breakthrough Management (BTM), a new approach for managing animal production units with the interest of keeping California dairy cows productive; Dr. Mary Ellen Sanders of Cal Poly San Luis Obispo, who discussed the benefits of probiotics in creating and marketing healthy, tasty dairy products; and Cal Poly's Dr. Phillip Tong, who talked about ways of identifying potential markets and extending the use of milk powders in foods.

Cullor says that implementation of the BTM concept developed at UC Davis is reliant upon "the ability to identify and prevent problems before they occur by putting in place a series of critical control point management processes."

Sanders observed that the association of dairy products with probiotics, bacteria that can play a role in human intestinal health, has stimulated numerous research projects, including a study at Cal Poly San Luis Obispo to determine the genetic code of the probiotic Lactobacillus acidophilus chromosome. "Information obtained from this detailed knowledge of Lactobacillus genes will enable scientists to better understand how this bacterium influences humans, how to encourage its desirable characteristics and how to discourage its undesirable ones," Sanders explained. For the past two years, scientists in the Dairy Products Technology Center (DPTC) at Cal Poly San Luis Obispo have been working closely with the state's milk powder manufacturers in an effort to improve opportunities in the worldwide marketplace. Numerous research projects related to milk powders are under way, and the DPTC is planning to establish a dairy ingredients applications program to transfer information and technology to manufacturers and end-users of dairy ingredients, including non-fat and whole milk powders.

"These projects demonstrate how academia and industry can effectively collaborate on science and technologies," observed Tong. Additional presentation topics included the role of milkfat in nutrient delivery and absorption; development of consumer-pleasing low-fat cheeses; use of whey protein to create edible film coatings for protecting foods; development of specialty cheeses; and application of genetics to change milk composition in cows to meet market needs. Keynote speaker Dr. Bill Aimutis, director of research for Land O' Lakes, also offered a perspective on how dairy research is preparing the industry for the 21st century by answering the needs for healthier, safer and less expensive products.

According to Aimutis, "strong, technology-driven dairy companies will be able to deliver higher quality, value-added products to the market more quickly, thereby improving consumer satisfaction."

Event organizer John Bruhn, director of the Dairy Research and Information Center at UC Davis, was pleased with the results of the conference. "This is a meaningful step in connecting

producers and researchers," he said. "It's important for the future of dairy research for producers to understand how research affects them in order to prepare them for the emerging global economy."

Producers who were unable to attend the October conference can obtain a copy of the program's proceedings with detailed abstracts of the presentations by contacting Corinne Esser at the CDRF, (530) 753-0681 or by e-mail at esser@cdrf.org.

36th Marschall Cheese Seminar comes to the Golden State

The California Dairy Research Foundation (CDRF) and Rhodia Inc. will bring together cheese processors and suppliers in Santa Clara, Calif., Sept. 9 and 10 for the 36th annual Marschall Cheese Seminar.

The seminar, previously held in Wisconsin, is a forum for the introduction of new information and technology to stimulate growth in the domestic Italian and specialty cheese markets. The 1999 theme, "Gateway to Success," will focus on educating cheesemakers and maintaining the continuing popularity of Italian and specialty cheeses.

Joseph O'Donnell, executive director of the CDRF, believes the seminar is an important educational opportunity for cheesemakers. "By bringing the seminar to California, we hope to expose a wider audience to the growth potential in the specialty cheese market as well as generate excitement and sharing among all cheesemakers throughout the country," he said. For more information on this year's seminar please contact Jo Ann Sterenberg at (574) 264-2557 or visit the Marschall Web site at www.marschalliscs.com.

Partnerships
Rhodia Foods: New name but longtime player in the food ingredients arena

Over the past 15 years a series of acquisitions by French company Rhône Poulenc has resulted in one of the largest food ingredients suppliers in the world. The most recognizable U.S. dairy supplier, Marschall Products, was acquired by Rhône Poulenc in 1990. In the summer of 1998, Rhône Poulenc decided to separate its food and specialty chemicals group from its pharmaceuticals unit by setting up a company called Rhodia. Within Rhodia is Rhodia Food, a global entity encompassing four market areas-dairy, baking, convenience foods and meats.

Rhodia supplies the dairy industry with cheese and dairy cultures and media as well as a variety of functional ingredients (e.g. coagulants, natural colors and food protectants). Rhodia's food protectants, which are fermentation-derived and microbiology-oriented, are produced by the company's dairy ingredients group. Bill King, a member of CDRF's Technical Advisory Committee and a UC Davis food science graduate, heads up Rhodia's global business on food protection. "In the dairy industry, our MicroGARD product line protects about 40 percent of all of the cottage cheese made in the U.S. market," said King. "We've extended shelf life by an average of 10 days or more, allowing introduction of single-serving portions and reducing costly product returns."

Rhodia's bioprotectant products are also used in the dairy industry to prevent lactobacillus gassing in high-moisture, low-fat cheeses. The company is in the process of introducing long shelf-life technology for chocolate, strawberry and other flavored refrigerated milks. The technology is already being used widely in yogurt, sour cream and dips to control mold growth.

Another area in which Rhodia is making inroads is the emerging field of probiotics. In the 1980s Marschall Products commercialized the first sweet acidophilus, following its development in the late 1970s by Dr. Martin Speck at North Carolina State University. Through Marschall Products, Rhône Poulenc has had a long-term relationship with North Carolina State University, paying royalties to the North Carolina Dairy Research Foundation for acidophilus used in dairy products.

"Probiotics is a bet on the future for Rhodia," said King. "Very few people know what a probiotic is or what a culture is and whether or not it can be good." About five years ago, King participated in a focus group coordinated by the California Dairy Research Foundation (CDRF) to determine public response to probiotic cultures. "Through participation in that focus group I realized that the public is not really ready for or aware of the benefits of lactic cultures," said King, who says the industry needs to identify tangible benefits of consuming dairy products made with probiotic cultures before it can educate the public.

King notes that Europeans have a very different attitude toward consumption of probiotic cultures. "They believe that if they can get more culture in their diet, they'll be healthier, feel better and have better digestive processes," said King. "We clearly have to show the rest of the world that probiotics aid digestion or that they have some other long-term health benefit, and the CDRF is paving the way for that by supporting research toward that goal."

Dairy Management Inc. and CDRF are currently supporting probiotic projects at Cal Poly's Dairy Products Technology Center in San Luis Obispo and at North Carolina State University to understand the genetics and genome of probiotic cultures. Lead researchers Raul Cano at Cal Poly and Todd Klaenhammer at North Carolina State are collaborating very closely on their work in the area of probiotics. "The work at Cal Poly will give us an understanding of what genes are in these probiotic strains, and the North Carolina work will give us some tools so we can do scientifically controlled tests on measuring the benefits. That way we can track the effects of probiotics to specific genes and mechanisms and be very intentional about them in the future," explained King.

King is convinced that dairy products provide the best means for delivering live cultures to the digestive system. Milk acts as a buffer and protects cultures from stomach acids so it can be delivered to the intestines. Milk also provides nutritional support for the probiotic culture once it gets into the stomach. "If you take a pill, chances are less than 50 percent that the 'bugs' [cultures] will encounter the growth nutrients they need to proliferate in the stomach," said King. "That's why the dairy industry has a potential advantage if these bugs are proven to provide measurable health benefits."

King received his undergraduate degree in microbiology in 1977 and a master's degree in food microbiology in 1980 at the University of California, Davis. He then joined the Dairy and Food Labs (now Silliker DFL), where he developed new culture strains and testing technologies. When DFL sold the culture service to Rhône Poulenc, King came with the acquisition.

"California is a hotbed for innovation," said King. "But I think that California has been underrepresented at a national level. There is a lot of good technology being developed at the dairy research centers both in Davis and at Cal Poly, and I give a lot of credit to CDRF for helping to steer California's research efforts in the direction of value-added technology for the dairy industry."

Research Update: Researchers issue food safety cautions

Study shows chlorine sanitation is ineffective against cryptosporidiosis, but ice cream freezing provides protection

By Dean Cliver

Nearly six years have passed since the massive outbreak of 403,000 cryptosporidiosis cases in Milwaukee, Wisc. Although concern focused on contamination of the water supply, the incident required measures more encompassing than merely boiling the water.

Fearing that contamination by Cryptosporidium parvum oöcysts might have entered processing facilities through publicly supplied water, the U.S. Food and Drug Administration advised processors of several types of foods to withhold sales or even to recall their products. Oöcysts are the small (millionths of a meter), oval transmission form of Cryptosporidium parvum shed in the feces of humans and several other animal species. When ingested with food or water, oöcysts open in the small intestine and releases four sporozoites, which penetrate the cells lining the intestine and multiply. FDA officials who believed that oöcysts were capable of surviving exposure to chlorine in the sanitizing solution asked a large dairy processor to recall yogurt and ice cream, on the chance that oöcysts remained on the stainless steel contact surfaces and contaminated incoming milk or ice cream mix.

Because scientific evidence of that assertion was lacking, the California Dairy Research Foundation agreed to support investigation of oöcysts in my food safety laboratory at the UC Davis School of Veterinary Medicine. There my colleagues and I recreated conditions in the dairy processing facility, and soon demonstrated that C. parvum oöcysts in tap water readily withstood a solution of 5 percent laundry bleach and remained viable on stainless steel surfaces, from which they could transfer to water or milk.

Milk that had been contaminated with the oöcysts, inoculated with starter, incubated at 99°F for 48 hours, was then stored for eight days at 40°F. Following that eight-day storage period, about half of the inoculated oöcysts were still viable in the yogurt, suggesting that the contamination threat was real and that the company's agreement to recall the yogurt was wise.

The ice cream mix, however, proved easier to decontaminate. A 30-minute freezing cycle killed 80 percent of the inoculated oöcysts, and none could be found alive after overnight hardening at -4°F, whereas 8 percent of oöcysts frozen 24 hours in water at this temperature were still viable. That finding suggested that the company's decision against recalling their ice cream was correct. It now appears that C. parvum oöcysts occur at low levels in other public water supplies from time to time, so it is important to know that freezing and hardening of ice cream offer a critical control point to protect against consumer crypto-sporidiosis, whereas chlorine sanitation of milk contact surfaces and yogurt making do not. These results will appear soon in the International Journal of Food Microbiology.

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Use of herbs, decorative flowers and oils on specialty cheeses can create potential health hazards

By Linda Harris

Novel presentations of traditional foods are often used to stimulate consumer interest. Accordingly, marketers are reporting increasing popularity of soft, fresh or aged cheeses that are wrapped with leaves or fresh herbs and immersed in oil or are decorated with edible flowers. However, the visual appeal of these products disguises a number of potential safety issues that can arise during their production.

Identification of botanicals
The safety of these products is dependent upon careful verification by producers that only edible plant materials are being used. Precise determination must be made before production begins. Accurate identification of plant materials is very difficult once they are chopped or otherwise prepared. Producers have the responsibility to ensure that only edible plant materials are used for flavoring or decoration.

Pesticide residues and microbial contamination
Pesticide application and use for plant foods is well defined. Production procedures ensuring conditions rendering decorative botanicals acceptable for food use are of critical importance. Fresh plant materials can be a source of enteric pathogens and manufacturers of cheeses using these products should pay attention to growing conditions. The U.S. Food and Drug Administration recently published "Guidance for Industry: Guide to Minimize Microbial Food Safety Hazards for Fresh Fruits and Vegetables." These guidelines, available through the Federal Register, should also apply to fresh decorative botanicals. The guidance document provides suggestions for growing fresh fruits and vegetables under conditions that would minimize microbial contamination. I would suggest that cheese manufacturers examine their sources of fresh botanical to make sure they are grown using these guidelines.

Herbs, oils and the risk of botulism
Outbreaks of botulism have occurred with chopped garlic and oil mixtures as well as with roasted eggplant covered in oil. Evidence suggests that any low-acid plant material is capable of supporting growth of Clostridium botulinum, especially given the anaerobic conditions provided by the oil. While refrigeration provides some barrier to the growth of C. botulinum, manufacturers should not rely on refrigeration alone. Low-acid plant materials should be dried or properly acidified, following FDA guidelines, to below pH 4.6 before being used in oil-covered products.

The need for conclusive scientific evidence has prompted further study. Researchers at UC Davis in cooperation with the UC Laboratory for Research in Food Preservation in Dublin, Calif., are currently evaluating the risks of botulism associated with flavored or infused oils. The results of that study, funded by the California Division of Agriculture and Natural Resources, should be of paramount importance to dairy food processors.

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