DAIRY FOODS
Summary of Conclusions from a Consensus Panel of Experts on Health Attributes of Lactic Cultures: Significance to Fluid Milk Products Containing Cultures

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ABSTRACT
A panel of experts sponsored by the California Dairy Research Foundation was convened on January 31, 1992 to discuss the effect of the consumption of lactic cultures on human health. The panel was composed of 10 scientists with diverse applicable specialties. Topics discussed included lactose digestion, diarrheal diseases, chronic kidney disease, cancer, adherence, immune system stimulation, cholesterol reduction, constipation, and safety. Legitimacy of health claims and research needs for these areas were determined. The panel noted the promising results in the areas of positive effects of ingestion of lactic cultures on lactose digestion, some diarrheal illnesses, small bowel overgrowth associated with chronic kidney disease, and reduction of fecal enzymes that may play a role in colon cancer. However, additional research is necessary to confirm the effects in all of these areas. A coordinated research effort between microbiologists and clinicians is essential for the most effective research to ensure the choice of best available strains, the best conditions of analysis, and the best clinical models.

(Key words: panel, experts, health, lactic cultures)

INTRODUCTION
In attendance at the consensus panel were moderator, Mary Ellen Sanders (consultant); coordinator, Joseph O'Donnell [California Dairy Research Foundation (CDRF)]; panel members: Dwaine Savage (Department of Microbiology, University of Tennessee; expertise in intestinal microflora of humans and animals); Kent Erickson (Department Cell Biology, UC Davis, School of Medicine; expertise in dietary fat, breast cancer, immunity, and human gene regulation); Michael Levitt (Minneapolis Veterans Administration Hospital; expertise in clinical evaluation of lactose absorption); Dennis Savaiano (University of Minnesota; expertise in lactose digestion and fermented dairy foods); Robert Russell (Tufts University; expertise in aging and the gastrointestinal flora); Todd Klaenhammer (North Carolina State University; expertise in development of genetic systems in intestinal Lactobacillus strains and bacteriocins of lactobacilli); William Sandine (Oregon State University; expertise in starter cultures and the role of lactobacilli in cholesterol assimilation); Michael Simenhoff (Jefferson Medical College; expertise in the production of uremic toxins in the bowel, kidney failure, and the role of lactobacilli in alleviating the symptoms of these diseases); Rangne Fonden (Swedish Agricultural University, Sweden; expertise in dairy microbiology and special products fermented with Lactobacillus acidophilus); Anatoly Bezkorovainy (Rush-Presbyterian-St. Luke's Medical Center; expertise in bifidobacterial physiology and special interest in iron and other metals and metabolisms); observers, Adri Boudewyn (California Milk Advisory Board); Ron Bowen (Mid-Valley Dairy); Bob Boynton (Dairy Industry of California); Cal Crandall (California Milk Advisory Board); Kristen Dahl (California Department of Food and Agriculture); Brenda Hoeman (FDA); Bill

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King (Marshall Products); Matt Mathison (Sanofi Bio-Industries); Tom Rank (Chr. Hansen’s Laboratories, Inc.); Moshe Rosenberg [University of California, Davis (UC Davis)]; Jerry Scott (McCann-Erickson); Kent Sorrells (Alta Dena Dairy); Richard Tate (California Department of Food and Agriculture); Susan Taylor (Chr. Hansen’s Laboratories, Inc.); Phil Tong [California Polytechnic State University (Cal Poly)]; Doug Willrett (Marshall Products).

Background and Purpose

Joseph O’Donnell presented an overview of the California Dairy Foods Research Center (CDFRC) and its function in research activities for the California dairy industry. The CDFRC consists of three components: UC Davis, Cal Poly, and CDRF. The UC Davis focuses primarily on basic research, and Cal Poly focuses primarily on applications research; CDRF is the dairy industry’s representation in the CDFRC. The CDRF oversees the administration of the CDFRC and ensures that communication with all aspects of the dairy industry is facilitated. The mission of the CDFRC is to provide the California dairy industry the means to generate, to acquire, and to deliver the basic scientific information and applied technology needed by dairy producers and manufacturers to increase the marketability of their products.

The CDFRC Board establishes the priority research areas. The universities respond to these areas with proposals. Center subcommittees (both technical and industrial) review research project proposals and make recommendations to the Technical Advisory Committee. The Technical Advisory Committee discusses industry research priorities, reviews the subcommittee proposal recommendations, and makes final recommendations to the CDFRC Board. The Board then gives ultimate approval for funding of research projects.

The dairy industry, through the CDFRC Board and the subcommittees, has directed that scientific rationale be the basis for consumer benefit claims about dairy products. Therefore, the forum for the present panel meeting was established to review the scientific validity of health claims that have been attributed specifically to fluid milk products with added cultures. The conclusions from this meeting will be referred to the CDFRC industry subcommittee, the Fluid Milk Subcommittee, and ultimately to the CDFRC Board. Because the consumer trend is toward individualization, the dairy industry is positioning itself to respond to individual consumer needs by promoting a balanced diet and by providing a wide variety of products.

FORMAT

The panel was instructed to consider various health claims and to attempt to arrive at a consensus on the scientific validity of each health claim at present. The panel was requested also to give suggestions regarding research needs that may be pertinent to substantiate each claim further and to identify the necessary microbiological criteria needed by the culture to support the claim. Health claims were focused on the effect of ingested lactic cultures on lactose digestion, diarrheal diseases, chronic kidney disease, cancer, adherence, immune system stimulation, cholesterol reduction, and constipation. Audience participation was restricted until the question and answer period at the end of the day. Panel members and observers were provided a copy of a manuscript “Effect of Consumption of Lactic Cultures on Human Health” prepared by M. E. Sanders (Advances in Food and Nutrition Research 37:67). The panelists’ suggestions and the panel conclusions were considered before the review was submitted for publication. Following is a summary of discussions of possible health claims regarding addition of cultures to fluid milk products and the final consensus on claims.

SCIENTIFIC DISCUSSION AND CONSENSUS ON CLAIMS

Lactose Digestion

A sound body of information is available on the ability of fermented milk products to enhance lactose digestion. This information is less widespread for fluid milks containing cultures than for fermented milks and yogurts. Some published studies conclusively demonstrate an effect on alleviation of breath hydrogen symptoms with some fluid milk prepara-
tion containing cultures. The panel agreed that confirmatory studies were needed in more diverse populations and with more people.

Product application varies in different populations. R. Russell mentioned that *L. acidophilus* ADH is effective at decreasing breath hydrogen in aged populations who have low amounts of stomach acid (atrophic gastritis), although strain ADH has only been marginally successful in D. Savaiano's studies. Specific target subpopulations need to be defined and clinically tested.

The importance of symptoms of lactose intolerance was questioned. Frequently, biochemically lactase-deficient people may be symptom-free when they consume only limited amounts of lactose.

*Lactobacillus acidophilus* in general is bile-resistant, has low concentrations of lactase, and is therefore a poor enzyme deliverer, although strain differences exist. *Lactobacillus delbrueckii* ssp. *bulgaricus* and *Streptococcus salivarius* ssp. *thermophilus* have higher concentrations of lactase, are bile-sensitive, and are therefore much better for enzyme delivery applications. Strain criteria identified for lactose intolerance cultures were as follows.

- Populations of efficacious culture should be added at $10^7$ to $10^9$/ml in milk (concentrations depend on β-galactosidase activity and cellular permeability).
- Strains must be permeable so that lactase can be released. Although in vitro assays may predict this behavior, clinical studies need to be conducted to confirm this effect for specific strains.
- Enzyme survival in the stomachs of the elderly with low acid outputs and in stomachs of populations with normal gastric acidity should be identified.
- A standard for microbial concentrations of lactase needed for an effect is difficult to establish because the amount of lactase released into the gastrointestinal tract is dependent on the intrinsic lactase activity of the microbe and its bile sensitivity. How much lactase activity can be delivered in an available form to the intestine is more important than how much lactase is present in any particular strain.
- Studies indicate that, at a culture dose of $10^8$, optimization of intracellular bacterial lactase did not enhance clinical effects. At lower culture dose, optimized lactase is likely to be more important. However, the influence of a logarithmic increase of microbes compared with an arithmetic increase of enzyme activity must be considered.

Research needs for the area of lactose intolerance and health cultures were identified as follows.

- Identification of strains containing β-galactosidase resistant to bile acids is needed.
- Additional clinical studies with regard to the diversity of the population and the number of people studied should be obtained for confirmation of these results.
- Microbiological issues related to culture and enzyme survival should be researched. Culture conditions affecting β-galactosidase are important to define. The dependence of the physiological state of the microbe on media, growth phase, and storage conditions must be determined.
- The relative importance of lactobacilli versus *S. salivarius* ssp. *thermophilus* for lactase delivery should be determined.

Panel Consensus on Claims for Lactose Digestion

Claims acceptable to the panel on lactose digestion are listed.

- Consumption of *S. salivarius* ssp. *thermophilus* and *L. delbrueckii* ssp. *bulgaricus* enhances lactose digestion in lactase-deficient people, assuming that microbial enzyme concentration and stability are adequate.
- Lactose digestion can be improved in lactase-deficient subjects by consumption of some lactic cultures in milk instead of plain milk.
- An unfermented, culture-containing milk could be formulated to increase lactose digestion if the cultures used for this product contain suitable selected strains.

Diarrheal Diseases

The panel agreed that no published data exist to prove conclusively that culture-
containing milk controls diarrhea. Although some legitimate research has been done in this area, difficulties with identification of causes of diarrhea, lack of suitable numbers of subjects in the studies, strain selection, and antibiotic resistance of the therapeutic culture has made the body of available research in this area quite disjointed. Therefore, no confirmed health claim can be made in this area. Studies in Japan with bifidobacteria may be more conclusive, but, because many studies are published in Japanese or in journals that are not peer reviewed, the value of the results is difficult to assess.

Studies have been conducted pertaining to the control of side effects of antibiotic-associated diarrhea with yogurt products, but fluid milk products present specific obstacles. R. Russell noted that it may not be feasible to treat diarrhea associated with antibiotic treatment (including _Clostridium difficile_ diarrhea, because antibiotics are the treatment of choice) with cultures carried in milk. A milk delivery system for a culture targeted for diarrhea may not be prudent because temporary lactose intolerance may occur in the patient. The most marketable treatment would most likely use pill form. The panel agreed that diarrhea prevention (not cure) may be a more appropriate target for culture-containing milks.

Research needs for the control of diarrheal diseases include the following.

- The investment to determine properly the effect of lactic cultures on diarrhea would be monumental and is best performed at a dedicated clinic with those who are expert in recognizing diarrheal causes.
- Strains must be properly selected prior to clinical evaluation. Because many causes of diarrhea exist, different strains are appropriate for different applications.
- To screen candidate strains for potential in vivo antipathogenic activity (e.g., against enterotoxigenic _Escherichia coli_), two criteria could be used: 1) adherence in the best available in vitro assays and 2) in vitro production of bacteriocin-like inhibitory substances that may act against the pathogen in vivo. Although these in vitro activities are not necessarily indicative of in vivo efficacy, follow-up clinical studies are necessary to determine the usefulness of the selected strains and the in vitro assays.
- Research must recognize the different causes of diarrhea, including rotavirus, antibiotics, and _E. coli_, because not all may be reasonably affected by lactic cultures.
- Some organizations deliberately infect patients with enterotoxigenic _E. coli_. These patients may be a good target population for quick studies to determine an effect.
- A target of prevention is better for a milk delivery system. Research shows that cultures may have a positive effect on decreasing the duration and severity of diarrhea, which may also be indicative of positive effects on prevention. Use of milk as a carrier may be encouraged for these types of prophylactic effects. The optimal prophylactic culture may be mixed; different strains can be targeted toward different ailments and can be blended into one preparation.
- Studies should focus on diseases of the small bowel predominantly (populations of lactobacilli are relatively small in large bowel), e.g., rotavirus, _E. coli_, and traveler’s diarrhea.
- Continuous feeding is important for preventive effect.
- Pseudomembranous colitis caused by _C. difficile_ infection is a large bowel disease. This disease is also difficult to study because most cases are treatable by a second antibiotic. Cases are rarely persistent, and such subjects tend to be scattered. But pseudomembranous colitis is an important hospital problem. For this application, an antibiotic-resistant lactic culture is likely to be necessary.
- Cultures for certain applications should be resistant to therapeutic antibiotics (but sensitive to multiple others). Otherwise, the lactic cultures administered during therapy are likely to be killed. Antibiotic resistance is not important in a prevention application, however, in instances in which antibiotic therapy is not being used. Because the causative agents of many diarrheas are not determined, some bacterial diarrheas are not treated with antibiotics, and viral diarrheas should not be treated with antibiotics. Therefore, antibiotic resistance markers carried by therapeutic lactobacilli may only be necessary in very specific applications.
- Epidemiologic studies may give an indication of the likelihood of success of this research.

Panel Consensus on Claims of Alleviation of Diarrhea

Acceptable to the panel was the statement that, in certain subpopulations, properly selected live lactic cultures may help control the incidence, severity, or duration of diarrheal diseases, but more research needs to be done, especially concerning preventive effects of added lactic cultures.

Chronic Kidney Disease

Patients with end stage kidney failure almost universally have bacterial overgrowth of the small bowel. Both anaerobes and aerobes are involved in this overgrowth and are present at $10^6$ to $10^8$/ml of small bowel fluid. Patients with end stage kidney failure accumulate large amounts of toxic and carcinogenic compounds. Overgrowth correlates with presence of these toxic compounds in blood. Also, some abnormal encephalographic patterns emerge in these patients. The abnormal concentrations of bacteria in the small bowel likely produce these toxic metabolites. If unabsorbable antibiotics are administered against these bacteria, these blood compounds decrease, and encephalographic patterns improve. However, long-term administration of unabsorbable antibiotics is not feasible. Probiotic cultures were tested to control small bowel overgrowth and to prevent the accumulation of these toxins. Because people with end stage kidney failure have no urine, the volume required for adequate amounts of milk or yogurt precludes its use as a carrier for the culture. Therefore, a study was conducted by M. Simenhoff in which a freeze-dried, enteric-coated preparation of L. acidophilus NCFM was administered. Three patients were selected and were fed $2 \times 10^{10}$ cfu/d of this culture. Amines and nitrosamines decreased, and breath hydrogen concentrations improved. The breath hydrogen returned to prefeeding concentrations within 1 mo after culture feeding was stopped. The overgrowth may occur because of a depressed immune system in these patients or perhaps because of decreased intestinal motility. Treatment can begin earlier in the course of the disease so that accumulation of these toxins can be prevented. There is no indication of liver malfunction in these patients, but results are preliminary. Results are only available for three subjects, and the work has not yet been published as a refereed publication.

This model system may be very fruitful for future research. The system focuses on the small bowel, an area where significant effects are more likely to occur. Furthermore, because the efficacious microorganisms are associated with dairy products, positive results can be translated back to the dairy products later. Patients with end stage renal failure are not a large or a reasonable target group for culture-added milks. However, the research system for those patients may provide some quantifiable impact, and, if that can be done, then other studies may be built upon those results. If those results can be further substantiated with end stage patients, then more common conditions, such as diabetes, high blood pressure, heart disease, and intrinsic kidney disease, which may have similar associated symptoms, can be studied using a similar model. The importance of funding support for the potential opportunities of this study was noted.

The panel's conclusions on research needs for patients with small bowel overgrowth include the following.

- Studies should focus on the small bowel.
- Current projects should be continued and expanded to include less advanced patients who can tolerate milk as a culture carrier.
- Success with this focused model suggests a long-term approach to more general models that may be fruitful. This system gives quantifiable responses, so the model system gives good opportunity to study in depth the effects of cultures in human systems.
- This research should be expanded to include patients who have liver disease because bacterial overgrowth can be problematic in those patients. However, no data suggest that cultures have a role in this disease.
- Bowel motility and stasis may need to be explored further in patients with small bowel overgrowth.

Panel Consensus on Claims of Chronic Kidney Disease

At present, only preliminary scientific proof exists that freeze-dried lactic cultures can re-
duce bacterial toxins that result from small bowel overgrowth in patients with chronic kidney failure. More research is needed to confirm the effects in a larger number of subjects and to extend the findings to culture-containing milks.

Cancer or Antitumor Effects

The research area of lactic cultures and their effect on humans can be divided into four major areas: bacterial therapy, modulatory effects, epidemiology studies, and mutagen absorption.

Bacterial therapy involves injection of bacteria into tumors to provide a local modulatory effect. The panel agreed that this approach is 10 to 20 yr behind the use of other bacterial products. Also, approval of this type of experiment for humans is very unlikely. Therefore, this approach may not be relevant.

The modulatory effects of lactic cultures may be more fruitful. The strongest evidence for the role of lactic cultures in cancer is with affecting enzymes that may play a role in tumorigenesis. A few studies have been published that show a decline of carcinogen-producing fecal enzymes after subjects are fed with some lactic cultures. These studies are encouraging in that they have been repeated in several labs. The difficulties with these experiments are that 1) the physiological significance of the degree of enzyme inhibition is not known, 2) the role of these enzymes in the etiology of colon cancer is not substantiated, 3) the mechanism of action of the lactic cultures to decrease the enzyme concentrations is not known, and 4) reliable measurement of fecal enzymes is difficult. Although diet clearly influences colon cancer, it is supposition that the biochemical activity of the microbial flora is responsible for this. Therefore, work to study modulatory effects of lactic cultures is encouraging but is not sufficiently substantial to support an anticancer claim.

Epidemiological studies, although useful, are sparse. One study correlated the consumption of yogurt with incidence of breast cancer in The Netherlands. Although the correlation was negative between cancer incidence and yogurt consumption over 3 to 6 mo prior to diagnosis, because breast cancer develops slowly, diet over this period is unlikely to have any significant effect. This avenue of study can be defended, however, because generally speaking, fermented milk consumption is a pattern that is established for life. Therefore, patterns revealed by dietary recall over several months may be indicative of lifelong patterns. This assumption should be scientifically established in a publication and addressed in future substantiation of this research.

The work done on mutagen absorption was questioned by the panel. The in vitro assays have limited significance, because they are unlikely to occur physiologically. There is no evidence for any specificity in mutagen binding. However, R. Fonden discussed a small study that examined the levels of mutagens in urine and feces in 10 subjects fed fluid milk plus either milk fermented with *L. acidophilus* or mesophilic starters. A statistically lower level of mutagens was found in subjects consuming the *L. acidophilus* product, which provides some credibility to the possibility that this mechanism functions in vivo. However, the study was small, lacked some desirable controls, and needs to be repeated by others to gain acceptance by the panel. Skepticism was expressed over reports of "magic bullet" theories of delivery of agents to tumor cells. This topic was not discussed at any length.

The influence of lactic cultures on potential cancer-promoting enzymes may be greater in people with a degree of small bowel overgrowth. R. Russell noted that, in recent studies conducted with T. Klaenhammer, elderly patients with atrophic gastritis were given *L. acidophilus* ADH and yogurt. In the elderly without atrophic gastritis, azoreductase, nitroreductase, and β-glucuronidase were markedly reduced by 50 to 75%. For patients with the atrophic gastritis, reductions were even more profound, likely because of greater survival of the administered culture. Microbial survival may be a factor in people with other causes of small bowel overgrowth as well. In fact, M. Simenhoff has shown that nitrosamine was reduced in patients with bacterial overgrowth of the small bowel (from end stage kidney failure) that were fed a freeze-dried *L. acidophilus* preparation. This result supports the therapeutic efficacy of certain cultures in patients with bacterial overgrowth of the small bowel.

An epidemiological approach to the effects of cultures on cancer was recommended. Some
longitudinal studies in progress on diet and chronic disease in the elderly and middle-aged population can provide a means for testing these effects.

Research needs for cancer or antitumor effects were identified.

- Studies concerning Lactobacillus modulation of tumorigenesis should include the effects on primary tumor growth and metastasis.
- Patients with small bowel overgrowth, such as those with renal failure or achlorhydria or who are elderly should be used as models for the effect of lactic cultures on mutagen uptake and on levels of cancer-promoting enzymes.
- Animal studies should be done to gain an understanding of the mechanism of action of lactic cultures on the levels of these enzymes.
- Fecal enzyme studies should be repeated to gain consensus on results for specific strains and enzymes.
- The in vivo mutagen-binding study needs to be confirmed.
- Coordination is needed with the ongoing massive longitudinal studies, which are already being conducted on thousands of people to obtain epidemiological data for potential correlation of decreased fecal enzymes with cancer incidence or cultured dairy food consumption with cancer incidence. These studies may be conducted less expensively if they can be added onto existing studies.

**Panel Consensus on Cancer and Antitumor Effects**

A lactic culture can reduce certain fecal enzymes that may play a role in colon cancer. A lactic culture can bind mutagens in vitro. The physiological significance of this finding is not known.

**Adherence**

No evidence exists that lactobacilli adhere either to the human stomach or to the human small bowel. Organisms residing in the upper part of the gastrointestinal canal must colonize because the rate of passage of luminal content exceeds their rate of multiplication. In the colon and cecum of the human bowel, the rate of passage does not exceed replication rates of even some of the slowest anaerobes, which replicate with 10- to 12-h generation times. Those organisms can still multiply because lactobacilli have no trouble surviving as long as they find compatible physiological and nutritional conditions. A distinction was made between association (a loose, nonspecific attachment), replication (rates of which exceed colonic emptying), colonization (growth after adherence), and adherence (prior to colonization). Adherence may occur but is difficult to detect unless colonization also occurs. With colonization, replication ensues, and the chances of finding the microbe are increased. Colonization accounts for long-term persistence; otherwise, cells are completely sloughed off with time. Colonization may not be necessary if culture is regularly fed. Also, retention time can be increased strictly from association with the mucous (this may not strictly be adherence). Competitive exclusion is a mechanism that may be exerted by lactic cultures.

The involvement of microorganisms in colonizing the lower part of the small bowel was questioned. Evidence is mixed. The organisms in the small bowel are very representative of fecal flora, and they exist in the mucous and in association with the epithelium. No direct evidence exists that lactobacilli are adherent, but, if a strain were available that did associate with the mucous or the epithelium of the small bowel, its retention time could be enhanced, promoting likelihood of competitive exclusion. These are useful characteristics. Some advances in in vitro techniques for potential use to evaluate adherence have been made, such as the development of a mucous-secreting line of tissue culture cells, the CaCo-2 and HT29 cells, and actual scraping of intestinal cells from humans fed lactobacilli. Some in vivo techniques are also available. Biopsy of humans uses a Carey capsule or suction biopsy. Suction biopsy can disrupt the epithelium, making differentiation between adherence and association difficult. Carey capsule biopsy is better, but problems with small sample size and disruption of the mucous tag make this procedure less than ideal. All noninvasive, direct measurements of microbial adherence in humans can be criticized for inaccuracies.
herence of microbes in humans remains very difficult to study reliably. Perhaps the only way to approach this problem is a combination of the best available in vitro and in vivo techniques coupled with clinical efficacy studies. Adherence information on some intestinal pathogens is available because they are so adherent that results are more easily obtained. Furthermore, for diseases that carry a threat of mortality, autopsy data can confirm intestinal adherence. Even with difficulties of adherence assays, a properly controlled study, for example, with isogenic strains, one that adheres in vitro and one that does not, could provide a good indication of adherence. No adherence information on bifidobacteria exists. In conclusion, measurement of adherence is very difficult in humans; some direct measurements can be made but must be confirmed with clinical studies.

If adhering strains were administered, could they colonize an already fully colonized, adult human? Because the small intestine is so sparsely populated, it may be a site where an input of large numbers of live bacteria could more likely have an effect. Displacement of normal colonized bacteria in the large intestine is not likely. Whether a tenaciously adhering *Lactobacillus* is desirable or undesirable in a human system is not known.

The research needs for adherence were identified as follows.

- Correlate in vivo with in vitro adherence capability.
- Define adherence factors.
- Conduct competitive exclusion studies with adhering and nonadhering strains in in vitro model systems. These studies may not be very useful because studies to date suggest that the organism added first to these systems is the exclusionary strain. Therefore, the pathogen could just as likely exclude the lactic culture. This result is a justification for continuous feeding so that the fed microbe would initially occupy sites and potentially decrease pathogenicity of a subsequently ingested pathogen.
- Challenge humans with enterotoxigenic *E. coli* after administration of adhering *L. acidophilus* and quantify symptoms and adherence. Studies can be approved. Alternatively, once in vitro assays are verified, this study could be done with competitive challenge experiments in tissue culture.
- Determine whether adherence or transient passage is responsible for positive effects in lactase delivery, small bowel overgrowth, levels of cancer-promoting enzymes, and effects on intestinal pathogen populations.
- Use recombinant DNA techniques for genetic characterization of strains. Safety should be studied.
- Use the Carey capsule to verify whether lactobacilli adhere. Isogenic strains would ensure the best controlled study.
- Investigate colonization of the human large intestine by biopsy with focus on diseased humans (e.g., use studies of patients with pseudomembranous colitis).

**Panel Consensus on Adherence**

Adherence of ingested cultures may be necessary for some beneficial effects (except lactase delivery). Transient passage of ingested cultures is sufficient to elicit some healthful effects, assuming regular consumption. A dietary culture adjunct may achieve adherence in the small bowel of a healthy human, assuming suitable physiological characteristics. Confirming experiments must be done. In vitro assays may be useful for determining adherence characteristics of lactic cultures.

**Immune System Stimulation**

Research is too preliminary to make a claim about lactic culture stimulation of the immune system. Studies in germ-free animals may not reflect any significant effects. It is important to determine whether the need is to assess the systemic immune or the focal immune response. The normal human's immune system is constantly stimulated throughout life by the gut flora. The GALT (gut-associated lymphatic tissue) mediates this occurrence. The effect of the addition of large numbers of microbes to this system is not known.

Whether chronic stimulation of the immune system is good or bad is not known. A primed immune system may be advantageous, but a fully active immune system may result in autoimmune disease and is not desirable.

It was questioned whether immune system studies should be a high priority, considering
the need for AIDS (autoimmune deficiency syndrome) research currently being undertaken by other research facilities. Because of this need, research on the effect of lactic acid bacteria on the immune system may be low priority. However, studies on the immune system can be done relatively quickly if rat models are used prior to human studies.

The elderly exist in an immunosuppressed state, yet they also have increased incidence of autoimmune diseases, possibly because gastrointestinal tract leaks increase with age. Initial immune stimulation is easy to induce (vitamin E and others), but the effects may be short lived and may be ineffectual against disease.

Research needs for immune system stimulation were suggested.

- Focused animal studies should be conducted prior to human studies until a working hypothesis is obtained because immune system stimulation is too broad.

- When research advances to human studies, specific subpopulations such as the elderly and the immunosuppressed should be targeted.

Panel Consensus on Immune System Stimulation

No claim on the effect of lactic cultures on functioning of the immune system can be made.

Cholesterol Reduction

No statement linking lactic cultures with reduced cholesterol can be made. Research to date that has demonstrated cholesterol binding in vitro systems is likely just reflecting an affinity of cells in an aqueous environment to associate with other hydrophobic molecules in a micellar fashion. Some lactobacilli deconjugate bile acids. Although deconjugatation of bile acids interferes with cholesterol absorption, this activity is undesirable because it would also interfere with absorption of fat soluble vitamins.

Suggested research approaches to determine a link between ingestion of lactic cultures and cholesterol reduction are to feed strains selected for in vitro cholesterol-binding capabilities to ileostomy patients, to check for cholesterol concentrations from ileum contents to determine whether cholesterol absorption is changed, and, possibly, to rescue the bacteria with bound cholesterol, providing mechanistic evidence for cholesterol absorption by the lactobacilli.

Panel Consensus on Claims on Cholesterol Reduction

At present, no scientific research exists to support cholesterol reduction claims for fluid milk products with cultures added.

Panel Consensus on Constipation

At present, no scientific research conclusively supports prevention or relief of constipation because of fluid milk products with cultures added.

General Health Claims

A general statement accepted by the panel was that culture-containing milk may reduce undesirable intestinal microbes after use of antibiotics. Other general claims must be qualified. Evidence for a normalization or restoration function of health cultures is weak. Measuring restoration is difficult. Furthermore, the gastrointestinal system has an inherent normalization capability. Therefore, studies on normalization are difficult. Some evidence for hastening the decrease of Candida, coliforms, and staphylococci in healthy, antibiotic, and colon cancer patients is available. The effect is more dramatic in infant populations. Daily variation in microflora in the gastrointestinal tract is so large that causal effects of fed cultures are difficult to assess. The panel agreed that specific beneficial effects of bifidobacteria are even less clear than those for lactobacilli.

Safety Concerns

How the addition of large numbers of viable bacteria affects the gastrointestinal tract ecosystem is unknown. Genetic exchange in this ecosystem is difficult to control. Although there is a history of safety with strains used in traditional fermented foods, the supplementation of foods with strains of intestinal origin, capable of replicating in the intestine, may have undesired ramifications. Information should be obtained on the genetic capability of
intestinal bacteria as recipients or donors of genes. The concern is not that disease will be caused by these microbes, but rather that transfer will occur among the populations of potentially dangerous genes, such as virulence and antibiotic resistance factors. The years of safe use of bifidus-supplemented milks in Japan and acidophilus-fermented milks in Europe should help alleviate some of the concerns over safety.

The introduction of chimeric intestinal strains may also be a problem.

**Future Directions**

The consensus of the scientific panel provided some claims that could be made on promotional material for culture-containing milks. In general, the panel noted that, although these claims are scientifically valid, the use of “may” in any message to the public may be misinterpreted. Claims made about benefits of cultures are based on research that has been done on specific strains, and no one or two strains are available today for which all these claims can be made. Although a significant body of research has been conducted, the research area as a whole has suffered for the lack of a coordinated, national effort. There is a need for a sustained effort combining the skills of microbiologists and clinicians. Studies should be done with high levels of the best defined strains known today.

A commitment for future research dollars in this area could provide much more solid ground for expanded claims in the future. The panel nicely described some focused studies that should provide more definitive answers to many of these health attributes. Some means of tapping into foreign research results may expand some of these claims. Meetings with the culture suppliers should be conducted to determine what they are able to substantiate about their cultures. A list of strain criteria must be developed for a unified approach to culture-supplemented milk.