Probiotics in Transition

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Abstract

Despite the hyperbole often linked with a popular research field, the scientific rationale for probiotics is sound. The probiotic concept is not new but is undergoing transition as knowledge of the gut microbiota in health and disease becomes translated to the clinic. Operationally, a probiotic represents a mimic of and/or supplement to the normal gut microbiota. Much confusion has arisen among consumers because of media misportrayals of probiotics as all being the same. However, with clarification of the molecular basis of host-microbe interactions, the selection criteria for probiotics and the delineation of their distinct mechanisms of action are improving. Most probiotics are from the genus Lactobacillus or Bifidobacterium; this is likely to change and diversify. Similarly, the development of new therapeutic strategies, such as the development of phagebiotics, psychobiotics, and genetically modified pharmabiotics, is poised to become a therapeutic reality.

Probiotics represent one part of a bigger story—the gut microbiota. This unfolding story promises much, including scientific and societal impact. Few areas in biology have generated as much interest as the gut microbiota during the past few years, regularly adorning the covers of the top peer-reviewed journals. The significance of the field has not escaped the lay press and business world; one business magazine proclaimed on its cover that “Microbes maketh man.”[1] The attention to microbe-host interactions is due to the impact of the former on the development of the latter, the contribution of the microbiota to health maintenance, and the evidence linking changes in the microbiota with gastrointestinal and extraintestinal disorders such as asthma, obesity, and other metabolic disorders.[2–4] In addition, there is potential to manipulate the microbiota, not only by probiotics but also by basic dietary measures.[5,6] For example, a recent study of the composition of the gut microbiota in elderly people showed a strong correlation with diet and health including inflammation and frailty and underscores the importance of a diversified diet to maintain a diversified microbiota.[7] Because the microbiota represents a health asset, with some microbial

constituents becoming a liability in susceptible hosts, the rationale for probiotics is to enhance microbial assets and to offset liabilities. Although often defined as "live microorganisms, which, when consumed in adequate amounts, confer a health benefit on the host,"[5] a probiotic may be operationally defined as a mimic of the beneficial effects of the gut microbiota.[6,7] A less restrictive term, such as pharmabiotic, seems preferable to embrace all therapeutic microbial components, live or dead, including metabolites and bioactive fragments, genetically modified organisms, or even transplants of whole microbial communities.[6] This field is in transition, as the molecular basis of host microbe and of microbe-microbe interactions unfold, and has entered the realm of evidence-based medicine. Like many an expanding field, the probiotic concept has suffered from extravagant and unsubstantiated claims. However, there is solid science underlying the concept with several success stories. Analysis of the normal microbiota will yield new candidate strains, but criteria for selection of a probiotic or pharmabiotic should be based on demonstrable mechanisms of action, matched for a specific clinical indication.[8] In addition, the host-microbe interface has widened to include the brain-gut-microbiota axis, which may also be a target for therapeutic manipulation.[9] Evolving concepts will be addressed in this overview; comprehensive reviews of the microbiota may be found elsewhere.[2–4]

What Are the Findings? Mimicking the Microbiota

Consumption of a probiotic will have minimal impacton the composition or diversity of the resident microbiota, but murine studies have shown that it may have a profound influence on gene expression and metabolic activity of the microbiota.[10] Probiotic metabolites and signaling with the host may be more important. Signals from microbiota provide trophic, nutritional, metabolic, and protective input to the development and maintenance of the host digestive, immune, and neuroendocrine systems. Although the definition of a core microbiota in health is unclear,[11] it seems intuitive that broad biodiversity ensures optimal competition against pathogens and maximal microbial stimulation for the developing host. In clinical situations where there is a low microbial diversity (such as in preterm neonates colonized before maturation of the mucosal barrier, immune system, or blood-brain barrier), a high bacterial intake (from contaminated water), or a major disturbance of the microbiota (after antibiotics), a probiotic may serve as a safe, nonspecific supplement to compete with opportunistic pathogens (Figure 1). In such circumstances, one or more of a diversity of probiotic strains might suffice. In contrast, when a specific effect (antimicrobial, anti-inflammatory, or metabolic) is required, the precise selection of probiotic strain is critical (Figure 2).

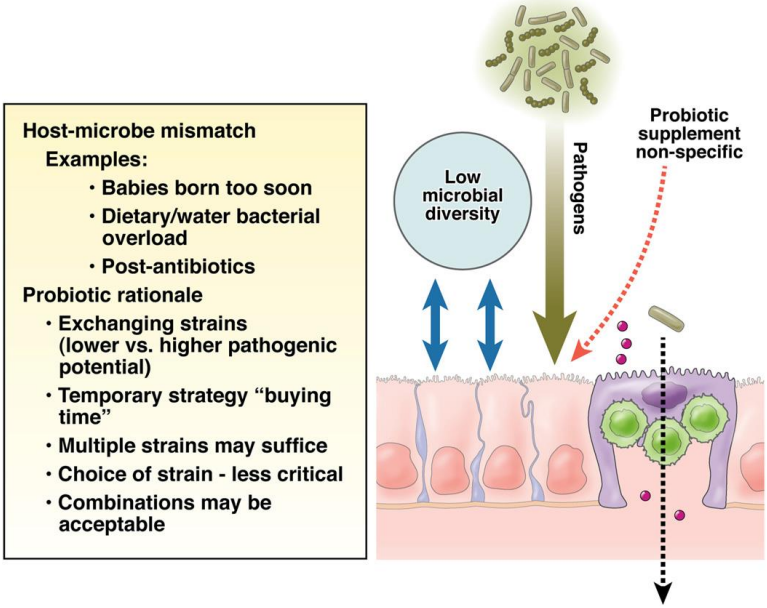

Figure 1. Probiotic rationale in settings of low microbial diversity. Low microbial diversity favors ingress of pathogens and limits the protective signaling from the microbiota to the host; host-microbe mismatch may also arise with bacterial overload such as might occur in undeveloped countries (modified with permission from Shanahan[6]).

From Mimics to Mechanisms

Elucidation of distinct mechanisms of action for different probiotics has provided reassuring support and insight. For example, in seeking an antimicrobial protective effect, a screen of different probiotic candidates revealed one (*Lactobacillus salivarius* UCC118) that was highly effective against *Listeria monocytogenes*. The anti-*Listeria* effect was mediated by an antibacterial peptide (bacteriocin) that is specific to that probiotic. Its action was demonstrable in vivo by live animal imaging with a luminescence-tagged inoculum of the pathogen.[12] Curiously, the same probiotic also protects against infection with salmonella but by a different mechanism, because, in contrast to the anti-*Listeria* effect, protection against salmonella was retained by a bacteriocin-negative mutant.

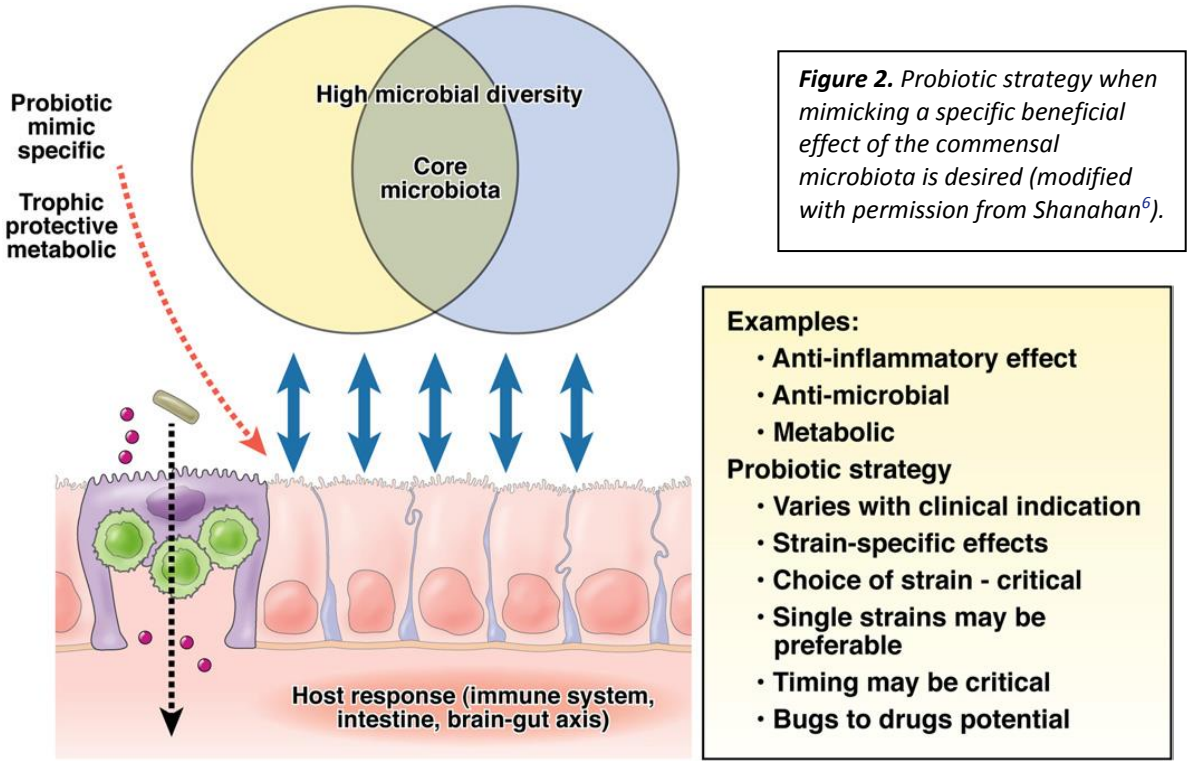

Figure 2. *Probiotic strategy when mimicking a specific beneficial effect of the commensal microbiota is desired (modified with permission from Shanahan[6]).*

Visualization of an anti-inflammatory mechanism of action has been equally impressive with other probiotic strains. The anti-inflammatory effect of orally administered *Bifidobacterium infantis 35624* was imaged in live animals engineered to elicit a luminescent signal on activation of nuclear factor kappa B in inflamed tissues.[13] In contrast to placebo-fed controls, the probiotic- fed animals exhibited an attenuated signal. Furthermore, the anti-inflammatory mechanism was due to induction of regulatory T cells that limited the activation of nuclear factor kappa B, and the effect was transferrable by adoptive transfer of these cells to naive animals.[13] The molecular underpinning of how probiotics engage in crosstalk with the host exemplifies the divergent functional properties of closely related strains. Although probiotics express surface molecules similar to those of other bacteria, including commensals and pathogens,[14] differential responses from the host have been elicited on exposure to different probiotic strains.[8,15] Furthermore, the anti-inflammatory capacity of some, but not all, lactobacilli has been shown to be dependent on NOD2 recognition of a peptide constituent of the probiotic cell wall.[16] Thus, structural differences in cell wall peptidoglycans may account for some of the strain specificity of the anti-inflammatory lactobacilli.

Intriguingly, other lactobacilli lacking in anti-inflammatory capacity can deploy separate mechanisms to exhibit antinociceptive properties.[8] With regard to bifidobacteria, the expression of pili is required for colonization and engagement with the host,[17] but a probiotic effect requires the additional expression of an exopolysaccharide to elicit anti-inflammatory properties.[18] Thus, as with commensals, variability across probiotics is reflected in the diversity of their signalling with the host.

Probiotic Selection

Failure to appreciate the importance of strain specificity for different probiotic effects is the single greatest problem relating to the interpretation of data on probiotics. In the same way that all pills or tablets are not the same, all bacteria are not the same, and all probiotics could not be the same. Like other microbes, probiotics are defined by their genus (eg, Lactobacillus), species (eg, L salivarius), and strain name (L salivarius UCC118). Lumping all lactobacilli together as probiotics is folly; some have no probiotic effect, and those strains that do differ profoundly in their genotype and functionality. Similarly, the effect of one probiotic strain cannot be extrapolated to another microbe, even if it belongs to the same genus or species. Several reports attest to the strain specificity of different probiotic actions in vitro and in vivo,[8,15] and where head-to-head comparisons have been performed in humans,[15,19] differences in responses and efficacy have also been evident. At present, yeast (eg, Saccharomyces boulardii) and bacteria comply with the current definition of probiotics, with the latter
including species of Lactobacillus, Streptococcus, Enterococcus, Bifidobacterium, Propionibacterium, Bacillus, and Escherichia coli. This degree of heterogeneity underscores the problem of using a collective generic name. The safety and efficacy of drugs are not addressed generically; similarly, probiotics need consideration on a strain-by-strain basis.

What to Expect: the Gastroenterology Experience

Probiotics are not drugs. Like many naturally occurring agents, modest rather than robust effects should be anticipated. Although a multitude of meta-analyses are available for probiotics in different conditions, the literature contracts when one looks for evidence for a specific strain in a specific indication. The European Food Safety Agency has rejected health claims for probiotics, in most cases because of inadequate characterization of the microbe. However, despite a lack of uniformity in quality of clinical trials and much hyperbole, there have been successes, and the following conservative conclusions seem reasonable. The most impressive results have been in protection against infection, particularly at the extremes of life and in other vulnerable groups.[6,20] Although not without some controversy because of the variable efficacy of different probiotics, the evidence favoring probiotics to reduce the risk of necrotizing enterocolitis and mortality in preterm neonates is impressive. Similarly, in elderly people and for prevention and treatment of antibiotic- associated diarrhea, the efficacy of probiotics, albeit modest, is persuasive,[21] as is their role in prevention of childhood diarrheal disease, particularly in developing countries.[6] Although several studies support a role for some but not all probiotics in irritable bowel syndrome,[19,22] the evidence in inflammatory bowel disease has been disappointing.[6] The microbiota in inflammatory bowel disease has been reviewed elsewhere.[23] There is no evidence for any probiotic in Crohn's disease at present. The evidence in ulcerative colitis is more encouraging but is not as impressive as results in animal models of acute colitis, and a recent meta-analysis of probiotics for maintenance of remission is inconclusive, highlighting the need for more trials.[24] Trials of a cocktail of 8 different bacteria (VSL #3) in pouchitis showed efficacy in treatment and prophylaxis,[25,26] but subsequent reports and anecdotal experience have been inconsistent and disappointing.[6]

What Clinicians Should Advise

The concept of probiotics has been embraced by consumers, and clinicians need an informed, measured response to patients seeking guidance. For specific indications, the specific probiotic strain with proven efficacy for that indication should be selected. Otherwise, general advice should include the following. First, patients should understand that probiotics are not alternatives or substitutes for conventional therapy; they are supplements. Second, although there is a good safety record for probiotics, there is no such thing as zero risk. Rare unexpected outcomes may occur, particularly in those with severe illness or other susceptibilities. As with other interventions, risk should be balanced against evidence of benefit. Third, in addition to variability in action and in efficacy across different probiotic strains, an important concern is the lack of uniform quality control in probiotic products. To offset this, consumers should opt for a reputable manufacturer. Fourth, it should not be assumed that combinations are synergistic; they may be antagonistic. Fifth, although dosimetry of probiotics is often unclear, increments of bacteria are log-fold, and more than once-daily consumption of any product should be unnecessary. In adulthood, probiotics seldom colonize the host and, therefore, must be taken indefinitely for their desired effect. Finally, for the worried well and those patients seeking only to optimize their intestinal microbiota, a probiotic should not be a substitute for a diversified nutritious diet, which is a major determinant of the composition of the gut microbiota.[7]

A Glimpse of the Future

Establishing the molecular mechanisms of action of probiotics opens the prospect of translating "bugs to drugs." Microbial-derived antimicrobial, immunoregulatory, or anti-inflammatory molecules promise new natural therapies (pharmabiotics). Similarly, emerging evidence for a brain-gut-microbe axis[8] has therapeutic implications for the development of psychobiotics, whereas the selective elimination of components of the microbial population with phage viruses is an old concept that may now be ready for exploitation (phagebiotics).

What Are the Roadblocks and/or Limitations?

More research, bigger and better clinical trials, and enhanced understanding of host-microbe interactions in health and disease are self-evident requirements for progress. More specific impediments at each step linking the science with the consumer must be tackled. First and most culpable are the media, with continual misinterpretation of the research and, in particular, inaccurate portrayal of probiotics as if all strains were the same. Second has been a lack of specific guidelines from regulatory agencies concerning the establishment of health claims for probiotics and, in some instances, inadequate policing of spurious claims. Third, the oversight of probiotic quality control by commercial suppliers is variable; indeed, this may be a more important safety consideration than concerns regarding the active strain. Hence, there is the recommendation to use probiotics only from a reputable supplier. Fourth, clinicians and scientists have a responsibility to avoid hyperbole in describing their results. For example, documenting host-microbe interactions in vitro, regardless of their intrigue and scientific merit, is not the same as demonstrating a probiotic effect in vivo and cannot be used to infer a probiotic effect. A related problem is repetition of flawed or unproven concepts or imprecise language. An example of the former is the notion of using probiotics (or anything else) to "boost the immune system" in the normal population.

An example of the latter is the clichéd term dysbiosis, which gives the vague impression of an understanding of some microbial imbalance, when neither the understanding nor the imbalance exists. The word is unnecessary, often used inaccurately, and introduces bias if the intent is merely to describe a change in the microbiota. Fifth, at consumer level, there are the poor comprehension of risk/benefit analysis and inadequate critical appraisal of evidence for health claims. This is part of a

bigger problem of science awareness and inadequate scientific thinking in society. Finally, managing patient expectations is a lingering caveat, particularly in relation to the notion of trying to reverse chronic disease, the risk for which might relate to the impact of the microbiota on the developing immune system in early infancy.
Because immunologic priming by the microbiota occurs in early life, the potential for altering immunoallergic disorders in adulthood by manipulating the microbiota is limited.

Conclusions

The rationale for probiotics is scientifically sound, and in some instances, mechanisms of action have been demonstrated at a molecular level. The selection of probiotic should be matched with the clinical indication. Probiotics are not alternative medicine; they are adjuncts or supplements to conventional medicine, not substitutes. The benefits are modest, as might be expected for a naturally occurring agent. While acknowledging that there is no such thing as zero risk, there is a long safety record with probiotics. However, quality control of probiotic products is nonuniform, and consumers should select a reputable supplier with a product for which there is supporting science.

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