Probiotics: Definition, Sources, Selection, and Uses

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Interest in probiotics is at an all-time high in the United States, driven in part by new products emerging in the market, by US researchers eager to evaluate efficacy claims rigorously, and by consumers interested in potential therapeutic and preventive health benefits. The US marketplace is a mixed bag of products, some well-defined and properly evaluated in controlled clinical studies and others with unsubstantiated claims of efficacy. Validation of probiotic contents in commercial products is needed to ensure consumer confidence. The term “probiotic” should be used only for products that meet the scientific criteria for this term—namely, products that contain an adequate dose of live microbes that have been documented in target-host studies to confer a health benefit. Probiotics must be identified to the level of strain, must be characterized for the specific health target, and must be formulated into products using strains and doses shown to be efficacious. Several characteristics commonly presumed to be essential to probiotics, such as human origin and the ability to improve the balance of the intestinal microbiota, are discussed.

DEFINITION OF PROBIOTICS

The internationally endorsed definition of probiotics is live microorganisms that, when administered in adequate amounts, confer a health benefit on the host. Other definitions advanced through the years have been restrictive by specification of mechanisms, site of action, delivery format, method, or host. Probiotics have been shown to exert a wide range of effects. The mechanism of action of probiotics (e.g., having an impact on the intestinal microbiota or enhancing immune function) was dropped from the definition to encompass health effects due to novel mechanisms and to allow application of the term before the mechanism is confirmed. Physiologic benefits have been attributed to dead microorganisms [1]. Furthermore, certain mechanisms of action (such as delivery of certain enzymes to the intestine) may not require live cells. However, regardless of functionality, dead microbes are not probiotics.

The term “probiotic” is sometimes erroneously used as a synonym for putatively beneficial members of commensal microbiota. The context for this misuse is the assertion that certain dietary or environmental factors may “encourage your native probiotics.” Members of human commensal microbiota are often sources from which probiotics are isolated, but, until such strains are isolated and then adequately characterized for content, stability, and health effects, they are not probiotics.

The US Food and Drug Administration (FDA) uses other terms for live microbes for regulatory purposes; live microbes used in animal feeds are called “direct-fed microbials” [2], and, when intended for use as human drugs, they are classified as “live biotherapeutics” [3]. However, no legal definition of probiotics exists in the United States or in other countries, which allows the marketing of products labeled as “probiotics” that do not meet the fundamental criteria stipulated in the scientific definition. As public awareness of probiotics increases, the use of the term has implications both for the violation of the standard of truthful and not misleading labeling and for consumer confidence in this product category. In the absence of a legal definition of the term “probiotic,” it is incumbent on industry participants to adopt and adhere to the scientific definition of the term. If the products are not properly characterized and validated, the category will suffer.
VALIDITY OF COMMERCIAL PRODUCTS

Reports that the labels on commercial probiotic products are inaccurate with regard to the identity and potency of the contents are numerous [4, 5]. However, care must be exercised in interpreting some of these published reports, since not all arrive at accurate conclusions, because of inappropriate methods used to address the research question being asked [6–8]. Studies are further complicated by the need to obtain statistically representative samples, by the difficulty in differentiating multiple species and strains that may be combined into one product, by the presence of injured cells that may be viable but not culturable under certain circumstances, and by the need to use DNA-based approaches to properly identify species of some probiotic genera, including Lactobacillus and Bifidobacterium. Consumer Reports reported that yogurts are better sources of probiotics than are supplements [6]. But this conclusion may have been in error. Although methods were not disclosed, the results suggested that no differentiation of the different types of bacteria present in yogurt was conducted. Reporting the total number of bacteria in products containing multiple strains can be misleading. In the case of yogurt, both starter cultures (Lactobacillus delbrueckii subsp. bulgaricus and Streptococcus thermophilus) and extra bacteria added for health effects (e.g., strains of other species of Lactobacillus or Bifidobacterium) can be present. If the health effects are derived from these additional bacteria, then these need to be specifically enumerated before a judgment can be made on the potency of the product. A total count does not indicate the levels of each of the added probiotic strains. For any product containing multiple strains of probiotics, counts of each strain should be provided so that consumers are clear on the relative potency of each added strain.

REGULATION OF PROBIOTICS

It is often misstated that probiotic products are unregulated. Clearly, the FDA has regulatory authority over probiotic products and regulates manufacturers’ responsibilities, including the labeling and safety of these products, whether in food, supplement, or drug form. Of note, on 24 August 2007, the FDA issued regulations that require current good manufacturing practices for dietary supplements to be phased in over the next few years. Although these regulations do not address verification of efficacy claims, hopefully they will improve the compositional quality (identity, purity, and strength) of probiotic supplements in the US market. However, manufacturers of foods and supplements are not required to obtain premarket approval of claims of efficacy or safety. In practice, the FDA has never challenged the labeling or safety of a probiotic product except in cases where the product is represented as a drug (i.e., to treat, cure, prevent, mitigate, or diagnose disease) and lacks approval as a drug. Usually, these cases result from incorrect product labeling by manufacturers that were intending to market a dietary supplement. Lack of meaningful FDA oversight of claims of efficacy makes it difficult for consumers to distinguish probiotic products that are properly formulated and labeled from those that are not.

A reasonable approach for manufacturers marketing a product that contains a probiotic is to use guidelines established by a working group convened jointly by the Food and Agriculture Organization of the United Nations and the World Health Organization [9], which include the following.

1. Proper identification to the level of strain of all probiotics in the product, with deposit of all strains in an international culture collection
2. Characterization of each strain for traits important to its safety and function
3. Validation of health benefits in human studies, including identification of the quantity of the microorganism required to provide the benefit
4. Truthful and not misleading labeling of efficacy claims and content through the end of shelf life

REQUIRED ATTRIBUTES OF PROBIOTICS

Often, assumptions are made about mechanisms of action and the “essential” characteristics of probiotics on the basis of supposition more than rigorous science. For example, it is commonly stipulated that probiotics must adhere to intestinal cells. However, data that support adherence of probiotics are mostly derived from in vitro assays, which have limited predictability for the in vivo situation [10], and must be reconciled with the fact that, in general, probiotics persist only short term in the host after feeding has stopped. The nature of the association of probiotics with the epithelial cell surfaces or mucous layer remains to be determined.

Although it often suggested that probiotics for human use must be of “human origin,” some strains that are not normally isolated from humans have been shown to be effective probiotics (e.g., strains of the species Bifidobacterium animalis), which negates this requirement [11].

The statement that probiotics “improve the balance of microbiota” is often made. However, it is not clear what this assertion means or how it is measured. Probiotics have been shown to alter populations or activities of colonizing microbes, but does this correspond to an “improved balance”? Improved balance is often equated with increased fecal levels of lactobacilli or bifidobacteria. This is a measure not of balance but of fecal microbiota alteration. Since no scientific consensus exists on the composition of a “healthy microbiota,” the health implications of such microbiota alterations remain unclear. Fur-
thermore, it is difficult to measure intestinal microbiota; fecal microbiota is not equivalent to intestinal microbiota, and luminal microbiota is not equivalent to epithelial microbiota [12]. Probiotics may, in fact, facilitate a return to normal status after a perturbation of the microbiota (e.g., because of the use of antibiotics or illness) or may reduce the degree of change invoked by such challenges. This function more closely supports the concept that probiotics can improve the balance of microbiota. A few studies have measured a probiotic-enhanced return to baseline levels after antibiotic use in humans [13]. The concept of probiotic-induced improved balance of microbiota would benefit from further study.

When probiotic strains are selected, attributes important for efficacy and technological function must be assessed. Because the range of targets for in vivo function is broad, spanning oral, stomach, respiratory, intestinal, vaginal, and immune functions, it would be a daunting task to develop a list of characteristics required for all probiotic functions. Basic initial characterization of strain identity and taxonomy should be conducted, followed by evaluation with validated assays both in studies of animal models and in controlled studies in the target host. In vitro assays are frequently conducted that have not been proved to be predictive of in vivo function [14]. Technological robustness must also be determined, such as the strain’s ability to be grown to high numbers, concentrated, stabilized, and incorporated into a final product with good sensory properties, if applicable, and to be stable, both physiologically and genetically, through the end of the shelf life of the product and at the active site in the host. Assessment of stability can also be a challenge, since factors such as chain length and injury may challenge the typical assessment of colony-forming units, as well as in vivo function.

**THE ROLE OF STRAIN, DOSE, AND PRODUCT FORMAT IN PROBIOTIC FUNCTION**

Strains of the same probiotic species can be different, which has been demonstrated both in vitro and in animals, although similar data in humans are rare. Thus, clinical results from one study are applicable only to the strain or strains being evaluated in that study. Given this, however, different strains may have the same effects, and similar immune effects have been documented for different strains [14]. As research continues, certain species- or genus-specific attributes are likely to be found. Identification of genes or gene systems may make it feasible to predict in vivo function, and documentation of expression of these genes may become adequate substantiation for in vivo function. Functional genomics may greatly aid this research, which is progressing at a rapid pace [15]. Currently, however, research specific to the strain or strain combinations should be used to substantiate claims of physiological benefits. Furthermore, the genus, species, and strain designations should be specified for all probiotic strains in a product.

Dose levels of probiotics should be based on levels found to be efficacious in human studies. One dose level cannot be assumed to be effective for all strains. For example, the efficacy of *Bifidobacterium infantis* 35264 has been documented at 10^8 cfu/day [16], whereas the recommended dose of VSL#3 (VSL Pharmaceuticals) is 1.8 × 10^10 cfu/day, a 4-log cycle difference [17].

The impact of product format on probiotic function has yet to be explored in depth. The common quality-control parameter of colony-forming units per gram may not be the only parameter indicative of the efficacy of the final product. Other factors, such as probiotic growth during product manufacture, enteric coating, preservation technology, metabolic state of the probiotic, and the presence of other functional ingredients in the final product, may play a role in the effectiveness of a product. More research is needed to understand how much influence such factors have on in vivo efficacy.

**CONCLUSIONS**

The field of probiotics is growing quickly in the United States, as is evidenced by the increasing interest of industry, consumers, and researchers. As professionals embrace this burgeoning area, they need to approach it in a manner that will ensure that product formulations and communications about these products are done responsibly and with the primary objective to benefit consumers. Both the science and its limitations should be communicated with precision. Improper use of the term “probiotic” and failure to recognize the importance of the dose specificity and strain specificity of effects is a concern. There is a great need for controlled studies in humans to further document the health benefits of probiotics as part of the human diet. Important target groups for such studies include healthy people, people at elevated risk for developing a disease, and people searching for dietary-management techniques to control symptoms. All these groups would benefit from publicly funded research of probiotics as foods or supplements. Probiotics could also be studied for use as drugs.

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References