Critical Overview of Probiotics Efficacy on Health and its Safety

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ABSTRACT

The review is to analyse the therapeutic benefits of probiotics in various chronic disorders. All the data were identified using PUBMED (2000-2020) and bibliographic reviews of recent and old articles from an English literature search. After independent analysis by co-authors of the identified articles, data were analysed and extracted for the stated purpose. The most-studied species include *Lactobacillus*, *Bifidobacterium*, and *Saccharomyces*. Probiotics have an essential role in maintaining immunologic equilibrium in the gastrointestinal tract through direct interaction with immune cells. There is high-quality evidence that probiotics are effective for acute infectious diarrhoea, antibiotic-associated diarrhea, *Clostridium difficile* associated diarrhoea, hepatic encephalopathy, ulcerative colitis, irritable bowel syndrome, and necrotizing enterocolitis. Probiotics are safe for infants, children, adults, and older patients. Therapeutic applications of probiotics have been widely studied to treat and improve the gut's health; however, choosing a probiotic for a specific condition is a challenging task that requires parsing of the data for strain-specific efficacy and evaluation of product quality. It appears likely that more national organizations will be conducting evidence-based research, and the pharmacist and clinicians should pay attention to this ever-changing field.

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INTRODUCTION

Probiotics are live microorganisms when administered in adequate amounts; confer a health benefit on the host¹. They are one of the most accepted dietary supplements in the United States, and their use continues to expand. According to the National Health Interview Survey, four times as

many adults reported taking a probiotic or prebiotic in 2012 than in 2007². Among non-mineral, non-vitamin dietary supplements, probiotics ranked third behind fish oil/omega-3 fatty acids and glucosamine and chondroitin in usage³. After primarily appearing in yogurt and kefir, probiotics are now marketed in

foods ranging from chocolate to gummies. In 2015, probiotic supplements found in the pharmacy aisle accounted for only 9% of the market, dwarfed by sales of probiotics in foods⁴. This is poised to change, fuelled by basic research about the human gut microbiota from endeavors such as the Human Microbiome Project, many probiotic clinical trials, and a groundswell of consumer awareness⁵. According to a recent survey, 65% of consumers believe probiotics improve health conditions. Simultaneously, the number of products has grown, including enteric-coated and microencapsulated formulations. Probio, a new international database that catalogs information about commercially available or investigational probiotics, currently lists 329 probiotics marketed for human use⁶. Because probiotics are marketed as dietary supplements in the United States, labeling often does not provide consumers with the information they require to select the best product for their needs. Based on the trends already discussed, pharmacists will face more questions from patients about the benefits of probiotics, which one they should use, and how they should be administered. This article intends to provide the background that community pharmacists need to answer these questions and to review the conditions that have the most evidence of benefit from probiotics, especially common diseases encountered by community pharmacists⁷.

GLOBAL NEED AND DEMAND OF PROBIOTICS⁸

- Probiotics are a boon for increasing the health of the gut and reduce intestinal inflammation. They also help in enhancing the strength of the immune system.
- In 2018 it has been estimated that the market of probiotics globally would be USD 48.38 billion and it has been expected that it would expand CAGR of 6.9% in the coming days.
- The market of probiotics is being designated by the existence of demographic-specific probiotics such as geriatrics and genders. These niche markets provide the manufacturers of prebiotics to embrace a variety of strategies to increase their product portfolio and to increase their resources in a great amount. Due to awareness and knowledge for healthy diet gained from the different sources by the people it has been expected that the global market of probiotics would further raise.

- Because of the increase in demand, a huge investment is being done in Research and Development activities to produce probiotic strain. Also, it has been seen that there is a great advancement in the delivery system, which would help to increase the delivery of probiotics in the animal and human. Probiotics demand is greatly seen not only in the aging population but also in the young people who are conscious of healthcare.
- The manufacturers developed a new product which is given along with probiotics and in addition to other dietary supplements.
- Probiotics are available in various forms like tablets, capsules, drops, etc.
- The market of Asia-Pacific probiotics witnesses innovative new products. In February 2018, signature probiotics drink was launched in India by Yakult and Danone which contain Vitamin D and Vitamin E along with *Lactobacillus casei* strain in the formulation.

HIGHLIGHTS OF THE REPORT OF PROBIOTICS⁹

- The probiotics beverage turned up to be the largest segment in 2018 in terms of product type with USD 39.56 billion.
- It is anticipated that in terms of ingredient bacteria segment would control the market in coming years.
- Due to the increased focus on enhancing the health of the animal the demand for probiotics for animal nutrition is increased in which animal probiotics segment is anticipated to register relatively faster growth of over 7%. Some of the key market- players are Aria Food Inc, Biodata AB, General Mills Inc.

PROBIOTIC

The International Scientific Association for Probiotics and Prebiotics (ISAPP) defines probiotics as, "live microorganisms that, when administered in adequate amounts, confer a health benefit on the host". By industry standards, an effective dose is generally expressed as the number of colony-forming units (CFUs). Based on this definition, some experts do not consider fecal microbiota transplant a probiotic because it is not standardized with identified microorganism strains¹⁰. It is important to note that the terms "live" or "active" do not imply probiotic activity. A product containing live

microorganisms must demonstrate a health benefit to the host to qualify it as a probiotic. Generally, probiotics need to survive the gastrointestinal (GI) tract to render a health benefit, but this is not a requirement. To deliver live microorganisms to the lower GI tract, probiotics must survive proteolytic enzymes and the acidic environment of the stomach, as well as bile salts in the duodenum¹¹. For example, *Saccharomyces boulardii* and *Saccharomyces cerevisiae* are considered acid- and bile-resistant¹². *Lactobacillus* is considered relatively acid-resistant, as are many *bifidobacteria*¹³. *Bacillus coagulans*, a spore-forming probiotic, survive exposure to gastric acid and bile salts by generating spores that resist degradation to germinate in the small intestines¹⁴. Enteric-coating and microencapsulation may increase survival through the upper GI tract. It is essential to understand that although the genus may generally survive, the specific strain must be shown to survive in a human study¹⁵.

PREBIOTIC

By definition, prebiotics indirectly benefits the host by enhancing the growth or activity of beneficial microorganisms in the digestive tract¹⁶. The International Scientific Association for Probiotics and Prebiotics (ISAPP) recently revised the definition to specify that a prebiotic is a substrate that is selectively used by host microorganisms and confers a health benefit¹⁷. Prebiotics are not alive or digested by the host. Like probiotics, orally administered prebiotics must survive passage through the upper GI tract¹⁸. Prebiotics include conjugated linoleic acid, polyunsaturated fatty acid, human milk oligosaccharides, and oligosaccharides (e.g., fructooligosaccharides, galactooligosaccharides, inulin, mannan-oligosaccharide, and Xylo oligosaccharide). Most of them stimulate the growth of *bifidobacteria* by acting as a food source. A product that contains a prebiotic and a probiotic is called a symbiotic¹⁹.

WORKING OF PROBIOTIC

Probiotics have many complex proposed and proven mechanisms of action^{20,21}. They can temporarily change the composition and activity of the indigenous microbiota. Still, short-term administration does not appear to cause lasting changes in colonic bacterial composition or diversity in healthy adults²². Correction of dysbiosis is a proposed mechanism of action for probiotics in disorders such as antibiotic-associated diarrhea (AAD), irritable bowel syndrome (IBS), and

inflammatory bowel disease (IBD)²³. Probiotics can enhance epithelial barrier function, compete with pathogens to prevent their attachment, and stimulate the production of substances that inhibit pathogen growth^{24,25} (Fig 1).

For example, some *bifidobacteria* inhibit adherence of pathogenic *Escherichia coli* and *Clostridium difficile* to intestinal epithelial cells²⁶. Some probiotics produce short-chain fatty acids that appear to have anti-inflammatory effects and may modulate metabolic effects in conditions such as type 2 diabetes and hyperlipidemia. Members of a genus such as *Lactobacillus* or *Bifidobacterium* may share a mechanism of action, in the same way, drug classes share a mechanism of action²⁷. For example, all *Bifidobacterium* species produce short-chain fatty acids. Some mechanisms appear specific to a species or subspecies. For instance, *Bifidobacterium longum* subsp *infantis* metabolizes human milk oligosaccharides while *Bifidobacterium* subsp *lactis* does not, leading to better gut colonization in premature infants by *B. longum* subsp *infantis*²⁸. This could have implications for probiotic use in infants, such as prevention of necrotizing enterocolitis in premature infants²⁹. Probiotics can reduce intestinal transit time, primarily in patients with constipation. *Bifidobacterium lactis* HN019 and *Bifidobacterium lactis* DN-173 have medium to large effects on intestinal transit time compared with other strains³⁰.

Until recently, they were modulating the immune system with probiotics was a far-fetched idea. Now, substantial research shows that the gut microbiota interacts with the immune system in many ways throughout our lifespan³¹. In the GI tract, it interacts with gut-associated lymphoid tissue. The gut microbiota improves T cell-mediated immunity (Th1) by enhancing the activity of circulating phagocytic neutrophils and natural killer cells. Effect on humoral immunity(Th2) is reflected by higher levels of immunoglobulins³².

Effects like these have led to the evaluation of probiotics for indications such as enhancing the immune response to vaccines in elderly persons and preventing upper respiratory tract infections³³. Development of the infant immune system requires stimulation from the gut microbiota. In early infancy, the general immune response tends to be Th2. Exposure to normal gut flora soon after birth tips the balance toward a response that includes Th1. If the normal gut flora is not present, an immune response that remains skewed toward Th2 may increase the risk of allergic disorders. Understanding this process

has resulted in studies evaluating whether giving a probiotic during pregnancy and infancy can prevent the development of immune-mediated diseases, often with positive outcomes³⁴.

USES OF PROBIOTICS IN CLINICAL PRACTICE

A probiotic, by definition, confer a health benefit to the host. Therefore, what constitutes proof of a probiotic health benefit in humans is a key question. According to ISAPP, human clinical studies are needed, the gold standard being randomized controlled trials (RCTs) that are adequately powered. Meta-analyses are also useful because probiotic clinical trials are often underpowered. The outcomes of meta-analyses evaluating probiotics in

Cochrane reviews are summarized satisfactorily³⁶. Cochrane reviews systematically assess the evidence and results from clinical studies. Although probiotics have been evaluated for a wide range of diseases, GI disorders have the most evidence of benefit³⁷. Conditions for which recent meta-analyses have found promising but insufficient substantiation of the high level of evidence (i.e., Cochrane level type) of treatment benefit include treatment of hypertension, hyperlipidemia, obesity, allergic rhinitis, atopic dermatitis in infants, and type 2 diabetes³⁸⁻⁴¹. Additional conditions include prevention of asthma in children and the prevention of dental caries^{42,43}. Most probiotics are marketed as dietary supplements^{44,45}. Product labeling generally makes a structure-function claim such as “to support digestive and immune health”^{46,47}.

Fig 1: Role of probiotics in the amelioration of gut functions. (1) probiotics could protect from pathogenic microbes in a diversified manner. (2) probiotics can secrete antimicrobial compounds and metabolites such as lactic acid, enzymes, and bacteriocins which can alter the pH that ultimately can inhibit the growth of pathogens. (3) probiotics maintain the gut mucosal barriers, i.e., the chemical barriers such as antimicrobial peptides (AMPs), which can inhibit the invading microorganisms, and the physical barriers including cellular junction and the mucus layer that repel the invading microorganisms. (4) alliance of microbiota with the immune system allows the maintenance of regulatory function involved in the conservation of tolerance to safer antigens and induction of protection to the pathogens³⁵.

Without regulatory guidance on probiotic efficacy, guideline recommendations from medical organizations take on added importance. A study⁴⁸⁻

⁵⁰ summarizes clinical practice guideline recommendations for the use of probiotics in specific conditions. It includes recent meta-analyses results

that estimate the size of the benefit when possible^{51,52}. Online resources such as The Clinical Guide to Probiotic Products and can also help to identify products with clinical trial data for specific conditions⁵³.

Antibiotic-associated diarrhoea

The Infectious Diseases Society of America (IDSA) and the American College of Gastroenterology (ACG) support the use of probiotics to manage AAD⁵⁴. A meta-analysis of more than 11,000 patients in sixty-three studies found that using a probiotic during a course of antibiotics reduced the risk of AAD by about 40%. In a Cochrane review, prophylactic use of probiotics reduced the risk of AAD in children by 54%, with an NNT of 55. Most studies have been conducted with *Lactobacillus rhamnosus* GG, *Lactobacillus casei* DN114001 and Shirota, and *Saccharomyces boulardii* [*cerevisiae*]⁵⁶. Based on moderate-quality evidence, the European Society for Paediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) prefer *L. rhamnosus* GG or *S. boulardii* for the prevention of AAD in children regardless of whether the antibiotic regimen is for *Helicobacter pylori* eradication or other reasons. Although the Toronto Consensus does not routinely recommend probiotics as an adjunct to *H. pylori* eradication regimens in adults, the group acknowledges that probiotics are unlikely to be harmful and may have benefits for patients at high risk of adverse effects (AEs)⁵⁷⁻⁵⁹.

Irritable bowel syndrome

The American College of Gastroenterology (ACG) gave probiotics a weak recommendation based on low-quality evidence for treatment of IBS⁶⁰. The reason for the low quality of evidence was due to inconsistencies between the studies, as most studies evaluated different probiotics⁶¹. This is based on a meta-analysis of 23 RCT in almost 2600 patients in which probiotics significantly improved global symptoms, abdominal pain, bloating, and flatulence scores⁶². It reported a number needed to treat (NNT) of 7 for symptom improvement and a number needed to harm of 35 for probiotic AEs^{63,64}. A Cochrane review reported an NNT of 8 for IBS pain to improve with probiotic treatment in children⁶⁵.

Clostridium difficile-associated diarrhea (CDAD)

A Cochrane review found that probiotics significantly reduced the risk of CDAD by 64% (2.0% with probiotic vs 5.5% without), but did not affect the

incidence of *C. difficile* infection (CDI)⁶⁶. A plausible explanation is that probiotics suppress symptoms of infection but do not prevent *C. difficile* colonization. This confusing picture has led to somewhat conflicting guideline recommendations. Infectious Disease Society of America (IDSA), American College of Gastroenterology (ACG), and American Academy of Pediatrics (AAP) do not recommend probiotics to prevent CDAD⁶⁷. ACG and the American Society of Colon and Rectal Surgeons support adjunctive probiotics to reduce recurrences in patients with recurrent CDAD⁶⁸. Based on low-quality evidence, The European Society for Paediatric Gastroenterology Hepatology and Nutrition (ESPGHAN) suggests using *Saccharomyces boulardii* if a probiotic is used to prevent CDAD in children⁶⁸.

Infantile colic

Pharmacists can offer a probiotic to the desperate new parents they encounter wandering the pharmacy aisles in search of an infant colic remedy. In theory, inflammation triggered by abnormal gut microbiota may cause colic⁶⁹. Although several probiotics have been evaluated, *Lactobacillus reuteri* 17938 is the best-studied strain. In a meta-analysis of 4 RCTs (N = 345), the mean difference in change of crying/fussing time from baseline was reduced by about 25 minutes more in infants treated with *L. reuteri* 17938 than with placebo at 21 days. The NNT was 4 for an infant to have a 50% or a more significant decrease in crying or fussing at two weeks. It appears the positive results are primarily of breastfed infants. An RCT that included formula-fed infants did not demonstrate benefit, raising questions about generalizability to formula-fed infants⁷⁰. Although the evidence is insufficient to establish efficacy definitively, the Canadian Paediatric Society recommends that clinicians consider recommending probiotics to treat colic^{71,72}.

Health benefits of probiotics on other diseases like Acute Pancreatitis

Probiotics are defined as live microorganisms that promote the healthiness when they are administered into the body by improving the flora of the gut. Probiotics could be given with prebiotics, on digestive fiber to increase the effectiveness of the probiotic activity⁷³.

Cause of Acute Pancreatitis

Acute pancreatitis is caused due to Necrotic Tissue infection. The main reason is a dysfunction of the

intestinal barrier and shifting of the bacteria from the bloodstream to the necrotic tissue which later causes infection to the Necrotic tissue⁷³.

Remedy

Upon administration of probiotics, it was found that the translocation of the bacteria is reduced via three levels of host bacteria; infection. They are:-

- The intestinal lumen
- The intestinal epithelium
- The immune system

In the intestinal lumen, the overgrowth of the bacteria is arrested by a direct antimicrobial effect and competitive growth. It prevents the adhesion of the bacteria by competitive exclusion to the epithelial surface and also the permeability of the epithelial by the influence of pathogen. The Epithelial function is preserved by regulating enterocyte gene expression which is involved in the maintenance of the mucosal barrier⁷⁴. It has been also found that probiotics could inhibit local pro-inflammatory in enterocytes following the adhesion of the pathogenic bacteria. Thus in vitro probiotic strain could produce anti-inflammatory cytokine IL-10. It is believed that such a similar effect has a governing effect on the mucosal and systemic immune system too⁷⁵.

SAFETY OF PROBIOTICS

Probiotics are live organisms that pose a theoretical risk of infection for patients who are immunosuppressed, critically ill, and either very young or old⁷². For immune-competent consumers, probiotics are generally well tolerated, except for mild GI complaints early in use (e.g., bloating, flatulence, diarrhea, and hiccups)⁷⁶. An analysis by the Agency for Healthcare Research and Quality (AHRQ) of almost 450 RCTs enrolling more than 24,000 patients found no evidence of harm. A caveat is that safety assessments in the trials that AHRQ reviewed lacked the rigor needed to conclude that probiotics have no risks definitively⁷⁷. In a 10-year observational study at the Fred Hutchinson Cancer Research Centre, a bloodstream infection due to a probiotic organism occurred in 0.5% of hematopoietic cell transplant recipients. Other infectious complications in case reports include endocarditis, abscesses, and aspiration pneumonia⁵⁸. In some cases, contamination of a central venous access port was the likely route of entry⁷⁸. In theory, active inflammatory bowel disease could increase the risk of probiotic bloodstream infections because mucosal ulcerations in the lower

GI tract increase the risk of bacterial translocation into the systemic circulation⁷⁹. However, these are case reports and not RCTs. Probiotic bacterial strains can carry and transmit antimicrobial resistance genes to other bacterial species⁸⁰. In one case, resistance genes in *Lactobacillus reuteri* ATCC were removed to create a daughter strain; *Lactobacillus reuteri* DSM 17938⁸¹. A rodent study has documented the transfer of a plasmid carrying the vancomycin resistance gene from *Enterococcus faecium* to a *Lactobacillus acidophilus* probiotic strain. Concerns about antimicrobial resistance gene transfer by probiotics is a theoretical risk that deserves attention as more probiotics are consumed⁸².

PROBIOTIC QUALITY ISSUES

Although they are typically marketed for use by healthy people, pharmacists are increasingly responsible for selecting probiotics to treat or prevent medical conditions in patients who are not healthy. Product quality is an important consideration when choosing a supplement like a probiotic. In 2014, the issue was brought to the forefront when a premature infant died from a rare fungal infection that was likely caused by mold contamination of a probiotic^{83,84}. In another case, milk protein contamination of a probiotic was linked to anaphylaxis in an infant with cow milk allergy⁸⁵. Egg protein and gluten has also been detected as unlabelled ingredients in probiotic products^{86,87}. Gluten was detected in 8 of 15 probiotic supplements that were labeled gluten-free, including two that contained more than 20 parts per million^{88,89}. Dietary supplements containing gluten, mainly probiotics, have been linked to a higher rate of celiac symptoms in patients with celiac disease⁸⁹. Additional probiotic quality problems have included misidentification of bacterial strains, sub potent doses, poor shelf-life stability, and reduced functional properties⁹⁰. Reassuringly, a recent study performed viable colony counts on some popular probiotics and found they were similar to the CFUs stated on the product label⁹¹. Probiotic integrity can be damaged by conditions such as temperature extremes, humidity, and exposure to oxygen. The susceptibility to damage from temperature extremes varies between products. VSL#3 and Visbiome require refrigeration during shipping and storage to preserve bacterial potency⁹².

Bacillus coagulans MTCC 5856 has demonstrated viable spore counts at ambient temperatures for up to 3 years⁹³. *Saccharomyces boulardii* CNCM I-745 is stable at room temperature for three years,

and should not be refrigerated out of concern that moisture may affect potency⁹⁴. Microencapsulation may help to extend the shelf-life of some probiotics⁹⁵. Pharmacists should check the product labeling for storage requirements because they are product specific and educate consumers accordingly. Although an expiration date is not a legal requirement, a high-quality probiotic supplement should have one^{96,97}. Probiotics should contain enough viable CFUs to provide an adequate dose at the end of the product's shelf-life⁹⁸. Therefore, ISAPP advises consumers to avoid products that state the number of CFUs at the time of manufacture⁹⁸.

PROBIOTIC PRODUCT SELECTION

To ensure they are recommending high-quality probiotic products, pharmacists can ask manufacturers directly for product quality data, or they can use third-party verification programs⁹⁹. A growing number of United States Pharmacopoeia (USP) monographs have been published in the *Food Chemicals Codex* to establish strain-specific testing standards for identification, concentration, and contamination^{100,101}. In the future, pharmacists may rely on the USP Dietary Supplements Verification Program to help ensure probiotic product quality. This certification program includes rigorous product testing and chemistry, manufacturing, and controls documentation review¹⁰¹. Unfortunately, it does not list any probiotic products yet. The first probiotic product facility successfully passed the USP good manufacturing practices (GMPs) audit in September 2017. The Natural Products Association also certifies that supplements meet GMPs for dietary supplement manufacturing. Specifications include identity, purity, strength, and composition. Companies that receive an "A" compliance rating after third-party auditing of manufacturing facilities can use The Public Health and Safety Organization, formerly known as the National Sanitation Foundation (NSF) GMP certification seal¹⁰². Other verification programs that certify probiotics include ConsumerLabs.com and NSF. ConsumerLabs.com, a subscription-based service, purchases samples in stores and tests them annually for content and contamination, while USP and NSF usually obtain them directly from manufacturers¹⁰³. In addition to these seals and others, probiotic supplements often carry seals indicating the product is manufactured in an FDA-registered facility, gluten-free, genetically modified-organism-free, and dairy-free. It is reasonable to ask manufacturers for documentation to support the claims made on seals when choosing a probiotic for a patient with dietary restrictions¹⁰⁴.

INTERACTIONS OF PROBIOTICS WITH FOOD AND DRUG

Consumers often ask whether they should take a probiotic with food or on an empty stomach. In theory, certain foods may buffer gastric acid and improve the chance of a probiotic reaching the colon¹⁰⁵. In an in vitro model, *Lactobacilli* probiotics' survival (*Lactobacillus helveticus* R0052, *Lactobacillus rhamnosus* R0011, *Bifidobacterium long* R0175) was most significant when given with a meal or 30 minutes before a meal. It was improved by administration with dairy products compared with apple juice or spring water^{106,107}. Survival was worse when taken 30 minutes after a meal. Survival of *Saccharomyces boulardii* strains was generally not affected by in vitro meal conditions¹⁰⁸. In a clinical trial, giving a *Bifidobacterium bifidum* strain Bb probiotic capsule on an empty stomach did not prevent colonization in the lower GI tract¹⁰⁹. Collectively, these studies suggest that some probiotic strains may be taken without regard to meals, while others may benefit from administration right before or with a meal. For now, the best advice to consumers may be to follow instructions on the product label. Administering specific formulations as they were given in clinical trials is also reasonable. Patients taking a probiotic to prevent AAD often ask if antibiotics destroy the probiotic. There is no definitive answer, but they can be reassured that probiotics reduced the risk of AAD in clinical trials despite being taken during a course of antibiotics¹¹⁰. In one study, co-administration of oral doxycycline did not impact the survival of a probiotic containing *Lactobacillus acidophilus* LaCH-5 and *Bifidobacterium animalis* subsp *lactis* Bb-12. Empirically, a probiotic bacterial strain is unlikely to be killed if it is not susceptible to the antibiotic the patient is taking. This type of data can help to address antibiotic-specific concerns. For example, in vitro susceptibility studies have found that most *Lactobacilli* are variably sensitive to penicillins, cephalosporins, fluoroquinolones, and macrolides. Most are not susceptible to cotrimoxazole, metronidazole, or (except *L. acidophilus*) vancomycin¹¹¹. Destruction of *Saccharomyces boulardii* by antibacterial agents is not a consideration because it is a fungus, but it is likely susceptible to destruction by azole antifungals such as fluconazole. Some resources empirically recommend separating the administration of probiotic bacterial strains from antibacterial drugs with activity against them by at least 2 hours¹¹². Product labeling for VSL#3 recommends avoiding antibiotics altogether, while the manufacturer of Vis biome advises separating doses by at least 4 hours.

Probiotics are not known to interact with immune suppressants directly. However, the presence of immune-suppressing drugs on a patient's medication profile should alert pharmacists to consider whether the patient may be at risk for infection from probiotic use. Patients such as these may want to seek advice from their health care provider before using a probiotic¹¹³.

COUNSELLING PATIENTS ABOUT PROBIOTICS

When helping consumers select a probiotic for a specific condition, direct them towards products containing strains and doses shown to have benefit, based on the intended use. Review product-specific instructions for administration and storage, and avoid products that are near or past their expiration date¹¹⁴. For chronic conditions, counsel consumers that continued use of a probiotic are usually advised because probiotics do not cause long-lasting changes in the gut microbiota^{35,115}.

LIMITATIONS OF PROBIOTICS

Mainly probiotics are associated with four types of side-effects viz systemic infections, deleterious metabolic activities, excessive immune stimulation in susceptible individuals, and gene transfer. Some factors like temperature, moisture, and air affect during storage of probiotics. Oxygen toxicity is a major problem in the survival of probiotics bacteria in dairy foods. Elevated levels of oxygen in the product are fatal to the viability of these anaerobic bacteria¹¹⁶. Probiotics are limited to products that-

1. Consists of live microorganisms (e.g., fermented products)
2. Stabilized the health, growth, and well being of humans or animals and
3. May destroy the host mucosal surfaces.

It was found that all probiotics strains are not effective, and considerable strain-to-strain variation in properties related to probiotic efficacy is observed within bacterial species.

CHALLENGES OF PROBIOTICS

Probiotics can associate with commensal bacteria and may also have a direct action on the host. To know this association is one of the main challenges for future research. Others include knowing the mechanism of action of probiotics to elucidate more specifically which probiotic strains possess which

health benefits and to explain the quantity needed to attain those effects¹¹⁷. Clinical and mechanistic studies are needed to better understand the interface between the microbes, host cells, mucus, and immune defences and to form efficacious interventions. Such studies comprise of molecular examination of the intestinal flora and long term (5-10 years) effects of probiotics microorganisms¹¹⁸. Retention of the viability of the probiotics bacteria is a major challenge for marketing and technological application of probiotic cultures in functional foods¹¹⁹. The production of probiotics in large scale industries faces mainly two main challenges

1. The low-cost production of probiotics.
2. Better probiotic viability after storage, during the manufacturing process of the functional food, and during transit through the stomach.

FUTURE PERSPECTIVES OF PROBIOTICS

To access the safety and efficacy of probiotics various *in-vivo* and/or *in-vitro* studies were done. Presently, probiotics like *Lactobacillus*, *Bifidobacterium* do not need FDA approval and are present in our daily diet. Though the safety of probiotics for food application has been confirmed by several authorities worldwide, few studies found the incidence of bloating, flatulence, and high osmotic pressure which leads to gastrointestinal discomfort¹²⁰.

The genetic interactions between ingested probiotics and the native intestinal microbes may be considered a topic of interest. The genetic materials may be altered by transduction, conjugation, and transformation. The transformation of intestinal microflora by DNA may be increased by the ingestion of bacteria, which leads to genetic rearrangements. Besides this, the transmission of antibiotic-resistant genes in beneficial bacteria and pathogens could interact with a complex microflora colony in the Gastrointestinal tract. This process can, in turn, leads to the evolution of antibiotic-resistant probiotics and the potential emergence of resistant pathogens¹²¹.

Probiotics in Clinical Studies

A study demonstrated that *Bifidobacterium* BB-12 reduces symptoms of atopic eczema, immune-modulating influence, increases the possibility of H.Pylori eradication, functions as a shield against mild diarrhea¹²². Another study on Probiotic strain

Lactobacillus casei 431 has shown that the strain modulates the immune response following vaccination and promotes fast recovery from diarrhoeal attacks in infants¹²³. Similarly Probiotic strain *L. Acidophilus* NCFB 1748 decreases fecal enzyme action, lowering fecal mutagenicity, protection of radiotherapy-related diarrhea, enhancement of constipation¹²⁴. *Lactobacillus* F19 enhances gastrointestinal flora and boost immune health. *L. Casei* protects intestinal disturbances, decreases fecal enzyme activities, positive effects on downregulating the recurrence of superficial bladder cancer¹²⁴. *L. Rhamnosus* GG is used to treat diarrhea. Another probiotic strain *Lactobacillus salivarius* WB21 regulates oral health in subjects at a high risk of periodontal disease¹²⁴.

CONCLUSION

Therapeutic application of probiotics has been widely studied to treat and improve the health of the Gastrointestinal tract, however, choosing a probiotic for a specific condition is an increasingly complex task that requires parsing of the data for strain-specific efficacy and evaluation of product quality. However, due to much new research, reviews, and evidence-based guidelines, it is becoming much more comfortable to choose an appropriate probiotic. Pharmacists provide a valuable service by helping their patients decide when a probiotic is worth trying, recommending which product may be helpful, and advising how to use it. It appears likely that more national organizations will be conducting evidence-based research and the pharmacist and clinicians should pay attention to this ever-changing field.

CONFLICT OF INTEREST

Nil

AUTHOR'S CONTRIBUTION

BD, BS, BB, and AMAI have contributed to the preparation of the manuscript under the supervision of Dr. Biswajit Dash. All the authors approved the manuscript for publication.

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