Digestive and Liver Disease 49 (2017) 1177–1184

Contents lists available at ScienceDirect

Digestive and Liver Disease

journal homepage: www.elsevier.com/locate/dld

Review Article

A consumer's guide for probiotics: 10 golden rules for a correct use

Marco Toscano[a], Roberta De Grandi[a], Luca Pastorelli[b,c], Maurizio Vecchi[b,c], Lorenzo Drago[a,d,*]

[a] Laboratory of Clinical Microbiology, Department of Biomedical Science for Health, University of Milan, Milan, Italy
[b] Department of Biomedical Science for Health, University of Milan, Milan, Italy
[c] Gastroenterology and Gastrointestinal Endoscopy Unit, IRCCS Policlinico San Donato, San Donato Milanese, Italy
[d] Laboratory of Clinical-Chemistry and Microbiology, IRCCS Galeazzi Institute, University of Milan, Milan, Italy

ARTICLE INFO

Article history:
Received 15 February 2017
Received in revised form 19 July 2017
Accepted 20 July 2017
Available online 1 August 2017

Keywords:
Anti-inflammatory response
Gut microbiota
Multistrain products

ABSTRACT

Probiotics are used all over the world as their beneficial effects on the human organism have been widely demonstrated. Certain probiotics can down-regulate production of pro-inflammatory cytokines and promote intestinal epithelial barrier functions, increasing an anti-inflammatory response and contributing to the host's overall health. The main mechanisms by which probiotic microorganisms can interact with the host are by modulating the immune system and the epithelial cell functions and interacting with intestinal gut microbiota.

To date, hundreds of different microorganisms are used for the formulation of numerous probiotic products; therefore, it is very difficult to choose the best probiotic product for specific or more general needs. Therefore, physicians are getting more and more confused due to the high number of commercial products which are often lacking healthy effects on the host. Therefore, the aim of this paper is to demonstrate the main characteristics that probiotic microorganisms and products should possess to have a positive impact on the host's health. To this purpose, this review suggests "10 golden rules" or "commandments" that clinicians should follow to properly select the optimal probiotic product and avoid misidentifications, mislabelling and "*pie in the sky*" stories.

1. Introduction

In the last few years, a growing interest in studying and using probiotic microorganisms has been observed, not only for the treatment of gastrointestinal diseases but also for the improvement of overall human health. Indeed, several studies highlighting both the systemic activity of probiotics and their beneficial role in ameliorating diabetes and allergic diseases management have been published [1–4].

The leading mechanisms by which probiotics are thought to be effective on the host's health are their ability to modulate the intestinal immune system, to improve intestinal physical and immunological barrier functions [5,6] and displace potential pathogenic microorganisms by competitive exclusion through the production of antimicrobial peptides [7]. Recently, there has also been a great interest in studying the effect of probiotics in modulating the gut microbiota composition [8–11]. Commensal bacteria belonging to intestinal microbiota protect the host from the action of pathogens, regulate the host's fat storage, stimulate intestinal angiogenesis and the immune system and aid the digestion of numerous dietary components [8,11]. However, gut microbiota is often associated to many conditions of clinical interest, in particular dysbiosis [11]. Consequently, it is clear that the manipulation of intestinal microbiota composition by means of probiotics may be a promising approach to ensure the correct maintenance and improvement of human health.

To date, hundreds of different intestinal microbial species are used in the preparation of probiotic supplements and, for this reason, it is very difficult to choose the probiotic product that is best suited to a patient's needs.

The common approach to this issue should be one that follows a set of specific guidelines, which guarantee a probiotic product's quality and efficacy, such as:

- proper microbial species and strain identification of all microorganisms contained in the product, with the deposit of all strains in an international culture collection;
- characterization of the safety and probiotic efficacy of each strain;
- evaluation of the microbial beneficial effects on the human host.

However, today it is very difficult to identify which probiotic formulation is the best one to improve the human health, mostly

* Corresponding author at: IRCCS Galeazzi Orthopaedic Institute, Milan, Italy.
E-mail address: lorenzo.drago@unimi.it (L. Drago).

http://dx.doi.org/10.1016/j.dld.2017.07.011
1590-8658/

Table 1
The 10 recommendations of probiotics.

1st	Knowing the correct definition of probiotics
2nd	Microbial lysates, non-living bacteria and non-colonizing spores cannot be considered probiotics
3rd	Getting an exhaustive probiotic identikit
4th	Monostrain or multistrains products: making the correct choice
5th	Avoid antibiotic resistance genes in probiotic strains and products
6th	Choose probiotic strains resistant to gastrointestinal environment
7th	Probiotic strains must be able to colonize the gut
8th	Preferring probiotics that are able to positively interact with gut microbiota
9th	Be sure about the safety of probiotic strains and evaluate the subject health status before probiotic administration
10th	Preferring probiotics with a demonstrated clinical efficacy

because of the lack of knowledge of specific probiotic features, which should be considered.

The present paper aims to clearly inform Manufacturers and Clinicians on the basic characteristics that probiotic microorganisms, and above all, probiotic products should possess to be used as positive bio-modulators of human health. This study also aims to provide a useful and quick "instruction kit" for physicians to follow, in order to give an easy and immediate interpretation of the probiotic(s) under consideration. The 10 "commandments" or "recommendations" clinicians should never forget when dealing with probiotics are listed in Table 1.

2. 1st commandment: know the correct definition of probiotics

The classic definition of probiotics is "live microorganisms which, when administered in adequate amounts, confer a health benefit to the host" [12]. Interestingly, bacteria belonging to *Lactobacillus* and *Bifidobacterium* genera are the most used probiotic microorganisms in the food industry, due to their probiotic and beneficial effects. However, also non-bacterial microorganisms, such as *Saccharomyces boulardii* and *Saccharomyces cerevisiae*, are often used as probiotics because of their postulated activities and beneficial effects both at intestinal and systemic level.

Sometimes, probiotics are referred to as "biotherapeutics" or "pharmacobiotics", leading to a profound misunderstanding of what probiotics are [13]. Indeed, the term "therapeutic" indicates the handling of a disease and, therefore, should be referred only to a drug, while probiotics can have a broader meaning [13]. Conversely, the term "pharma" is mainly related to pharmaceuticals, either drugs or chemical components [13]. On the contrary, probiotics are not drugs but, instead, they are "biomodulators" that must be resistant to gastric juice and bile salts to arrive intact to the intestinal environment and exert their beneficial activity on the host's organism [7]. Indeed, a probiotic microorganism should also be able to positively influence the host's health, leading to beneficial effects after its intake [7].

2.1. Recommendation

Probiotics are live microorganisms acting as powerful biomodulators, with a positive impact on human health. Indeed, the official definition of probiotics given by FAO/WHO Committee is "live microorganisms which, when administered in adequate amounts, confer a health benefit to the host."

3. 2nd commandment: microbial lysates, non-living bacteria and non-colonizing spores cannot be considered probiotics

In the last years, there was an increasing interest in studying microbial components (e.g. proteins, lipids or nucleic acids) and non-living bacteria for their beneficial effects on the human organism. Several evidences highlighted the ability of dead cells to induce a wide range of biological responses in the host enhancing the anti-inflammatory response [14–16]. The administration of heat-killed bifidobacteria and lactobacilli, for instance, has been observed to induce a pronounced increase in the production of anti-inflammatory cytokines and modulate positively allergic symptomatology [16,17]. Consequently, the immunostimulatory effect of probiotics seems not to depend on the cells being alive but it may be linked directly to the biological and physical nature of specific microbial components. Indeed, there is also evidence that metabolites and cell fractions of probiotics can exert a positive function on the host [14]; DNA, lipopolysaccharides, peptidoglycans and cell homogenates have all a strong immunomodulatory effect acting on the innate immune system and they have been demonstrated to decrease the risk of atopic dermatitis onset in children [18–21].

Although microbial lysates have a beneficial impact on the host health, they cannot be considered as probiotics; indeed, they are effective on the organism only if continually administered not being able to induce biological responses for a long period. Conversely, only alive microorganisms can have a lasting impact on the organism being able to colonize the gastrointestinal environment and so persist in the host.

Regarding microbial spore formers, instead, only spores that can germinate in the gastrointestinal tract and colonize the environment can be considered as probiotics and used in the formulation of products [22]. Spores can survive transit across the stomach barrier and are more resistant than vegetative cells; moreover, they can germinate in presence of nutrients and favourable environmental conditions becoming able to colonize the gut and exert their probiotic effects [22]. However, if spores do not germinate they are not active at intestinal level and no beneficial effects on the host will be highlighted.

3.1. Recommendation

Although microbial components have been observed to have a beneficial impact on the host's immune system and health, they cannot be considered as probiotics, as well as non-colonizing spores.

4. 3rd commandment: get an exhaustive probiotic identikit

In the last few decades, advances in molecular biology and microbiology techniques have allowed a clear identification and characterization of bacterial strains, avoiding any confusion about probiotic identity [23]. Identification and tracking of individual strains is essential when microorganisms are used for the formulation of probiotic products; to this purpose, phylogenetic analysis is the most powerful tool for bacterial taxonomic classification, as ribosomal RNA sequences provide detailed information for a comparative identification of probiotic microorganisms [24]. When a microorganism is used for the formulation of food supplements or other products for human consumption, its full characterization at both genomic and physiological level is critical. Each microorganism contained in probiotic products must be identified at species and strain level, according to the International Code of Nomenclature and its microbial genome must be completely sequenced. This allows the identification of every single gene involved in bacterial metabolism and its function. Consequently, the safety of food products and, above all, of commercial probiotic strains should be evaluated before their launch on the market. Generally, the guidelines require: