



An Overview of Guidelines and Regulations for Probiotics

Virender K. Batish

Molecular Biology Unit

Dairy Microbiology Division,

NDRI, Karnal (Haryana) 132 001, INDIA



Probiotics – the magic bugs

- **Boom in probiotic based functional / health foods**
- **Catchy slogans – billion a day keeps Dr. away**
- **Probiotics as alternative therapy**
- **Exaggerated health claims without scientific evidence**
- **Optimism versus skepticism**
- **Unreliable, inferior and inconsistent quality / spurious products**
- **Consumer confidence and acceptability ?**
- **Urgent need for guidelines / regulations for probiotic management at global level**

World Scenario of Regulatory approaches on Probiotics

- **Probiotic standards / regulations differ among different countries**
- **No harmonization of standards among EU countries and USA/ Canada**
- **As foods/ functional foods/ Novel Food/ Natural remedy (Denmark/ Sweden/ Finland)**
- **As natural health product (Canada)**
- **As Dietetic food (Italy)**
- **Dietary supplement (USA)**
- **Biotherapeutic / Pharmaceuticals (Canada, China, East European countries / France/ Germany / Belgium/ Austria / Italy)**

Current Status of National/ International Regulatory Agencies Standards for Evaluation and Safety of Probiotics in commercial products

- **FAO / WHO (www.fao.org) (www.fao.org) – Worldwide**
 - Developed guidelines for evaluation of probiotics
- **IDF (www.fil-idf.org) - Worldwide**
 - Joint action team on establishing methods to determine functional / safety attributes of probiotics in food in line with FAO/WHO
- **EFECA (www.efecca.com) – Europe**
 - Developed guidelines for use of probiotics in foods
- **Codex standards for fermented milks (243-2003) (www.codexalimentarius.net) – Worldwide**
 - Minimum number of characterizing and additional level of microbes in yoghurt / acidophilus milk / kefir / kumiss and others
- **NYA (www.aboutyoghurt.com) - US**
 - Minimum level of viable cultures
- **FDA (www.fda.org) - US**
 - Minimum level of viable cultures

FAO/WHO Standards/ guidelines on Probiotic s

- Joint FAO/ WHO Expert Consultation on Evaluation of Health and Nutritional properties of Probiotics in foods

(Argentina, 1-4th Oct., 2001)

Working group	guidelines, criteria / methodology for evaluation of Probiotic
	Identify and define data for substantiating Health Claims
	Minimum requirements for Probiotic status
Scope	Probiotics as foods only
	Excluded GMOs / Bio-therapeutic agents

Definitions of Probiotics

No universal legal definition

‘Probiotics as live micro -organisms which when administered in adequate amounts confer a health benefit on the host’

(FAO/WHO, 2001)

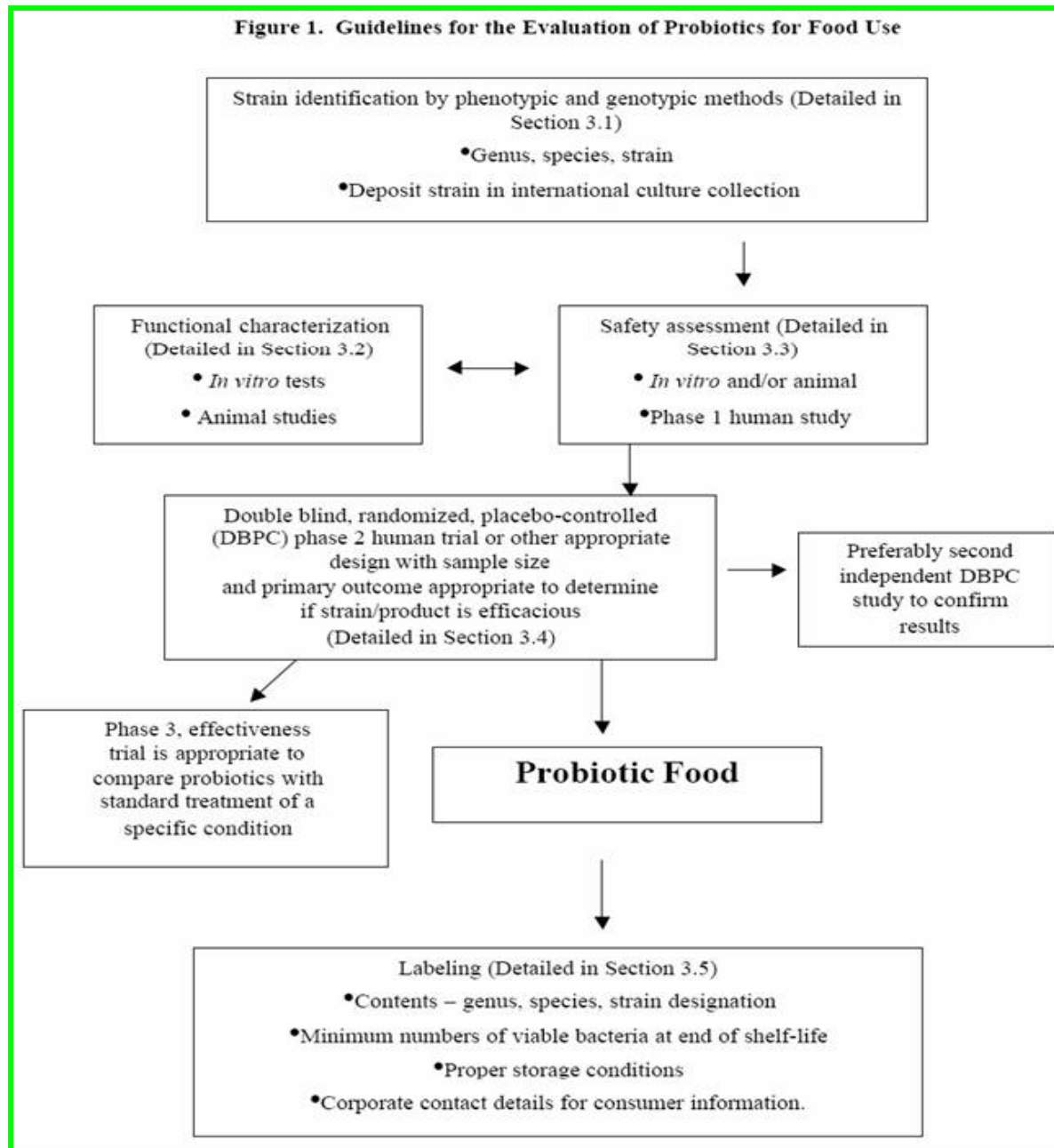
Probiotics are microbial cell preparations or components of microbial cells that have a beneficial effect on the health & wellbeing of the host.

(Salminen *et al.*, 1999)

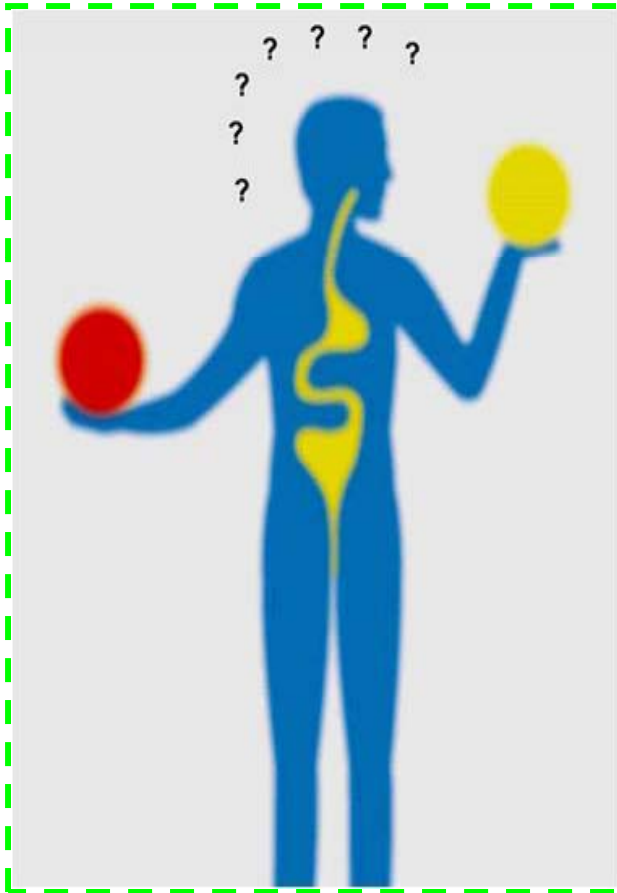
FAO/WHO- Guidelines for the assessment of Probiotics

- **Definition of Probiotics**
- **Identification at Genus, species and strain level (Phenotypic and genetic Tests)**
- **Probiotic Function – strain specific**
- ***In vitro* tests for evaluation of probiotics**
- **Safety Assessment- *In vitro* / animal studies/ clinical studies (Human) Phase I/ Phase II/ Phase III**
- **No adverse effects related to Probiotic administration to occur**
- **The outcome of human studies (even with negative results) to be published in a peer reviewed journals**

Figure 1. Guidelines for the Evaluation of Probiotics for Food Use



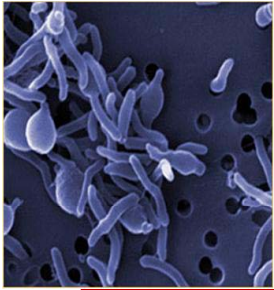
Selection of probiotic strains for human use



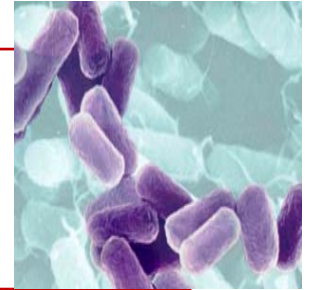
Specificity of the action of probiotic bacteria –not the source that is more important

Viability at the target site and effectiveness

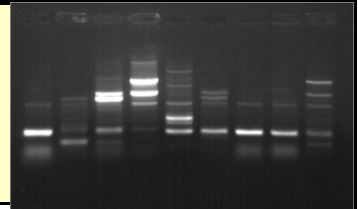
Refinement of *in vitro* tests to predict the ability of probiotics to function in human



Classification and identification at strain level



The probiotic functions – highly strain specific



Identification as per international code of nomenclature at strain level

Potential molecular tools for probiotic identification – DNA-DNA hybridization, 16S rRNA sequencing, RAPD, RFLP, FISH, probes as a complement to phenotypic tests

Defining and measuring the health benefits of probiotics

✓ Dose and duration of probiotic use – critical for measuring their efficacy to confer health benefits on the host

✓ Minimum daily amount required to confer the purported health benefits is to be defined explicitly based on *in vitro* and *in vivo* studies on cell lines and animal models as well as humans clinical trials

Testing methods for establishing health benefits conferred by probiotics

- **Well standardized *In vitro* studies for establishing the health benefits prior to *in vivo* trials**
- **Battery of tests such as acid and bile tolerance, antimicrobial production and adherence ability to be conducted depending on the purported health benefits**
- **In-vivo testing – RDBPC human trials by enrolling adequate number of subjects to achieve statistical significance**

Safety considerations – specific tests for assessing probiotic safety

- ✓ **Antibiotic resistance patterns**
- ✓ Undesirable metabolic activities (D-lactate, bile salt deconjugation)
- ✓ **Side effects during human studies**
- ✓ Epidemiological surveillance of adverse incidents in consumers (post market)
- ✓ **Toxin production**
- ✓ Hemolytic activity
- ✓ **Lack of infectivity in immunocompromised animal models**



FAO/WHO recommended In vitro tests for Probiotics

- **Human Origin**
- Resistance to gastric acidity
- **Bile salt resistance (Colonization)**
- Adherence to mucus / human epithelial cells
- **Antimicrobial activity against pathogens**
- Ability to reduce pathogen adhesion to surfaces
- **Bile salt hydrolase activity (As a Probiotic Marker)**
- Resistance to spermicides (vaginal application of Probiotics)



Probiotic product specifications – Quality Assurance and Regulatory Issues



Regulatory issues

Regulations differ among countries

Status of probiotics as a component of food not established on International basis

Disease reduction claims to be permitted for specific probiotics if demonstrated successfully

Appropriate labeling

Identity of microbial strains to be stated on the label

**Indicate viable concentration of each probiotic
at the end of shelf life by enumerating probiotic
bacteria in food products**

Manufacturing and handling procedures

Stock cultures be maintained under appropriate conditions to ensure that the given culture maintains the beneficial properties

Check purity, viability and probiotic activity throughout processing, handling and storage of the food product containing probiotic and verify it at the end of shelf life (follow QA)

Post market surveillance

All the stake holders to develop some form of system to monitor the health outcome of long term probiotic administration

Potential side effects and long term benefits to be recorded and documented

Proper trace back system – a pre-requisite for surveillance of probiotic products

*Regulating Pre- and Pro-biotics:
a US FDA Perspective*

Objectives

- **Review the terms probiotic, prebiotic, and biotherapeutic.**
- Review the regulatory definition of drug, biologic, dietary supplement and GRAS.
- **Discuss the regulatory difference between a dietary supplement and a drug/biologic.**
- Focus on the regulation of probiotics as biologics.
- **Discuss regulatory considerations in the development of probiotics for clinical indications.**

Non-regulatory Terms

- **Probiotic**
- **Prebiotic**
- **Live Biotherapeutic**

“Live Biotherapeutic”

- **CBER/OVRR Working Definition**
- **Live microorganisms with an intended therapeutic effect in humans.**
 - **May include bacteria or yeast**
 - **May be used in disease prevention or treatment**
 - **Intended local or regional action**
 - **Includes “probiotics for clinical use”**
- **“Biotherapeutic Agent”**
 - **“Microorganisms having therapeutic effects in humans.”**
 - **Excludes “probiotics,” defined as “microorganisms having general beneficial effects on the health of animals or humans.”**

*How are probiotics and
prebiotics regulated in the US?*

Intended use determines how a substance is regulated?

Regulatory Definitions

**Drug – article intended for use in the diagnosis, cure, mitigation, treatment, or prevention of disease
(Food, Drug & Cosmetic Act of 1938).**

**Biological product – a “virus,” (i.e., bacteria, fungi, etc.) “...etc.... applicable to the prevention, treatment, or cure of a disease or condition of human beings
(Public Health Service Act of 1944, 42 USC §262).**

**Dietary supplement - a product taken by mouth that contains a “dietary ingredient” intended to supplement the diet
(DSHEA of 1994, amendment to the Food, Drug & Cosmetic Act).**

GRAS

**“Generally Recognized As Safe”
21 CFR 170.3 and 170.30**

A food ingredient classification that distinguishes a substance from a food additive on the basis of common knowledge about safety for its intended use.

Pertains to the use of a food substance, rather than the substance itself.

Dietary Supplement v/s Biological Product

A probiotic may be marketed and regulated as a dietary supplement and/or a biological product. It all depends on how it is intended to be used.

Biological products require premarket review and approval by FDA. Dietary supplements do not.

The safety, purity and potency, as well as efficacy, of a biological product must be demonstrated for approval. Dietary supplements need not demonstrate any of these to be marketed.

FDA's Center for Food Safety and Applied Nutrition (CFSA) regulates probiotics and prebiotics marketed as dietary supplements or food ingredients.

When is a probiotic a drug/biological product?

A probiotic used for diagnosis, cure, mitigate, treat, or prevent disease is a drug and a biological product.

FDA's Center for Biologics Evaluation and Research (CBER) regulates probiotic products when used for clinical indications

CBER's Office of Vaccines Research and Review has regulatory jurisdiction over most probiotic products for clinical use.

Currently, CBER/OVRR views probiotics for clinical use as live biotherapeutics

CBER/OVRR has vast experience in regulating products that contain microorganisms and/or their components

Focus and experience to date has been on the safety and activity of 'live' agents

Regulating the Safety of Probiotics – The European approach

- **Probiotics for human foods not governed under specific EU regulatory frame works.**
- **Novel Food regulation EU 258/97 relevant in some specific cases.**
- **Probiotics as feed additives – regulated by Council Directive 70/254/EEC & in accordance with the guidelines of SCAN**
 - Transmissible antibiotic resistance markers?
 - Production of harmful metabolites?
- **EU guidelines do not differentiate between species & strains with long history of safe use.**
- **Concern regarding over-regulation of Probiotics for human food use.**
- **Concept of Qualified Perception of Safety (QRS)**
 - allow strains with established safety status to enter the market without any testing requirement.
- **EFSA – Central role in the regulation of both human & animal probiotics.**

Japanese perspective

- **Japan – a key player in probiotic based functional foods accounting for more than half of global probiotic foods market.**
- **Japan – the only nation that has legally defined functional/health foods.**
- **Introduction of FOSHU system to regulate foods for specific health use.**
- **Unique features of FOSHU**
 - Voluntary with government approval
 - Specific health claims approved prior to use.
 - Approval based on documented scientific evidences.
 - FOSHU Label on the approved products.
- **MHLW – new regulatory system – Foods with health claims (FHC) and foods with nutrient functions (FNFC) in 2001.**

Japanese perspective

- **New Subsystems of FOSHU**
 - Standardized FOSHU
 - Qualified FOSHU
 - Disease risk reduction claims for FOSHU.
- **336 probiotic based products approved by FOSHU.**
- **FOSHU – a role model for other countries.**
- **Application for grant of FOSHU Label submitted to local regional authorities/MHW (1 year processing time) with following inputs :**
 - Scientific evidence for health claim.
 - Recommended dose of the functional food/ingredient.
 - Safety of the ingredient.
 - Physical & chemical characteristics.
 - Relevant test methods.
 - Compositional analysis.
 - Scientific paper that substantiate health claims.

Canadian perspective

- **In Canada, probiotics regulated by natural health product Directorate (NHPD) which became a law in 2004**
- **Covers therapeutic claims, risk reduction and structure / function claims**
- **Under item 8 (schedule 1) – the term probiotic is defined as monoculture or mixed culture of live micro-organisms that benefit the microbiota indigenous to humans**
- **Provision of dead microbes or other microbes not labeled as probiotics**
- **Direct fed microbes for animal nutrition covered under the jurisdiction of Canadian feed inspection agency (CFIA)**

Contd....

- **Health claims based on standard of evidence presented by NHPD's clearly defined criteria**
- **New NHPD guidelines allow for qualified claims based on the weight of evidence provided**
- **The design of clinical trials to assess probiotics follow the ICH and GCP guidelines used for pharmaceutical products**

Indian Scenario

- **Regulatory standards and guidelines of probiotics no formulated as yet.**
- **India – a potential market for Probiotic foods**
- **Entry of spurious products with false claims.**
- **Non-availability of proven and well characterized Indian Probiotic cultures.**
- **Poor colonization of commercial strains of western origins in Indian gut due to short transit time.**
- **Need for development of indigenous probiotic strains for expressing optimal functionality.**
- **Holistic approach for formulating guidelines and regulations for evaluating the efficacy of Indian probiotics in harmony with international standards.**

Indian Scenario

- **Currently foods regulation by PFA and drugs by FDA in India.**
- Initiatives being taken by DBT/ICMR/MFPI to formulate guidelines on Probiotics.
- **Food safety and standards Act passed in parliament (2006) to regulate foods, dietary supplement & nutraceuticals.**
- Creation of Food Safety and Standards Authority (FSSA, 2005)– drafting rules and regulations in the food sector
- **Liberal regulations permitting claims if not misleading**
- Stringent regulations for Food supplements/biotherapeutics for ensured quality and safety.

Risk concerns associated with Probiotics – Regulations

- **Regulation of Probiotics as Foods**
- **Food/dietary supplement**
- **Pharmaceutical a biological products**
- **No stringent requirements when used as food/dietary supplements**
- **Consistencies between claimed & actual contents.**
- **Legal requirements when intended for use in infants/children in Europe.**
- **In U.S., no pre-market review & approval by FDA. FDA approval needed when marketed as biologicals specifically for treatment/prevention of target disease.**
- **IN Australia – Probiotics as complementary medicine for specific health benefits require pre-market review by Therapeutics goods Administration.**
- **Japan – General probiotics products have FOSHU status for specified health use – premarket review by Health Ministry.**
- **Probiotics – GRAS status – safety concerns in particular population (sepsis, bacteriocin, endocarditis).**

Current status of Marketing of Probiotics

- Marketing of Probiotic products concurrent with science based substantiation of product efficacy.
- **Enforced regulation on Probiotic quality & efficacy - non existent worldwide.**
- Recall of Probiotic products in USA due to non allowable disease claims.
- **Mislabeled products wrt genes, species, cell count or unsubstantiated structure – function statements invariably found in the market in almost all the countries.**
- Consumer disillusionment & loss of confidence – declining market.
- **Regulating approaches differ among different countries & hence no harmonization.**
- Regulations & guidelines yet to be formulated in developing countries including India.
- **Holistic approach on guidelines and regulations needed for each country for judicious & effective use of Probiotics.**

•

Key Debatable Issues

- **Scope for coining a new definition of “Probiotics” to address legal implications (Live versus dead organisms)**
- **Human origin versus nonhuman origin ?**
- **What should be the Gold Standard for identification of probiotics at strain level? (16s rRNA and other house keeping genes)**
- **Minimum criteria for qualifying the status of “Probiotics” based on specific well defined standard tests in quantifiable terms**
- **Lack of correlation between the results of historical *In-vitro* / *In-vivo* methods for probiotic selection and clinical manifestations in humans**
- **Selection criteria should differ with intended probiotic applications at different human ecosystems (oral / gut / vaginal)**
- **Use of probiotics with multiple antibiotic resistance for human application when used in conjunction with drugs to manage diarrheal diseases**
- **Is adhesion to epithelial cells a desirable criteria for probiotic selection to confer positive health effects? (strong versus poor colonizers)**

Contd....

- Identification of universal probiotic markers for mass screening of microbial cultures from different ecological niches
- No solid documentation that strains performing well in physiological tests perform any better in human evaluations than those performing poorly in these tests
- Establishing standard operating procedures for limited number of functional assays is premature considering the current status of research
- Concept of consortia of probiotic strains versus individual /single strains?
- No clarity on the effective doses of probiotics . May vary from strain to strain and the food and the host (age, sex and target specific)-healthy and immunocompromised
- Need for multicentric approach for human clinical trials for validating the specific health claims of probiotic strains for high level of confidence and statistical significance
- Formulating uniform Standard Operating Procedures (SOPs) and analytical assays for assessing the quality, safety and efficacy of probiotics

Conclusion

Regulatory standards and guidelines at National/International level are extremely important for assessing the efficacy and safety of probiotics

However, there is need for harmonization of these regulations and guidelines on probiotics at the global level to ensure the quality and safety of probiotic foods for their effective utilization in different countries

A Quote on Probiotic regulations & guidelines

We need someone like Bono to sit down with politicians & tell them that probiotics are integral to health, to the budget & for the future of the country

(Prof. G. Reid, 2007)