

## PŮVODNÍ PRÁCE

Evaluation of labeling and content of probiotics available in the Czech Republic

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### Summary

Probiotics are live cultures of mikroorganisms which are good for human health and are used in human medicine as a treatment. Advantageous effect on a human health was proved in many studies in Europe and in the USA. On the other hand, many studies described lack of information about products containing probiotics - in particular the real bacterial strain or quantity of bacteria. We tried to evaluate thirteen products containing probiotics available in the Czech Republic. In all cases the leaflets contained information about the species and amount of bacteria (from seven products containing more than one species of bacteria were only in two cases declared the quantity of particular bacteria, in five cases was declared just the total quantity of bacteria). The declared species of bacteria were cultured in all tested products. Concluding; the information on the leaflets of probiotic products is sufficient for a clinical practice and the real species and quantity of bacteria come up to declared data.

Key words: probiotics - information about a drug - cultivation

Obsah bakterií v probiotických dostupných na českém trhu

### Souhrn

Probiotika jsou živé kultury mikroorganismů příznivě působící na lidské zdraví a používané v léčbě mnoha chorob. Řada článků z USA i západní Evropy však popisuje nedostatečné informace o obsahu a množství bakterií v případě některých probiotik, a především fakt, že řada preparátů živé mikroorganismy buď vůbec neobsahuje, obsahuje jiné, či v menším množství. Vzhledem k tomu, že zprávy týkající se probiotik dostupných u nás chybějí, zhodnotili jsme informace a kultivačně vyšetřili 13 u nás dostupných probiotik. U všech byly informace o rodu i druhu obsažených bakterií, údaj o množství (ze 7 kombinovaných preparátů však jen u 2 o množství jednotlivých druhů; v ostatních případech bylo udáno jen celkové množství bakterií). Ze všech preparátů byl vykultivován udaný rod minimálně v udaném množství. Informace jsou pro klinickou praxi tudíž dostatečné a obsah i množství bakterií odpovídá deklarovaným množstvím.

Klíčová slova: probiotika - informace o léku - označení - kultivace

### Introduction

Probiotics are live microbiological substances which have been used as a treatment for over one hundred years. An increase of interest in probiotics was noticed in the last decade, particularly in the sphere of business. Although an advantageous effect on a human health was proved in more than fifty randomised studies in human medicine, a certain scepticism still exists. Both, specialists and patients, sometimes mistrust these products and do not believe what the producers declare. Some studies have documented discrepancies in information marked on the official leaflets, actual species and amount of contained bacteria.

With regard to the above mentioned facts we decided to evaluate information and content of thirteen products containing probiotics available in pharmacies in the Czech Republic.

### **Material and methods**

Thirteen commonly available products containing probiotics were evaluated in April 2005 and, after a week of recommended storage, examined using bacteriological culture. Products were assigned numbers 1 to 13.

The data was obtained from official leaflets or information on the package where the leaflet was missing. The products contained: once yeast fungus *Saccharomyces boulardii*, three times one bacterial strain (*Lactobacillus acidophilus*, *Escherichia coli*, *Enterococcus faecalis*), for four times it was a mixture of species (*Lactobacillus*), for three times a mixture of *Lactobacillus* with another strain and once *Lactobacillus* with vitamins (Tab. 1). The tablets and capsules were dissolved in broth and homogenized properly. Afterwards the broth was dissolved to obtain ten-fold dilutions with each dilutions being cultured using a calibrated inoculating loop (1 µl) on blood agar and VL agar for anaerobes. We kept the culture media at 37 °C for 48 hours under aerobic as well as anaerobic conditions. Then the growing colonies were identified according to their growth, morphology, microscopic findings and essential biochemical characteristics.

### **Results**

All products declared the expiry date and the amount of bacteria contained in one capsule. The supposed loss of the quantity of microorganisms during storage was evaluate in two products. Only in two products which included more than one species of bacteria (mixtures of probiotics) there was established the quantity of each particular bacteria and in six products was established the total quantity of bacteria. There was not a recommendation to take probiotics with food in six cases.

All products we examined contained the declared strain and quantity of bacteria indicated on the leaflet or package. In some products there was cultured even more bacteria (Tab. 2).

### **Discussion**

Yogurt has been considered desirable for more than a thousand years. The modern history of probiotics begins with a Russian scientist Ilja Metchnikoff who promoted eating yogurts and fermented milk products because it should have prolonged lives of certain ethnic groups using these food [1]. Germans as the first founder of probiotics consider Döderlein (1892) who suggested more than sixteen years before Metchnikoff to use live bacteria producing the lactic acid for vaginal infection by inhibiting the growth of pathogenic bacteria.

Originally the probiotics were defined as “microorganisms promoting the growth of other microorganisms“. Later they were defined as “live microorganisms causing or promoting the useful equilibrium of the autochthonous microbial population of the gastrointestinal tract“. These microorganisms do not need to be necessarily a permanent component of a gastrointestinal tract but should serve to promote the general state of health [4,5]. Nowadays probiotics are defined as “mono or mixed cultures of live microorganisms with a positive effect on a human health and microflora in the human intestine“ [6]. Benefits from using probiotics were published in more than fifty clinical studies. Probiotics are used in the therapy of diarrhoea of various etiology, inflammatory bowel diseases, liver damage - steatohepatitis, liver cirrhosis, or in alergology and so on [8,9,10].

In the last ten years we have noticed a huge increase of interest in using probiotics and it is very popular to produce them. There are currently plenty of products on the European and American market. Products are advertised and recommended to the public because of their benefits on human health. However, probiotics are only very slowly becoming part of official

human medical treatment - evidence based medicine. A scepticism still exists. There are of course a few reasons: extensive randomised studies of companies are missing, pharmaceutical companies do not invest in research, because it is too difficult to patent these products. In western Europe and also in the USA products containing probiotics are generally for sale without prescription. The evaluation according to the requirement of the evidence-based medicine is difficult because there are plenty of probiotic strains with a large spectrum of efficacy used in different branches of medicine and many combinations of bacteria can be used. But sometimes even medical doctors cannot be sure about the composition of these products because the information on the leaflets is not precise and adequate. The problem also is that for example in the USA products containing probiotic strains are considered as food additives and not as real drugs (medicaments).

There was published a study where the authors evaluated information about forty four commercial human and veterinary products (probiotics) and total information about the strain and quantity of bacteria was declared only in two human and nine veterinary products and none contained data about the amount of present microorganisms on the expiration date [11]. A more serious omission, however, was the content and quantity of bacteria. Another study tested food additives, the presence of live and active cultures of bacteria. However, the data differed a lot. One tested product, yogurt, did not contain any marked bacteria [12]. In some cases differences in the contents of bacteria in products from the same producer were found. Majaama et al. in their study administered a product which should contain *Lactobacillus acidophilus* in the quantity of  $1 \times 10^9$  but they found out *Lactobacillus casei* in the quantity of  $4,4 \times 10^7$  [13]. Moreover advertisements of some producers are misleading. For example, a company marketing a new probiotic product containing several bacterial strains showed data confirming the efficacy of the product. None however concerned studies including their product [14].

In the European Union the situation is rather better than in America because official projects occupying with probiotics exist [15] and also in veterinary medicine (probiotics are often used as food additives for animals) it is necessary to be declared a new strain as an official strain which can later be signed as a probiotic strain [16]. However, there are always found some mistakes in data - quantity and strain of bacteria. In a recent study information about a quantity of strains in seven of ten products was missing. By using PCR there was a disagreement in a fifty percent of products and a different strain of *Lactobacillus* was found in three cases [17]. Similar examination was not done in our country before, this fact and general scepticism against probiotics inspired us to evaluate products containing probiotic strains available on the Czech market. We established a strain and in case of *enterococcus* also the species.

All products we examined contained the indicated value on the leaflet (strain and quantity of bacteria). On the products containing more than one strain of bacteria was the quantity of bacteria noted mostly summarily for all bacteria. Only in two products was the quantity noted separately. From a practical point of view it is acceptable but it makes it more difficult to evaluate the effects of particular strains of bacteria for human health - respecting evidence based medicine. The missing information about the way of using probiotics together with the meal can sometimes reduce the effect of probiotics as well.

Finally - we can say that the labelling and signs of all tested products was sufficient for clinical practice. All tested products contained a declared strain of bacteria and a minimal quantity of bacteria in the capsule. This seems to be very stimulating and surprising on the one hand. In the USA and the Western Europe the situation is quite different and the declared composition of the probiotic products is often not adequate.

It means that in the Czech republic if we decide to use probiotics we can be sure about their adequate composition. We might use their physiological and nontoxic profits in curing diseases. Nevertheless many extensive randomised studies have to be carried out in order to assess optimal bacterial strain, dose, combination and curative conditions.

### **Acknowledgements**

The study was supported by the Internal Grant Agency of the Ministry of Health of the Czech Republic (NK 7366-3)

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*Doručeno do redakce: 22. 6. 2005*  
*Přijato po recenzi: 11. 7. 2005*