

Elaboration of synbiotic preparations for swine health prophylaxis

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Abstract

In the European Union Member States, the use of antibiotic growth promoters for livestock breeding has been prohibited since the 1st of January 2006. As a result of this regulation, a reduction in animals' growth efficiency could be observed, as well as an increase in the abundance of pathogenic microorganisms in the environment. Although, the development of antibiotic resistance among pathogens also pose a great threat for, both, animals and humans. Therefore, the vital aspect of livestock breeding, such as swine, is to elaborate and implement alternative feed additives that could stimulate the growth of animals and have a positive impact on their health.

It is acknowledged that the most beneficial effect on swine is attributed to probiotics and prebiotics. Preparations, which comprise both of these components, namely, synbiotics, have a considerably stronger positive impact on the host's organisms, than using probiotics or prebiotics individually. Hence, the research aimed to establish synbiotic preparations for swine health prophylaxis that would be more effective than commercially available probiotics. The first stage of the study was in vitro analyses, which purposes were to choose lactobacilli and *Saccharomyces cerevisiae* strains with probiotic characteristics and preparation of an alternative cost-efficient fermentation medium for their growth, as well as select a prebiotic component.

Preliminary selection of probiotic lactobacilli or *S. cerevisiae* strains was conducted based on their antagonism against pathogens and antibiotic resistance or amylolytic activity, respectively. Subsequently, the survivability of selected strains in low pH or the presence of bile salts was analysed, followed by an evaluation of mycotoxins' detoxifying properties, which resulted in the selection of the strains intended for the development of multi-strain probiotic preparations. Next, the safety of chosen isolates was tested, including the study of mucolytic and haemolytic activities, as well as the cytotoxicity of strains. Additionally, the hydrophobicity, auto-aggregation and co-aggregation abilities were studied, as well as the adhesion capability of strains to the intestinal epithelium and other biotic or abiotic surfaces. The potential of strains to inhibit the attachment of pathogenic bacteria to Caco-2 cells was also analyzed. As the consequence of mentioned analyses, 5 lactobacilli strains, namely, *Lacticaseibacillus (Lc.) paracasei* ŁOCK 1091, *Lactiplantibacillus (L.) pentosus* ŁOCK 1094, *L. plantarum* ŁOCK 0860, *Limosilactobacillus reuteri* ŁOCK 1092 and *Lc. rhamnosus* ŁOCK 1094, as well as *S. cerevisiae* ŁOCK 0119 with probiotic potential were chosen.

Afterwards, the cost-efficient alternative fermentation medium, composed of natural ingredients commonly used in swine feeding, was elaborated. To accomplish that, growth kinetics of selected probiotic strains in a mixture of different flours and water in adequate ratio and optimal temperature and pH were tested. Fermentation medium consisting of flours, such as wheat (40%), barley (30%), maize (20%), as well as rye (10%) and water in the ratio of 1:1.5, met the nutritional requirements of selected strains and stimulated their growth.

The final step in the in vitro stage was to select the prebiotic substance for chosen probiotics. The impact of 5 compounds, namely, inulin, maltodextrin, apples' pectin, β -glucan and corn starch, as well as glucose comparatively on the growth, metabolism and enzymatic

profile of lactobacilli were studied. Moreover, the antagonistic properties of probiotics towards pathogenic bacteria in relation to the presence of different prebiotics were tested with the use of the co-culture method. Only inulin had a positive influence on the production of lactic acid and short-chain fatty acids, simultaneously enhancing the antimicrobial potential of probiotics without having a negative effect on their enzymatic profile.

Conducted in vitro studies lead to the elaboration of 5 synbiotic preparations comprised of inulin, *S. cerevisiae* ŁOCK 0119 yeast strain and 3 – 5 lactobacilli strains. In the second phase of the research, the influence of these synbiotics on pigs' faecal microbiota and its metabolism was analysed in vivo. Furthermore, the potential of synbiotic preparation to reduce the genotoxicity of swine faecal water was studied. It was shown, that newly elaborated synbiotics had a positive influence on the dominant faecal microbiota of pigs in each breeding stage, as well as induced beneficial changes in concentrations of organic acids in swine faeces were observed. No impact of synbiotic preparations on pigs' faecal water genotoxicity was established. The effect of synbiotics on pigs was more substantial than observed when commercially available probiotics, namely, BioPlus 2B® and Cylactin® LBC, were used.

Based on the conducted research it was concluded that elaborated synbiotic preparations could have a positive influence on the well-being and growth of swine through modulation of intestinal microbiota. Therefore, they could be used as an alternative to antibiotic growth promoters.