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## International Journal of Probiotics & Prebiotics 6(1): 1-12

- 1-12 **IN VITRO CHARACTERIZATION AND SAFETY OF THE PROBIOTIC STRAIN *LACTOBACILLUS REUTERI CARDIOVIVA* NCIMB 30242**  
WB Branton, ML Jones, C Tomaro-Duchesneau, CJ Martoni and S Prakash

**ABSTRACT:** *In-vitro* characterization and safety evaluations were performed for *Lactobacillus reuteri cardiovascularis* NCIMB 30242 using molecular and metabolic analysis techniques to investigate the safety of the strain for human consumption. First, the identity of the strain was confirmed using a combination of metabolic profiling and 16s rDNA sequencing techniques. The genome of the strain was sequenced by a shotgun sequencing strategy and annotated using RAST software. Results show that the strain does not have antimicrobial resistance genes or other virulence factors uncommon to other probiotic *Lactobacilli* or associated with mobile genetic elements. This genetic analysis was further confirmed by phenotypic testing. The minimum inhibitory concentration (MIC) of a panel of antibiotics showed that this strain is susceptible to the 8 antibiotics tested. The ability of the strain to produce potentially harmful by-products and antimicrobial compounds was also tested. Results show that the strain does not produce biogenic amines above the detection limits for plate screening and HPLC based methods. In addition, the strain was shown to produce lactic acid and hydrogen peroxide and did not show bacteriocin activity or reuterin production. These results confirm that *Lactobacillus reuteri cardiovascularis* NCIMB 30242 is a safe probiotic strain; however, *in vivo* validation is required.

## International Journal of Probiotics & Prebiotics 6(1): 13-20

- 13-20 **SELECTION OF PROBIOTIC *LACTOBACILLUS* SPECIES TO ERADICATE RESISTANT UROGENITAL PATHOGENS IN PREGNANT WOMEN**  
Pratima Pradhan, Rama Charan Mohanty and Aruna Mishra

**ABSTRACT:** *The aim of this research was to evaluate some probiotic traits of Lactobacillus strains isolated from different sources (dairy foods, vagina, stool and common probiotic drugs) and also to investigate the antibacterial properties of these strains against the urogenital pathogens isolated from urine and vaginal swabs of pregnant women. For this purpose 13 Lactobacillus strains were screened for their probiotic traits and tested against the multi-drug resistant urogenital pathogens. In the present study 2 strains of E. coli, 2 strains of Staphylococcus aureus (MRSA) and one strain of Streptococcus agalactiae were found to be resistant to all the antibiotics tested. 5 Lactobacillus strains (45.4%), L. fermentum, L. plantarum, L. acidophilus, L. rhamnosus and L. gasseri were found to have good probiotic traits and showed broad antibacterial spectrum against E. coli and S. aureus. All the selected Lactobacilli were observed to produce lactic acid, H<sub>2</sub>O<sub>2</sub> and bacteriocin. Streptococcus agalactiae was inhibited by only L. plantarum (21±6.16 mm). Six Lactobacillus strains were effective against E. coli and eight Lactobacillus strains were found to be effective against S. aureus respectively.*

## International Journal of Probiotics & Prebiotics 6(1): 21-38

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### A REVIEW OF THE EVIDENCE AVAILABLE FOR THE USE AND EFFECTIVENESS OF PROBIOTIC DRINKS AND SUPPLEMENTS FOR THE TREATMENT OF IRRITABLE BOWEL SYNDROME

Brinda Botschinsky, David Botschinsky and Amalia Tsiami

**ABSTRACT:** *Irritable Bowel Syndrome (IBS) is a substantial burden on healthcare systems. There are a plethora of probiotic products on the market that target gastrointestinal problems. This review aims to guide the healthcare practitioner to make an informed judgment when prescribing probiotic products for alleviating the symptoms of IBS, as conventional medication has been found to have a few adverse effects. Six recent systematic reviews and 27 clinical trials were analysed. The microbial content of twelve commercial products was examined. All the extracted evidence was summarised and critically reviewed. The quality of the research was found to be limited and often contradictory. A need for studies of longer duration and of larger sample size was identified. Dosages in clinical trials varied greatly as did the use of multi or single strain products. Two parameters were selected (global improvement and abdominal pain) and probiotic species were scored according to their performance in clinical studies. *Lactobacillus rhamnosus* scored the highest for improvement of global symptoms and *Lactobacillus acidophilus* scored the highest for improvement of abdominal pain. Probiotics have few adverse effects, and although they may not supply the cure for all the symptoms of IBS, they could provide a way to self-manage the condition. The increase in research dedicated to understanding and proving the efficacy of probiotics should mean that before long current inconsistencies in research methods are removed.*

## International Journal of Probiotics & Prebiotics 6(1): 39-42

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### ASSESSMENT OF $\beta$ -GALACTOSIDASE PRODUCTION BY *LACTOBACILLUS* SPECIES ISOLATED FROM OGI PRODUCED FROM COMPOSITE GRAINS

VO Oyetayo, TA Olofin, OV Olaiya and OA Oseni

**ABSTRACT:** *The potential of lactobacilli isolated from Ogi, obtained from fermented composite grain, *Zea mays* and *Sorghum vulgare*, to produce  $\beta$ -galactosidase was investigated. Three lactobacilli viz: *Lactobacillus fermentum*, *Lactobacillus plantarum* and *Lactobacillus brevis* isolated from ogi were assessed for their ability to produce  $\beta$ -galactosidase.  $\beta$ -galactosidase activity was measured by the release of  $\alpha$ -nitrophenol (ONP) from  $\alpha$ -nitrophenol- $\beta$ -galactopyranoside (ONPG). The production of  $\beta$ -galactosidase by the lactobacilli was highest at the pH range 6.5 to 7.6. Among the three lactobacilli isolates obtained from ogi, *Lactobacillus fermentum* exhibited better  $\beta$ -galactosidase production (249.78  $\mu$ mole/min/ml) at pH 7.5. *Lactobacillus plantarum* and *Lactobacillus brevis* had 153  $\mu$ mole/min/ml and 142  $\mu$ mole/min/ml respectively at the highest level of production. The present study reveals that lactobacilli isolated from Ogi is capable of producing appreciable quantity of  $\beta$ -galactosidase which can be used in addressing incidences of lactose intolerance in individuals.*

## International Journal of Probiotics & Prebiotics 6(1): 43-48

- 43-48      **SAFETY STUDIES OF *BACILLUS COAGULANS* Unique IS-2 IN RATS: MORPHOLOGICAL, BIOCHEMICAL AND CLINICAL EVALUATIONS**  
Ratna M. Sudha, M. Sunita and Babu M. Sekhar

**ABSTRACT:** *Probiotics are vital bacteria that colonize the intestine and modify its microbial flora with benefits for the host. Food products containing lactic acid bacteria have been available in the market including health supplements. However, there is not much information available regarding the toxicity of probiotic bacteria present in such preparations. In the present study acute and sub-acute oral toxicity of a new probiotic strain, Bacillus coagulans Unique IS-2 (MTCC- 5260) was conducted in Sprague Dawley rats. The rats were orally fed with a single dose of 3250 and 6500 mg/kg b.w/day ( $5 \times 10^9$  spores/g in water) dose of test organism for acute and sub-acute toxicity studies respectively. While the experimental rats received doses of 130, 650, 1300 mg/kg b.w/day ( $5 \times 10^9$  spores/g) for 14 consecutive days, other control animals received only water. The follow up study was carried out till 28 days. The results of this toxicity assessment indicate that there were no treatment related changes such as clinical signs, bodyweight, food intake, parameters pertaining to urine, hematological examinations, clinical chemistry, gross pathology and histopathology exhibited by experimental rats at both time intervals. Strain B. coagulans Unique IS-2 did not exhibit any clinical symptoms up to 1300 mg/kg b.w dose ( $5 \times 10^9$  CFU/gram), when administered for 14 days which was considered as 'No Observed Adverse Effect Level' (NOAEL). These results suggest that the strain B. coagulans Unique IS-2 may be considered as a non-pathogenic and safe for human consumption.*

## International Journal of Probiotics & Prebiotics 6(1): 49-56

- 49-56      **A PREBIOTIC OLIGOFRUCTOSE-ENRICHED INULIN PREPARATION STIMULATES INTESTINAL GROWTH AND FUNCTION IN SUCKLING RATS**  
Frida Fåk, Elin Johansson, Siv Ahrné, Göran Molin and Björn Weström

**ABSTRACT:** *Postnatal development of the gastrointestinal (GI) tract in mammals is influenced, to an unknown extent, by the diet and the microflora. The aim of the current study was to clarify the role of a dietary fibre on postnatal GI development, by studying the effects of a prebiotic oligofructose-enriched inulin preparation (Synergy1) on GI growth and function in a suckling rat model. Between 10 and 16 days of age, pups were daily gavaged with Synergy1 (2.0 mg/g b. wt), or Synergy1 together with the probiotic bacterium Lactobacillus plantarum 299v (Lp299v,  $5.0 \times 10^6$  CFU/g b. wt), while control pups only received tap water. At 17 days of age, the prebiotic-treated pups showed an increased growth of the small intestine, along with increased mucosal lactase activity as compared to controls. Furthermore, prebiotic-treated pups had an increased proportion of mature (adult-type non-vacuolated) enterocytes in the distal small intestine as compared to the control pups. In the synbiotic group, the addition of Lp299v did not appear to enhance the effect of Synergy1. The prebiotic oligofructose-enriched inulin preparation had a stimulatory effect on intestinal growth and maturation in young developing rats, probably through changes in SFCA concentrations or bacterial composition.*

## International Journal of Probiotics & Prebiotics 6(1): 57-64

### 57-64 USE OF STATIC BATCH CULTURE SYSTEMS TO INVESTIGATE THE FERMENTATION EFFECTS OF SELECTED OLIGOSACCHARIDES AND FIBRES BY THE CANINE FAECAL MICROBIOTA

VL Inness, C Khoo, KL Gross, L Hoyles, GR Gibson and AL McCartney

**ABSTRACT:** *To date, the majority of studies assessing the prebiotic potential of substrates for canine use have been performed using dietary intervention. However, such studies are expensive and generally rely on the use of prebiotics that have been previously proven to have a use in humans. Consequently, cheap and rapid in vitro methods are required that allow initial screening of acknowledged and novel prebiotics for use in canine dietary intervention studies. Anaerobic static batch culture fermentation systems may provide a valuable preliminary screening tool for prebiotic selection for canines, thus reducing the number of clinical trials required for product testing. Here, the effects of eight different oligosaccharide and fibrous substrates on canine faecal microbial populations and short-chain fatty acid (SCFA) production were investigated in vitro using static batch culture systems. Changes in the canine microbiota, as determined by fluorescence in situ hybridization analysis, and SCFA ratios were observed during the fermentation of all the tested substrates. Rice bran was the only substrate to have a pronounced bifidogenic effect, accompanied by increases in lactic acid bacteria. All substrates examined produced significant quantities of acetate and butyrate as products of bacterial fermentation, with mannanoligosaccharide (MOS) fermentation generating the highest amounts of SCFAs. The results show that the static batch culture system was useful for examining the fermentation of substrates by the canine faecal microbiota and could be used to screen candidate prebiotics for further use in canine foods. Of the substrates screened, rice bran and MOS exhibited the greatest prebiotic potential.*

## International Journal of Probiotics & Prebiotics 6(1): 65-72

### 65-72 IMPACT OF GANEDENBC30 (*BACILLUS COAGULANS* GBI-30, 6086) ON POPULATION DYNAMICS OF THE HUMAN GUT MICROBIOTA IN A CONTINUOUS CULTURE FERMENTATION SYSTEM

Harue Honda, Lesley Hoyles, Glenn R. Gibson, Sean Farmer, David Keller and Anne L. McCartney

**ABSTRACT:** *Interest in Bacillus probiotics, for use in humans and other animals, is increasing. GannedenBC<sup>30</sup> (Bacillus coagulans GBI-30, 6086) is a commercially available probiotic that is considered safe for human use. Few data are available in relation to the impact of Bacillus coagulans supplementation on the gut microbiota of animals, with none available for humans. In vitro single-stage continuous culture fermentation experiments were performed to examine the effect of GannedenBC<sup>30</sup> supplementation on the human gut microbiota and to determine persistence of the probiotic strain upon cessation of supplementation. Fluorescence in situ hybridization and denaturing gradient gel electrophoresis were employed to monitor bacterial populations within the fermentation systems, while cultivation work was performed to examine the aerobic spore-formers in samples. GannedenBC<sup>30</sup> supplementation had no major effect on the microbiota in vitro, although post-treatment clustering was seen for DGGE profiles. Both cultivation and fluorescence in situ hybridization analysis (with probe Bcoa191) demonstrated persistence of*

*GanedenBC<sup>30</sup> in vitro post-treatment and indicated germination of spores in vitro. GanedenBC<sup>30</sup> supplementation did not cause dybiosis of the human gut microbiota in vitro, indicating GanedenBC<sup>30</sup> is microbiologically safe for probiotic application in humans. Future work should examine the potential probiotic function(s) of GanedenBC<sup>30</sup>, including its immunomodulatory and antimicrobial effects.*