### **Results:**

Healthy and dysbiotic intestinal models have been established. One of the culture media used allowed maintaining initial healthy bacterial distribution up to 4-hours, while the other culture media favours Enterobacteriales growth, which is an indicator of dysbiosis in the small intestine. Amino-acids absorption is significantly different in the Caco-2 model that contains the 6 different representative strains from small intestine, compared to Caco-2 models without those microbial strains.

# **Discussion:**

Absorption results demonstrate the influence of the selected microorganisms in the in vitro Caco-2 models. The metabolism diversity of the 6 strains used in the in vitro Caco-2 models is capable of transforming and favour the absorption of a large part of the nutrients studied. Thus, the implementation of characteristic microorganisms from the small intestine to in vitro Caco-2 absorption models provides added value compared to the currently available models. It offers a new tool to study the effect of new bio-surfactant compounds included in MARISURF project.

**Keywords:** Caco-2 model, Small intestine microbiota, host-microbe relationship, Amino acid metabolism, Absorption

# DEVELOPMENT OF A NON-DAIRY PROBIOTIC FERMENTED RICE FOOD WITH BIFIDOBACTERIUM ANIMALIS AND STREPTOCOCCUS THERMOPHILUS

Gomes A.; Rodrigues D.; Baltazar B.; Sousa S.; Vasconcelos M.; Pimenta A.; Freitas A. Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina – Laboratório, Associado, Escola Superior de Biotecnologia, Porto, Portugal

#### Introduction:

Fermented foods provide health benefits well beyond their starting food materials, enhancing their nutritional and functional properties, in particular due to the presence of beneficial microorganisms. The search for new non-dairy foods, that may suit vegan lifestyle or lactose intolerants, has been gaining interest, in particular those based on legumes and/or cereals. Rice is a cereal produced and consumed worldwide playing important roles in dietary health and this work aimed to develop a non-dairy fermented rice food with probiotic *Bifidobacterium animalis BB-12* and yoghurt starter *Streptococcus thermophilus*.

# **Methods:**

Japonica rice grains (Oryza sativa L.) were milled for 10 min in a Vorwerk Bimby TM5 and slurries with different rice flour:water ratios (1:10/1:15/1:20, w/w) were sterilized (15 min/120°C). After rapid cooling, slurries were inoculated with 2% (v/v) of nu-trish® *B. animalis BB-12*® and *S. thermophilus (YoFlex® YF-L01 DA)*, both from Christian Hansen, in 1:1 proportion. Fermentation was performed at 40°C for 6h, in duplicate. Inoculated and non-inoculated (control) slurries were analysed at 0, 1.5, 3, 4.5 and 6h for microbial counts, pH variation and organic acids. Enumeration was performed according to Miles & Misra method on MRS agar containing 0.5 g/L L-cysteine•HCl and on *Streptococcus thermophilus* agar incubated at 37°C for 48h under anaerobic and aerobic conditions for *B. animalis BB-12* and *S. thermophilus*, respectively. PCA incubated aerobically (37°C/48h) enabled microbiological control. Organic acids were measured according to Sousa et al.[1] with some modifications. Viscosity was measured after 0 and 6h of incubation by performing frequency sweeps (0.1-10 Hz) at a strain of 0.1 using an oscillatory Gemini Advanced rheometer.

## Results:

Both bacteria grew well in rice slurries and viable cell numbers increased 2 log cycles within 3h of incubation and were maintained practically constant thereafter independently of the slurry: 5.5 to 7.4 log cfu/g for *B. animalis* and 5.4 to 7.4 log cfu/g for *S. thermophilus*. pH values were decreased accordingly, on account of high yield in organic acids (lactic and acetic acids): after 3h initial pH values of 6.4-6.5 decreased to 5.0, 4.7 and 4.8 in 1:10, 1:15 and 1:20 slurries, respectively; values were maintained until 6h. Higher rice flour concentrations originated higher viscosity values; although temperature (40°C) led to higher values (in particular 1:10 slurry), fermentation caused a slightly less viscous matrix, particularly for 1:10 and 1:15 slurries. Fermented rice slurries using probiotic strain and yoghurt starter generated yoghurt-like organoleptic attributes.

# **Discussion:**

Rice-based slurries favored bacterial growth to values above the minimum probiotic threshold and shortened

the time needed to reach pH 4.5. Recently, Giri et al.[2] reported that Lactobacillus plantarum L7 reached 8.98 log cfu/mL after 4d of fermentation in rice-based beverage. Although growth herein was less high, fermentation capacity was much faster: 3h vs 4d. Compared to the controls, fermentation altered the physical characteristics of the matrix, consequence of the starch hydrolysis into smaller sub-units. Results are promising toward development of alternative probiotic rice-based fermented beverage/food.

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Keywords: non-dairy fermented foods, rice-based products, probiotics, viability, fermentation capacity

# EFFECT OF *BIFIDOBACTERIUM SP* AND INULIN ADMINISTRATION ON RATS IN A HEPATOCELLULAR CARCINOMA EXPERIMENTAL MODEL

#### Gutierrez A.

Universidad Autonoma Metropolitana, Mexico

# **Introduction:**

The liver receives approximately 70% of blood directly through the gut by means of the portal vein, which exposes the liver continuously to issues regarding the gut, including bacteria and their metabolites. Currently, a close relationship between gut microbiota and chronic liver diseases is recognized. Therefore, the use of probiotics as an alternative to reduce the risk of certain diseases is being widely studied.

#### Methods

Different concentrations of *Bifidobacterium sp.* and inulin as prebiotic, were administered to male Wistar rats in a hepatocellular carcinoma experimental model. Hepatocarcinogenesis was induced receiving weekly intraperitoneal injections of diethylnitrosamine. Enzymatic activity of GGT was measured in liver tissue as damage control. DNA from the fecal contents of each animal were used to detect levels of Bifidobacterium by quantitative PCR (qPCR). Liver and cecum samples were stained with Hematoxylin & Eosin and Masson's trichrome, for histological analysis.

## **Results:**

The analysis of GGT on liver tissue showed that *Bifidobacterium sp.* administration at different concentrations reduces tissue damage and nodules were of smaller size compared to the positive control. Changes were observed on liver and gut structures on different experimental groups. Bifidobacteria were detected in fecal samples and it was observed that the consumption of prebiotic promotes their development in the intestine.

### **Discussion:**

It is important to study the effect of probiotics on health due to the increase in consumption of these microorganisms. When administered in adequate amounts, probiotics can be used to prevent some liver diseases.

**Keywords:** Bifidobacterium, probiotics, prebiotics, hepatocellular carcinoma, GGT