







# Nutritional enhancement of plant-based products by lactic acid bacteria fermentation

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#### Introduction

Legumes and cereals have an important role in the shift towards more sustainable, plant-based diet. However, the bioavailability of proteins, minerals and vitamins can be impaired by the presence of anti-nutrients in these plants. Moreover, proteins of plant origin have different functional and sensory properties compared to their animal counterparts and may require additional processing steps in order to be successfully applied in new products. In the SIMBA project, the objective is to upgrade the nutritional value and safety of the selected pulses and cereals and develop new product prototypes by fermentation.

### Materials and methods

Raw materials in the study include faba bean, oat, pea and lentil. To select the most suitable or promising microbes and microbial consortia for fermentations, screening of strains belonging to LAB and propionibacteria (PAB) was performed using genomic databases, published data and previous studies. Species with antinutrient degrading activity (phytates, galacto-oligosaccharides, vicines) or ability to produce vitamins B12 and vitamin K were targeted.

In the raw material pretreatment of the selected plant based materials soaking, grinding and heating were applied. Fermentations were optimized for the selected bacterial strains. Analyses of vicines, oligosaccharides and vitamin B12 were performed by liquid chromatography techniques and phytic acid by Phytic Acid Assay Kit (Megazyme). Sensory evaluation test was implemented to verify the effect of fermentation on the organoleptic properties of the selected raw materials.

The selected LAB strains were also characterized for their ability to survive during digestion in presence and absence of the fermented food matrix using a static model for in vitro digestion.





## Results

Bacteria belonging to the genera *Lactiplantibacillus*, *Limosilactobacillus*, *Leuconostoc*, *Levilactobacillus*, *Pediococcus*, *Lactococcus*, and *Propionibacterium* were selected for further fermentation studies. Selected pretreatment and fermentation conditions vary and were based on the characteristics of raw materials and bacterial species. Microbial consortia were designed based on the results of antinutrients degradation and vitamin production in the preliminary studies on single strain cultivations. Four different consortia were

Good growth of the LAB and PAB strains was evidenced in all plant-based materials, and cell densities of  $10^6 - 10^8$  CFU/g were obtained after fermentation (fig. 1).

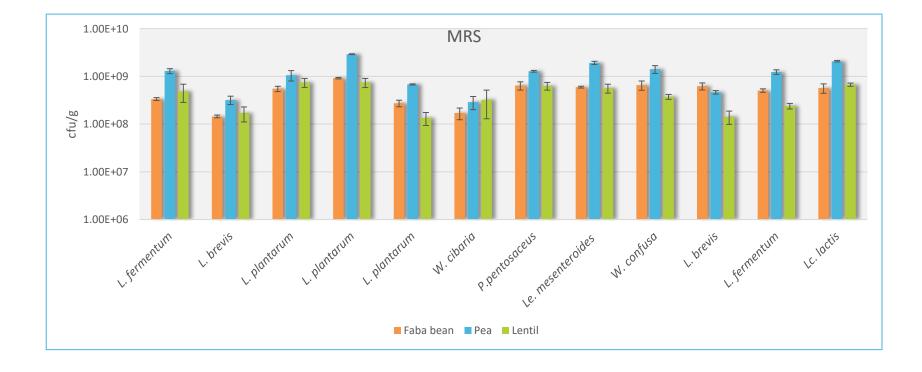


Fig. 1 Single strains of LAB were cultivated in the selected pulses at 30 °C for 2 d and growth was evaluated on MRS agar

The results of antinutrient contents showed significant reduction in contents of vicines in faba bean (fig. 2) and degradation of oligosaccharides in faba bean, lentils and pea (fig. 3), while slight reductions in content of tannins were found in fermented products. Levels of phytates in the selected materials were not affected with the selected LAB consortia. Preliminary results also show the potential of the selected microbes to produce vitamin B12 during the fermentation of pulses.

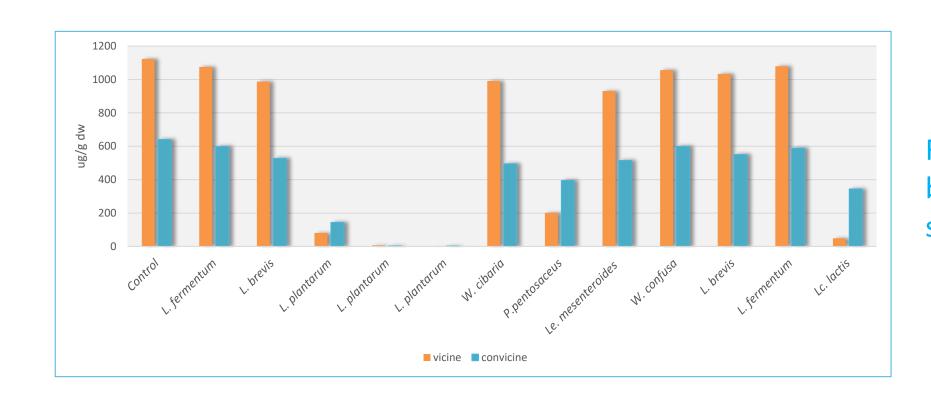


Fig. 2 Degradation of vicines in faba bean fermentations by single LAB strains (2 d fermentation at 30 °C)

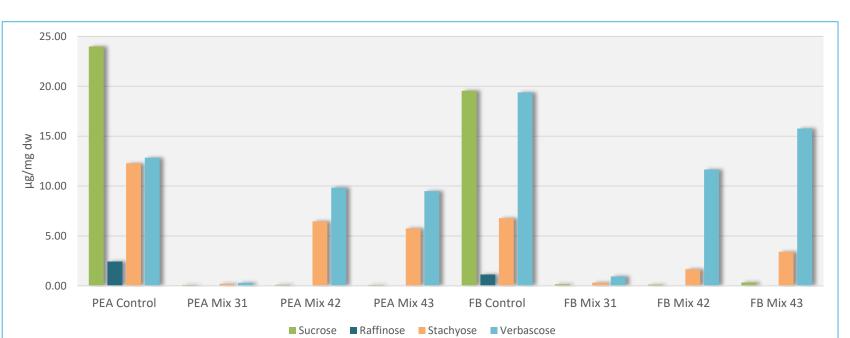


Fig. 3 The amount of sucrose and galacto-oligosaccharides in pea and faba bean (FB) after 2 d fermentation at 30 °C using selected LAB mixtures (31, 42, 43)

Among the strains tested in a static model for in vitro digestion, two strains, namely *Levilactobacillus brevis* and *Limosilactobacillus fermentum*, showed promising survival rate during in vitro digestion, both through the gastric and the intestinal phase (fig. 4).

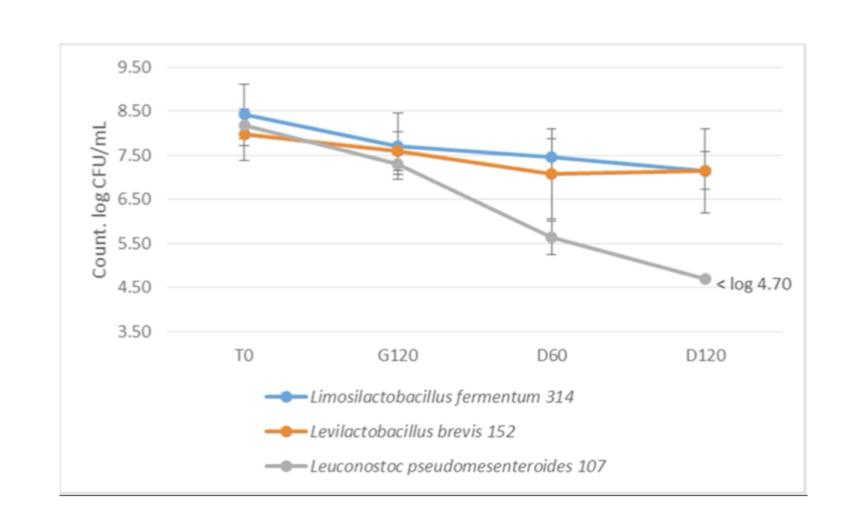


Fig. 4 Survival of LAB strains during in vitro digestion. To starting point; G120: gastric phase after 120 minutes; D60: intestinal phase after 60 minutes from the start of the intestinal phase; D120: intestinal phase is the plating result after 120 minutes from the start of the intestinal phase.

## Conclusions

Strains selected in the study showed promising characteristics for enhancing the nutritional value of the plant based raw materials by reducing the level of antinutrients. The work continues by studying the effect of fermentation on bioactive compounds and protein digestibility.

The microbial consortia developed have potential to be utilized in the fermentation process of proteinaceous food leading to prototypes with improved nutritional value, digestibility, sensory properties, texture and food safety for human consumption.