The gastrointestinal tract harbours a diverse and dynamic microbial community that directly impacts human health. Prebiotics, such as fructo-oligosaccharides (FOS), play a crucial role in the modulation of colonic microbiota, reducing pathophysiological disorders and associated chronic diseases.

The prebiotic potential of FOS produced by a newly isolated strain – *Aspergillus ibericus* was studied. FOS fermentability by the probiotic *Lactobacillus rhamnosus* was evaluated. 

*L. rhamnosus* was grown in de Man-Rogosa-Sharpe (MRS) broth, with different carbon sources: glucose (positive control), no sugar (negative control), microbial-derived FOS from *A. ibericus* and Raftilose®P95, a non-microbial commercial FOS sample (from Beneo-Orafti, Belgium). A final concentration of 2 % (w/v) in sugar was used. Fermentation was carried out in a 96-well microplate and a shake flask, for 24 h, at 37 °C, with an agitation of 120 rpm. Biomass growth was analysed by optical density at 620 nm. The consumption of sugars and the production of short chain fatty acids (SCFA) and lactate was quantified by HPLC.

Maximum cell growth was reached at approximately 12 h, for all carbon sources. The highest growth was achieved for glucose samples, followed by the microbial-derived FOS, then Raftitose and finally the negative control. Although the microbial-derived FOS promoted great cellular growth, only kestose (GF2), together with residual amounts of glucose and sucrose presented in the sample, were consumed. This may explain the two different slopes exhibited during the exponential phase growth. Most likely hypothesis is that probiotic bacteria was cleaving GF2 in the first hours of fermentation, using only the smallest sugars present for growing. And Nystose (GF3) and fructofuranosylnystose (GF4) were not consumed, even when prolonging the fermentation up to 48 h.

SCFA identified were valerate and propionate, as well as succinate, formate, acetate, iso-butyrate and n-butyrate, although in lower amount. Higher amount of SCFA and lactate were determined while growing in the microbial-derived FOS, as compared to the commercial sample. Overall, lactate was the main metabolite produced during the fermentations.

In conclusion, the prebiotic potential of microbial-derived FOS synthesized by *A. ibericus* was demonstrated, providing promising indication of its usability as food ingredient with strong prebiotic features.