## Microbial fructo-oligosaccharides as emergent prebiotics

Nobre, C.

CEB - Centre of Biological Engineering, University of Minho, 4710-057 Braga, Portugal E-mail address: clarissenobre@deb.uminho.pt; Phone: (+351) 253 601 986

Introduction. Fructo-oligosaccharides (FOS) have been associated to prevention and treatment of many XXI century's diseases. There is therefore a huge interest in successfully developing bioprocesses for their production. FOS have been industrially produced via fermentation, by several microorganism enzymes, with low yields and purities in a two-step bioprocess. To obtain high-content FOS we explored an integrated strategy using a co-culture of an Aspergillus ibericus as FOS producer strain, with Saccharomyces cerevisiae, with the gene responsible for sucrose hydrolysis disrupted, as small saccharides removal. The functionality of the microbial FOS produced as prebiotics was further assessed. Methodology. The whole-cell microorganisms were used in a onestep bioprocess. Parameters such as initial yeast concentration, inoculation time, fermentative broth composition, temperature and pH were optimized in shake-flask previously scale-up to a lab bioreactor size. FOS prebiotic potential was evaluated in anaerobic batch cultures for 24 h using human faeces from 5 healthy volunteer individuals. The dynamic bacterial populations changes were assessed by PCR-real time, as well as the production of short chain fatty acids (SCFA) by HPLC. Sugars were analysed by HPLC. Results. Fermentations run under 30 °C, initial pH of 6.0 and 17 g.L<sup>-</sup> <sup>1</sup> yeast extract led to FOS mixtures with 97.4  $\pm$  0.2% (w/w) purity in shake-flask. The scaled-up fermentations in bioreactor yielded  $64 \pm 2\%$  of FOS, after 141 h, with a FOS content of  $118 \pm 5$  g.L<sup>-1</sup> and a purity of  $93.0 \pm 0.5\%$  (w/w). The microbial FOS triggered a beneficial effect on gut microbiota composition. SCFA including succinate, acetate, propionate and valerate were produced by the five faecal inoculum tested, at high concentrations. Lower amount of formate and butyrate were also produced. Conclusions. The one-step fermentation using the specific co-culture studied showed to be more efficient, economical and fast than the conventional two-step bioprocesses, thus avoiding the need of the conventional downstream treatments. The microbial FOS produced exhibited promising potential as nutraceutical ingredients for gut microbiota modulation with likely prebiotic features.

**Keywords:** Fructo-oligosaccharides, prebiotics, one-step fermentation, human microbiota.

**Acknowledgements** This study was supported by the Portuguese Foundation for Science and Technology (FCT) under the scope of the strategic funding of UIDB/04469/2020 unit and BioTecNorte operation (NORTE–01–0145–FEDER-000004) funded by the European Regional Development Fund under the scope of Norte 2020–Programa Operacional Regional do Norte and the Project ColOsH PTDC/BTM–SAL/30071/2017 (POCI–01–0145–FEDER–030071).