# In vitro evaluation of the prebiotic potential of new carbohydrates extracted from agro-industrial byproducts

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Prebiotics are indigestible carbohydrates (CHO) which fermentation in the human gastrointestinal tract (GIT) shifts the intestinal microbiota composition in favor to health. Appraisal of potential in a given CHO requires animal studies using either invasive techniques such as fistulation or slaughtering to sample intestinal chyme. In this study, we show that an in vitro model of the pig GIT totally replacing animal studies can be used to assess the prebiotic potential of 20 new CHO: pectic oligosaccharides (POS), pectins, cellobiose, gluconic acid (GA) and isomaltooligosaccharides (IMO). Inulin was included in the experiment because of its well-documented in vivo prebiotic attributes.

The CHO fermentation characteristics were assessed using sow fecal bacteria cultures (39°C; pH 6.8; 72 h) at a concentration of 0.667 w/v of CHO. Fermentation kinetics and short-chain fatty acids (SCFA) production were compared. The total microbiota and four genera of bacteria (*Lactobacilli, Bifidobacteria, Bacteroides, Clostridium* Cluster 1) were quantified after 12 and 24 h fermentation using qPCR and a prebiotic index (PI) was calculated (Olano-Martin *et al.* 2003, FEMS Microbiology Letters, 243, 101-105).

The IMO ranked amongst the CHO with the highest PI (0.2 *vs*. 0.1 for inulin). They showed bifidogenic and butyrogenic effects close to inulin. The fermentation kinetics was similar to inulin. GA ranked behind IMO (PI=0.12). It proved to be butyrogenic, but sustained less *Bifidobacteria* than inulin and IMO. GA produced more gas and fermented slightly later than inulin. Cellobiose seemed less promising. It was butyrogenic, but it did not favor a healthy microbiota (PI=-0.2). Cellobiose fermented later than inulin but its fermentation rate was higher. The less effective prebiotics were the pectins and POS which showed slow and late starting fermentation. Their butyrogenic activity was the lowest amongst the CHO tested. Their PI scores were close to the cellobiose (PI=-0.05). Finaly they produced less *Bifidobacteria* than inulin.

The in vitro method showed to be an interesting tool to replace animal models in a first screening of prebiotics candidates. The IMO and GA show similar prebiotic activities as inulin. The other CHO were not so promising but their effects on some parameters such as butyrate or *bifidobacteria* may be promising if mixtures of CHO are to be developed.