Bioconversion of lactose from Greek yoghurt acid whey effluents into prebiotic galactooligosaccharides via a novel hyperthermophilic ?-glucosidase from Thermotoga neapolitana

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The steadily increasing global popularity of Greek strained yoghurt has necessitated alternative approaches of valorization of the acid whey (AW) effluents from the straining process, amounting to twice the volume of the final product. In this context, prebiotic oligosaccharides can be enzymatically synthesized from AW lactose, via conventional and novel glycosyl hydrolases (GHs). Increased thermostability of such hydrolases can be a crucial asset in a combined concentration/oligomerization process.
In this research, the gene encoding a ?-glucosidase from the hyperthermophilic bacterium *Thermotoga neapolitana* was cloned and the recombinant enzyme (*Tn*bGal1) was heterologously expressed in *Escherichia coli* and biotechnologically characterized. Enzyme activity and thermostability of *Tn* bGal1 was studied in the temperature and pH ranges of 60-100 °C and 4.0-8.0. Thereafter, *Tn*bGal1 was applied in non-concentrated and concentrated acid whey with lactose concentration of 3.5 to 20 % w/w. The production of galactooligosaccharides (GOS) was monitored over time in relation to lactose concentration and enzyme load, at the optimum reaction conditions. Reaction products were analyzed via High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection and GOS yield in relation to initial lactose content was quantified.
*Tn*bGal1 is a protein of 444 amino acids with a molecular weight of 52 kDa. Optimum catalysis conditions of *Tn*bGal1 were found at 90 °C and pH=5.5, in which enzyme was stable for more than 10 h. Transgalactosylation efficiency of *Tn*bGal1 applied on acid whey was found significant, reaching up to 15.6 % in non-concentrated whey, after 8 h of reaction, using enzyme load of 1 U/mL.
*Tn*bGal1 is a novel, thermostable GH that demonstrated great potential for the oligomerization of AW lactose to produce prebiotic GOS at high temperatures of a combined concentration/oligomerization industrial process. Further research towards optimization of lactose oligomerization will allow efficient, cost effective production of valuable prebiotics in the framework of circular economy.

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