HYDROLYSIS OF AGAVE FRUCTANS BY FUNGAL EXTRACTS PRODUCED BY SOLID FERMENTATION

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Introduction. Fructans are polysaccharides of fructose usually with a terminal glucose. They are classified into linear: inulin (β-2,1 bonds), levan (β-2,6 bonds); branched (β-2,1 and β-2,6 bonds); and fructooligosaccharides (linear or branched with β-2,1 and β-2,6 bonds). Agave tequilana Weber var. azul produces highly branched mixed fructans with a degree of polymerization between 3 and 29 moieties [1]. It is possible to obtain fructose syrup with a high sweetness from the hydrolysis of agave fructans (AF). There are studies about the fructose syrup production from chicory inulin and levans by enzymatic action [2]; but so far there are no studies about enzymes with specificity for AF, so the production of fructanhydrolyases with specificity on these fructans produced by filamentous fungi is interesting.

Methods. Two fungi, F1 and Penicillium comumne (PC), were selected by their ability to grow up on AF as the solely carbon source. The fructanhydrolyase activities were produced in SSF column reactors, using agave bagasse as support and AF as carbon source. Enzymatic broths were obtained after SSF columns extraction with phosphate buffer. The fructanhydrolyase activities were evaluated on AF, chicory inulin (I) and sucrose (S), respectively. The extracts with higher AF/S ratio were selected for further studies. The extracts were mixed with the commercial cocktail Fructozym 960® in an activity relation of 1:1. The products were analyzed by TLC and quantified by HPLC.

Results and discussion. For the PC fungus, the highest activities on the three substrates (30 U/g for S, 5 U/g for I and 2.5 U/g for AF) were produced after 24 h (fig 1). Whereas for the F1, the highest activities were 9 U/g for S at 72 h, 3 U/g for I at 48 h and 1.5 U/g for AF at 24 h. All these activities are low compared to the reported by Penicillium sp. and Aspergillus niger on sucrrose and inulin [3,4]. However, it is important to point out, that our fungi were chosen by their higher AF/S activity ratio and not by the absolute activity. The TLC analysis of the hydrolysis products, showed that both enzymatic extracts produced only fructose (data not show), probably due to the predominant presence of exo-fructanhydrolyase activity. On the other hand, HPLC analysis (fig. 2) showed that the concomitant use of the commercial Fructozyme 960 cocktail with PC and F1, increased by 1.5-fold the AF hydrolysis, probably due to the synergistic effect of endo-inulinases (Fructozym 960®) with exo-fructanhydrolyases (PC and F1).

Fig. 1. Production of fructanhydrolyases by the fungi P. communel (left) y F1(right).

Fig. 2. Percent of hydrolysis of AF to fructose by the extracts and cocktails.

Conclusions. It was possible to produce by column SSF, two fungal enzymatic broths with exo-fructanhydrolyaseactivity. Both cocktails showed an important (1.5-fold) AF hydrolysis rise to fructose, after addition of endo-inulinase activity (Fructozym 960®). This result shows a synergistic effect of endo and exo fructanhydrolyase activity on AF, as previously reported for inulin. This work may be of special interest for the production of fructose syrup from AF.

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References.