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## **Enzymatic hydrolysis efficiency of different industrial hemp biomass fractions: A comparative study of two cultivars**

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Industrial hemp is a promising, renewable feedstock for biorefineries. Low agricultural needs, and considerable biomass yield from a hectare of the plantation every season make industrial hemp an interesting raw material for renewable feedstock utilization studies. Particularly hemp fibers, being a lignocellulosic biomass, may be enzymatically broken down to monosaccharides. These hydrolysates, rich in glucose and xylose, after appropriate supplementation can serve as a culture medium for microorganisms able to produce useful chemical compounds. In this study, an enzymatic hydrolysis of two cultivars of the industrial hemp, Santhica and Futura, was conducted on three separate fractions of these plants: stems, leaves and spent flowers. The flowers were previously extracted with supercritical carbon dioxide to obtain pharmaceutically useful compounds e.g. cannabinoids, terpenes etc. The aim of this study was to compare the enzymatic saccharification efficiency of different waste hemp fractions. Enzymatic hydrolysis was preceded by chemical pretreatment of dry hemp material. First the stems were milled, leaves and flowers were crushed. Then all the fractions were treated with sodium hydroxide solution for 20 minutes at 121°C, 0,1 MPa. After the hot stage, the treated hemp fractions were washed with distilled water and dried in room temperature before hydrolysis. The hydrolysis took 72 h in citrate buffer at 50°C and pH=5 with sodium azide as an antimicrobial agent, sampled twice a day and the monosaccharides concentration was measured using HPLC method. The initial biomass loading was 60 g/dm<sup>3</sup> and the enzyme concentration (Cellic CTec2, SigmaAldrich) was 9% (w/w). The obtained results exhibit a strong advantage of hemp stems over other fractions, due to the higher content of hydrolysable polysaccharides, with final glucose concentration after 72 h of hydrolysis reaching >30 g/dm<sup>3</sup>. Glucans hydrolysis efficiency reached 74,33±1,15% for Santhica and 79,68±2,10% for Futura, and for xylans' efficiency reached 82,39±2,10% and 95,73±6,17% for Santhica and Futura, respectively. Efficiency of the hydrolysis never reached over 20% for both glucans and xylans in the leaves, furthermore the saccharides concentration increased for the first 24 h and then started to decrease to reach zero after 72 hours.