Abstract

The paper presents a review of the literature on the application of plant biomass in biotechnology, as well as the results of experiments aimed at enhancing the enzymatic hydrolysis step of this biomass by using recycling of enzymes adsorbed on the solid fraction of lignocellulosic material, improving the pretreatment process of plant biomass by using vacuum expansion of biomass or microwave radiation, and using lignocellulosic hydrolysates for the synthesis of carotenoids by yeasts of the genus *Rhodotorula*.

The results of the study confirmed that the use of enzyme recycling allows increasing the final efficiency of enzymatic hydrolysis of giant miscanthus and sugar miscanthus biomass, and the efficiency of biomass hydrolysis increased with increasing cellulase activity in the range of 18 - 58 FPU. The results of the study also confirmed that vacuum expansion allows increasing the efficiency of enzymatic hydrolysis of sugar beet biomass compared to mechanical grinding. The most effective process option for processing sugar beet biomass is to use vacuum expansion followed by enzymatic hydrolysis at 50°C, using enzyme compositions: Rapidase® and Accellerase 1500 with complex of supporting enzymes (Accellerase: XY, XC, BG). On the medium prepared in this way, it was possible to reuse the enzymes in the form of immobilized fractions on undecomposed lignocellulose. The highest average ethanol concentration in the post-reaction medium was obtained after sequential fermentation of hydrolysates obtained from mechanically crushed and vacuum-exploded beet biomass by *Saccharomyces cerevisiae* 7, yielding 50.5 and 53.6 $g \cdot dm^{-3}$, respectively. After using the simultaneous process, the attenuation level was lower than that obtained in the SHF system, and the alcohol content was lower by 19% (when shredded biomass was used) and by 15% (after vacuum expansion). The *Mucor indicus* filamentous fungus was found to be unsuitable for ethanol fermentation of sugar beet biomass.

In experiments using microwave-assisted pretreatment of giant miscanthus biomass, the highest hydrolysis efficiency occurred in samples with sulfuric acid (VI) at 90 Psi (58.3%), but fermentation of these hydrolysates yielded only 13.1 mg of ethanol per gram of dry biomass. The opposite relationship occurred in samples treated in an acidic environment at 30 Psi. At the relatively low hydrolysis efficiency of these samples (30.1%), 23.6 mg of ethanol per gram of dry biomass was obtained. Microwave processing parameters should thus be chosen as a trade-off between high enzymatic hydrolysis efficiency and ethanol fermentation productivity.

The results of the experiments demonstrated that adapting yeasts of the genus *Rhodotorula* to the use of lignocellulosic hydrolysates from refined purple willow bark for growth and multiplication will result in more efficient carotenoids biosynthesis. Laboratory adaptation made it possible to obtain *Rhodotorula* strains with increased growth capacity on medium with the addition of lignocellulosic hydrolysate. The obtained modified strains (*R. glutinis* 50H, *R. rubra* KKP350 50H and *R. glutinis* KKP347 50H) showed more efficient synthesis of carotenoids compared to the corresponding test strains. The use of lignocellulosic hydrolysate as a substrate additive increased the amount of carotenoids obtained, even in native strains. After biomass extraction of *R. glutinis* strain KKP347 50H after culture on medium with lignocellulosic hydrolysate, the highest concentration of carotenoids (130 $\mu g \cdot g^{-1}$ d.m.) was obtained.

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