EXTRACTIVE AMMONIA (EA): A NOVEL AMMONIA-BASED PRETREATMENT TECHNOLOGY FOR LIGNOCELLULOSIC BIOMASS

Large scale production of economically viable biofuels can only be achieved from widely available resources, notably lignocellulosic biomass. This feedstock is largely composed by complex carbohydrates, which can be enzymatically hydrolyzed and converted to fuels and chemicals by fermentative organisms. Lignocellulosic biomass is recalcitrant to enzymatic degradation and therefore, a pretreatment step is required for achieving acceptable sugar yields. Recent research has improved our fundamental understanding about the physico-chemical events occurring during ammonia pretreatment of biomass that correlate with enzymatic hydrolysis yields. From this understanding, a novel Extractive Ammonia (EA) pretreatment was developed and is presented herein for the first time. This technology allows the conversion of naturally occurring cellulose I (CI) to cellulose III (CIII) and the selective extraction of lignin from the plant cell wall with liquid ammonia. These extractives are collected in a separate stream as a valuable byproduct. While CIII is known to improve enzymatic hydrolysis rates up to two fold compared to native CI, lignin is acknowledged as a major inhibitor for both enzymes and microbes. Other key events include ester-bonds cleavage via ammonolysis and hydrolysis reactions during EA. Ester bonds play an important role in cross-linking lignin and carbohydrates and therefore, their cleavage disrupts the complex cell wall architecture, allowing carbohydrates to be more easily accessible by enzymes.

The effect of pretreatment variables on CIII conversion was studied using isolated cellulose from commercial sources (Avicel). These studies revealed that samples of higher CIII crystallinity can significantly increase enzymatic activity, which contradicts the current paradigm based on CI...
results, which show that higher CI crystallinity reduces enzymatic activity of cellulases. In the present work, EA performance was evaluated using corn stover (CS) as primary biomass feedstock. An empirical model was created using statistical design of experiments to predict EA pretreatment effectiveness on lignin extraction, ester-bond cleavage and sugar yields as a function of pretreatment conditions. The results show that EA allows extraction of up to ~ 50 % of the lignin present in corn stover, while cleaving about 70 % of the ester bonds. These effects, in synergy with cellulose III formation, allow maximum monomeric glucose and xylose conversions of 93 % and 79 %, respectively, using 15 mg/g glucan of enzyme for 24 h, at 1% glucan loading enzymatic hydrolysis. At 15 % to 20% solid loadings, EA allows up to 2.7 fold reduction of enzyme loading during enzymatic hydrolysis compared to AFEX™-CS. The benefits of EA on fermentation were also explored using the novel Fast Separated Hydrolysis and Fermentation (FSHF) process, which is capable of doubling biofuel productivity while decreasing enzyme loading by 30 % compared with traditional SHF. By coupling EA and FSHF technologies was possible to efficiently generate 191 g ethanol/Kg biomass in 48 h. using 7.5 mg/g glucan of enzyme loading.

The viability of the EA process depends on enzyme savings and on the utilization of lignin streams. From this perspective, EA lignin extracts were fractionated by sequential precipitation generating a fraction composed by ~92 % lignin, which represented about 32 % of the lignin initially present in CS. Techno-economic evaluation of the EA biorefinery was also performed and will be presented along with recommendations for further improvement of this technology.

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