# Nutrients Supply was Reduced to Half in Mixed Feedstock Fermentations

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## Overview

Mixed feedstocks can help reduce the risk associated with feedstock availability for bio-based production of fuels and chemicals. Through this study, we not only want to evaluate cellulosic hydrolysates for fermentation to biofuels but also probe the possibility of reducing nutrient concentration in the broth media. Three feedstocks were treated with alkali and ionic liquid with parameters were generated by SAS JMP®, as given in Table 1. After pretreatment, enzymatic hydrolysis was conducted, glucose and furfural values were measured, and these values were fed back into SAS JMP® to generate a predictive model. The model was validated and the ideal mixed feedstock compositions predicted by the model were then tested for ferment-ability to ethanol. During fermentation, we were primarily focused on the impact of nutrients on ethanol yield. Results indicate that fermentation can be effectively performed with half the nutrients recommended by standard media recipes.

## Methods

- Feedstock: Ratios of Corn stover (CS), Switchgrass (SG), Energy Cane (EC)
- Temperature (1-100%): Alkali: 55-120°C, Ionic liquid: 120-160°C
- Time (1-100%): Alkali:1 to 24 hours, Ionic liquid: 1 to 3 hours
- Enzymatic hydrolysis: 10 mg protein/g glucan in untreated biomass mixture, 50°C, 120 hours, 250 RPM
- Fermentation: *Saccharomyces cerevisiae* at 5% (v/v), 250 RPM, 30°C

Table 1. Pretreatment Design of Experiments

| Pretreatment | CS | SG | EC | Temperature% | Time% |
|---|---|---|---|---|---|
| Alkali | 0.0 | 1.0 | 0.0 | 100 | 1 |
| Alkali | 0.0 | 0.0 | 1.0 | 100 | 1 |
| Alkali | 1.0 | 0.0 | 0.0 | 100 | 1 |
| Alkali | 0.9 | 0.1 | 0.0 | 15.6 | 25 |
| Alkali | 0.2 | 0.4 | 0.4 | 80 | 80 |
| IL | 0 | 0 | 1 | 1 | 100 |
| IL | 0 | 1 | 0 | 100 | 60 |
| IL | 0.33 | 0.33 | 0.33 | 100 | 60 |
| IL | 0.5 | 0 | 0.5 | 100 | 1 |
| IL | 1.0 | 0.0 | 0.0 | 80 | 80 |

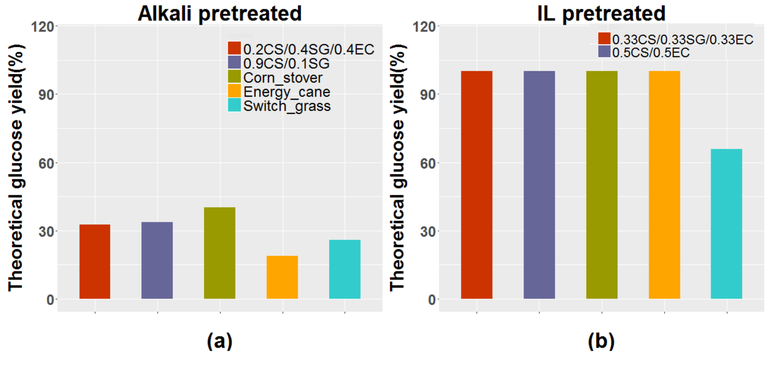
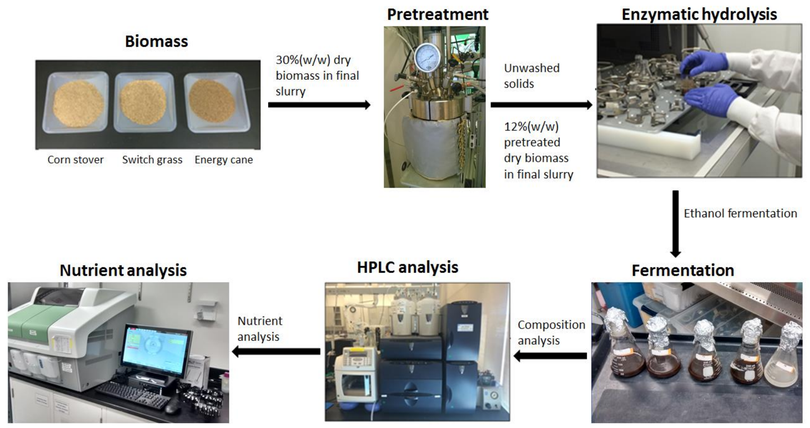



Figure 1. Glucose yield from (a) alkali and (b) IL pretreatment

## Deconstruction and Fermentation Process




## Mixed feedstock with 20% Corn Stover can deliver 100% theoretical ethanol yield

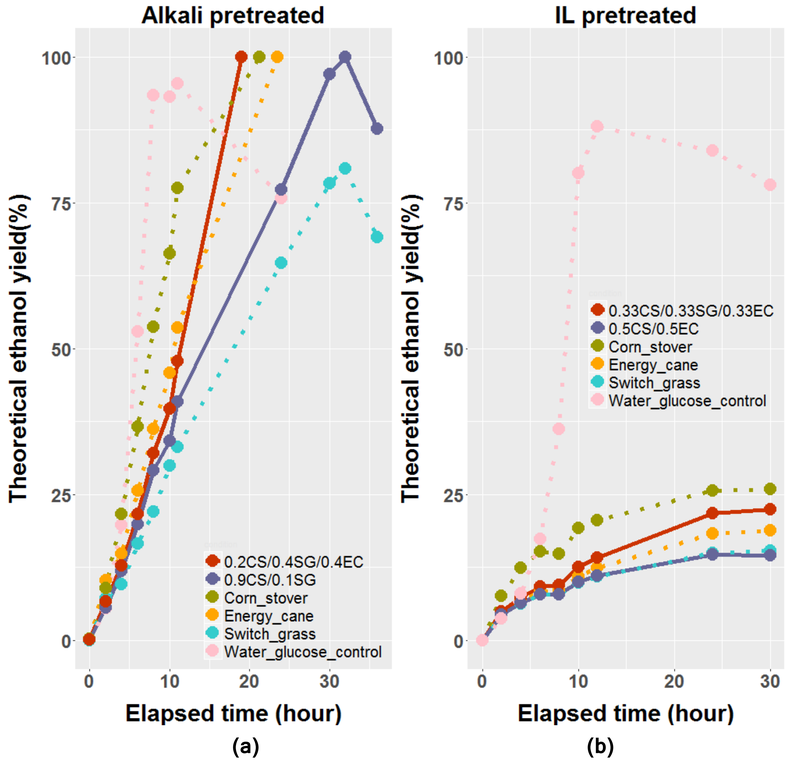



Figure 2. Comparison of ethanol yield from (a) alkali and (b) IL pretreated mixed feedstocks

## Same ethanol yield with half the nutrients in an extended fermentation

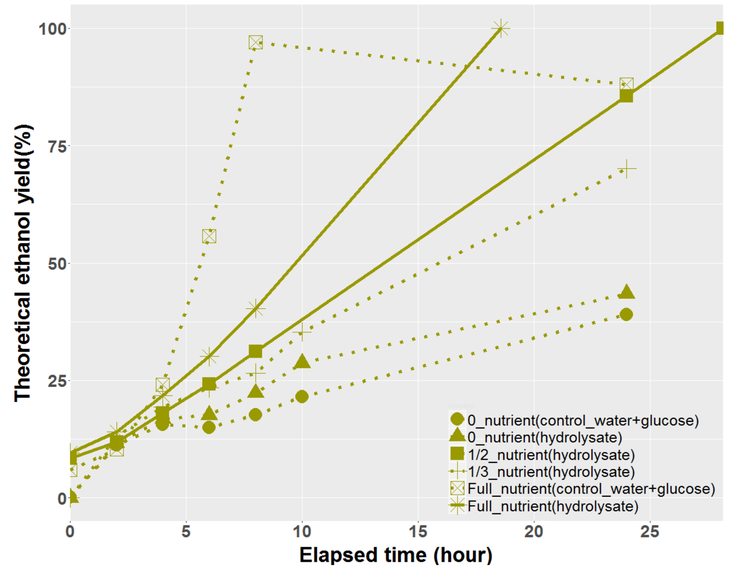



Figure 3. Theoretical ethanol yield with different amounts of nutrients applied

## Summary / Conclusion

- Mixed feedstocks can release 80 -100% of the sugar that is obtained from corn stover alone, and 100% of the released sugars from mixed feedstocks can be converted to ethanol
- Alkali pretreated mixed feedstock has higher ethanol yield but lower glucose yield compared to IL pretreated mixed feedstock due to inhibition of microbial growth by residual EmimAcetate.
- Same ethanol yield can be achieved with lower nutrient supplied but with longer fermentation time
- Future work includes producing hydrocarbons (e.g. bisabolene)

## Acknowledgements

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