Swatchgrass is a promising dedicated bioenergy feedstock with numerous environmental benefits, due to its low fertility requirement, tolerance of poor soils, and high yield. Swatchgrass is composed of 30-35% cellulose, 20-25% hemicellulose, 18% lignin, and 24 % other. If swatchgrass is processed without pretreatment, the maximal conversion achieved at an enzyme loading of 15 FPU/g glucan (5 FPU/g biomass) is less than 5%. When the swatchgrass is pretreated in liquid hot water, the conversion increases by 25-fold, resulting in 80% glucose yield. Arabinoxylan in grasses is extensively esterified with phenolic acid groups which act as cross-linking agents between polysaccharides and lignin in cell wall structure. The complex structure of the hemicellulose in swatchgrass necessitates use of various enzyme activities for efficient hydrolysis of the bonds in the hemicellulose, as well as cellulases for converting cellulose to glucose. We report the enzymatic effect of hemicellulolytic enzymes on enhancing the yield of fermentable sugars derived from enzymolysis of liquid hot water pretreated swatchgrass. We have observed that the addition of hemicellulolytic enzymes increases not only xylose but also glucose yield. Hydrolysis of liquid hot water pretreated Dacotah swatchgrass at 15% w/v dry solids loading in 80% glucose yield and 10% xylose yield at a total protein loading of 11 mg protein/DG where the total protein consists of cellulases combined with supplementary enzymes.

## Results

### Enzyme Activity

![Figure 1. Glucose yields; (B) xylose yields; (C) total combined sugar yields by enzymatic hydrolysis of LHW pretreated swatchgrass.](image)

#### Cellulose Dose vs Sugar Yields

![Figure 2. Effect of enzyme dosages used in this study is shown in Table 1. Experiments were also conducted to examine the effect of supplementary xylanase enzymes on yields of sugars. In addition, an enzyme dose-response curve was generated to determine hydrolysis efficiency of the LHW pretreatment at various loadings of cellulases.](image)

#### Effect of Supplementary Xylanase on Sugar Yields

![Figure 3. Hydrolysis of LHW Pretreated Dacotah Switchgrass at 15% w/v DS Loading](image)

### Conclusions

LHW pretreatment improves enzymatic digestibility of swatchgrass. Optimal LHW pretreatment conditions need to be determined by considering energy requirement and enzyme dose. Optimal enzyme mixture for swatchgrass hydrolysis should contain various activities necessary for hydrolysis of complex structure of heteroxylan. Addition of hemicellulose increases not only xylose but also glucose yield. Hydrolysis of LHW pretreated Dacotah swatchgrass at 15% w/v dry solids loading and a total protein loading of 11 mg protein/DG resulted in 80% glucose yield and 90% xylose yield.

## Material and Methods

### 1. Liquid Hot Water Pretreatment Optimization

**Substrate:** Shawnee switchgrass
**Pretreatment:** 15% dry solids (150 g/L)
160, 190, 200°C
5, 10, 15, 20 min
**Hydrolysis:** whole pretreated slurry
4% dry solids
Spzyme CP (15 FPU/g cellulose)
Novozym 188 (40 CBU/g cellulose)

**Substrate:** Dacotah Switchgrass (40 mesh)
**Pretreatment:** 200°C, 10 min or 20 min; 5 min 150 g dry solids/L
**Hydrolysis**:
- whole pretreatment slurry:
  - 4% dry solids
- Spzyme CP (15 FPU/g cellulose)
- Novozym 188 (40 CBU/g cellulose)
  - pH 4.8, 200 rpm, 50°C, 48 hr.

**Temperature**
- 5°C, 10°C, 15°C, 20°C

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### References