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Effect of particle size on enzymatic hydrolysis of pretreated miscanthus

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Effect of Particle Size on Enzymatic Hydrolysis of Miscanthus

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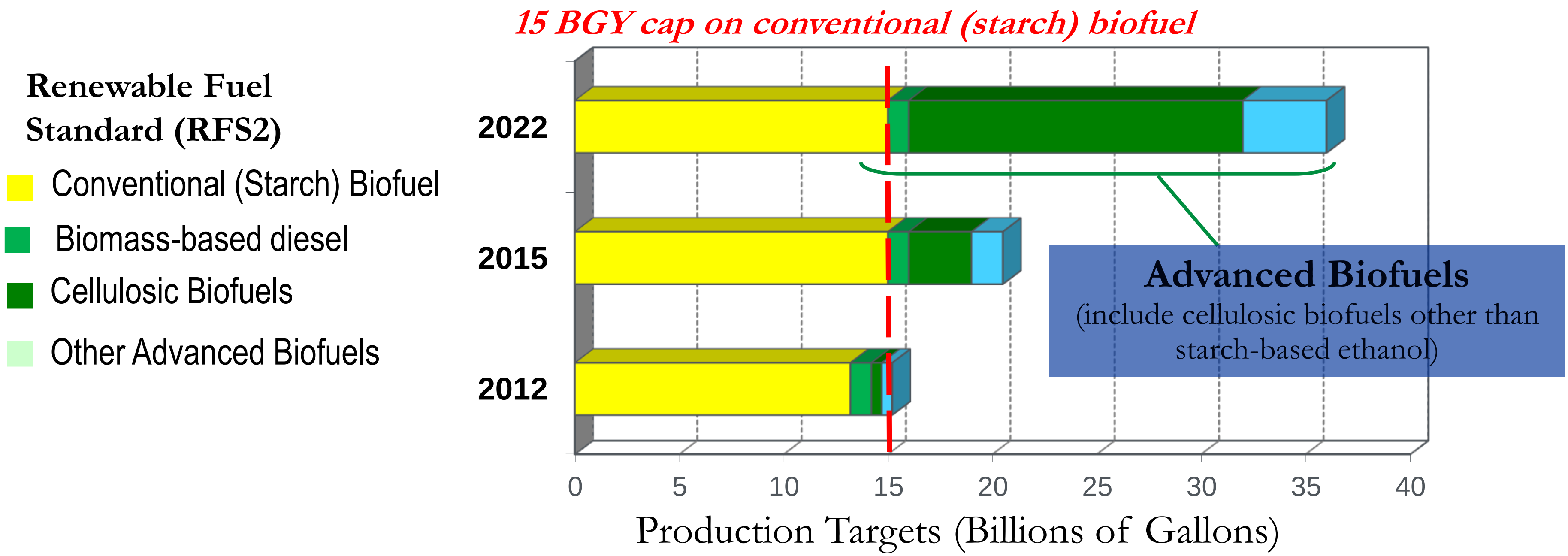
illinois.edu

Cellulosic Feedstocks for Fuel

- Leading contender for ethanol production
  - Relative abundance
  - Low energy inputs
  - Ability to grow on marginal lands
  - Sustainable production
- Uncertainties
  - Establishment: yields with low fertilizer application, marginal lands
  - Lack of technology
  - Biomass availability, seasonality of crop
- 1.3 billion tons of biomass available

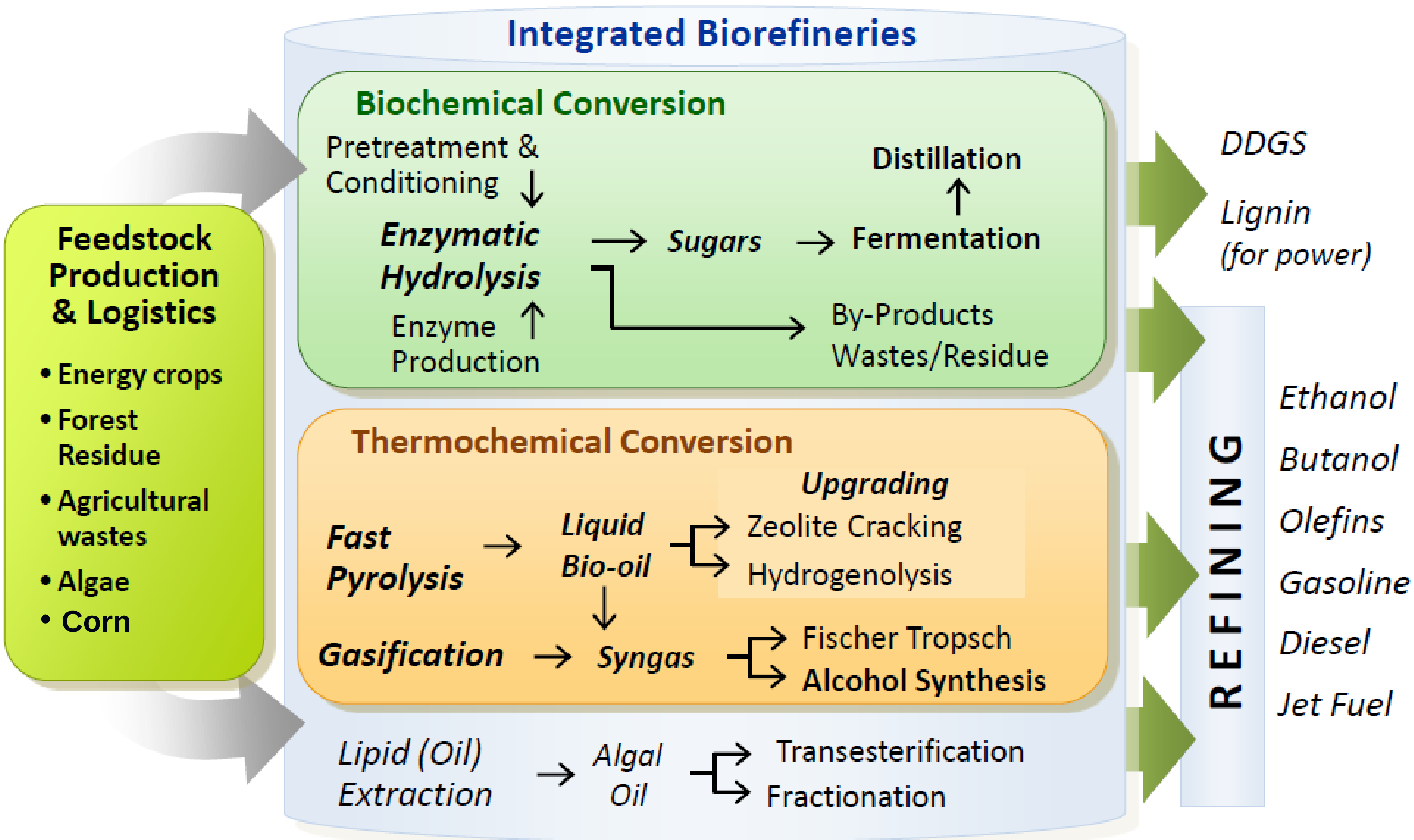
EISA Mandated Biofuel Production Targets



EISA defines ***Cellulosic Biofuel*** as "renewable fuel derived from any cellulose, hemicellulose, or lignin that is derived from renewable biomass and that has lifecycle greenhouse gas emissions…that are *at least 60 percent less* than baseline lifecycle greenhouse gas emissions." The EPA interprets this to include cellulosic-based diesel fuel.

EISA defines ***Advanced Biofuel*** as "renewable fuel, other than ethanol derived from corn starch, that has lifecycle greenhouse gas emissions…that are *at least 50 percent less* than baseline lifecycle greenhouse gas emissions." This includes biomass-based diesel, cellulosic biofuels, and other advanced fuels such as sugarcane-based ethanol.

Different Integrated Biorefineries



Energy Biosciences Institute

MAIN AREAS OF RESEARCH

The EBI's multidisciplinary research teams explore total-system solutions to global energy problems.

FEEDSTOCK DEVELOPMENT

BIOMASS DEPOLYMERIZATION

BIOFUELS PRODUCTION

FOSSIL FUEL BIOPROCESSING

ENVIRONMENTAL, SOCIAL & ECONOMIC IMPACTS

Miscanthus x giganteus

- Perennial grass
- High yielding (20 to 25 dry ton/hectare)
- Less inputs

Lignocellulose Structure

- Cellulose (40 to 50%)
- Hemicellulose (25 to 35%)
- Lignin (15 to 20%)
- Miscanthus
  - 40% cellulose, 18% hemicellulose, 25% lignin

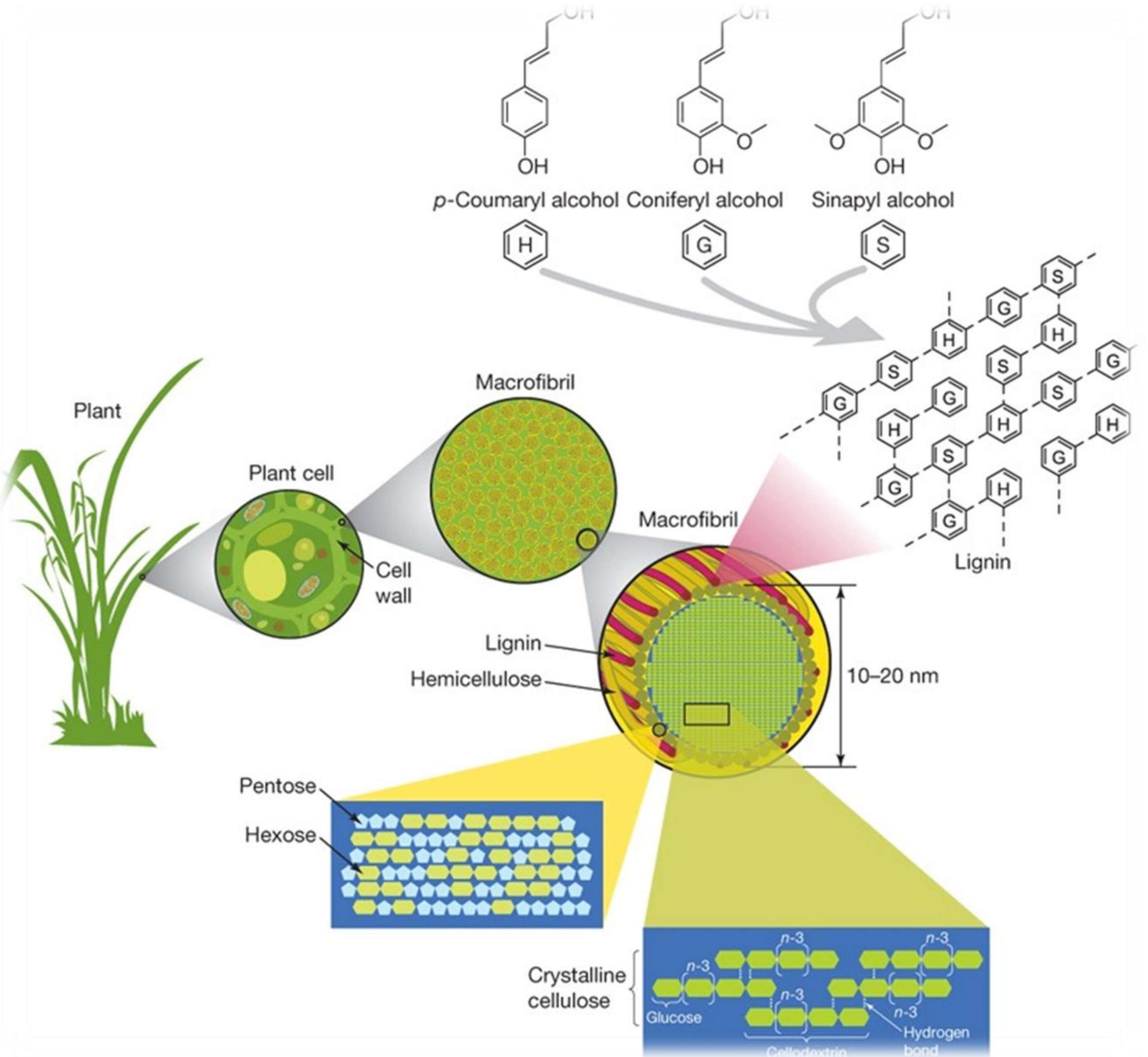
Microfibril

- 36 glucan chains, DP 500 to 14000
- Intra- and inter chain hydrogen bonds

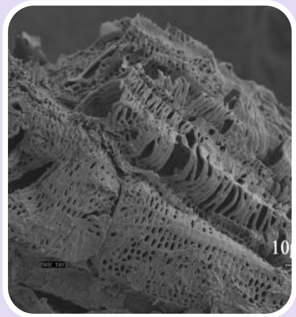
Macrofibiril

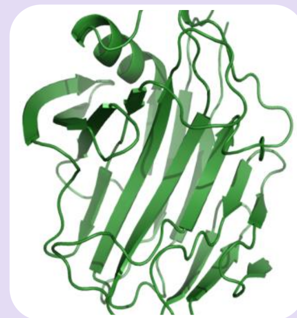
- Ribbon like bundles
- Hemicellulose coats microfibrils, hydrogen bonds with cellulose

Lignin deposited in final stages and enclose microfibrils and polysaccharides



Biochemical Conversion





Feedstock

- Production
- Logistics
- Storage
- Handling

Pretreatment

- Physical
- Chemical
- Biological

Enzyme Hydrolysis

- Cellulase
- Hemi-cellulase
- Other

Fermentation

- Engineered microbes
- C5 and C6 sugars

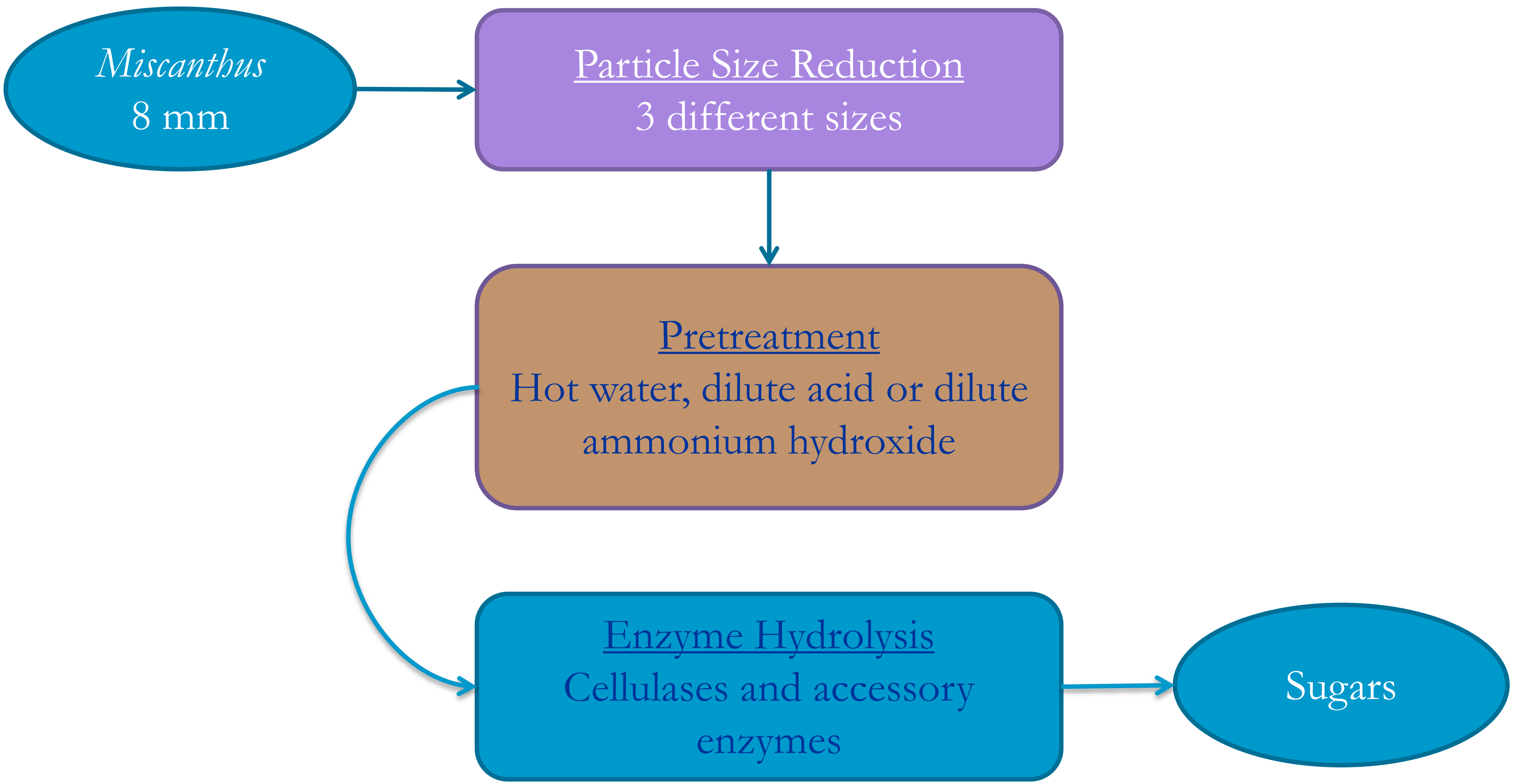
Product Recovery

- Ethanol
- Residues

Objective

Effect of particle size on enzymatic hydrolysis of pretreated *Miscanthus*

Experimental Design



Particle Size Reduction

- Hammer mill
- Sieve sizes: 0.08, 2.0 and 6.0 mm
- Particle size distributions and geometric mean diameters

Pretreatment

- Tubular reactors in a fluidized sand bath
- Reactor fitted with thermocouple for internal temperature measurements
- 10% solids content (d.b.)
- Conditions
  - Hot water: 200°C, 30 min
  - Dilute acid: 160°C, 10 min, 1% w/w sulfuric acid
  - Dilute ammonium hydroxide: 160°C, 5 min, 5%
  - Unpretreated

Enzyme Hydrolysis

- Enzymes
  - Accellerase 1500: exoglucanase, endoglucanase, hemicellulase and beta-glucosidase
  - Accellerase BG: beta-glucosidase
  - Accellerase XY: hemicellulase
  - Accellerase XC: endoglucanase and xylanase
- Conditions
  - 10% solids content
  - 50°C, 75 rpm
  - 72 hr (samples at 3, 12, 24, 48 and 72 hr)
  - HPLC determination of sugars

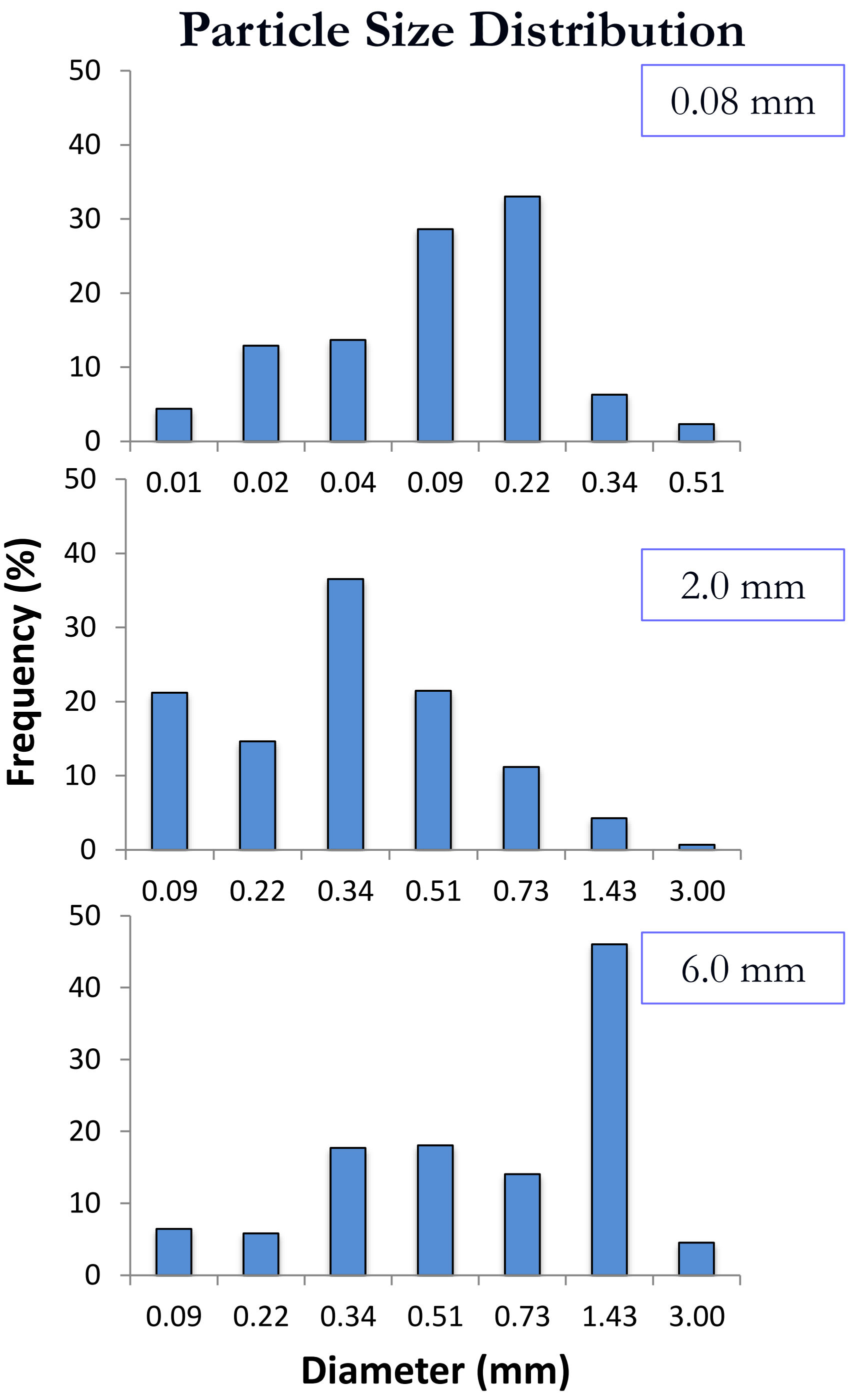
Data Analysis

- Full factorial, completely randomized design
- Particle size analyses, pretreatments and enzyme hydrolysis conducted in triplicates
- Glucan, xylan and total polysaccharide conversion (%)

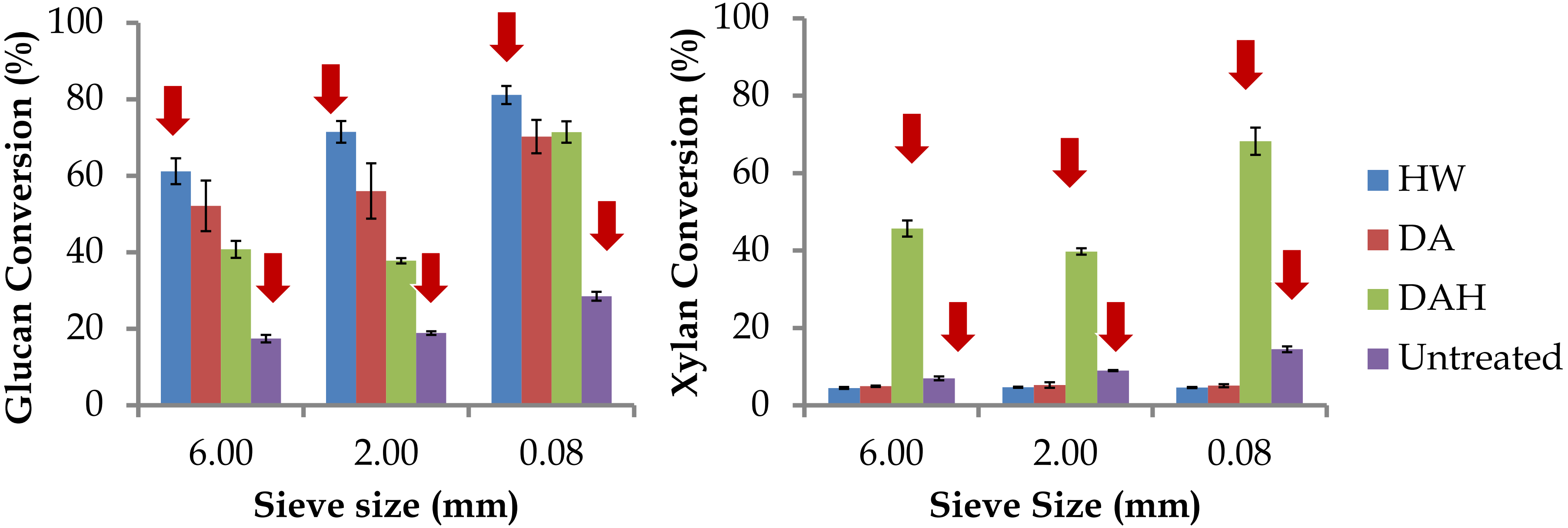
Results

Geometric Mean Diameters

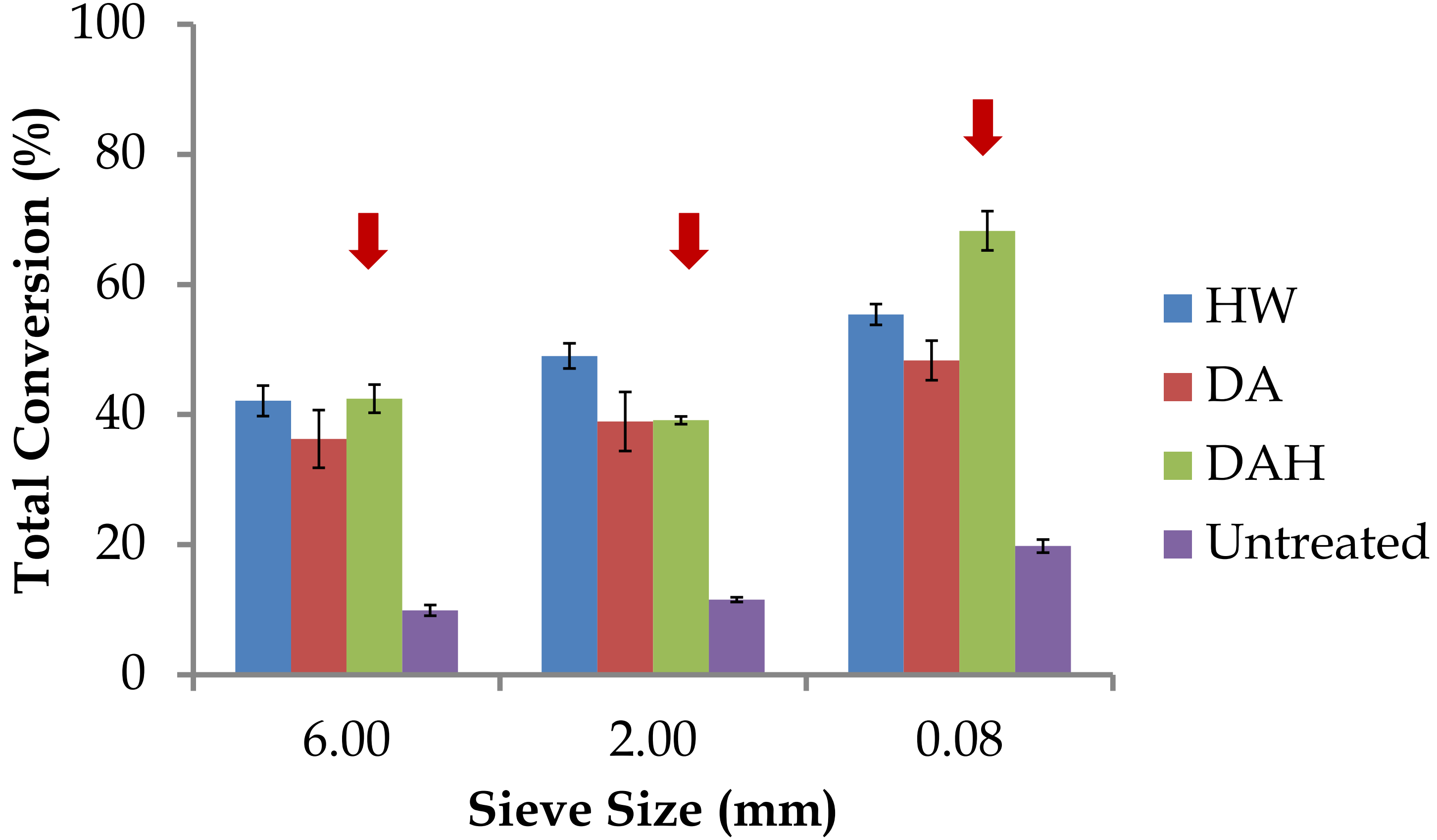
Sieve Size (mm)	Geometric Mean Diameter (µm)
0.08	56.00 ± 0.54 C
2.00	300.5 ± 4.10 B
6.00	695.3 ± 69.1 A



Glucan & Xylan Conversion (%)



Total Conversion (%)



Conclusions

- Decreased particle size increased total polysaccharide conversion for
  - all pretreatments
  - unpretreated *Miscanthus;* 20 to 60% lower than chemical pretreatments
- Sample ground using 0.08 mm sieve screen was used for further studies

Objective

Optimization of hot water pretreatment for *Miscanthus*

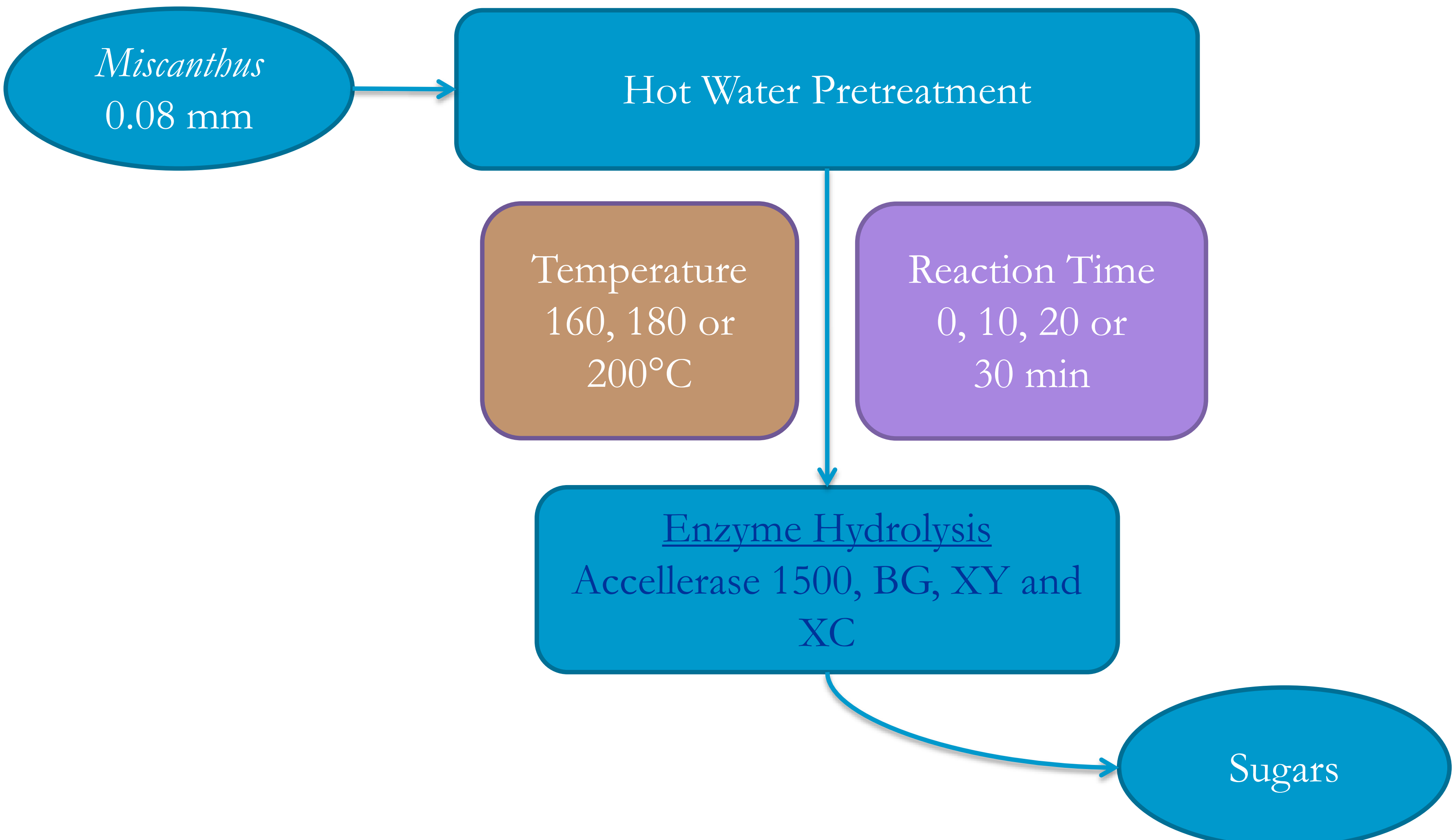
Hot Water Pretreatments

- Water at 160 to 240°C
- At high temperatures,
  - water acts as a weak acid
  - solubilizes hemicellulose as oligosaccharides
  - loosens lignin
- Advantages
  - No chemicals; lessen need for expensive reactors, eliminated need for recycling chemicals or catalysts
  - No neutralization
  - Minimizes monosaccharides from hydrolysis; minimizes inhibitor production

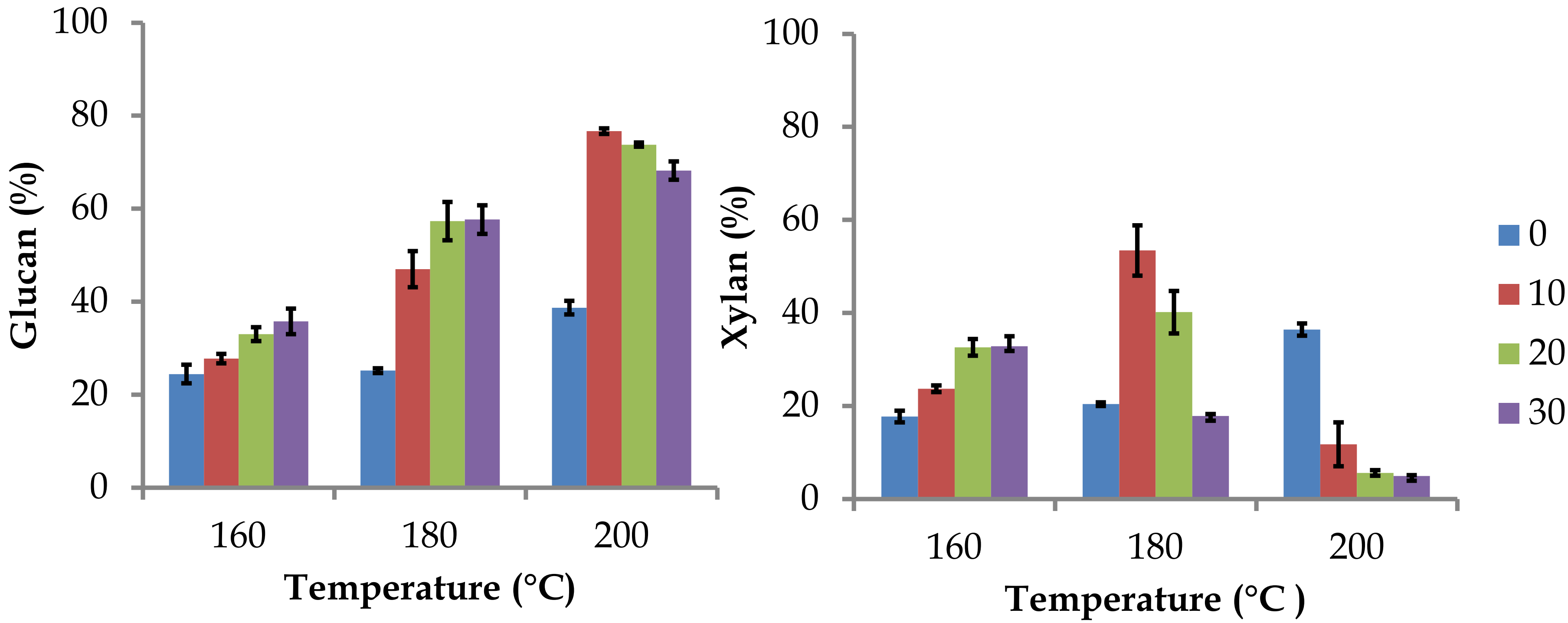
Hot Water Pretreatments

- Have been applied to many substrates
  - Corn stover
  - Switchgrass
  - Yellow polar sawdust
  - *Eucalyptus grandis*
  - Corn fiber
  - Sugarcane bagasse
  - Wheat straw
  - Alfalfa fiber
  - Prarie cord grass
- Not for *Miscanthus x giganteus*

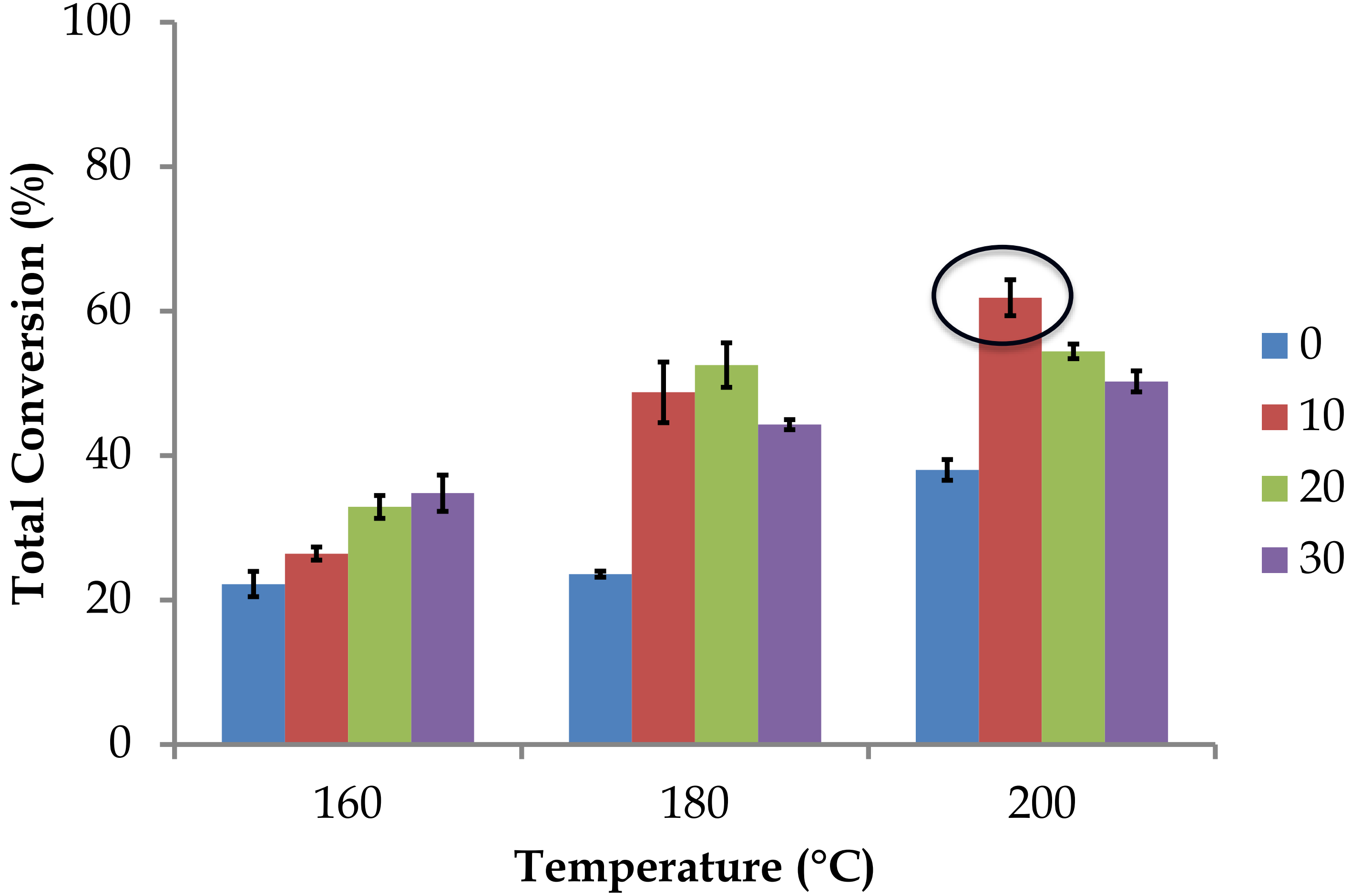
Experimental Design



Glucan & Xylan Conversion (%)

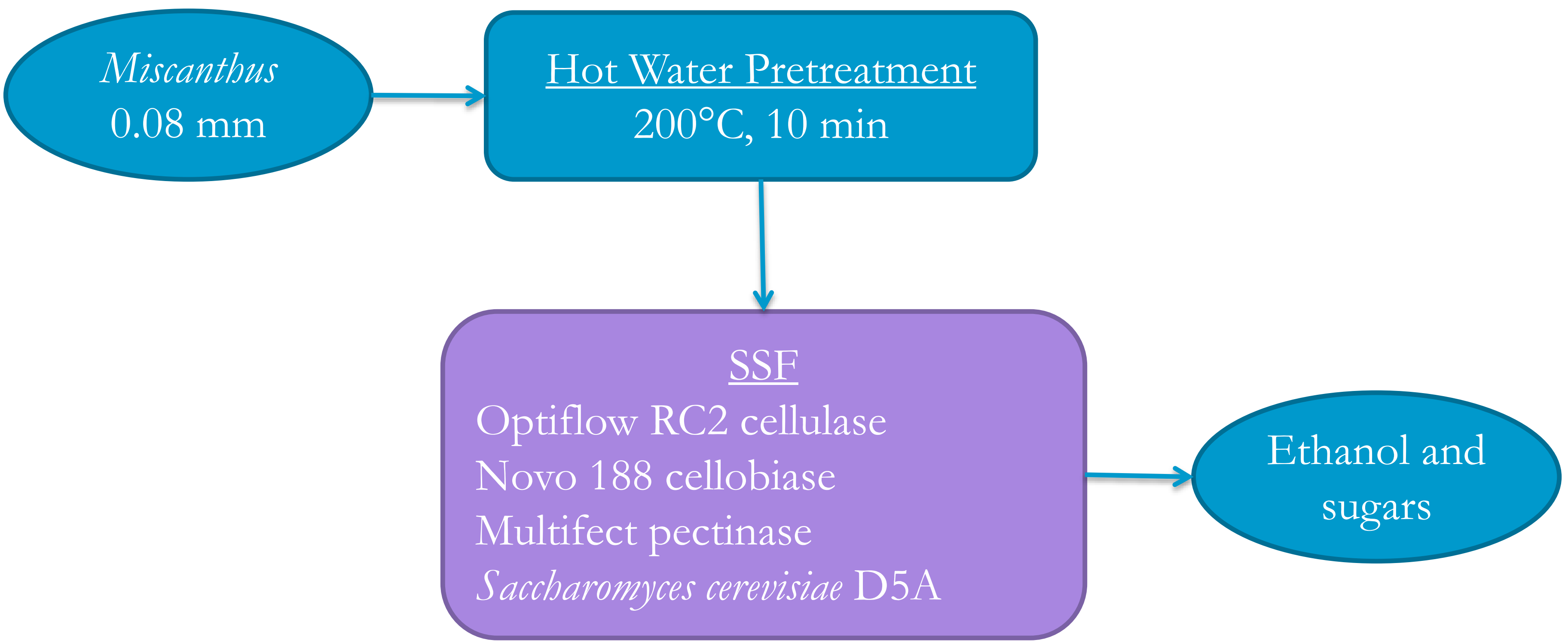


Total Conversion (%)



Simultaneous Saccharification & Fermentation (SSF)

- Optimized pretreatment conditions used for SSF experiment



SSF Results

- Final concentrations at 72 hr
  - Ethanol: 2.04%w/v
  - Glucose: 0.050 %w/v
  - Xylose: 0.093 %w/v
- Ethanol yield was 70%

Conclusions

- Optimized conditions were 200°C for 10 min
- At optimal conditions, pretreated washed solids had 77% glucan, 12% xylan and 62% total conversion
- SSF of pretreated solids resulted in 70% ethanol yield

Thanks!

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