

Non-equilibrium Oscillatory States promote Lignocellulosic Biofuel Production

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Highlights

- Spatiotemporal oscillations observed in lignocellulose hydrolysis in batch reactors
- Enzymatic and catalytic hydrolyses produce limit cycles and chemical chaos, respectively
- Oscillatory dissipative states substantially increase the soluble sugar yields
- Employing non-equilibrium oscillations improves cellulosic biofuel production

1. Introduction

Biological self-assembly renders the cellulose polymers in plant cell walls resistant to deconstruction to monomeric sugars. Here we show experimentally that non-equilibrium oscillatory dissipative states formed via autoregulatory (autocatalytic/ auto-inhibitory) reactions – either catalytic or enzymatic – in batch reactors fuelled by external energy sources result in faster hydrolysis of lignocelluloses leading to higher production of cellulosic biofuels. Enzymatic hydrolysis of hemicelluloses [1] in unmixed tubular reactors lead to activator-inhibitor type unsteady state limit cycles, while ionic-liquid mediated catalytic conversion of non-edible lignocelluloses [2] in batch reactors heated by oil-baths result in chemical chaos, characterized by chaotic strange attractors with fractal dimensions and positive Lyapunov coefficients.

2. Methods

A. Catalytic conversion of lignocelluloses to biofuel precursors: The experiments are carried out in 15 ml glass vials heated in a temperature-controlled oil bath. 150 mg of lignocellulosic Sunn hemp fiber and 3 gm of ionic liquid ([Bmim]Cl) are added to the glass vial and the contents are mixed with a magnetic stirrer at a mixing speed of 500 rpm and a temperature of 160°C. The pre-treatment process consists of stirring the mixture for 6 hours at 110°C. 8 mg of CuCl₂ and water are added to the reaction vial after pre-treatment. Water is added for 16 hours at every half-hour interval at the rate of 25, 28.33, 33.33, 37.5 and 42.8 µl/gm/hr, with the reaction temperature and the mixing speed being maintained at 110°C and 500 rpm, respectively. The samples are collected after pre-treatment every half-hour, and the concentrations of glucose, hydroxymethyl furfural (HMF), levulinic acid (LA) and formic acid (FA) are measured. The glucose separated from the reaction medium is fermented to bioethanol using the yeast *Saccharomyces cerevisiae*.

B. Enzymatic deconstruction of hemicellulose: Batch experiments are carried out with a mixture of hemicellulose (xylan loading = 5 mg/ml) and endo-xylanase (0.25 mg/ml) in 15 ml of sodium acetate buffer solution at 40°C and 150 rpm in an incubator. After mixing for 10 min, the reaction mixture is transferred to tubular reactors of 10 mm diameter, and rest of the hydrolysis is carried out without any mixing (at 0 rpm). Samples are collected at regular time intervals from the different longitudinal locations at an interval of 3 cm along the length of the tubular reactor. The concentrations of the activators and the inhibitors (monomer and dimer) are measured by the Dinitrosalicylic Acid method and using HPLC, respectively.

3. Results and discussion

Figure 1 shows chaotic strange attractors with fractal dimension (1.0305) and positive Lyapunov coefficient (0.3031) appearing on the glucose vs. HMF phase space for catalytic hydrolysis at 110°C and 1 atm in batch reactors. The strange attractors originate from aperiodic (i.e., non-repetitive) temporal oscillations, where all product yields peak at 5 hours for all water-addition rates, suggesting 5 hours to be the optimum hydrolysis time in the domain of chemical chaos. 37.5 µl/gm/hr of water-addition maximizes the concentrations of three of the products – glucose, LA, FA – at all times, with their average yields peaking at 5 hours to 67.45%,

12.42%, and 4.97%, respectively. The equilibrium yields of glucose are about half the maximum non-equilibrium yields: 36.5%, 33.7%, 32%, 28.3%, 28%, 17.2%, at water addition rates of 50, 58.33, 66.67, 75, 83.33, 90 $\mu\text{l/gm/hr}$, respectively. Bioethanol yield of 78% is produced after 15 hours of batch fermentation reaction from glucose separated from the ionic liquid medium using the yeast *Saccharomyces cerevisiae*.

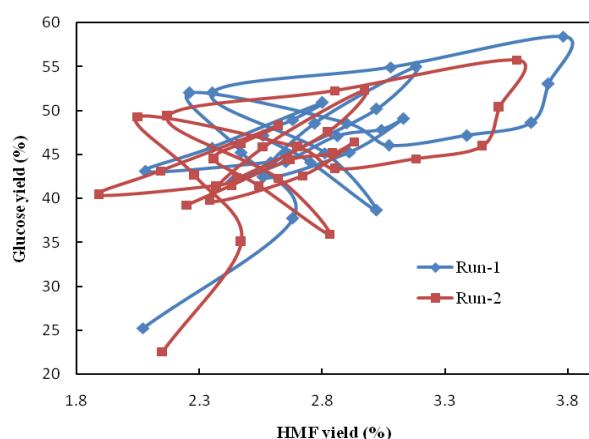


Figure 1. Strange Attractors with fractal dimensions and positive Lyapunov coefficients appear in glucose vs. HMF phase space at 110°C and 1 atm in batch reactors.

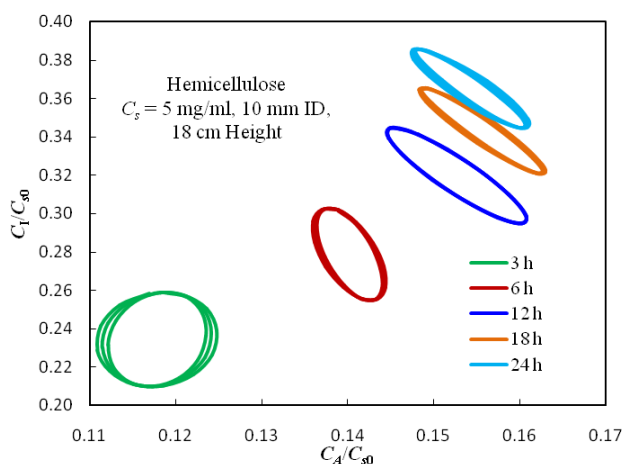


Figure 2. Limit cycles in activator (C_A)-inhibitor (C_I) phase space show oscillatory dynamics during enzymatic depolymerization of hemicellulose in tubular reactor.

Figure 2 shows the limit cycles at various times in the activator-inhibitor phase space during enzymatic hydrolysis of hemicellulose, where the activators are the soluble sugars with Degree of Polymerization of 3 to 11, while the inhibitors are the monomer (xylose) and the dimer (xylobiose). These limit cycles correspond to the unsteady-state oscillatory spatial patterns obtained along the length of the tube due to the inhibitor-activator reaction kinetics and the faster longitudinal diffusion of the inhibitor in the tubular reactor. As the hydrolysis time increases, the limit cycles change from circular to elliptical, with the amplitudes of the oscillations of inhibitor formation and activator depletion representing the major and minor axes of the ellipse, respectively. Initially, the two rates are nearly equal, resulting in circular limit cycles, but as the hydrolysis progresses, the rate of inhibitor formation exceeds that of activator depletion, resulting in elliptical limit cycles. The limit cycles disappear after 3 days of hydrolysis, due to the complete consumption of the activator in the reactor. We observe that the non-equilibrium oscillatory patterns in the tubular reactor enhance the yield of reducing sugar by 8.7% and 14.1% over no mixing and continuous mixing at 24 hrs, respectively, while xylose yield increasing by 3.6% and 7.9%, respectively.

4. Conclusions

For the case of ionic liquid mediated catalytic conversion of lignocellulosic Sunn hemp fibres in batch reactors, we show that non-equilibrium oscillatory states that almost double the yields of biofuel precursors over equilibrium yields can be accessed for a small range of water addition rate ($<50 \mu\text{l/gm/hr}$), with peak product yields observed at $37.5 \mu\text{l/gm/hr}$. In enzymatic hydrolysis of hemicelluloses, the non-equilibrium spatial oscillations are observed to increase the soluble sugar yield by 14% in 24 hrs over continuously mixed spatially homogenous states. This new technology of substantially increasing the yields of biofuel precursors by employing non-equilibrium oscillatory states enhances the production of cellulosic biofuels.

References

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Keywords

Lignocellulosic biofuel; non-equilibrium dissipative states; spatiotemporal oscillations; autocatalysis