

formation, and monitoring protein folding and membrane insertion processes. This dynamic information can be a valuable complement to the more detailed structural information produced by crystallography and NMR spectroscopy. Synchrotron Radiation Circular Dichroism (SRCD) spectroscopy, which uses the intense light of a synchrotron for the measurements, has a number of advantages for membrane protein studies over conventional CD spectroscopy: The higher penetration of the light means proteins can be examined in detergents and lipid environments, as well as in high salts and buffer conditions used for crystallisation, so comparisons can be made as to the physiological relevance of structures. In addition it permits the use of high lipid-to-protein ratios which are more similar to native membranes. The higher signal-to-noise levels in SRCD enable the use of smaller amounts of protein and the detection of smaller conformational changes, as well as the detection of faster dynamic processes over a wider wavelength range. The lower wavelength data measurable improve the accuracy of secondary structure determinations and provide additional information on supersecondary motifs and folds. Plus, using oriented SRCD it is possible to determine the dispositions of different structural elements with respect to the membrane.

I will use voltage-gated sodium channels as a case study demonstrating the types of information that can be gleaned from CD and SRCD studies, including ligand and drug binding, thermal stability, and comparisons of wildtype, modified and mutant proteins.

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#### 1091-Wkshp

##### Using Atomic Force Microscopy for Membrane Structural Analyses Andreas Engel.

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No Abstract.

## Workshop 3: Biophysics of Renewable Energy and Cellular Power Plants

#### 1092-Wkshp

##### The Development of Cellulosic Fuels Chris Somerville.

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Because plants can be deployed inexpensively on a large scale to capture and store solar energy in biomass, one way of moving toward the development of carbon neutral fuels is to use plant biomass for production of fuels. The efficient production of biofuels by routes other than gasification will require innovation in three main areas: sustainable production of feedstocks that do not compete with food production, depolymerization of feedstocks, and conversion of feedstocks to fuels. At present, it is expected that gasoline and diesel replacements will ultimately be derived from cellulosic biomass. In this respect there is renewed interest in identifying plants that have optimal biomass accumulation and understanding the production issues associated with large-scale cultivation and sustainable harvesting of such species. Additionally, the importance of enhancing soil carbon and nutrient retention while minimizing inputs will require an integrated approach to the development of cellulosic energy crops. The challenges on the processing side include the development of improved catalysts for polysaccharide and lignin depolymerization and conversion to fuels as well as the development of microbial strains that can convert a wide range of sugars to next generation fuels under harsh conditions.

#### 1093-Wkshp

##### Ultrastructural Plant Cell Wall 3D Organization and Microbial Deconstruction Manfred Auer.

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No Abstract.

#### 1094-Wkshp

##### Biophysics in Cellulose Biosynthesis and Biodegradation Shi-you Ding.

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Cellulose is considered to be one of the most abundant biopolymers on earth. Although the chemical composition of cellulose,  $\beta$ -1,4 linked linear glucose polymer (glucan), is relatively simple compared to other plant cell wall poly-

saccharides, the physical structure of cellulose is complex. It is generally believed that the  $\beta$ -1,4-glucans are synthesized by 36-unit synthase rosettes, each of which forms a 36-chain cellulose elementary fibril (CEF). Our preliminary results based on nanoscale imaging of samples from living maize cell walls suggest that several CEFs may then coalesce to a bundle, termed a macrofibril, which eventually splits at the end to form parallel microfibrils with concurrent deposition of other cell wall components (i.e. hemicellulose and pectin) secreted from Golgi apparatus. Cellulose microfibrils may twist during dehydration process of natural senescence. High resolution surface measurements suggest that native plant cellulose is a well-organized bundle of  $\beta$ -1,4-glucans. Celluloses can be crystalline, para-crystalline, and even amorphous, depending on their tissue source in native plants, or the way that cellulose is isolated. The structural integrity of cellulose is believed to be one of the major causes of resistance to chemical and enzymatic hydrolysis. Imaging of chemically pretreated corn stover has revealed that enzyme digestibility is positively correlated to the degree of disorder of plant cell wall microfibrils. The ordered nature of the cell wall microfibril probably represents the last and most crucial biological barrier to enzyme hydrolysis. Chemical pretreatment may cause both disordering of native cellulose structure and increased surface accessibility; different pretreatment approaches may be effective in bringing out either of these two changes, or both. Our current work is also focused on investigating the specific interaction between individual enzymes and biomass substrates using single molecule spectroscopy.

#### 1095-Wkshp

##### The Genus *Prevotella*, A Resource of Enzymes for Hemicellulose Degradation

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Hemicellulose and cellulose constitute two major targets in plants for cellulosic ethanol production. Whereas cellulose is a highly homogenous polymer of glucose joined in  $\beta$ -1,4-glycosidic linkages, hemicellulose is mostly a heterogeneous polymer of xylose and arabinose. Thus, the common arrangement of sugars in hemicelluloses, such as xylans from bioenergy feedstocks, is a  $\beta$ -1,4-linked xylose backbone with side chains of arabinofuranosyl, acetyl, and 4-O-methyl glucuronyl groups. Complete hydrolysis of hemicellulose, therefore, requires a complex set of enzymes. Nature has selected for microorganisms that derive their carbon and energy sources from hemicellulose by an enzymatic action that deconstructs the polymer into its component sugars. Such microorganisms include the genus *Prevotella*. We are, therefore, using genomics, bioinformatics, biochemical, and structural analyses to unravel the strategies used by *Prevotella* spp to break down hemicellulose. Our ultimate goal is to rationally assemble enzyme cocktails from these microorganisms for use in the bioenergy industry. We have demonstrated that *Prevotella bryantii* grows rapidly on hemicellulose. Furthermore, the sequencing of its genome has allowed identification of genes likely to encode products for deconstruction of hemicellulosic substrates. Several of these genes have been expressed as recombinant proteins in *E. coli*. Experiments that aimed at examining the synergistic activities of the *P. bryantii* enzymes have led us to reconstitute an enzyme complex that degrades wheat arabinoxylan into its component sugars. This enzyme mixture is a promising product in our effort to deconstruct hemicellulose. We are currently using transcriptomic analyses to improve the enzyme cocktail, and structural analysis is also being applied to rationally synthesize new carbohydrate active enzymes with enhanced activities.

#### 1096-Wkshp

##### Engineering Feedstocks for Biofuel Production Pamela Ronald.

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Researchers in JBEI's Feedstocks Division are developing plants whose lignocellulosic biomass can be more economically and efficiently deconstructed into fermentable sugars for the production of biofuels. Achieving this requires a far better scientific understanding of plant cell wall structure, as well as identifying all the genes and enzymes involved in making lignocellulose. JBEI Feedstocks Division researchers focus their studies on rice, a genetic model for switchgrass and *Miscanthus*, two perennial grasses with great potential as energy crops; and on *Arabidopsis*, a small flowering plant related to mustard, which is a model for poplar, a tree that's also touted as a future source of biofuels. Rice and *Arabidopsis* go from seed to maturity in a matter of weeks, as compared to the year or more required for the biofuel plants they model.