Summary Proceedings

The World Congress on Industrial Biotechnology and Bioprocessing

Orlando, FL, April 21–23, 2004

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Dear World Congress Attendees:

It is with great pleasure that I welcome you to the first ever World Congress on Industrial Biotechnology and Bioprocessing. This conference is the result of discussions that began about a year ago between the Biotechnology Industry Organization (BIO), the American Chemical Society, and the National Agriculture Biotechnology Council. We think it important to bring together leaders from diverse industries to highlight industrial biotechnology applications in manufacturing, agriculture processing, energy and chemical production and to provide a forum for information sharing and business development activities.

Industrial biotechnology is the "third wave" in biotechnology and, with advances in this field accelerating, the need for education and collaboration is obvious. This new conference is designed to be a significant step towards stimulating dialogue, collaboration and fostering the diffusion of industrial biotechnology throughout the manufacturing sector on an international scale. We hope to make it an annual event.

For those of you who are unfamiliar with BIO, we provide advocacy, communications and business development services to more than 1,000 biotechnology companies and some chemical companies, academic centers, and state and international biotechnology associations. Our six-year-old Industrial & Environmental Section represents over 50 innovative companies and organizations that bring biotechnology techniques to large-scale manufacturing, chemical synthesis and bioremediation. You will meet executives from many of those firms over the next three days during breakout sessions and workshops. They will articulate why industrial biotechnology is providing new tools to create economic value while at the same time enhancing environmental protection and sustainable development.

I am confident we will emerge from this event better informed and energized to harness biotechnology's diverse tools for mutual economic and environmental benefit.

I look forward to meeting you and to forging new and productive relationships.

Sincerely,

Carl B

Carl B. Feldbaum
President

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April 8, 2004

The National Agricultural Biotechnology Council (NABC), a consortium of 37 not-for-profit research and/or teaching institutions in Canada and the United States, welcomes participants to the first World Congress on Industrial Biotechnology and Bioprocessing. We are pleased to be conference co-organizers with BIO and ACS to link biotechnolgy, chemistry and agriculture to create new value chains. NABC has been a pioneer in expanding the vision of agriculture beyond food, feed and fiber to energy, chemicals and materials. NABC focused its 2000 annual meeting in Orlando on "The Biobased Economy of the 21st Century: Agriculture Expanding into Health, Energy, Chemicals and Materials." The panels and workshops at this Congress will document expanding national and international commitment and progress from research to product development and commercialization in the biobased economy.

Ralph W.F. Hardy
President, NABC

American Chemical Society
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Madeleine Jacobs
Executive Director & Chief Executive Officer

Dear World Congress Attendee:

Welcome to the first annual World Congress on Industrial Biotechnology and Bioprocessing. We at the American Chemical Society are proud to partner with the Biotechnology Industry Organization (BIO) and the National Agricultural Biotechnology Council (NABC) on this first-ever meeting.

More than ever before, chemistry is deeply entwined with other disciplines, such as biotechnology, engineering, physics, and computer science. The World Congress on Industrial Biotechnology and Bioprocessing provides a unique forum for those practitioners working at this multidisciplinary frontier. We know this meeting will provide you with valuable information, and, more importantly, with a valuable forum for interaction and discussion. Personally, i view this meeting as a catalyst for change in this important new field. We hope the meeting addresses your needs and the needs of the technology that it is meant to serve.

The American Chemical Society is the largest, single-discipline scientific society in the world, with over 159,000 members. We provide a host of products and services to chemical professionals, from technical meetings and publications to education and career services. We also advocate on behalf of the profession, science funding, and education in the sciences. It we can be of service to you, please contact me at executivedirector@acs.org .

Sincerely

Madeleine Jacobs

Madeleine Jacobs
Executive Director & Chief Executive Officer
American Chemical Society

Foreword

The first World Congress on Industrial Biotechnology and Bioprocessing convened in Lake Buena Vista, Florida, April 21–23, 2004, jointly sponsored by the Biotechnology Industry Organization (BIO), the American Chemical Society (ACS), and the National Agricultural Biotechnology Council (NABC). Some 100 presentations were made in three plenary and four parallel "break-out" sessions, and ten workshops were convened for discussion of ancillary topics. Over 400 attendees provided very positive feedback—the congress was deemed an outstanding success and a second is planned for April 20–22, 2005.

The chief organizers were Brent Erickson (Director, Industrial and Environmental Biotechnology Section, BIO), Peter Kelly (Manager, Industry Member Programs, ACS) and Ralph Hardy (President, NABC), who thank the World Congress Progam Committee for their invaluable assistance: David Bransby (Auburn University), Doug Cameron (Cargill, Inc.), Bruce Dale (Michigan State University), David Glassner (Cargill-Dow), Jack Huttner (Genencor International) and Mahmoud Mahmoudian (Eastman Chemical Company). The success of the meeting was due in no small measure to the unflagging, careful planning of Lauren Lamoureux (BIO).

Special thanks are due the US Department of Energy's Genomes to Life program for being a sponsor of the congress and especially of this publication.

Without the excellent assistance of Sean Gorman (University of Florida), Erin Krause and Scott Pryor (both of Cornell University) as recorders of the breakout sessions, and of Arvid Boe, William Gibbons, Kevin Kephart, Padu Krishnan and Vance Owens (all of South Dakota State University), Larry Drum (BIOLarry Consulting), Stephen Eule, John Houghton, George Michaels, Andrew Paterson and Ari Patrinos (all of the Department of Energy), Paula DeGrandis (Cargill, Inc.), Lila Feisee (BIO), Marc Henniker (Strategic Decisions Group), James Hettenhaus (CEAssist), Blair Hughes (McDonnell, Boehnen, Hulbert and Berghoff), Michael Knotec (consultant), Richard Powers (Dorsey and Whitney LLP) and Jerry Warner (Defense Life Sciences) as moderators and recorders of the workshops, this publication would not be possible. Page-layout work was by Susanne Lipari (NABC).

Allan Eaglesham

Allan Eaglesham
Executive Director, NABC
Summary Proceedings Editor
October 2004

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Opening Remarks

Carl Feldbaum, President of the Biotechnology Industry Organization (BIO)

In his opening remarks, Carl Feldbaum reminded the audience that the first BIO annual meeting a decade before attracted an audience of about 500—similar to that assembled for this first World Congress. He predicted that **interest in industrial biotech will grow quickly and that World Congress attendance will increase, within ten years, to the current number participating in the main BIO annual meeting, approximately 15,000.**

Feldbaum offered three pieces of advice. First, he stressed the **need to develop biotech products that directly benefit consumers**. Agricultural biotechnology's focus on the farmer has led to the anti-biotechnology sentiment so prevalent in Europe and to a lesser extent here in the United States. Rather than risk more "seismic shocks" in the future, he suggested that effort be exerted in reaching out to the public to anticipate how new products will be received.

Secondly, he suggested that **possible controversies over safety and ethical concerns raised by new technologies and products should be dealt with honestly and transparently**. He recalled that, in the 1970s, scientists and policymakers alike worried about the safety of genetic engineering and openly established self-regulatory protocols—to develop the technology safely—which proved in due course that the technology is safe. Now high-school students do recombinant-DNA experiments. Not only safety, but also ethical issues have also been of concern. Today, all of BIO's members adhere to a strict bioethics code. Industrial biotechnology has received little public scrutiny and almost all media coverage has been positive. Yet a misstep could change things overnight. Dolly the cloned sheep came as a surprise and discussion developed—almost overnight—from the cloning of animals to the cloning of humans. This global ethical sensation surpassed media attention to the first moon landing. Similar reactions are possible to products currently under development, and it is important that they be considered in advance and accommodated. For example, gene-assembly technology is being developed for the creation of microorganisms. Experiments should be planned with an eye to how the public may receive them; consideration should be given in advance as to how to reach out to bioethicists, NGOs, *etc.*

Thirdly Feldbaum suggested **reaching out to the agriculture and environmental communities**. They are economically and politically powerful groups and potentially invaluable allies in earning government support for R&D and in gaining market and public acceptance for products. Although benefits from industrial biotech may seem obvious, its broad potential is still missed by sections of the media: they do not see it as their beat. But their beat will expand. Industrial biotech has already garnered coverage in the trade and national media. Like biotechnology as a whole, it will move from the science column to the business page to the front page. Fundamental to this is the establishment of a positive reputation. "Today's public opinion, though it may appear as light as air, may be tomorrow's legislation, for better or worse."

With industrial output in populous nations, such as China and India, growing rapidly new technologies are desperately needed to enable sustainable growth. At this stage, **biotechnology's** greatest uses are in medicine and agriculture, but its **greatest long-term impact may well be industrial.**

Plenary Sessions

Opening Plenary Session: Creating Bio-Sustainability in the Twenty-First Century

Rick Oliver (American Learning Solutions), Patrick Moore (Greenspirit), David Morris (Institute for Local Self-Reliance)

Rick Oliver began by asking, "Which is smarter, a personal computer or an ear of corn?" He returned to the question later. All products and technologies follow a predictable cycle—an S-curve—from inception through growth to maturity to decline. This cycle applies also to groups of technologies that characterize an economic era. Seven thousand years ago, the plough led to the growth phase of the agrarian age; the industrial age started in the eighteenth century with the harnessing of energy. In Oliver's opinion the information age began in 1947 with the invention of the transistor, which led to the development of the computer chip.

The strategic inflection point—the beginning of the growth phase—causes individual products, whole technologies and economies to move from one phase to another. In the industrial age, inflection points occurred with the steam engine, the harnessing of electricity, and the jet engine. Landing on the moon marked the maturity of the industrial age: energy, rather than computer power, underpinned Apollo 11. Inflection points within the information age were marked by the transistor, the computer, and the Internet. Oliver suggested that the Internet is to the information age as the moon-landing was to the industrial. If so, **we stand at the beginning of a new economic era, for which he has coined the word "bioterials" (biotechnology / materials). Bioterial developments will affect everything in the economy: the environment, energy, information and, most importantly, manufacturing.**

Each era is driven by a key technology: the plough for the agrarian age, plastics for the industrial age, the personal computer for the information age, and now it is protein. Key inflection points within the bioterials age can already be seen: healthcare, then agriculture and finally—the greatest from an economical point of view—industrial biotechnology. Oliver suggested that the global scope of each bioterial product will be inversely proportional to its size; the smaller the technology the greater its potential economic impact. The bioterials age will last about 25 years and its global economic impact will be greater than those of the agrarian, industrial and information ages combined.

In every other economic era, benefits have come at the end, *e.g.* the moon landing and the $300 personal computer. In contrast, benefits from biotechnology are already accruing. It took 70 years for railroad managers to accept diesel engines and it took 10 years for business managers to accept PCs, whereas it took farmers just 2 years to accept *Bt* corn.

Rather than being seen as the "bad guys," in due course bioterial industrialists will be viewed as protectors and enhancers of the environment.

Value creation will be transformed from the physical to intellectual. **The information content of the bioproduct will eclipse its physical value.** It will be the most information-rich technology known to humankind.

The number of people attending this meeting reflects that we are already in the growth phase of the bioterials S-curve—about 10% of industrial producers are moving to bio-related technologies. There is no time to waste! **A simple ear of corn will prove to be so smart as to make the PC look lame. The PC simply processes whereas protein creates, which is an economic miracle.**

Patrick Moore first heard the term "sustainable development" in 1982. It was a compromise between environmentalists from industrial countries and environmentalists from developing countries. The term—meaning development that is environmentally sustainable—came into general use 5 years later with the UN

Brundtland Commission report on the environment and development, which provided a picture of a sustainable future for society and the world economy. The word "biodiversity" was coined also about twenty years ago; since these terms entered the lexicon, it is noteworthy how far we have progressed in our thinking, our actions and our environment.

Sustainable development is a relative concept. There is no such thing as a perpetually sustainable state; things change all the time. Sustainability is actually an on-going process. Even as the sun is not indefinitely sustainable, neither is any species. We must change our perspective from six-months to a 50- to 100-year period and be aware of the implications of what we are doing now for the distant future. Sustainable development involves balancing the environmental, economic and social frontiers.

A weakness of the environmental movement is that it is fixated solely on environmental considerations, almost as if the 6 billion on earth do not exist and do not continuously need food, energy and materials. However, **sustainable development tries to incorporate environmental values with social and economic priorities that influence decision-making at the highest level and on a personal basis daily.** Therefore, sustainability is a balancing act rather than a single issue: maintaining and improving our civilization—including making more energy and materials available for people in the developing world—while reducing our negative environmental impact. To many in the environmental movement, these goals are mutually exclusive.

It has been claimed that, because Americans on average use eighty times more material and energy daily than people in some developing countries, they have a commensurate negative impact on the environment. In fact, the wealthiest countries have the strictest regulations and the cleanest environments. Human activity is not necessarily bad for the environment: it is possible to improve our lifestyle and culture while reducing the environmental impact, and industrial biotechnology has an important role to play in this regard. It can help sustainability in a number of ways. **The single greatest contribution will be in transportation fuels, which, being non-renewable, constitutes one of the most difficult components of sustainable development**. National security issues are also involved, in terms of our dependence on unstable countries for oil. Attention should be focused on fuels made from biomass and on improved fuel efficiency through hybrid technologies. Hydrogen-powered transportation has a 25- to 50-year time frame; the next phase will be gas-electric and diesel-electric technologies, followed by biofuel hybrids.

Moore is "big on trees." Agriculture production has come at the cost of forest coverage, albeit that **the same area of forest exists in the United States today as 100 years ago. We are growing five times as much food on the same area of land as 50 years ago.** Thus, campaigns of environmental activists against genetically modified (GM) crops are misguided in declaring that the forests can be preserved while we return to less-productive organic farming.

Trees have tremendous potential as a feedstock source for cellulose, hemicellulose and lignin. Rather than clearing land for agricultural crops, we should develop ways of using trees to keep the land forested. In a 10-year rotation tree farm, at least 90% of the land is tree-covered while 10% is harvested every year, thus also providing an environment favoring biodiversity **"Trees are the answer."**

It is unfortunate that the organic farming movement is aligned with the environmentalist anti-GM stance. The fact is, **genetic engineering is a purely organic science that should have been embraced by the organic farmers.** They use seed varieties produced by chemical and nuclear mutagenesis, yet refused the much more precise, 100% organic method: genetic engineering.

By embracing the precautionary principle, environmental activists do not have to demonstrate harm to lobby for banning something. There hasn't been a single stomach ache from GM food, yet environmentalists have a zero-tolerance policy—even with respect to golden rice, with its potential to prevent blindness in vitamin-A deficient children in developing countries.

Despite this irrationality, **the industrial biotechnology industry must engage the environmental movement.** The best approach is in long-term forums, *e.g.* in a round-table format, that include people from many walks of life so that the environmentalists are exposed to diverse opinions. One-on-one negotiations often achieve little because their objective is to maintain confrontation.

In a recent verbal presentation, author Michael Crighton stated that environmentalism has become a religious movement, no longer based on science and logic. They believe that genetic engineering is bad, and their influence includes African countries that refused GM maize as food aid even though it is sold there as cornflakes and many other processed foods available on supermarket shelves to the wealthy—"better dead than GM-fed." Hopefully,industrial biotechnology will avoid such entanglements and will live up to its tremendous promise for the betterment of human welfare and environmental health.

David Morris stated that 150 years ago industrial economies were based on carbohydrates. **In the United States in 1820, 2 tons of plant material were consumed for every ton of minerals.** The chemical industry was based on two waste materials: tar from coal and linters from cotton. There was parallel growth between the biotech industry and the petrochemical industry in the nineteenth and twentieth centuries. Even before the civil war, ethanol made from grain was the country's best-selling chemical, used for fuel and other purposes. The first plastics were made from cotton. Cellanese is a contraction of "cellulose" and "ease" (of use). Cellulose was the basic ingredient in film synthesis and is the origin of the word "celluloid.The main problem in the nineteenth and early twentieth centuries was largely self-inflicted, when public policy applied hobbling taxation and then banned the production of the commodity chemical that was the basis of the biotechnology industry: ethanol. Thus, fifty years of chemical engineering development were thwarted. Even so, in the late 1930s and early 1940s, the first injection molded plastics were made from cellulose acetate. In World War II, rubber was synthesized in breweries.

Plastics made from soy in the 1940s gave way to plastics synthesized from petroleum in the 1960s, and **by the mid-1970s, 8 tons of minerals were being consumed for each ton of plant material in the production of chemicals, *i.e.* 97% were produced from petrochemicals and only 3% from plants. Since then, there has been a turn-around: 2% of our transportation is run on biofuels, 5% of industrial chemicals are from biological sources, and 3% of our electricity is from biomass.** "We are at the end of the beginning," and agriculture will be the key to the carbohydrate or biobased economy. Two to three times more plant matter will be needed. Farmers will need to be involved in true partnerships—enabled by state and federal policies—including those in developing countries.

In the United States, ethanol production has increased enormously in the past 5 years, increa-sing the price of corn by eight to ten cents per bushel. However, if the farmer owns a share in the ethanol plant, then (s)he will earn extra dividend from the sale of the final product. In Minnesota a policy was created, similar to that of the federal government in the early 1980s, to have a state excise tax exemption, like the federal excise tax exemption, which created a market but did not significantly help the farmer—the ethanol was transported to large ADM plants in Illinois and Iowa. In the mid-80s the incentive—$0.20/gallon—was changed to favor the producer rather than the consumer; the incentive applied only to production capacities of up to 15 million gallons/yearAs hoped, ethanol-producing plants sprung up—fifteen in Minnesota—with capacities of up to 15 million gallons; they produce 10% of the US transportation fuel, and have the capacity for 25%. As a result of competition among these producers, efficiency has increased and price of ethanol has decreased. Nine thousand farmer-partners in Minnesota now profit from the sale of the ethanol as well as of the grain. Thus, when we think about industrial biotechnology we must also think about how it integrates with agriculture. If farmers view a new effort as another industry gimmick, they will not buy into it.

Morris drew a distinction between change and progress. The former occurs whether we wish it or not, whereas the latter comes only by changing policies to channel our scientific ingenuity and entrepreneurial energies in directions that are compatible with society's needs and values.

Second Plenary Session:
Industrial Biotechnology and Biomass: From Awareness to Capturing the Value

Jens Riese (McKinsey and Company)

Currently, 5% of industrial chemicals are biobased: alcohols, amino acids, vitamins, pharmaceuticals, and special chemicals. This may increase to 10% by 2010; it could even go to 20%, depending on feedstock prices, consumer acceptance, government policies and support, and investment levels. (Riese's earlier "bullish" projections for 2010 have become somewhat less aggressive.) The field is taking off at this time because hundreds of enzymes are available—it takes only weeks to develop a new one—and because yields are higher and costs are lower with biological systems. Biological systems contribute to sustainability, particularly from the environmental standpoint, *e.g.* fewer and lower emissions including greenhouse gases. Cost advantages include lower capital investment.

Biotechnology holds particular promise for polymer production. Development of novel polymers from petroleum peaked in the 1950s, yet new polymers continue to be needed. The new polymers produced from lactic and succinic acids are good examples of what is possible, but monomers must be produced at low cost: waste biomass holds potential with conversion to fermentable sugars—renewable and eco-friendly.

Iogen in Canada, the world's first biorefinery, is now fully operational, converting cellulosic material—wheat straw—into etha-nol for blending with gasoline. With modifications, production costs could fall to 50% of what they are today. By 2010 bioethanol will constitute 6% of transportation fuel used in Europe, and it is expected that 50% of fine chemicals will be biobased. Also three-fold more bulk chemicals will be biobased than today.

Challenges include determining how and where to compete, identifying the right opportunities, managing a portfolio of costly and risky R&D projects, and market development is often underestimated in terms of complexity and time required. Riese recommended joining forces with complementary entities to create favorable consummer perceptions and regulatory boundary conditions.

Third Plenary Session: Technotrends

Daniel Burrus (Burrus Research Associates, Inc.)

To ensure a sustainable planet even as the Chinese give up bicycles for automobiles, we have to work beyond personal and organizational egos. Cooperation to make the pie bigger for all will entail collaboration and communication. Biotechnological progress is in the "big deal" phase as a result of the availability of computers for genomics work. We need to tell the right story to sell it. A problem is an opportunity when you see it coming; we must identify the real problems to change the role of chemicals/materials/energy industries, not only with technology but also with integrity.

Breakout Sessions

Track 1: Manufacturing and Synthesis

Enzyme Development for Bioprocessing Applications

Philippe Soucaille (Metabolic Explorer), Reinhard Rosson (Bio-Technical Resources), Richard Burlingame (Dyadic International)

An efficient, low-cost method for the production of pharmaceuticals and chemicals in a continuous culture has been developed. MetEvol, an *in vivo* molecular technique, takes advantage of the rapid growth rate and high natural mutation frequency of microorganisms: recombinations are not enhanced via other methods. Examples of MetEvol include glycerol production in *E. coli*, evolution of a methionine pathway in *E. coli*, and evolution of NADPH-over-producing strains. Since microorganism growth rate is fast, pathways can be enhanced or newly developed in only a few months.

Fermentation processes for the production of glucosamine and N-acetylglucosamine are being developed with metabolically engineered strains of *E. coli*. Glucosamine, used to treat osteoarthritis in humans and animals, is currently manufactured from chitin, which is allergenic for some. Over-expressing the gene and reducing feedback inhibition led to a 200-fold increase in glucosamine production in shake flasks. Since glucosamine is unstable at neutral pH, the focus was shifted towards the production of N-acetylglucosamine, which can be easily converted to glucosamine.

An integrated technology platform has been developed for gene discovery, gene expression, and protein production in the fungus *Chrysosporium lucknowense*. Seventy thousand clones can be screened weekly. The system has many advantages, including efficient intron processing, no hyperglycosylation, high transformation efficiency, high levels of protein production, amenable laboratory conditions such as no well-to-well contamination and no clogging, along with versatile fermentation conditions such as a broad pH range, low viscosity, wide temperature range, and short time cycles. Using a single strain host system for gene discovery and expression allows a more streamlined, efficient approach; the probability of success is increased, and is generally more cost-effective. Products are being developed to serve the nutrition, pulp and paper, and pharmaceutical industries, among others. Currently an endoglucanase and a beta-glucanase are on the market

Advanced Biocatalysis in the Chemical Industry: Fine Chemical and Pharmaceutical Production

Mahmoud Mahmoudian (Eastman), Robert DiCosimo, (DuPont), Ramesh Patel (Bristol-Meyers Squibb), Mani Subramanian (Dow)

Biocatalysis is increasingly being used in the pharmaceutical industry to produce chemical building blocks and intermediates. Since many compounds under development in the pharmaceutical sector are chiral and pose significant challenges for industrial production, the industry has begun to outsource much of this work to the biotechnology and academic sectors. One example of a collaboration that has emerged for the production of a bioproduct from biocatalysis is Cargill-Dow LLC, which produces polylactic acid from corn and other plants.

DuPont is developing biocatalytic processes that include the following steps: prediction of expected market volume over time, calculation of fermentation requirements, identification of contract manufacturers, calculation of production costs, optimization of the process for available equipment, and preparation of a smooth technology-transfer package. **Biocatalyses are under study for production of the pharmaceutical intermediate (R,S)-1, *cis*-4-hydroxy-D-proline, 5-cyanovaleramide, the industrial solvent dimethyl-2-piperidone, and 3-hydroxyalkanoic acid for the synthesis of co-polyester polyols.** These had significant manufacturing hurdles using chemocatalytic methods, whereas biocatalytic methods—entailing optimization of enzyme selectivity, stability, or specific activity, reduction of waste streams, or enhancement of reaction productivity—enable cost-effective production. Some recombinant microbes are being used, but only if wild-type strains fail to meet economic goals. The screening is done with high-throughput assays and colorimetry.

Biocatalysis using, for example, *E. coli* (in which enzymes are expressed in large amounts) for the production of chiral pharmaceutical intermediates has clear advantages over chemical routes, including stereoselectivity of biocatalysts, increased yields, ambient temperature, waste minimization, and reusability of biocatalysts. This approach **is being used to produce key intermediates for the development of drugs for the treatment of Alzheimer's disease, hypertension, and AIDS. The use of genetically modified organisms has led to even greater yields.**

In an initiative to develop new specialty products for pharmaceutical and agricultural applications, Dow set out to enzymically hydrolyze DOWANOL PMA into pure enantiomeric forms using lipase B from *Candida antarctica*. Maximizing substrate loading and minimizing enzyme loading while optimizing other process parameters established a cost-effective process. Potential added-value applications include chiral solvents for asymmetric synthesis and chromatographic separation.

Biocatalyst Engineering for Synthesis of Pharmaceutical Intermediates

Gjalt Huisman (Codexis), Jim Lalonde (Altus), Wen Chen Suen (Schering-Plough Research Institute)

Using evolution technology developed by Codexis, biocatalysts can be developed with increased volumetric productivity, yields, and/or ability to withstand higher substrate loading than their native forms. The platform provides rapid, practical, and clean, economically feasible processes. It can be applied to single genes, operons, or entire genomes. DNA-family shuffling is used to make the recombinant libraries, and screening is performed to select mutants that are more active and/or less product-inhibitive. It has been developed for nitrilases, lipases, and ketoreductases. As a model for the system, the activity of glucose dehydrogenase has been increased. Since many of the changes made to the enzymes are far from the active site, it is presumed that many of the improvements result from changes in tertiary structure.

In an attempt to develop a practical biocatalytic pharmaceutical process, Altus has focused on the utilization of **cross-linked enzyme crystals (CLEC) for the bioconversion of substrates to usable end-products.** The CLEC approach has many advantages, including increased enzyme stability, recyclability, lower cost, a highly porous network (maximizing surface area), and the ability to catalyze reactions in organic and mixed aqueous/organic media for the production of a broad array of catalysts such as antibodies and therapeutic enzymes. TheraCLEC lipase, an oral drug that survives the low pH in the stomach, is now entering large-scale trials.

Error-prone PCR and DNA-family shuffling of homologous lipase B genes were used successfully with *Candida antartica* to create a variant with increased thermostability and improved activity (increased k_{cat}).

Federal Policy and Programs for Biorefinery Development

Doug Kaempf (US Department of Energy), Roger Conway (US Department of Agriculture), Lee Lynd (Dartmouth College)

In an effort to decrease US dependence on foreign oil, **the Department of Energy implemented the Biomass Research and Development Act of 2002 and created the Biomass Research and Development Initiative**, a multi-agency effort guided by a technical advisory committee that aims to accelerate biobased industrial development. This initiative provides funds for research to overcome the technological barriers to the development of biobased industries, specifically in the areas of biomass harvesting, storage, and collection. Other challenges associated with the emergence of a biobased economy are process integration, risk, and attracting investors. The DOE is also building industrial linkages with the forest and petroleum industries, in an effort to transform them into more sustainable, biobased entities. **Twelve fundamental chemicals have been identified that can be produced by biobased processes.**

The Energy Title of the 2002 Farm Bill, and

specifically Title 9, creates new opportunities for agriculture and for biobased products, power, and other renewable resources. Included in Title 9 are initiatives such as a federal procurement program for biobased products, a program to develop applications for hydrogen and fuel-cell technologies co-sponsored by the USDA and the DOE, and incentives for production of bioethanol and biodiesel. Competitive grants for biorefinery development, biodiesel fuel education, renewable energy and energy efficiency, and biomass research and development are included. The Energy Title includes programs for rural development, marketing and regulatory programs, farm and foreign agricultural services, research, education, and economics, as well as on natural resources and the environment. The USDA encourages cooperative participation from farm communities in the development of biobased business endeavors.

Developing a biobased economy will be vital for a sustainable and energy-secure future for the United States. Many fuels and non-energy products like animal feed, pulp and paper, and organic chemicals and polymers can be produced from renewable feedstocks such as corn and switchgrass. A high-impact scenario would include co-producing animal feedstock and cellulosic material for energy production, requiring no more land than is currently being used, and largely displacing US dependency on foreign oil. Highly productive crops, cost-effective and efficient processing of feedstocks, and fuel-efficient vehicles would be key parts of this scenario. Decreased air pollution from biobased fuels in comparison to petroleum-based should be considered in dollar form as a component of the economics. The Role of Biomass in America's Energy Future project was initiated to accelerate biomass use in our economy by identifying potential ways to integrate current knowledge and promote innovation.

The State of Industrial Biotechnology in Europe

Colja Laane (DSM Food Specialties), Dirk Carrez (BelgoBiotech), Kirsten Staer (Novozymes)

Due to the growing demand for innovation in the field of white biotechnology, **centers for public-private partnerships (PPPs) have been established in the Netherlands**: collaborations between industry and academia that attempt to bridge the gap between the short-term research and development that traditionally occurs in the private sector and the long-term R&D that traditionally occurs in the public sector. Two centers for PPPs now exist in the Netherlands: **the Kluyver Center for Genomics of Industrial Fermentation, which focuses on microbial genomics, and Biobased Sustainable Industrial Chemistry (B-Basic), which creates programs for the biobased production of bulk and fine chemicals and supports research on novel feedstocks**. These centers aim to translate new knowledge into products to further boost white biotechnology industry in the Netherlands. Companies and other interest groups in other European nations can join the Netherlands PPPs; however, increased fees may be required. Academia/industry partnerships are fostered when a defined research agenda is followed with appropriate funding.

Europe is the world's largest chemical producer, at one third of global production. Belgium generates 17% of Europe's chemical exports. The 2004 Industrial Biotechnology and Sustainable Chemistry Report published by the Royal Belgian Academy Council of Applied Sciences cited many public-policy recommendations that will facilitate a smooth transition to a biobased economy. These recommendations include more governmental support, more support for multidisciplinary research, attention to critical mass for R&D, promotion of knowledge and awareness of industrial biotechnology, development of political and fiscal support measures, less taxation of bioenergy, and the promotion of market penetration of sustainable bioproducts and bioprocesses. In addition, the Belgian Interdisciplinary Platform for Industrial Biotechnology was created with distinct task forces charged with creating visibility and awareness, and facilitating ease into markets for areas such as biomass, bioprocesses, manufacturing and wastes, bioenergy, and bioremediation.

White biotechnology represents a growing industrial base throughout Europe. **Genetically modified organisms are increasingly used for the production of enzymes and vita-**

mins. Enzymes are used to improve the quality of consumer products and food, and in the pulp and paper, textile, and leather industries. In contrast, concern remains over the safety of genetically modified foods, due to a general fear of biotechnology, distrust of authorities and industry, moral and ethical concerns, and the lack of choice for consumers. New, stricter regulations have been introduced requiring transparent labeling on all food and feed. Although labeling requirements are a step towards ensuring the safety of European consumers, more needs to be done to educate citizens about the potential benefits of biotechnology. Communicating to the public the economic and environmental advantages of using renewable resources and creating biodegradable products will be the key to the success of the biotech industry.

Federal Programs for Chemical Platform Development

Todd Werpy (Pacific Northwest National Laboratory on behalf of the US Department of Energy), Diza Braksmayer (Cargill), Patricia Nugent (Dow)

The goal of the Department of Energy's Energy Efficiency and Renewable Energy Office of Biomass Program (OBP), is to partner with industry to foster research in biotechnology. The development of an integrated biorefinery still has many barriers. The OBP is strategizing to overcome these obstacles by providing analysis and research in six program areas: a feedstock interface, a sugar platform, a thermochemical platform,products,integrated biorefineries, and program management. By assessing the petroleum industry and its ability to make many products from a few base compounds, the OBP has come up with a top-ten list of products that can be made from sugars and further transformed to myriad biobased items. Additional studies are underway assessing the potential for producing value-added products from oils and lignin. These products will be key to the economic viability of the integrated biorefinery.

In an initiative to create new value chains, Cargill is assessing the use of crops to produce **new carbohydrate feedstocks that can be integrated with downstream activities and minimize overall processing costs. 3-hydroxypropionic acid (3hp), which can be made by anaerobic fermentation of sugars, serves as an intermediate for the production of many other organic chemicals such as polyesters, monoesters, acrylic acid, 1,3-propanediol, as well as acrylamide and hydroxyamides.** 3hp has many direct uses in applications like water treatment where it has several distinct advantages over current products: it is less corrosive than citric acid due to a higher pKA, less toxic, water soluble, noncrystallizing, and has a great capacity to solubilize mineral salts. Applications of 3hp include use as a scale remover and corrosion inhibitor for boilers, heat exchangers, cooling and condenser systems, and water pipes, and as an ingredient in personal care products for anti-aging, skin lightening, moisturizing, dermatological conditioning, cosmetics, shampoos, and hair coloring. Cargill is participating in a jointly funded effort with the US Department of Energy to develop 3hp products.

In conjunction with the US Department of Energy, Dow is attempting to develop new, sustainable feedstock platforms for chemical production. These products must have characteristics that meet short- and long-term goals in terms of economic viability and sustainability. Currently, **Dow is partnering with the US Department of Agriculture and Castor Oil, Inc., to develop new oleochemical products from castor plants.** Castor serves as a model industrial crop because it is already a well established agricultural product, has well characterized lipid biochemistry, can grow on marginal land without a lot of water, and produces an abundance of cellulosic biomass that can be used in other biotech applications. The current research focus is on new catalyst synthesis and screening and process chemistry.

Biocatalyst Developments in Academia

John Frost (Michigan State University), Lonnie Ingram (University of Florida), Michael Flickinger (University of Minnesota)

The use of renewable resources in combination with microbial catalysis can provide a sustainable alternative to chemical means of conversion of nonrenewable resources. By manipulating native microbial metabolic pathways and introducing foreign genes, it is possible to maximize the number of chemicals that can be made in a sustainable fashion. **Microbial path-**

ways have been exploited to produce shikimic acid and amnoshikimic acid, 1,2,4-butanetriol, and Nylon66, illustrating the diversity of chemicals that can be so produced.

If biobased industries can address global warming and CO_2 issues, chemicals made from renewable resources may count as CO_2 credits. This needs to be addressed on national and international levels. Ultimately, it may be desirable to produce materials that are not biodegradable to act as sinks for carbon.

The United States imports over half of its petroleum needs, the majority of which is for transportation purposes. There is a growing need to replace petroleum with renewable feedstocks and to become more energy-efficient. With conversion to ethanol, lignocellulosics—of which there are diverse sources such as pulp mills, agricultural and forest residues—could supply 20 to 30% of US fuel uses.

Lignocellulosics can be broken down into pentoses and hexoses. Few microorganisms degrade both 5-C and 6-C sugars, therefore recombinant strains of *E. coli* have been produced that co-degrade pentoses and hexoses, and exhibit increased rates of glycolytic flux. **Large-scale microbial conversion of lignocellulosics to ethanol is not yet commercially viable. Developments in areas such as pretreatment of lignocellulosic biomass, harvesting technologies, and byproduct uses are required to justify construction of bioconversion plants.**

Biocatalytic coatings present an alternative to whole-cell biocatalysis. These coatings are thin, porous, and contain 50% (v/v) of non-growing metabolically active microorganisms. An example of one of the many applications of this technology is the use of inkjet printers to print microbial cultures directly onto Petri dishes, which will later be replaced with continuous tapes. The first commercial applications of this technology are expected to be as a means of supplying microbial cultures, and as phototrophic coatings for the production of gases such as hydrogen. Fermentors will still be required for the initial production of the cultures.

Development of New Biofactories

Tim Dodge (Genencor), Mark Finkelstein (National Renewable Energy Laboratory), K.T. Shanmugam (University of Florida)

The petroleum refinery is an extremely flexible facility. To replace the current petroleum-based economy with a biobased economy, it will be necessary to mimic this flexibility in the design of biorefineries. A wide array of organisms produce amylases or glucoamylases for the conversion of starch; however, few organisms possess the ability to effectively convert cellulases and hemicellulases to useful products. New biocatalysts are needed that can withstand the high-temperature environments of lignocellulosic saccarification and are simultaneously able to ferment the resulting sugars into products. Simultaneous saccarification and fermentation of lignocellulosic material will dramatically reduce the cost of the bio-conversion process.

The transition from a petrochemical based economy to a bioeconomy presents significant challenges. The intrinsic characteristics of biomass in comparison with petroleum impart distinct obstacles. Biomass has a low energy bulk density, is widely dispersed, is not always easily transported, and there is no infrastructure to develop a biobased economy from wheat straw, corn stover, *etc*. However, biomass does have a major advantage: a potentially large, sustainable supply. **The integrated biorefinery will make use of a thermal conversion pathway (producing syngas, heat, power, and electricity), a biochemical conversion pathway (producing sugars that can be converted to a variety of fermentation bioproducts) and existing technology. In terms of technological development, the following areas need attention: pretreatment of lignocellulosic material, cellulase enzymes, fermentative strains, saccarification/fermentation configurations, feedstock collection, and lignin utilization.** These challenges represent unique opportunities for industrial development in the biomass area.

Renewable resources such as lignocellulosics represent above-ground sources of carbon to displace below-ground sources such as oil. Lignocellulosic material must be depolymerized into sugars before it can be fermented to valuable end-products. Fungal cellulases are capable of depolymerizing cellulose at high temperatures to pentoses and hexoses. The biocatalysts that convert these sugars are not typically able to withstand high temperatures, which poses a challenge for simultaneous saccharification and fermentation. **New biocatalysts have been developed to withstand high temperatures and simultaneously convert both glucose and xylose to L(+)-lactic acid.** Being able to depolymerize lignocellulosics and ferment the resulting sugars in one reaction vessel will increase the economic feasibility of using lignocellulosic material as a biobased feedstock. Although lactic acid organisms are more temperature tolerant, they are not classified as thermophiles. **More focus is needed on thermophilic organisms.**

Track 2: Bioprocessing of Agricultural Feedstocks

Overcoming Barriers to Production, Harvest and Utilization of Biomass Feedstocks for Production of Bioethanol and Biobased Products

David Bransby (Auburn University), Tom Schechenger (Schechenger Consulting), Mark Downing (Oak Ridge National Laboratory)

Herbaceous biomass crops for energy production must be competitive with fossil fuels as a feedstock and with existing food, fiber, and feed crops, all of which are subsidized through various means. **The easiest and most likely policy changes that will allow herbaceous energy crops to compete equally with both fossil fuels and conventional crops are the creation of biomass energy subsidies to match those that exist for these other products.** The pulp/paper and sugar industries are good models for overcoming some of the technical barriers for biomass energy crops

The ideal residual biomass—dry, pure, baled, consistently available, and inexpensive feedstock—will be very difficult to achieve, but compromises and adaptations on the part of growers and industry can lead to economic benefits for both. Adapting to harvesting standing corn can lead to benefits in: broadening the harvest window, eliminating costs of gathering, raking, baling, *etc*., increased efficiency in use of equipment and labor, minimizing contamination, simplification of purchasing, and reducing transportation costs (if stover and grain are transported together). **Adapting to wet storage will lead to benefits in: increased harvest season, elimination of fire risks, decreased storage area requirements, and increased process automation.**

A 1,500-acre hybrid poplar demonstration project in Minnesota has shown that increasing yields by a factor of two decreased costs by 22 to 34%. Policy interventions that would increase the planting of dedicated woody biomass crops for bioenergy and bioproducts include: limiting external incentives, promoting market development, low-cost community-based exten-sion, focused research support, and decreasing federal and/or state regulatory disincentives. Production and consumption green power incentives, and dual risk/profit sharing structure are neces-sary conditions for woody biomass economic development, while technical demonstrations, state- and federal-level public/private coopera-tives, and annual producer payment programs to decrease farmer risk are sufficient conditions.

Nematode-control effects of rotating switchgrass (as a biomass crop) could have a large economic impact on farming systems. Corn stover and wheat straw will likely be the most plentiful forms of biomass feedstocks, but the chea-pest forms will vary with region. Some growers will be resistant to removing stover because of the soil organic matter (SOM) benefits it pro-vides if left on the field. On the other hand, no-till agriculture can improve SOM and still provide stover.

Upstream Biomass Processing for the Biorefinery Industry: Storage and Pre-Treatment Issues

James Hettenhaus (cea, Inc.), Susan Hennessey (DuPont), Gregory Lewis (Athenix)

The bulk of the available biomass (stover, straw, trimmings, and dedicated energy crops) for use as biorefinery feedstocks is located in and around the midwest. Growers have many concerns over stover removal and no-till agriculture including erosion, organic matter depletion, nutrient loss, moisture management, and feedstock collection logistics. These concerns are surmountable with significant economic benefits for farmers, especially if they are integrated further up the value-chain in the biorefinery industry.

In order to operate commercially successful biorefineries for the production of ethanol and valueadded chemicals, we must have:

- **low-cost, sustainable feedstock supplies,**
- **an infrastructure to support collection, transportation, and storage of feedstocks,**
- **a robust feedstock pre-treatment process that allows greater utilization of existing sugars from a variety of feedstocks and can be integrated with downstream processing.**

Athenix has developed a biomass pre-treatment method that is environmentally friendly, biologically compatible, simple, effective, and cost-competitive. Early data show that the method works on a wide variety of biomass feedstocks and allows greater than 90% hydrolysis of glucose and greater than 80% hydrolysis of sugars as a whole. An analysis with NREL's ASPEN Plus model shows that sugar production costs were $0.08/lb with projected costs of $0.05 to $0.06/lb after scale-up.

Novel Plant-Oil Derived Industrial Products Through Biocatalysis

Morton Wurtz-Christensen (Novozymes, Inc.), Geoffrey Hill (Ernst-Mortiz-Arndt University), James Iademarco (Diversa)

Novozymes has introduced a new immobilized lipase for the enzymatic interesterification of vegetable oils without producing the unhealthy *trans*-fats that form during the standard partial hydrogenation process. Fats produced through enzymatic interesterification have the same melting patterns as those conventionally produced, while the new process is simpler and cost-efficient. The technology can be easily integrated into the existing industry, but lack of equipment for and experience with enzymes means that much work is needed to show a conservative industry the benefits and ease of adapting the new technology.

Lipase technology uses commercially available enzymes to produce cosmetics and other important items using long-chain fatty acids, alcohols, amines, or other compounds. Enzymic production of these products provides advantages over chemical production with excellent conversion selectivity, minimal material loss, 60% less energy demand, and 80% less waste production. **Approximately 400 kg of lipase, produced from a genetically modified *Aspergillus*, can produce up to 2,000 metric tons of fat.** Although product quality is often much better using enzymic conversion processes and some economical advantages exist in recycling biocatalysts and in the lack of solvents or purification requirements, enzyme prices must still be reduced to bring production costs below current technology to speed transition.

Enzymic, fermentation, and transgenic technologies may provide the following benefits to plant-derived oil production: decreased usage of harsh chemicals and water, increased oil yields, and improved quality, stability, purity, and performance. Enzymic processing provides very selective transformation capabilities that may improve properties of existing products or create entirely new ones. Technologies such as high-throughput screening, directed evolution, and gene reassembly have produced enzymes that function under a wide array of temperatures and pHs with large increases in activity.

Enzymes for Bioprocessing Agricultural Feedstocks to Ethanol and Biobased Products

Sarah Teter (Novozymes, Inc.), Mike Lanahan (Syngenta), Rosa Dominguez (University of Yucatan)

Novozymes—with help from the National Renewable Energy Laboratory, and the US Department of Energy—**reduced cellulose production costs over a period of 3 years from $5.50/gallon ethanol to $0.30–$0.50/gallon ethanol, exceeding their goal of a ten-fold cost reduction.** Through an integration of bioinformatics, proteomic and microarray analyses, and a directed evolution program, significant improvements were seen in enzyme yield, activity, and thermostability. By cultivating the organisms on biomass rather than glucose media, novel fungal enzymes were expressed which may lead to further accessibility of biomass cellulose.

Syngenta has developed technology that **produces a variety of novel starch-hydrolyzing enzymes directly in corn grain, enabling the design of corn varieties especially suited to**

specific food, feed and processing applications. This development allows flexible and high-level enzyme expression in a dry, stable, pre-packaged formulation. The transgenic grain has the same starch, protein, and oil contents as its conventional counterpart and is expected to receive full approval as food and feed.

Waste wash waters from tortilla production in Mexico are being used to produce **amylase from *Aspergillus awamori***. This waste material—currently released to the environment—has high pH, temperature, organic matter concentration, and biological oxygen demand. **Technical feasibility, process simplicity, cost, and social/political factors make this technology adaptable to waste-streams from agricultural, food, and brewing industries.** Genetic modification of the fungus could lead to process improvements.

Update on Technology Development for Advanced Biorefineries

Dave Glassner (Cargill-Dow), Charles Wyman (Dartmouth College), Gerson Santos-Leon (Abengoa Bioenergy Corporation)

Commonly cited barriers to operational biorefineries—such as: 1) feedstock cost, availability and variability, 2) pre-hydrolysis reactor design and cost, 3) cellulose hydrolysis cost, and 4) the availability of mixed sugar fermenting biocatalysts—are perceived rather than real barriers to technological development. Tremendous progress has been made in all of these areas in the past 20 years, resulting in improved technologies and lower enzyme-processing and enzyme costs. **Proprietary intellectual property and its associated competitive advantage will be required to get beyond the real barriers of the high capital cost and high risk needed for biorefinery development.** Risk mitigation is critical for the development of large biorefineries as economies of scale require large capital investment in order to produce a cost-competitive product. Also, integrating several new technologies into a facility multiplies ultimate risk

Overcoming the challenges of the diversity and recalcitrance of sugars available in biomass feedstocks will provide sustainable and economic sources of specialty and commodity chemicals. **An economic model analysis shows that economies of scale associated with feedstock transportation decrease sugar costs for plants processing up to 10,000 dry tons per day with relatively small incremental benefits for increasingly larger plants.** The processing of those sugars into a combination of high-value, small-market chemicals and lower value, large-market commodities, such as ethanol, provide economic opportunities but come with significant economic and technological system complexities.

Abengoa Bioenergy Corporation has operational and developing ethanol plants in Europe and the United States. In addition to current dry-mill ethanol production technologies, the company is **working on advanced biomass-conversion processes, gasification, pyrolysis, and hydrogen production**. One goal for the dry mill industry is to increase production from 2.3 to 2.9 gallons ethanol/bushel. Goals for their co-product industry include increasing consistency, digestibility, and protein concentration of feed products derived from ethanol-processing residues.

State Initiatives to Jumpstart the Biobased Economy

Jill Euken (Iowa State University and BIOWA), Kevin Kephart (South Dakota State University), Larry Walker (Cornell University)

Following the development of a roadmap created by the Iowa Industries of the Future project, the **BIOWA Development Association has formed to implement the recommendations and to encourage the growth of Iowa's bioeconomy**. The not-for-profit group comprises, and has support from, government, industry, academia, university-extension services, growers, financial and environmental advisors, and the public. The group hopes to create jobs and investment opportunities both in larger-scale biorefineries and through creation of small businesses focusing on biobased products. **Up to 22,000 jobs are expected to come to Iowa within 10 years because of growth in biobased industries.**

The US Congress has passed legislation authorizing funding of the **Sun Grant Initiative** to encourage national energy security, rural economic development, and environmental sustainability through biobased-industry development. Working primarily through land-grant universities, the **legislation calls for the allocation of $375 million over 6 years to develop**

five biobased-industry regional centers and a competitive grants program within each region. Funds will be spent on research, extension, and education for technology development and implementation of biobased power, fuel, and products. **Up to 10,000 jobs could be added to the South Dakota economy if its agricultural resources remain in the state for bioprocessing.**

With a diversity in rural/urban areas and a wide variety of available biomass feedstocks, New York State can serve as a model for development of biobased opportunities in the northeast. **Several state (NYSTAR, genNYsis, NYSERDA), national/regional (Sun Grant Initiative), local (BioEconomy Partners in the Buffalo area), and university (SABBIC at Cornell University) research initiatives are helping NY progress as a leader in this field.** Genetic resources, basic and applied research, a systems development approach, educational outreach, and collaborations are key components to biobased-industry development initiatives.

Biopolymer Production in Three Platforms

John Pierce (DuPont), Pat Gruber (Cargill-Dow), Oliver Peoples (Metabolix)

With the help of Genencor, Dupont has created a biological process for production of 1,3-propanediol, a key component of their new Sorona polymer. Development of a genetically modified microorganism with the ability to synthesize 1,3-propanediol allows the production of a versatile polymer that has several commercially advantageous properties, but previously could not be economically produced. Biotechnology made this new development feasible, but the work is still an integration of knowledge and skill in biology, chemistry, physics, material science, and engineering.

Cargill-Dow is producing a new family of polymer materials under the tradenames of NatureWorks and INGEO that are manufactured from renewable sugar feedstocks. **The cost, petroleum usage, and greenhouse-gas emissions for the production of the new polylactate (PLA) polymers are now less than those from the competing polymer (polyethylene terephthalate, PET).** Production costs and the environmental footprint are expected to drop further with increased manufacturing and use of cellulosic biomass feedstocks.

Metabolix is a technology company that **has developed a process for producing a host of polyhydroxyalkanoate copolymers** from a range of biological feedstocks. Polymeric granules are fully formed within bacterial cells, simplifying the process and **bringing down overall costs to $0.60/lb with an expectation of further reduction to under $0.50/lb**. The new process uses standard fermentation and extraction equipment and the final product performs well in all downstream processing equipment.

Marketing issues concerning the use of genetically modified organisms have been low with this technology and appear to be surmountable with education regarding environmental benefits and differentiation from food products.

USDA's Biobased Products Rulemaking: Where Do We Go From Here?

Roger Conway (US Department of Agriculture), Mark Dungan (Consultant), Kim Kristoff (Biobased Manufacturers' Association)

The Office of Energy Policy and New Uses at the USDA has drafted a federal preferred procurement program for biobased products whereby federal agencies are required to purchase available biobased products when those products meet reasonable criteria of cost and performance. The program is intended to: spur demand for biobased products and agricultural commodities, encourage rural economic development, encourage environmentally sustainable manufacturing, and further the goal of national energy security.

Over the past 10 years, the Office of Energy Policy and New Uses has promoted biobased industries and will continue to accelerate the initiative with the new federal procurement mandate. With a federal procurement **market estimated at $280 billion to $300 billion covering a full range of products**, this program will be a powerful tool in industry support. The **USDA can further help by aiding product testing and certification and by the alignment of core farm-support programs to encourage grower involvement** and investment throughout the biobased product value chain.

The momentum of the mature petrochemical industry, along with a history of government subsidies and/or infrastructural support, brings formidable challenges to many small biobased manufacturing companies. **The Biobased Manufacturers' Association was formed to promote excellence in the manufacturing, sale, and use of biobased products.** Its methods of support include: an Internet support super-center, industry and market education, and a uniform content seal-labeling program.

Yeast and Fungi Expression Systems for Biomass Processing

Nancy Ho (Purdue University), Mariet van der Werf (TNO Nutrition and Food Research), Pengcheng Fu (University of Hawaii)

Saccharomyces cerevisiae does not possess the natural ability to ferment xylose due to the lack of xylose reductase and xylitol dehydrogenase. **Since the conversion both of glucose and of xylose is necessary for the efficient production of ethanol** from lignocellulosic biomass, **a recombinant yeast strain was created to ferment both sugars.** Through recombinations and large-scale screenings, a stable strain was identified with the ability to produce significant concentrations of ethanol. With further modifications, high-value coproducts are possible to further aid the profitability of lignocellulosics-to-ethanol production.

The traditional approach to microbial process improvement is to appraise on a trial-and-error basis selected target genes and/or metabolites. This method is rapid, but has a low success rate. An alternative approach, called **metabolomics**—requiring no prior knowledge of the specific metabolic pathways—uses multiple analytical methods to summarize the vast array of metabolites an organism creates and uses. This method **has been successfully applied to identify 96% of the commercially available *Bacillus subtilis* metabolites**. Sophisticated software and statistical tools have been developed to significantly increase the efficacy of this method.

Systems biology represents a new paradigm in that it allows all of the DNA and RNA in a genome to be looked at simultaneously to confer relevant information, in contrast to the conventional reductionist approach that focuses on a few genes and tries to interpret data without looking at the problem holistically. **In a systems-based approach, the first step is the development of a hypothesis, followed by information acquisition and the construction of information libraries.** The information is encoded via the construction of mathematical models. Information integration consists of quantitative analysis using statistical tools, along with visualization, comparison, and hypothesis-testing. Through this system-level analysis, the information content can be evaluated for contradictory issues.

Track 3: Sustainability Issues

Reconciling Private-Sector Needs with Academic Research and Curriculum in Biotechnology

Greg Stephanopoulos (Massachusetts Institute of Technology), Michael Betenbaugh (Johns Hopkins University), Kim Ogden (University of Arizona), John Pierce (DuPont), Doug Cameron (Cargill)

Over the past 50 years, the foundation for the biobased economy has been set, with physics and chemistry research benefiting biology. Academia needs to focus on educating people at all levels to ensure biotech/pharma's future. The academia/industry disconnect needs to be closed by refocusing bioprocessing research **to emphasize not only process optimization but also discovery of new processes** to strengthen the utilization of cells in processing. The **Society for Biological Engineering founded within the Society for Chemical Engineering offers a possible link to help reinvigorate industry-academia research collaboration**.

A decade ago, industry employment for BS chemical engineers was broken down to 45% chemicals and <3% biotechnology. Recently the industry employment ratio dropped to 31% chemicals, while biotechnology has taken off to account for >13%. **With biomedical engineering job growth projected to increase 26%**

by 2012 and chemical engineering projected to stay stagnant, current chemical engineering programs should include not only chemistry, physics, and mathematics, but increase the connection of their curricula to the biological sciences, including biochemistry, biology, and biophysics.

A solution for the academia/industry disconnect is illustrated by the University of Arizona's cooperation with the semiconductor industry benefiting their computer science students. **Courses co-taught by industry professionals, student internships in relevant industrial settings, weekly student presentations to their primary investigators and industrial mentors all contribute to preparing students for their careers.** The program also encourages new directions for student creativity for fueling academic R&D.

A common problem with academic industrial processing research is the feasibility of applying the research to industry. Research should focus on processes that have a legitimate commercial path as industry moves further into a multidisciplinary environment.

Cargill prefers to hire students with strong fundamental scientific understanding, excellent teamwork skills, and—most importantly—the ability to learn. **Industry is best at process optimization, but student understanding of patents and intellectual property is extremely important as 80% of biotechnological research is never published.** These are important factors for academia to consider when designing programs.

Biotechnology as an Enabler of Sustainability for the Chemical Industry

Darryl Banks, Dave Sherman (Sustainable Value Partners), Iain Gillespie (Organization of Economic Cooperation and Development), Jim Stoppert (Cargill)

The key element for sustainable manufacturing is to progress to technologies that are energy-efficient. Finding the solutions to this is the opportunity and challenge the biotechnology industry faces for the development of a sustainable chemical industry.

The chemical industry is challenged by stagnant financial returns and access to world capital markets, and societal expectations for sustainability need to be addressed. Corporations must consider impact on all stakeholders when making decisions, particularly in the face of globalization. **Biotechnology is a disruptive development unsought by the chemical industry, but which can greatly contribute to increasing value by addressing issues that concern all stakeholders, from the environment to process efficiency.**

In twenty-one OECD case studies, successful acceptance by industry of bioprocesses was consistently associated with decreased adverse environmental impact, and increased cost-efficiency. New skills need to be acquired in the adoption of a biotech process and partnerships with academia, consulting firms, or other companies will expedite adjustment and contribute to triple bottom line fulfillment.

The adoption of new tools needed to integrate new biotechnology with industry can be facilitated through government funding and partnerships. Biotech needs to be addressed as a disruptive innovation. It is important to avoid over-hyping its potential; recommendation of what it can do for industry must be realistic.

Environmental and Social Impacts of a Large-Scale Biorefining and Bioprocessing Industry

Robert Anex (Iowa State University), Bruce Dale (Michigan State University), John Sheehan (National Renewable Energy Laboratory)

All past economies since Mesopotamia have been biobased. Modern engineering capabilities now foster new uses of biomass that present the possibility of a return to a biobased economy and a sustainable society. **We do not understand all the complex issues of water supply, feedstock supply, petrochemical supply, and biotechnology's impacts on these resources; this is an important area for ecological study to assist in directing the rapid pace of biotechnological development.**

Biomass is inexpensive versus petroleum, but processing costs are high. **Biobased industry will boom with the development of efficient sugar-conversion technology.** Focus is needed on the best ways to utilize biomass on local geographical bases as a key component of the development of industrial bioprocessing.

Issues of environmental policy, ethics, social science, and biology need to be addressed when

conducting life-cycle analyses of energy sources. We must work towards understanding more of the technical impacts involved in biomass life cycle analysis. Educating not only industry but also the public will be needed for full discussion of the issues, to work together toward energy-sustainability.

Environmental Benefits of Industrial Biotechnology

Chris Hessler (AJW, Inc.), Peter Chant (FReMCo Corporation, Inc.), George Sugiyama (Dorsey Whitney)

The OECD case study (mentioned above) of twenty-one successful biotech-process integrations determined that each was environmentally friendlier than the process it replaced. **A new report by AJW, Inc., will address what pollution-prevention biotech can contribute if industry adopts it, along with upstream benefits from biotech processes.** It is important to engage all stakeholders in the industrial processes and build relationships with them to avoid GMO-type perception problems in industrial biotechnology.

Unbeknownst to many people and corporations, emission reduction credit (ERC) trading is a lucrative business. ERCs can be created by being able to show that emission reductions are quantifiable, verifiable, and that there is a real surplus during the manufacture, use, or disposal of a product. Even if the product costs more to produce than a less environmentally friendly process, the profit margin will often be equal or greater than the previous process when ERCs are included. In some cases, a product can even be given away, and the return from the ERC will provide a profit.

Biotechnology provides a great deal of promise for the **emerging and growing markets for biofuels**. However, as it progresses, **industry has to be aware of, and must confront, regulatory and legal matters**. Developing a biofuels infrastructure is fraught with difficulties, and we cannot simply scrap the current petrochemical infrastructure. Final commercialization of biotechnology will require close attention to the regulatory structure for all fields it operates in, which will be a significant challenge.

Harnessing Microbial Genomes to Address Climate Change

Jae Edmonds (Pacific Northwest National Laboratory), Mike Himmel (National Renewable Energy Laboratory), Karin Remington (Center for the Advancement of Genomics and Institute for Biological Energy Alternatives)

Addressing climate change is a long-term issue and large-scale solutions await many R&D breakthroughs. **Biotechnology could provide a broad suite of energy services from biofuel feedstocks to hydrogen production for fuel cells.** The combination of fuel cells and biotechnology solutions could make significant energy contributions, but for large-scale deployment to occur, we need to find more effective solutions, which microbial genomes may offer.

Approximately 360 glycosyl hydrolases—the main class of enzymes that break down cellulosic material—have been identified. However, cellulose is resistant to microbial degradation, and no superior enzymes or systems have been identified for its metabolism. **The DOE's Genomics:GTL program offers the best opportunity yet, using microbial genomes, to discover a system for efficiently breaking down cellulose to be utilized for biofuel synthesis.**

With over 1.28 million genes representing an estimated 1,800 new microbial species found from the first seawater sampling tour in the nutrient-poor Sargasso Sea, **the oceans promise gene discoveries that can be utilized for bioremediation and carbon sequestration among many of the possible applications**. The Institute for Biological Energy Alternatives has sampled the genomic makeup of seawaters from Nova Scotia to the Galapagos Islands and is now embarked on a round-the-world journey to reveal oceanic microbial diversity and possibly find new genetic tools for environmental sustainability.

Bioethics of Industrial Biotechnology

Mark Saner (Institute on Governance), David Castle (University of Guelph), Eric Mathur (Diversa)

Sustainability is desired by the public, politicians, industry, and NGOs. This unusual consensus provides a rare opportunity for biotechnology. **The biotechnology industry should be open**

to discussion and willing to address concerns of all sectors of society in order to operate in an effective triple bottom line manner. There is also the ethical voice that knows that we *can* engineer nature, but discussion should be opened in terms of *should* we engineer nature, and if so, how?

It is important to understand the ecological footprint that the cultural world has on the biotic world. Ecological studies measure the per capita "footprint" in hectares required to sustain a person's energy consumption per year. Biotechnology needs to be analyzed in these terms as to the benefits it provides for the world population as a whole, and questioned if it is truly a solution or if it only will create new problems. One such concern is the economic impact of biopiracy, as the most abundant resources are in regions of lowest socioeconomic status. Addressing such issues and applying distributional justice to problems like biopiracy are important for the socio-political success of the biotechnology industry.

Diversa provides a model for ethical bioprospecting. The corporation supports programs worldwide with organizations such as the National Institute of Biodiversity in Costa Rica to aiding PhD programs in Kenya. **It is important to support equitable benefit-sharing, from monetary donations to technology-capacity building** that contributes to people's understanding of biodiversity in their localities. Diversa has established relationships with governments and institutions worldwide to progress with biotic discovery and show that the environment has economic value.

Industrial Biotechnology Applications to Mitigate Greenhouse-Gas Emissions

Ranjini Chatterjee (Codexis), Mark Stowers (Michigan Biotechnology Institute), Robert Tabita, (Ohio State University)

Studies of cyanobacteria revealed that ribulose-bisphosphate carboxylase (rubisco) is the bottleneck in CO_2 fixation. **An engineered form of rubisco had a two-fold increase in rate of CO_2 fixation.** The carboxylation kinetics of rubisco were improved by DNA shuffling, which demonstrates that microbial systems can be manipulated for carbon sequestration and utilization.

Cellulose can be applied to many materials, such as nanofibers to replace fiberglass, which are safer to handle, biodegradable, and lightweight. New products can be made from succinic acid and novel polymers. The succinic acid market is limited now (~$2.5 million/yr), but as production costs decrease it will be more widely used and will contribute to numerous products while CO_2 is fixed, decreasing greenhouse gases.

Ribulose bisphosphate (RuBP) is a key acceptor of electrons in CO_2 fixation, and **mutant proteins have been isolated that confer increased affinity of rubisco for RuBP**. Manipulation of these genes has enabled researchers to engineer aerobic autotrophic microbes to increase their metabolism using CO_2 as the sole carbon source. This offers the possibility of engineering a transcription regulatory protein so that CO_2-fixing genes are always turned on, thereby increasing the carbon-sequestering capabilities of microbes.

Biomass Utilization and Ethanol Production: Mitigation of Greenhouse-Gas Emissions

David Pollock (BIOCAP), Jim Hettenhaus (cea, Inc.), Steve Eule (US Department of Energy)

BIOCAP is building a network encompassing federal and provincial Canadian governments, industry, academia, and NGOs to find least-cost, scientifically credible solutions to renewable energy consistent with Canadian policy. Results from laboratory research are being applied to national infrastructure as efficiently as possible. Forestry and natural ecosystems, agriculture, and biobased product sectors are working together to achieve the goal of overlaying natural science research with social policy for a sustainable energy future.

Soil quality is directly related to tillage, as tilling exposes the anaerobic subsurface to air and soil carbon is released as CO_2. If nitrogen is not added with plowed residues, an N deficiency may occur decreasing the next season's crop yield. By not tilling, there is less requirement for fertilization leading to better conditions for surrounding watersheds. **By converting 30% of corn stover to ethanol, the United States could reduce its greenhouse-gas emissions enough to meet 5% to 10% of the total offset required** to achieve the goals set out in the Kyoto Agreement.

Eighty percent of greenhouse-gas emissions are energy related, and emissions are increasing at a rapid rate. The solution to reducing such emissions is a century-long process, but the US government already supports development of an array of energy technologies from hydrogen production for fuel cells, biodiesel, and other renewable energy sources. An objective of DOE's Genomes to Life Program is to produce cellulases that are efficient enough to be used for large-scale industrial production of ethanol to significantly contribute to the country's energy supply.

Life Cycle Assessment and Sustainability of Bioprocesses

Robert Anex (Iowa State University), John Sheehan (National Renewable Energy Laboratory),
Albert Chan (National Research Council of Canada)

1, 3-propanediol (PDO) is used in producing solvents, adhesives, and laminates. In the PDO lifecycle, most of the fossil energy consumed is in the PDO production. **Biobased PDO production by fermentation of corn may be preferable and more environmentally beneficial than the petrochemical ethylene oxide pathway**, by reducing net energy requirements for production from 56.9 MJ/kg to 37.1 MJ/kg. Further analysis of competing methods will help determine which synthetic pathway is the most energy-efficient.

Life-cycle analysis (LCA) provides an comprehensive estimate of closeness to sustainability. LCA **modeling was applied to the entire state of Iowa for sustainable ethanol-production potential from corn stover.** If farmers used no-till cropping, the state would have significant ethanol-production potential. **The usage of bioethanol produced from corn stover has the potential to decrease coal consumption by twelve-fold, fossil-energy consumption by 102%, and natural gas usage by 200% in some localities.**

When conducting LCAs, it is important to have a combination of approaches by using systems modeling that looks at the long-term impact of a production process and not just at immediate production impacts.

Track 4: Novel Applications

European Advances in Marine Biotechnology for Food, Pharmaceuticals and Energy

Rene Wijffels (University of Wageningen), Detmer Sipkema (University of Wageningen), Peter Lindblad (University of Uppsala)

Large-scale "milking of microalgae" is done in ponds in Australia of up to 50 ha, for the production of beta-carotene. The productivity of such ponds is relatively low, therefore effort is being expended on the development of *in-vitro* systems. Using a biocompatible organic phase, *Dunaliella salina* can be cultured for relatively long periods with continuous synthesis and removal of beta-carotene in the organic phase, with significant improvement in carotenoid production over commonly used systems.

Because of the increasing problem of salinization of water in the Netherlands, ocean farming is being researched: the objective is to grow algae in saline water as feed for fish. The fish, in turn, are being "redesigned" to accept a vegetarian diet.

The lowly sponge is a source of over 5,000 unique compounds with potential uses as anti-viral, -tumor, -biotic, -flammatory agents. The potential for cultivating sponges (*Lissodendoryx* sp. and *Dysidea avara*) is being investigated as sources of halichondrin B (anticancer) and avarol (antipsoriasis), respectively. Although sponge growth rates can be increased sevenfold and seasonality eliminated in *ex-situ* culture, production costs of halichondrin B remain prohibitive. Avarol is more promising. A bacterial symbiont may be responsible for the synthesis of halichondrin B, in which case an alternative means of production may be possible. In the future, the oceans may assume the role of the rain forests, as potential sources of bioactive compounds. Sponges are a particularly fruitful source of compounds; being sessile, they have accumulated many chemical means of self-defense.

The production of hydrogen by splitting water using solar radiation is possible with

photosynthetic microorganisms. The development of reactors to produce bio-hydrogen is in the early research phase. Under the auspices of several international collaborative efforts, photobiologic and fermentative systems are under study, including the use of cyanobacteria that produce hydrogen as a by-product of nitrogen fixation. Another approach is to make the active site of photosystem II more efficient in the harnessing of protons. Although the current efficiency of bioreactors is low, significant improvements are expected over the next 20 to 40 years.

Microorganisms for Direct Energy Production

John Benemann (Institute for Environmental Management), K.T. Shanmugam (University of Florida), Juergen Polle (Brooklyn College)

The photobiological production of hydrogen is the "holy grail" of hydrogen production: photosynthesis with the hydrogenase deactivated. A major problem lies in keeping the oxygen separate from the reductant. Also, **photobioreactors are costly**.

Hydrogen is a by-product of cyanobacterial nitrogen fixation with light as the energy source. **Hydrogen may also be produced as a result of treatment of ethanol with heat over a catalyst.** The ethanol is produced by yeast provided with glucose produced by microbial degradation of lignocellulose.

Microalgae are attractive organisms: they are fast-growing and produce valuable commodities including biofuels and biomass. However, light harvesting in ponds is inefficient: cells on the surface are photo-inhibited—with 80 to 90% of the energy dissipated as heat—shading those below. Mutant strains are being developed with fewer antenna chlorophyll molecules. **It is expected that the current conversion of 2 to 3% of solar energy into biomass can be increased to 4 to 6%.**

Industrial Biotechnology and Bioprocessing for National Defense Applications

Rosemarie Szostak (Defense Advanced Research Projects Agency), James Valdes (Army Research and Development and Engineering Command), Harold Bright (Office of Naval Research), Michael Ladisch (Purdue University)

The Defense Advanced Research Projects Agency (DARPA) provides funds for universities and industry, and applies inventions and discoveries to improved defense capabilities, *e.g.* new materials for protection of soldiers on the battlefield. NASA and the Internet are spins-off. A major-risk/major pay-off approach is taken. Soldiers' waste packaging may in the future be used to generate energy on the battlefield; bioplastics may have greater fuel value than plastics from petrochemicals.

The Biotechnology Working Group of the US Army is a multidisciplinary entity that examines biotechnological approaches to solving military problems. There is interest in developing—*inter alia*—biological obscurants and using biological systems for detection of low-level toxicants.

The Office of Naval Research biomimetics program includes work on agile biosensors, energy harvesting, next-generation antibodies, and the green synthesis of energy sources. A biofuel cell has been developed, <1 mm^3 in volume and 5 μW in power, that can be placed inside a trained bee to detect explosives. Butanetriol can be synthesized by microorganisms from xylose and arabinose as a safer, more environmentally friendly alternative to nitroglycerin.

The possibility of microbial conversion of waste materials—food, human waste, and packaging—into ethanol for use by expedtionary forces is being examined. Conversion of these materials into electricity is also under study using biofuel cells. The electricity would be used to run sensors, *etc*. The concept is "add water and go."

Biotechnology at the Nano- and Micro-Scales for Drug Discovery and Functional Materials

Jonathan Dordick (Rensselaer Polytechnic Institute), Ping Wang (University of Akron), Alan Russell (University of Pittsburg)

Nanotechnology offers many new approaches to improving biocatalysis. Enzymes can be attached to single-walled nanotubes of 1 nm diameter and to multi-walled nanotubes of 20 to 40 nm diameter to generate biocatalytically active surfaces. The nanotubes are mechanically strong and are protective of proteins in the presence of chemical denaturants. Tube curvature stabilizes the enzyme, similar to what occurs inside cells. Embedded in a porous polymer, nanotubes can detoxify air of nerve-gas agents for example.

A critical issue in drug discovery is that high-throughput methods are missing for checking for toxicity to humans, for example. **A metabolizing enzyme toxicology assay chip (MetaChip) has been developed as a micro-scale alternative to *in vitro* screening methods.**

Silica glass nanoparticles contain pores of approximately 15 nm in diameter, large enough to accommodate a protein molecule, providing multiple attachment points that lend stability. **The half-life of glass-bound chymotrypsin, for example, is >1,000-fold longer than that of the native enzyme.** Binding of multi-enzyme complexes is possible, *e.g.* for the production of methanol from carbon dioxide, with regeneration of the NADH cofactor within the particle. Nanofibers are similar to nanoparticles in having a huge surface area with the advantage of easier regeneration. Beta-galactosidase conjugated to hydrophobic polystyrene particles had activity >100-fold higher at an organic/aqueous interface than the native enzyme in the aqueous phase.

There is need to learn how to build nanostructures to mimic those in biological systems. Biological components may be added to nanostructured materials to respond, for example, to toxins or pathogens by changing color. Polydiacetylene (PDA) sensors have been manufactured to change color in the presence of virus particles. Self-assembling PDA nanotubes can be manipulated for 100% uniformity in diameter. These adhere to the surface of bacterial cells, penetrate the outer membranes and are bactericidal. A single nanotube is lethal to a cell of *E. coli*.

Microbial Production Systems for Value Added Chemicals

Nicholas Ballor (Michigan Technology University), Regine Behr (Novozymes), Jan de Bont (TNO Environment, Energy and Process Innovation)

Ferric iron is an excellent oxidizing agent used for scrap recycling, wastewater treatment, *etc.* Current production methods entail boiling sulphuric acid, bubbling chlorine, and hyperbaric oxygen. The extremophile bacterium *Acidithiobacillus ferrooxidans* provides an attractive alternative; it uses ferrous iron as an energy source, is functional at 30°C and pH 2.0, and uses CO_2 as a source of carbon. **A stand-alone automated bioreactor for ferric iron generation (SAABFIG) has been developed, with low operating costs, requiring low capital investment, and modest operating conditions.** Using the SAABFIG bioreactor for scrap-iron processing, copper is captured and removed (otherwise steel production is compromised) and ferrous iron is recyclable.

Hyaluronic acid (HA) is a lubricant polysaccharide present in the capsules of some bacteria (*e.g. Streptococcus* spp.) that has many applications in cosmetics and pharmaceuticals including osteoarthritis treatment. Rooster combs and certain strains of *Streptococcus*, the current sources of HA, are less than ideal because of purification needs and other problems. ***Bacillus subtilis*, a non-fastidious bacterium used for production of many industrial enzymes, has been genetically engineered to secrete large amounts of high molecular weight HA, representing a new competitive system for large-scale synthesis of HA.**

Pseudomonas putida was chosen as a vehicle for synthesis of aromatic hydrocarbons that are toxic to most microorganisms because it is a versatile organism of known genomic sequence. **A solvent-tolerant strain** (the tolerance mechanism of which is poorly understood) **of *P. putida* was mutagenized and screened for overproduction of phenol from glucose as proof of principle.** Other aromatic compounds, alcohols, epoxides, terpenoids, *etc.* may be similarly produced by introduction of appropriate heterologous genes.

Industrial Biotech Applications in the Pulp and Paper Industry

Tim Presnell (MeadWestvaco), Geoff Hazlewood (Diversa), Art Ragauskas (Georgia Institute of Technology)

Xylanases are already used in pulp production to mitigate bleaching costs and pollutant production. Many opportunities for further enzyme use are under study, for even less chemical usage, less energy consumption, effluent amelioration, machine cleaning, de-inking for paper recycling, for improved lignin degradation, modification of fiber for improved paper quality, *etc*. Rejected wood chips and sawdust may be converted to value-added products like ethanol and levulinic acid by enzyme action. **Laccases and peroxidases hold promise for oxidation of lignin, lipases for**

pitch reduction, cellulases for cell-wall depolymerization, and pectinases for depolymerization of pectic polysaccharides. A search for improved xylanases from many locations globally resulted in more than 190 novel enzymes, some of which are effective at higher and lower pH values, and are effective with hard and soft woods with 22% less need for chlorine dioxide.

Industrial Biotechnology Applications for Fuel Cell Development

Michael Toney (University of California, Davis), Nick Akers and Shelley D. Minteer (St. Louis University), Bruce Logan (Pennsylvania State University), Robert Coughlin (University of Connecticut)

Biofuel cells, conceived in the 1960s, differ from traditional fuel cells in the catalyst used for oxidation of the fuel. Rather than a non-renewable precious metal, early types used live bacteria and lasted only up to 3 hours. **With polymerimmobilized enzymes, a 6-month longevity is now possible.** Highly selective enzymes are now available, able to utilize a variety of fuels. In ethanol fuel cells, alcohol dehydrogenase is immobilized in quaternary ammonium salt-treated Nafion membrane; NADH is regenerated within the membrane pores. Although the NAD is gradually degraded (over 45 days), it can be added back.

Microbial fuel cells (MFCs) offer a new approach to wastewater treatment. Thus, energy may be generated from a free "resource" that otherwise costs money to manage. Bacteria in the wastewater form a stable biofilm at the anode and transfer electrons obtained from the oxidation of the organic matter under anaerobiosis. It is possible to generate up to 150 mW/m^2 with 80% removal of the biochemical oxygen demand. **Approximately 1,000 mW/m^2 is thought to be achievable, in which case, wastewater from 100,000 people may be sufficient to power 300 homes.** It may be possible to link biohydrogen synthesis with electricity production for maximum capture of energy from wastewater, making this a practical technology.

MFCs use mediators of various kinds, which in the oxidized state must escape easily from the microbe. The mediator or the cell may be attached to the anode; if the cell is attached, a mediator may be unnecessary. **A special advantage of the MFC is fuel flexibility; electricity may be produced from acetate, lactate, succinate, glucose, ethanol, or methanol.**

Methanol and Methane as Carbon Sources for the Expression of Recombinant Proteins in Methylotrophs

Carlos Miguez (National Research Council of Canada), Colin Murrell (University of Warwick), Mary Lidstrom (University of Washington)

Methanotrophs are Gram-negative aerobic bac-teria that use methane for energy and as a carbon source. A subset of the methylotrophs that grow on single-C compounds, they are of interest as a source of single-cell protein. **Methane monooxygenase (MMO)**, the key enzyme, exists in membrane-bound and soluble forms; the latter (sMMO) **is** more versatile and **of interest for co-oxidation of alkanes, alkenes, aromatic and substituted aromatic compounds (>200 in total) with potential for biocatalysis (*e.g.* production of epoxides and methanol) and bioremediation (*e.g.* degradation of aromatic hydrocarbons).** Via gene transfer, "metabolic engineering" is now possible. New screening tools are needed to select superior sMMOs, *e.g.* for production of chiral alcohols.

Methanol is attractive as a feedstock: it is inexpensive ($226/tonne), non-corrosive, water-soluble, and renewable. ***Methylobacterium extorquens*, a methanotroph with a well characterized genome, is being developed as a model system for bioprocesses utilizing methanol.** High cell densities are possible in batch fermentation with the production of polyhydroxybutyrate. The central metabolism is being manipulated for overproduction of desired proteins and other compounds; stable vectors and efficient promoters have been identified, and green fluorescent protein (encoded on a plasmid) is being used to assay the effects of specific selection pressures.

Workshops

Positive and Negative Impacts of Agriculture Feedstock Utilization

Moderator: Kevin D. Kephart (South Dakota State University)

Recorder: Vance N. Owens (South Dakota State University)

In this two-session workshop, the group—representing various aspects of feedstock utilization—was charged with identifying and evaluating key issues involved with the use of agricultural feedstocks in large-scale biorefineries for biobased products. During the first session, more than twenty issues associated with agricultural feedstocks were identified, including production, effects on the environment and soil quality, how to include feedstock producers, and potential for research funding. During the second session, key issues, encompassing many of those identified in the first session, were discussed in detail.

Issue: Environmental Impact

The use of agricultural feedstocks in a biobased economy may have positive or negative effects on the environment. By consensus, it was concluded that any assessment of the use of agricultural feedstocks should be on a global scale, particularly as their use relates to the reduction in greenhouse-gas emissions and to global agricultural sustainability. However, it was recognized, as Bruce Dale (Michigan State University) suggested in an earlier breakout session, that "all biomass is local." Therefore, while the use of agricultural feedstocks is a global issue, it is critical that research, sustainability, producer involvement, and other factors associated with agricultural feedstock use be considered from local, national, and global perspectives.

Soil quality (fertility, organic matter content, tilth, particle aggregation, *etc.*) is a primary *vis-à-vis* production of agricultural feedstocks. Research has demonstrated, and producers in many countries have adopted, management techniques to improve soil quality. Soil erosion has been reduced and organic matter content increased on many acres in the United States through the Conservation Reserve Program (CRP). A significant number of CRP acres are reverted to annual crop production each year, and cultivation of these acres may dramatically reduce the environmental and conservation benefits gained during enrollment. Wise use of the species currently grown in CRP or use of reduced or no-till practices when converting to traditional crops, both of which may be used for bioenergy production, will help alleviate this concern.

Diverse species and types of agricultural feedstocks may be used as bioenergy crops including corn stover, wheat straw, perennial grasses, and poplar. By its very nature, this diversity helps reduce negative environmental impacts while improving long-term sustainability of various systems.

Recommendation Agricultural feedstock use may affect not only the local but also the global environment. However, the lack of representation from environmental groups was noted. Therefore, it was recommended that, for future meetings, greater effort should be made to include representatives from environmental organizations.

Issue: Public-Private Partnerships

In order to better understand the effects of agricultural feedstock use for bioenergy, vested interests must be present at the producer, public, and private levels. Agricultural feedstock research efforts must continue to:

- evaluate best-management practices for improved yield and sustainability,
- maintain breeding programs in which locally adapted species and cultivars are developed and tested, and
- determine the best methods for use of biotechnology.

As evidenced by attendance at this meeting, interest from the public and private sectors appears to be high. Partnerships between the public sector and private industry, however, must include feedstock producers.

Recommendations Producers must be included in all aspects of the development and use of biobased agricultural feedstocks. Producers must also capture a substantial proportion of the value generated by this new industry. Because they will supply the bulk of agricultural feedstocks, they may be most capable of providing public and industry personnel with insights into the challenges facing a large-scale agricultural feedstock industry. To enable potentially synergistic advances in knowledge and understanding of agricultural feedstock use, personnel at public institutions should continue to be encouraged to partner with private industry. Funding for research to improve our knowledge of agricultural feedstock utilization should be among the top priorities of public institutions.

Issue: Feedstock Diversity

Although agricultural feedstocks are abundantly available, the optimum methods required to harvest, store, and transport biomass crops must be ascertained. In addition, considering that feedstocks may come from crop residues, herbaceous plants, forest products, oilseeds, grain crops, and manure, supply and quality may be inconsistent. Quality and supply may also vary from year to year, by environment, by harvest and storage method, and region of origin. Diversity of feedstocks provides one method to manage risk and allows growers to produce crops best adapted to local conditions. The current biobased infrastructure largely supports feedstocks from which a consistent source of starch is the primary resource, however. Infrastructure and methods for processing and converting lignocellulosic biomass into biobased products are not as common or well defined. Therefore, in order to efficiently utilize diverse feedstocks, robust biomass conversion technology must be developed and tested on a large scale.

The need for a list of traits desirable for biobased agricultural feedstocks was discussed. For example, developing feedstocks with lower levels of lignin and protein while maintaining or increasing total yield and desirable structural carbohydrates would benefit many of the proposed conversion processes. Potential diseases and other pests must be studied, particularly if large areas are to be dedicated to biomass crops. Because of the diversity of feedstocks and the limited knowledge related to pests of some of them, breeders and geneticists should be aware of the potential pests and desired traits in their effort to develop cultivars adapted to the local environment and to the harvest, storage, and transportation methods available.

Recommendations Public institutions and private industry must increase research efforts and funding to address harvest, storage, and transportation of agricultural biomass. It was recommended that a committee be named to help elucidate the most desirable genetic and quality traits of various agricultural feedstocks. This committee should communicate this information to personnel in the public and private sectors to enhance resources allocated to improving agricultural feedstocks.

Issue: Policy

The use of agricultural feedstocks directly and indirectly affects current public policy and the development of future policies. Expanded development of biomass energy can help facilitate reduced dependence on foreign energy and improved national security.

Development of a biobased infrastructure may have dramatic positive effects on rural economies through creation of jobs, increased value of agricultural products, and ability to add value to existing agriculture crops or crop residues. Agricultural feedstock development may impact federal farm subsidies, but financial decisions related to the use of agricultural feedstocks should be made assuming absence of farm subsidy.

Recommendations A coalition representing producers, public institutions, private industry, and environmental organizations should be established to address the numerous public-policy issues. This coalition should work with personnel from state economic development offices and USDA Rural Development to encourage policy development for rural economies. This coalition should initiate a meeting in the near future to begin discussions regarding agricultural feedstock use.

Conclusions

To be successful, further efforts toward agricultural feedstock utilization must provide a method for greater inclusion of producers at all levels. In addition, producers, representatives from environmental groups, and personnel from the private and public sectors must develop and maintain excellent communication channels in order to optimize potential benefits and avoid possible pitfalls in the utilization of agricultural feedstocks.

If correctly implemented, biomass systems can provide reliable means of energy production with concomitant environmental and social benefits, including enhanced conservation of natural resources, agricultural sustainability, enhanced rural economies, and improved national energy security. Areas of emphasis for feedstock-production programs should include sustainability, efficient harvest and transportation systems, and marketing to provide incentive and opportunity for producers.

State and Federal Funding Opportunities for Biomass Energy and Biomass Projects

Moderator: Richard Powers (Dorsey and Whitney LLP)
Recorder: William Gibbons (South Dakota State University

Five presentations were made, describing federal, state, and private sources of funding that may be leveraged to support research and commercialization of biobased products. A short Q&A session followed each, with general discussion after the final presentation.

Douglas Kaempf [Biomass Programs Manager for the US Department of Energy (DOE) [www.doe.gov] provided an overview of funding opportunities provided by his as well as other agencies. One DOE program, directed at the state level, is Deployment of Clean Energy Technologies, funded at $16.5 million. In 2004, this formula funding focused on biobased energy. DOE also supports an "ear-marked" Regional Biomass Program to support state priorities in energy.

Like other federal agencies, DOE participates in the Small Business Innovative Research program (SBIR, www.science.doe.gov/sbir); $95 million were available in 2004 for competitive grants to help small companies meet their R&D needs. Phase I awards provide up to $100,000 for 9 months for feasibility analysis. Phase II awards are for 2 years for up to $750,000.

The DOE's Office of Biomass Programs also offers competitive grants for research priorities listed in the multi-year technical plan; total dollar amount depends on annual appropriations (http://devafdc.nrel.gov/biogeneral/Program_Review/MYTP.pdf). Over the past 3 years, the Office of Biomass Programs has awarded $10–12 million/year through this program. In addition, DOE's Office of Science releases solicitations on fundamental R&D-related research in key targeted areas (www.sc.doe.gov). Currently, the focus is on plant genomes and cellulose biosynthesis.

DOE and the US Department of Agriculture (USDA) are in the second round of a joint solicitation on bioproducts and bioenergy, authorized by the Bio R&D Act of 2000, with $24 million of funding (dependent on appropriations, www.bioproducts-bioenergy.gov). DOE and USDA have developed roadmaps to help direct this research and have formed committees of industrialists, government representatives, researchers, and environmentalists to help focus these efforts.

Mr. Kaempf provided a brief update on programs of the National Science Foundation (NSF) related to the biobased economy (www.nsf.gov). Current programs (and funding) include: Biochemical Engineering ($15 million/year), Materials Use ($ 5million/year), Plant Genome ($13 million), and Functional Genomics ($3 million).

Frank Flora, Chair of the Biobased Products and Bioenergy Coordination Council (USDA, www.usda.gov) expanded on the USDA's programs, which range from research and extension to support for industry and commodity groups through special grants and Commodity Credit Corporation programs. Two new programs were included in the 2002 Farm Bill. Section 9002 authorized the Federal Procurement of Biobased Products program (funded at $1 million/year), while Section 9006 authorized the Biodiesel Fuel Education program ($1 million/year) and the Renewable Energy Systems and Energy Efficiency Improvements program ($23 million/year). The Bio R&D Act of 2000 (see above) directed the USDA to contribute $14 million annually to the joint DOE/USDA bioproducts program and authorized the USDA to contribute up to $150 million annually to producer payments for expanded biomass utilization. The Healthy Forest Restoration Act provides $5 million/year to expand utilization of small-diameter woody products for bioenergy and bioproducts. The USDA SBIR program contains various sections specifically directed to biobased products and energy. USDA programs can be explored at www.usda.gov/edcc.

Alison Schumacher is Director for the Clean Energy Group (CEG, www.cleanegroup.org), a non-governmental organization that works to accelerate commercialization of clean energy technologies through advocacy, education, and partnerships with governments and private entitites. CEG works with states to more effectively use their Clean Energy Fund programs to develop niche renewable energy markets and foster climate-change action at the state level. Over the next decade, states will invest $3 billion in clean-energy technologies, with funding largely from systems benefits charges (utility settlement charges). State funds are typically used for grants, production/tax credits, green tags, loan programs, equity, and commercial financing programs. CEG will assist in a broad range of renewable energy projects, including: solar, wind, fuel cells, biomass, green buildings, energy efficiency, public education, strategic market research, niche markets, green power, and business development. In recent years, states have solicited proposals in the specific areas of biomass gasification and cofiring, land-fill methane, anaerobic digestion, wood-waste combined heat/power, biofuels for fuel cells, and integrated biomass gasification with fuel cells.

In summary, states are active players in innovative clean-energy deployment activity, and opportunities for partnerships focusing on technology, finance, and deployment matters are enormous. Links to various state programs are available at the CEG Web-site.

David Kolsrud (david@forfarmers.coop), a developer/consultant/farmer from Beaver Creek, MN, provided insight based on 10 years experience in developing farmer-owned cooperatives—from conception to completion—that have invested in value-added projects: ethanol production from corn, soybean crushing/biodiesel production, and wind-energy powered farms. Thirty steps are involved in the development process. He outlined a feasibility study approach for obtaining value-added development grants for preparing winning business plans and establishing successful value-added businesses, and briefly described his New Generation Cooperatives (NGCs) business model for farmer-owned cooperatives.

Bill Holmberg, Chair of the New Uses Council (www.newuses.org), discussed the formation of the Biomass Coordinating Council (BCC), which will serve as the operating arm for biomass for the American Coalition on Renewable Energy (ACORE). BCC's mission is to:

- provide encouragement and support to those involved in biobased products,
- promote optimization of energy efficiency, environmental enhancement, and long-term sustainability of biobased industry,
- gain public and governmental recognition and support for biobased industry,
- invite participation of industries and associations in BCC,
- develop liaison relationships with international biomass companies and organizations, and
- provide a registry of highly qualified consultants committed to the mission and goals of BCC.

The BCC will assist in advancing the full scope of the biomass industry, including preservation of soil quality and the full spectrum of biomass resources, the promotion of all industries involved in production of biobased products, and optimization of energy-efficiency and life-cycle benefits (biorefineries@aol.com).

Clearly, many different types and sources of funding are available for R&D on biobased products, and the list is expanding. There is a need to demonstrate that biobased products have benefits for society as a whole. There is a need also to inform federal and state appropriators and federal and state regulatory agencies—including DOE and USDA—on a continuous basis so that when projects and programs are being formulated and funded, the bio community is included.

Doing Business with the Department of Defense

Moderator: Jerry Warner (Defense Life Sciences)
Recorder: Vance Owens (South Dakota State University)

Increasingly, the Department of Defense (DOD) is turning to industrial biotechnology for solutions to problems. DOD is interested in the development of biobatteries, portable biorefineries, enzymes for chemical and biological weapon decontamination, new biopolymers and bioplastics for clothing, sleeping bags, meal packaging and utensils, and other technologies. Opportunities for biotech companies and researchers and details of how to work with DOD were the primary topics discussed.

Issue: Obtaining Funds from DOD

DOD is a large organization with an imposing bureaucracy. The following are key points for obtain grant funds:

DOD extends formal request for proposals (RFPs) as well as Broad Agency Announcements under the Federal Acquisition Regulation. Unlike other federal agencies, DOD is a customer for successful research; it consumes significant volumes of goods and services. Its budget for FY05 will be almost $400 billion.

The Small Business Innovative Research/Small Business Technology Transfer (SBIR/STTR) program Web-site (www.acq.osd.mil/sadbu/sbir) provides guidance on potential research and partnering.

Program managers should be consulted to determine DOD's potential interest in a specific research concept.

Recommendation To be of interest to DOD, proposed research should include a commercialization plan. Even basic research at an academic institution will be more favorably viewed if an industrial partner is involved.

Issue: What Traditional Procurement Methods and Public/Private Partnerships are Available?

Funding from DOD typically comes via one of the following:

- ♦ Traditional procurement methods
    - a. Federal Acquisition Regulation (FAR) contracts
        - I. For FAR contracts, the prime contractor must be registered (www.ccr.dlis.dla.mil) and in compliance with various rules.
        - II. For individuals/institutions/industries with no prior experience with DOD, it is recommended that the first award be obtained as a subcontractor rather than as the prime contractor.
        - III. Opportunities for FAR contracts may be found at www.fedbizops.gov.
    - b. SBIR, STTR, and Fast Track
        - I. SBIR provides up to $850,000 to small businesses for R&D.
        - II. STTR provides up to $850,000 per project for R&D to university/small-business teams.
        - III. Fast Track is a method of increasing the chance of obtaining SBIR or STTR awards and of obtaining continuous funding for small businesses that can attract outside investors.
- ♦ Non-traditional public/private partnerships, such as
    - a. Cooperative R&D agreements (CRADAs).
    - b. Patent licence agreements (PLAs).
    - c. Transportation Security Administration (TSA).
    - d. Cooperative agreements, enhanced use leases and other R&D transactions.

Overcoming Barriers to Growing a Biobased Economy

Moderator: James Hettenhaus (CEAssist)

Recorder: Padu Krishnan (South Dakota State University)

Barriers to growing a biobased economy include lack of technology validation, need for training, lack of available feedstocks, public perception/acceptance, government policies and the need to engage stakeholders in the agricultural community. Farmers' concerns in terms of issues and benefits must be addressed. Only conflicting, anecdotal information is available. A transparent information base is needed. There is also need for a "better" business model for the farmer.

Issue: Feedstocks

The magnitude of scale involved in the collection of feedstocks needs to be appreciated. The agricultural sector continues to be focused on the production of food, feed and fiber. Crops have not been bred specifically for biomass; bioenergy represents a significant shift in emphasis. The challenge is how to shift beyond food crops to energy crops.

Additional factors worthy of consideration include uncertainty in the energy market, problems inherent in pioneering activities, and the age-demographics of farmers. Questions exist on sustainability and environmental impact (no till, soil moisture, erosion, need for cover crops, topography and soil compaction). Storage stability of biomass must be addressed; product specifications will need to be standardized.

Farmers will need stable governmental agricultural policies. Studies will be required on equipment needs and tools for assessing sustainable biomass production and collection.

Issue: Business Models

Biomass production in significant amounts will require a long-term commitment from farmers. Large capital investments will be needed in processing plants. Parameters will need to be defined in terms of processing-plant size; subsidies may be needed for small plants. Exploratory studies will be needed on transportation, preprocessing and storage. There is need for boiler-plate technologies.

Issue: Policy

Government support is needed in the launching of any new technology. Social policies will need to keep pace with government policies to support the new effort.

How will equity be raised?

Issues requiring policy decisions include the rural economy, the environment, and fuel security.

Using Industrial Biotechnology to Improve the Bottom Line

Moderator: Marc Henniker (Strategic Decisions Group)
Recorder: Padu Krishnan (South Dakota State University)

Although the number of participants was small (seven) they were diverse in affiliation and experience. An informal discussion of experience in industry and government occurred. Participants were from Proctor and Gamble (P&G), the government of France, the government of Canada and the American Chemical Society. A representative of P&G used polylactic acid (PLA) and polyhydroxyalcanoate (PHA) as examples of biotechnology initiatives. P&G has been interested in a bleaching alternative in the context of the detergent phosphate ban.

A discussion of genetically modified organisms concluded that the public debate had been mishandled. Many consumers believe that little benefit will accrue from biotechnology. A government document that examined twenty-one biotechnology case studies showed a positive bottom line. Governments seemed to approve of the technology.

Examples of alternatives to current practices and uses were discussed, including zinc instead of gypsum, PLA in "Armani" products, and enzymes in baking. There is no good handle yet on economic impacts. Key will be people, products and profit.

Building a Biobased Economy: Policy and Financing Challenges (Sponsored by the US Department of Energy Office of Policy and International Affairs)

Moderators: Stephen D. Eule (DOE) and Andrew D. Paterson (Environmental Business International)
Recorder: Arvid Boe (South Dakota State University)

The objective of this workshop was to help identify the most critical business risks associated with building a biobased economy. It was designed to elicit industry input that could help guide the development of policies and tools to accelerate market penetration and address financing issues underpinning industrial biotechnology. Although industrial biotechnology has the potential to provide an alternative to conventional energy and chemical processes that are cost-competitive and have fewer environmental drawbacks, many hurdles remain to be overcome to fulfill this potential and make biorefining commercially successful.

The moderators distributed a document *DOE Risk Framework Study for Bio-refineries and Bio-processing: Review of Key Risks, Federal Incentives and Financing Altenatives*, by S.D. Eule and A. Paterson. The document was drafted specifically to stimulate discussion by attendees at this meeting. In it, the authors presented several leading questions:

- ♦ How have market factors and business risks shifted since 2000 to favor construction of biorefineries (*e.g.*, rise of oil prices and phasing out of MTBE)?

- Few biorefineries are being built. Is it primarily a matter of cost? Other business risks? Policies?
- Which risks and policies most deter construction and operation of commercial-scale biorefineries?
- Which policies could encourage wider commercial adoption of biorefineries (*e.g.*, environmental regulations, state and federal financial support)?
- How can risk-targeted incentives improve prospects for biorefineries and bioproducts?

The attendees were invited to describe their interests relative to the focus of this workshop:

- Flexible farmer-owned biorefineries (*e.g.*, investors receive money from sale of corn when corn prices are high and from ethanol when corn prices are low).
- Integrated multi-partnered biorefineries.
- New products from soybean.
- How to utilize government to enhance/support biobased programs.
- Ethanol production from cassava.
- Value-added benefits to farmers (*e.g.*, democratizing ownership of new industries, personal and community benefits, feedstock diversity).
- How to unite multiple stakeholders.
- How to finance multiple feedstock/multiple products businesses.
- Consumer willingness to pay premium for "greener" products.
- How to bring in farmers to maximize their involvement in the value chain [*e.g.*, producing crops with multiple uses (habitat for wildlife production and feedstock for biore-fining)].

Risks/concepts/issues presented in the document were discussed, more specifically:

- The roles of the various players (*e.g.*, DOE, regulators, enzyme sellers, labs/universities, chemical companies, growers) and varying views of risk.
- An overview and approach to risk framework with emphasis on "showstoppers" such as energy policy incentives, system performance, enzyme costs, price of fossil fuels, and agricultural policies.
- Risk framework, with different risks at each phase of investment.
- Reactions of a total of thirty respondents representing chemical/production companies, reagent suppliers/tech firms, labs/universities, and associations and government agencies to a Risk Questionnaire designed by the DOE Policy Office–Office of Climate Change and Environmental Business International. Biotechnology Risk Ratings were calculated using a probability x severity index for feedstock, delivery, bioprocessing, and market issues/concerns.

The ensuing discussion included the following questions/concerns/concepts:

- What are consumers willing to pay for "green power"?
- What will be the public's and environmental activists' reactions to genetically modified organisms being used in biorefineries?
- What are the prospects for governmental preferred procurement of biobased products (*e.g.*, ink made from soybean oil)?
- Farmer involvement/support is crucial to the success of biorefineries, without any major changes in public policy.
- Powerful momentum, driven by energy-security issues and climate change, exists for implementation of biorefineries.
- Financing structures and federal assistance should focus on key risks.

Patent Protection and Patenting Strategies for Industrial Biotechnology Inventions

Moderators: Lila Feisee (Biotechnology Industry Organization), Blair Hughes (McDonnell, Boehnen Hulbert and Berghoff), Paula DeGrandis (Cargill, Inc.)
Recorder: Arvid Boe (South Dakota State University)

The focus was on issues unique to patenting industrial biotechnology inventions, including identifying and accurately disclosing a biotech invention before it is proven or well understood. Discussion was expected to address:

- tactics for perfecting early invention rights and identifying the most valuable inventions to patent, and
- strategies for obtaining the broadest possible protection.

Attendee expectations included:

- to learn more about current trends in industrial biotechnology, intellectual property strategies, and protection vs. trademark vs. trade-secret approaches,

- better understanding of opportunities for patenting and protecting R & D entities,
- how to develop start-up companies from university-developed methodologies, products, *etc.*,
- expanded knowledge of patent law as it relates to biotech, and
- increased understanding of process patents.

The moderators described procedures necessary to draft a patent application that will result in a valid claim. They pointed out that a US patent is only enforceable in the US, and that US patent laws are quite liberal relative to those of most other countries. Biological materials (*e.g.*, processes, methods, biotechnology, DNA sequences, targets) and their uses inside and outside humans are patentable in the US. Many other countries do not allow patenting of bio-logical materials.

The moderators offered views on how to draft an application that will result in a valid invention claim. An invention exists when it has been reduced to practice (*i.e.*, when a definite and permanent idea of an operative invention is known, including every feature of the subject matter sought to be patented). A claim will be valid provided that it is sufficiently described and enabled in the patent specification. This requires, in most cases, detailed descriptions while not overreaching the claim (*i.e.*, claim only what is enabled).

The moderators initiated discussion on variations in patent laws among countries. BIO is pushing for international harmonization of patent laws since biotechnology is globally useful. Consensus is difficult to achieve, thus bilateral agreements are a place to start. Discussion addressed the European Union's progress in unifying patent laws and creating enforcement programs.

A question—what is a continuation-in-part (CIP)?—stimulated discussion on filing strategies. Hypothetically, a CIP in the biotech area may involve adding new data/claims to an initial discovery filed earlier. The actual filing date will depend on the nature of the new information. Dis-cussion continued on strategies for filing patent applications. Opinions varied, from keeping the invention a secret as long as possible before filing a broadly encompassing application to filing early which has become the prevalent cor-porate strategy. The moderators suggested filing a provisional patent application when commercial utilization is not imminent. However, if commercialization will likely occur in the near future, a provisional should not be filed. When filing for a patent of a process of lab-bench scale—although it may not totally apply to the commercial level—make the description broad enough to adequately support such a claim.

In response to a question on the relative value of trade secrets vs. patents, the moderators stated that in the health sciences, trade secrets are common; whereas, in biotech, platform technology is generally patented. If a trade secret is not created or a patent application made for a process, describing it in a published paper will preclude someone else from patenting it. Publishing a CIP also wards off competition.

Should an application contain the minimum amount of information or be a full disclosure of the process? As much detail as possible should be described. If unwilling to disclose information to competitors, don't file an application.

If in possession of the best current process, but it will be several years before commercialization, what are pros and cons of filing now? The first filing should describe the method. The process may be more complex several years later, but the basic steps will likely be the same. It depends on whether or not the invention is a single step or a complete process and whether the invention at laboratory scale will be carried through to commercial scale. From a research standpoint, file as early as possible; most frequently, advantage accrues to the first inventor. Patents in the United States are presumed to be valid, therefore there is incentive to disclose technology. When filing for a process or product, it's a good idea to involve commercialization people when constructing the application.

To what extent does a patent prevent a rival company using a proprietary process in their internal research? This would be infringement, but it is difficult to control. Negotiating research licenses is one approach, generating data in a country that doesn't recognize the patent is another.

DOE Genomics—the GTL Program and BIO—Forging a Critical Partnership in Industrial and Environmental Biotechnology

[Sponsored by the US Department of Energy (DOE) Genomes to Life (GTL) Project]

Moderators: John Houghton (DOE), Michael Knotek (Consultant for DOE), Ari Patrinos (DOE), and George Michaels (Pacific Northwest National Laboratory)

Recorder: Arvid Boe (South Dakota State University)

The focus was on microbes that maintain the planet's geochemistry and have diverse capabilities for using myriad energy sources. It was stressed by the moderators that, once deciphered, microbial capabilities will be the foundation for a new biotechnology that will meet DOE mission needs and will help fuel the Industrial and Environmental section of BIO. The result will be creation of new net-zero carbon-emitting energy sources, reduction in dependence on foreign oil, sequestration of greenhouse gases, and remediation of contaminated environments. The workshop introduced DOE mission challenges that reflect pressing national and international needs, presented the DOE GTL program, and identified potential partnership opportunities and avenues for further discussion.

Presentations by each moderator, followed by discussions with the attendees.

John Houghton explained the US DOE's interest in clean-energy technologies and in developing biotechnological tools to mitigate the effects of global climate change.

Michael Knotek presented the potential role of biotechnology in the National Climate Change Technology program. He apprised the group of the national commitment to understanding and mitigating climate change, as indicated by the United States signing on to the United Nations Framework Convention on Climate Change and the formation of the Climate Change Science Program and the Climate Change Technology Program. He pointed out that microbes and biotechnology will play important roles in solving our energy, climate, and environmental problems. He identified the DOE Genomics: GTL program as a foundation for solving those problems and listed numerous way in which biotechnology may contribute.

Ari Patrinos discussed the DOE Genomics: GTL program and the DOE's high-throughput facilities. He stressed that the DOE started the human genome project and that their goal now is to "break the mold on how biological research is done." The GTL program takes a systems approach to understanding critical microbial processes useful for the DOE missions of clean energy, environmental remediation, and carbon sequestration. Biotechnology can play a major role in the global energy technology portfolio, and serious R & D efforts should be started immediately. The knowledge base resulting from GTL activities will transform the life-sciences landscape and provide a new foundation for biotechnology applications and solutions to DOE missions (*e.g.*, development of efficient ways to produce energy and removal of contaminants from the environment).

Patrinos explained why the DOE is focusing on microbes for its genomics work. They comprise about 60% of the biomass on Earth, and mediate our existence. Sequencing their genomes will increase understanding of the mechanisms they have harnessed and will stimulate new biotechnologies. Exploration of microbial systems has begun in the areas of industrial processes, radioactive-waste cleanup, and mitigation of global climate change (*e.g.*, the bacterium *Shewanella* can immobilize uranium from groundwater and precipitate it on its membranes).

Patrinos outlined the goals of the GTL program:

- to identify and characterize the molecular machines of life,
- to characterize gene regulatory networks,
- to characterize the functional repertoire of microbial communities, and
- to advance understanding of complex biological systems and predict their behavior.

These goals will be accomplished in association with computational tools needed to create useful databases. He stressed the importance of providing the right facilities to democratize access to system-biology resources and free scientists to get involved in biological discovery. Such geno-mics-user facilities, for which access would be awarded on a competitive basis, would include:

- production and characterization of proteins and molecular tags,
- characterization and imaging of molecular machines,

- whole proteome analysis, and
- analysis and modeling of cellular systems and microbial community dynamics.

George Michaels commented on DOE's plan to put in place facilities to enable scientists to work together and have access to instrumentation otherwise unavailable. Genomics would be treated as an information science, with DOE contributing the computational science to achieve whatever needs to be done (*e.g.*, access to data at an unprecedented rate, modeling of fundamental cellular processes).

Question from an attendee: How will GTL interface with the industrial community?

Moderator's answer: GTL will provide the fundamental discovery work. Microbes contain countless yet-undiscovered genes that have enormous potential for biobased industrial uses. DOE hopes to see the jump from science to marketplace dramatically shortened as the distance between scientist and technologist is correspondingly shortened.

Q: What is the role of the Industrial Advisory Board?

A: It will prioritize what research will be conducted at the facility. Scientists will submit applications for sequencing activities, and the scientific community will determine who will be allowed to use the facility. DOE would like to see an industrial coalition to help guide GTL and provide a level of validation.

Q: How do you use genomics to predict what a particular organism will do in a particular situation?

A: This will become possible once we understand how microbes interact with their environment.

Q: How do you plan to work with the international community?

A: Those possibilities are in place. DOE has the flexibility to fund almost anybody, anywhere.

Identifying and Overcoming Barriers to the Diffusion of Industrial Biotechnology in the Chemical Industry

Moderator: Larry Drum (BIOLarry Consulting)

Recorder: William Gibbons (South Dakota State University)

The following items were cited by the group as barriers to penetration of biotechnology in the chemical industry:

- lack of availability of capital for small companies,
- need for examples of successful programs with profitability for use by large and small companies to show that the technology can yield significant value,
- lack of cooperative efforts that can or have benefited industry,
- need for examples of deal structures that work for the chemical industry—value sharing vs. margin in deals,
- replacement technology examples vs. new product economics,
- large amounts of capitalization are needed to be successful in the industry:
  - biorefineries—scale vs. product mix
  - feedstock quality
  - infrastructure for feedstocks,
- industry development model—top down vs. bottom up (technology driven goes to the highest value targets first and a low-cost raw material driven effort goes to the large volume uses first),
- elucidation of the drivers that will compel the industry,
- for companies that have not been involved in the life sciences and even for those that have, in some cases, risk is perceived as higher with biology-based technology than with chemistry-based technology,
- for performance-based chemicals (and polymers) determining product value is an added hard-to-assess dimension,
- there is need for comprehensive studies:
  - value-chain benefits for all
  - need for inclusion of the consumer in the equation
  - full life-cycle analysis,
- there is need to shape policy with comprehensive information,
- isolation of the chemical companies from the end-consumer and the impact of that on product development—consumer companies are getting into the act (Toyota, P&G) because their suppliers are not responding to their needs and are not coming up with the solutions they seek—plastics traceability is an example,
- time to market should favor biotechnology as the technology advances,
- critical mass of product to make a complete

change (the plastics industry has many products and to make a complete change some companies will need many new product introductions in new polymers, not just one or two),
- vertically integrated companies are resistant to change because of sunk capital.

After much discussion, votes were taken and the most important were deemed to be:

- degree of capitalization needed,
- need for comprehensive studies,
- deal structures that work for the industry

When these issues were addressed, directions became less clear. We need a standardized process that can deliver low-cost raw materials to allow quick penetration of the technology. Certainly, sponsorship of research that allows the use of the lowest-cost raw materials is needed. Partnerships with farmers will also be needed. The development of scaleable processes and the use of dry mill concepts are details to be addressed.

On the need for studies and deal structures, industry, universities and government should work together. Many thought that universities should be at the center of the effort. Others thought it should be driven by industry.