

ABOUT CODEXIS TECHNOLOGY

Codexis' proprietary technology platform manufactures customized biocatalysts for use in making existing industrial processes faster, cleaner and more efficient than current methods and has the potential to make new industrial processes at commercial scale. We have commercialized our biocatalysts in the pharmaceutical industry and are developing biocatalysts for use in producing advanced biofuels under a multi-year research and development collaboration with Royal Dutch Shell plc. Codexis is also using our technology platform to pursue biocatalyst-enabled solutions in other bioindustrial markets, including carbon management, water treatment and chemicals.

1. ABOUT BIOCATALYSTS

Biocatalysts are enzymes or microbes that initiate or accelerate chemical reactions. Manufacturers have historically used naturally occurring biocatalysts to produce many goods used in everyday life. However, inherent limitations in naturally occurring biocatalysts have restricted their commercial use. Our proprietary technology platform is able to overcome many of these limitations, allowing us to evolve and optimize biocatalysts to perform specific and desired chemical reactions at commercial scale.

DIFFERENCES BETWEEN BIOCATALYST-ENABLED MANUFACTURING AND CHEMISTRY-BASED MANUFACTURING

Biocatalyst-enabled manufacturing processes may address a number of the drawbacks of conventional chemistry-based manufacturing. For example, unlike most chemistry-based manufacturing processes, biocatalysts can operate at or near room temperature and pressure, and often use manufacturing equipment that is less complex and expensive to build and operate. Biocatalyst-enabled processes can create products with the same or higher quality as chemistry-based manufacturing processes, while reducing risks associated with extreme manufacturing environments and without generating the high volumes of waste, some of it hazardous to health and the environment, typically associated with conventional chemistry-based manufacturing processes.

In addition, due to concerns about the environment and the scarcity and security of supply of petroleum, there is an increasing interest in using cellulosic biomass as non-petroleum-based feedstocks for a variety of products, including advanced biofuels and other chemicals. To date, conventional chemistry-based manufacturing approaches have not resulted in commercially viable processes for the conversion of cellulosic biomass to biofuels and other products. Biocatalysts have the potential to enable processes for the development of products, such as cellulose-derived biofuels, that cannot currently be manufactured using alternative techniques.

Despite their potentially significant advantages, biocatalysts have not achieved their full potential in industrial applications. Naturally occurring biocatalysts are often not stable enough to be used in industrial settings, where conditions may differ significantly from those in the biocatalysts' natural environments. The activity and productivity of these biocatalysts is often too limited to be cost-effective in commercial scale manufacturing. In addition, the activity of natural biocatalysts is typically inhibited by the end product of the reactions they facilitate. This characteristic of natural biocatalysts, which is

referred to as product inhibition, results in limited product yields in industrial settings. Moreover, for certain industrial applications, there are no known naturally occurring biocatalysts that catalyze the desired reaction.

Due to these limitations, other companies and researchers have tried to improve the performance of naturally occurring biocatalysts by directing their evolution through biotechnology techniques such as the random mutation of genes. However, to date, these techniques have had only limited success for a number of reasons. We believe there is a significant opportunity for novel technologies that can address the limitations of other biotechnology techniques and can substantially enhance the performance of biocatalysts in industrial settings.

HOW CODEXIS' TECHNOLOGY IS ABLE TO OVERCOME THE CHALLENGES OF BIOCATALYST-ENABLED MANUFACTURING

We believe that our proprietary technology platform can transform the industrial application of biocatalysts by improving their commercially relevant characteristics, such as stability, activity, product yield and tolerance to industrial conditions, while reducing product inhibition. In addition, our technology platform allows us to develop and optimize biocatalysts much more rapidly than is currently possible with alternative methods. Perhaps most importantly, we have demonstrated that our technology platform can enable the manufacture of products cost-effectively, at commercial scale and with significantly reduced environmental impact relative to conventional manufacturing processes.

Our proprietary technology platform uses advanced biotechnology methods, bioinformatics and years of accumulated experience and know-how to significantly expedite the process of developing optimized biocatalysts. Key components of our technology platform include gene shuffling, whole genome shuffling, multiplexed gene SOEing, and proprietary bioinformatic software tools that allow us to identify and quantify the potential value of beneficial mutations and avoid detrimental mutations.

2.CODEXIS APPLICATIONS IN THE PHARMACEUTICAL INDUSTRY

Our technology platform enables us to deliver solutions to our customers in the pharmaceutical market by developing and delivering optimized biocatalysts that perform chemical transformations at a lower cost, and improve the efficiency and productivity of manufacturing processes. We provide value throughout the pharmaceutical product lifecycle. Our technology platform allows us to provide benefits to our customers in a number of ways, including:

- reducing the use of raw materials and intermediate products;
- improving product yield;
- using water as a primary solvent;
- performing reactions at or near room temperature and pressure;
- eliminating the need for certain costly manufacturing equipment;
- reducing energy requirements;
- reducing the need for late-stage purification steps;
- eliminating multiple steps in the manufacturing process; and

- eliminating hazardous inputs and harmful emission by-products.

Early in the product lifecycle, customers can use our services to achieve speed to market and to reduce manufacturing costs. If an innovator incorporates our products or processes into an FDA-approved product, we expect the innovator to continue to use these products or processes for the patent life of the approved drug.

After a product is launched, customers also use our services to reduce manufacturing costs. At this stage, changes in the manufacturing process originally approved by the FDA may require additional review. Typically, pharmaceutical companies will only seek FDA approval for a manufacturing change if there is a substantial cost savings associated with the change. We believe that the cost savings associated with our products may lead our customers to change their manufacturing processes for approved products and, if necessary, seek FDA approval of the new processes which incorporate our biocatalysts. Moreover, we believe these cost savings are attractive to generics manufacturers, who compete primarily on price.

We are currently working with customers on approximately 35 pharmaceutical products in various stages of the pharmaceutical product lifecycle.

CODEXIS PRODUCTS FOR THE PHARMACEUTICALS MARKET

Codex™ Biocatalyst Panels. We sell Codex™ Biocatalyst Panels to customers who are engaged in both drug development and the marketing of approved drugs to allow them to screen and identify possible biocatalytic manufacturing processes for their drug candidates and their marketed products. Our Codex Biocatalyst Panels are plates embedded with genetically diverse variants of our proprietary biocatalysts, which allow our customers to determine whether a biocatalyst produces a desired activity that is applicable to a particular process.

For compounds that are in development, our Codex Biocatalyst Panels:

- allow innovators to rapidly and inexpensively screen and identify possible biocatalytic manufacturing processes for many of their drug candidates in-house, without the risks of disclosing the composition of their proprietary molecules before they have received patent protection; and
- generate data that we can use to rapidly optimize biocatalysts for a particular reaction, if necessary, reducing the time required to generate a manufacturing process capable of supporting clinical trials with inexpensively produced, pure drugs.

We believe that our Codex Biocatalyst Panels have helped us build early and broad awareness of the power and utility of our technology platform, and will increasingly lead to sales of our biocatalyst optimization services and biocatalysts, as well as intermediates and APIs made using our biocatalysts. We currently have over ten customers for our panels, including leading pharmaceutical companies such as F. Hoffman-La Roche Ltd., GlaxoSmithKline plc, Merck, Novartis and Pfizer. If our customers incorporate a biocatalytic manufacturing process early in a product's lifecycle, they can reduce their

manufacturing costs throughout that lifecycle, while we, in turn, could realize a long term revenue stream resulting from the use of our biocatalysts during that time. In addition, our Codex Biocatalyst Panels are increasingly used by our customers to evaluate the feasibility of changing the manufacturing process for their marketed products to a biocatalyst-enabled process.

Biocatalyst screening services. If a customer prefers, rather than subscribing to our Codex Biocatalyst Panels to use for their own screening, they can send us their materials to test against our existing libraries of biocatalysts. If we detect desired activity in a specific biocatalyst, we can supply the customer with this biocatalyst or perform optimization services to improve the performance of the biocatalyst.

Our screening services:

- allow innovators to rapidly and inexpensively screen and identify possible biocatalytic manufacturing processes through access to our extensive biocatalyst libraries; and
- generate data that we can use to rapidly optimize biocatalysts for a particular reaction, if necessary, reducing the time required to generate a manufacturing process capable of supporting the customers' particular needs, ranging from small quantities for clinical trials to full commercial production, in all cases providing inexpensively produced, pure drugs.

We have provided screening services to numerous innovator and generic pharmaceutical manufacturers.

Biocatalyst optimization services. We work with our customers throughout the pharmaceutical product lifecycle to customize proprietary biocatalysts, resulting in optimized biocatalysts that have been evolved specifically to perform a desired process according to a highly selective set of specifications.

Our biocatalyst optimization services:

- allow innovators to improve the manufacturing process as their drug candidates progress through preclinical and clinical development, deferring or reducing the need for significant manufacturing investment until the likelihood of commercial success is more certain; and
- enable manufacturing processes that are highly efficient, inexpensive, require relatively little energy, reduce the need for hazardous reagents, and reduce waste. For example, our activities with Pfizer have included developing an optimized biocatalytic manufacturing process for a key intermediate that eliminates three chemical steps.

Biocatalysts. We supply varying quantities of our proprietary biocatalysts to pharmaceutical companies, from small to moderate quantities while they are optimizing their production processes, to larger quantities during later-stage clinical development and commercial scale drug production.

Our biocatalysts:

- enable innovators to manufacture products more efficiently during preclinical and clinical development using optimized biocatalytic processes, with relatively low investment;
- eliminate the need for innovators to invest in the development of complex chemical synthesis routes during the development stage;
- allow innovators to achieve higher product purity during the development stage prior to investing in expensive late-stage clinical trials;
- reduce the risk of adverse effects arising from product impurities;
- allow the removal of entire steps from synthetic chemical production routes during commercial scale production, reducing raw material costs, energy requirements and the need for capital expenditures; and
- decrease the manufacturing costs for our customers.

For instance, as a part of our ongoing collaboration with Merck, we have developed a biocatalyst for use in a new manufacturing process for sitagliptin, the API in Merck's pharmaceutical product Januvia. Januvia is Merck's first-in-class medication for the treatment of Type II diabetes. Merck's current manufacturing process uses a high pressure chemo-catalysis platform, which requires the use of highly specialized equipment. The new biocatalyst-enabled process runs at atmospheric pressure, eliminates the need for certain highly specialized equipment and increases overall product yield.

Intermediates and APIs. We can supply our customers intermediates and APIs made using our biocatalysts throughout the drug lifecycle.

Our supply of intermediates has the following uses and benefits:

- lowers capital investment for innovators through outsourcing of manufacturing; and
- provides a source of less expensive, more pure products to innovator and generics manufacturers.

In the innovator market, we are currently supplying Pfizer with an intermediate in the manufacture of Lipitor. In February 2010, we entered into a collaboration with Dishman Pharmaceuticals and Chemicals, Ltd., or Dishman, a global manufacturer of intermediates and APIs located in India, to expand the application of our technology to a broader pipeline of innovator pharmaceutical products. Under our agreement with Dishman, we will work with Dishman exclusively, subject to certain exceptions, with respect to the manufacture and supply of intermediates and APIs using our biocatalysts for a select group of innovators. Dishman will also be our preferred contract manufacturing organization partner for new opportunities with other innovator pharmaceutical companies.

We have also developed biocatalysts for use in the manufacture of certain generic intermediates and APIs by various companies, including Arch and Teva Pharmaceutical Industries Ltd., or Teva. In addition, we have launched and are marketing several new intermediates and APIs for the generic equivalents of branded pharmaceutical products, including Singulair and Cymbalta, for sale in markets where innovators have not sought patent protection for their products and intend to sell these same

intermediates and APIs for use in markets where innovators have sought patent protection when the patent protection for each product expires.

3.CODEXIS APPLICATIONS FOR THE BIOFUELS MARKET

We believe that our technology platform will enable the development of biocatalysts that can be used to produce commercially viable, cellulose-derived biofuel alternatives to petroleum-based fuels. Since 2006, we have been engaged with Shell in a research and development collaboration under which we are developing biocatalysts for use in producing advanced biofuels.

Our advanced biofuels program focuses on two primary elements: (1) developing biocatalysts to convert cellulosic biomass into sugars; and (2) converting these sugars into two advanced biofuels, cellulosic ethanol and biohydrocarbon diesel. For the first element, we have used our technology platform to improve our cellulase and other biocatalysts. For the second element, we have developed a biocatalyst that converts sugars to diesel fuel, and are working on improving ethanol-producing yeast. We are using our technology platform to develop biocatalysts that we believe will:

- increase the rate at which cellulosic biomass is converted into biofuels;
- increase the yield of biofuels produced from cellulosic biomass;
- eliminate the need to use food resources for the production of biofuels;
- provide producers with more flexibility in designing processes to convert cellulosic biomass to biofuels, thereby reducing the costs associated with building and operating biofuel production facilities; and
- enable the production of new types of cellulosic biofuels that could be alternatives to petroleum-based fuels.

Under our research and development collaboration with Shell, Shell will have the right, but not the obligation, to commercialize any technology that we develop in our biofuels program. If Shell commercializes our biofuels technology, we will collect a royalty for every gallon of fuel that Shell produces using our technology. If Shell chooses to commercialize any biofuels products developed through our collaboration, we believe that the combination of our technology platform with Shell's proven product development capabilities and resources could enable a biofuels solution that extends from the conversion of cellulosic biomass into biofuels to delivery and distribution of refined biofuels to consumers at the pump.

CODEXIS PRODUCTS FOR THE BIOFUELS MARKET

Sugar Platform As part of our biofuels research and development collaboration with Shell, we are using our technology platform to develop a suite of cellulases and other biocatalysts to convert cellulosic biomass to sugar, which we sometimes refer to as our sugar platform. One of the goals of our sugar platform is to improve the performance and operational range of cellulases and other biocatalysts so that they cost-effectively function in industrial conditions. For example, we have developed several of our cellulase biocatalysts that now function at temperature and acidity levels that we believe are close to commercial production targets. The benefit of increasing the operational range of the cellulases is to

provide maximum flexibility in the design and function of the facility that is used to produce cellulose-derived sugars, thus decreasing the costs of production and lowering the cost of the end product to make it competitive with petroleum-based fuels.

Another goal of our sugar platform is to increase the rate and extent of conversion of cellulosic biomass to fermentable sugars. The more rapidly and efficiently biocatalysts convert cellulose and hemicellulose to sugars, the less expensive the biomass conversion process will be to operate. We are developing our biocatalysts to produce more sugar per unit volume. For example, we have developed a biocatalyst that we believe produces twice as much sugar from cellulose as a leading commercially available product. We believe faster sugar production from our biocatalysts will lower capital costs and production costs and result in lower-cost sugar to convert to an end fuel product.

We are developing a library of cellulases that have the potential to convert a wide variety of cellulosic biomass sources into fermentable sugars. The cellulosic biomass that we expect will be used to produce advanced biofuels is highly variable from region to region and can change over time. To optimize the local and seasonal conversion of biomass to fermentable sugars, we expect to use technology similar to our Codex Biocatalyst Panel of cellulases that Shell can use to customize the biocatalysts that they use at each advanced biofuel production facility. This technical innovation may ultimately make our sugar platform feedstock agnostic.

For example, based on our lab work, we believe that our cellulases have the potential to convert sugar cane bagasse or wheat straw to fermentable sugars. In addition, we licensed a commercial-scale enzyme production system from Dyadic in 2008 that we expect will enable the cost-effective production of the high-performing biocatalysts that we are developing for Shell. We believe that the combination of our high-performing cellulases and other biocatalysts, the feedstock flexibility that we expect our Codex Biocatalyst Panels will provide, plus the ability to produce these biocatalysts cost-effectively at commercial scale will enable us to develop a scalable, global sugar platform that will provide a competitive advantage in the advanced biofuels market.

Cellulosic Ethanol The goal of our cellulosic ethanol program is to develop commercial yeast that rapidly produces high levels of ethanol from cellulose-derived sugars. Cellulosic biomass produces a mix of several types of sugars, including glucose, xylose and arabinose. Glucose is the main type of sugar in the mix and it is readily converted to ethanol by fermentation using commercial yeast. Xylose is another significant component of the mix but is not converted to ethanol by the yeast currently used in today's first generation ethanol production. Therefore, it is important to develop yeast that can rapidly convert not only glucose but also xylose and other sugars into ethanol. The yeast that is developed must be sufficiently robust so that it can produce ethanol in the presence of a variety of chemical compounds that have been shown to directly inhibit yeast.

Using a number of our core technologies, including whole genome shuffling and cellular engineering, we are working with a variety of active industrial and laboratory yeast strains to develop a yeast strain that rapidly converts more of these sugars to ethanol under a range of industrial conditions, which should result in greater ethanol production and lower capital and ethanol production costs. Based on this lab

work, if the market opportunity presents itself, we believe that our technology platform can also be used to transform first generation yeast, which is currently used to convert sugars to ethanol at commercial scale.

Biohydrocarbon Diesel We have made significant advancements in our biohydrocarbon diesel fuel program, which is focused on converting cellulose-derived sugar into a fungible diesel blending stock. We also believe that diesel fuel will be able to be produced from cane sugar using our biocatalysts. Based on our testing to date, our biocatalysts rapidly produce high quantities of fuel product per unit volume, which has the potential to reduce production costs and increase the efficiency and productivity of the biohydrocarbon manufacturing process.

Our biohydrocarbon program has several additional advantages that could lower the production costs of diesel fuel. Our diesel-producing microbe secretes the diesel molecule from the cell, which then separates from the media in which the cell lives and grows. As a result, our production system can be run continuously without having to stop fuel production to harvest the fuel and purify the fuel product. We believe that many other comparable diesel-producing systems must isolate the fuel-producing cells, break-open the cells to release the fuel and purify the fuel from the resulting mixture, which significantly increase production costs for the end fuel product.

In addition, we believe that the biohydrocarbon fuel product that we develop will be able to be blended directly into existing diesel fuel with little or no additional processing at a refinery, which would further lower production costs. In contrast, existing biodiesel fuels that are derived from plant oils must be chemically modified before they are suitable for use as diesel components. These chemical modifications involve processing steps before such fuel is ready for use, which adds to the cost of producing the fuel. In addition, other advanced biofuel programs aimed at producing diesel alternatives require extensive and difficult hydrogenation reactions, which are expensive and require capital intensive facilities that are not widely available.

In contrast to biodiesel produced from plant oils, we expect that the diesel fuel that we develop will be compatible with the existing transportation infrastructure, including distribution systems. A new fuel that works in existing engines and fuel production and distribution systems will not require additional investment in infrastructure to deploy this new technology. As discussed above, we believe that the diesel fuel that we develop will be capable of being blended in conventional petrochemical refineries that are widely used across the globe. This production flexibility should reduce structural barriers to adoption of the molecule as a wide-spread petroleum alternative.

4. OTHER INDUSTRIAL APPLICATIONS OF CODEXIS TECHNOLOGY

We believe that our technology platform, together with the knowledge and experience gained from our efforts in the pharmaceutical market and in our biofuels development program, will allow us to capitalize on opportunities in other bioindustrial markets, including carbon management, water treatment and chemicals. Depending on the market, we may pursue collaborations with industry leaders to allow us to leverage their competitive strengths and resources in pursuit of these opportunities.

Carbon Management In the carbon management market, we are seeking to apply our technology platform to the management of carbon dioxide emissions from stationary point sources such as coal-fired power plants. As part of this effort, in December 2009, we entered into an exclusive joint development agreement with CO2 Solution under which we will combine our biocatalyst-enabled technology platform with CO2 Solution's proprietary enzymatic methods for the efficient capture of carbon dioxide from coal-fired power plants and other large sources of carbon dioxide emissions. We believe our biocatalysts have the potential to enhance the effectiveness of CO2 Solution's carbon capture processes in harsh industrial conditions.

To further our efforts in the carbon management market, we have filed provisional patent applications relating to biocatalysts that we believe may optimize the process of removing carbon dioxide from flue gases. These biocatalysts improve the effectiveness of amine solvents, one of the leading potential technologies to remove carbon dioxide from flue gas. A major drawback of amine solvent technologies is the additional "parasitic" energy required to operate them. Based on initial models, we believe that our biocatalysts may reduce this parasitic energy loss by up to 35%. In the laboratory, these biocatalysts have also exhibited increased tolerance for flue stack-type operating conditions, though not yet at target commercial levels. Although our research is in its early stages, we believe that it may be possible to cost-effectively utilize biocatalyst-enabled solutions to separate carbon dioxide from other exhaust gases and direct them to separate sequestration mechanisms.

Water Treatment The market for biocatalysts in water treatment is in a very early stage of development. However, new interest in biocatalyst-enabled solutions in water treatment has been sparked in part by concerns about possible contamination of drinking water from industrial and other sources. For example, a U.S. government report released in 2006 examined the potential of biocatalysts in the treatment of groundwater and drinking water in both civilian and military applications. The report concluded that biocatalyst-embedded water filters held significant promise for the treatment of agents, pesticides, or other chemical contaminants in drinking water systems, as well as for the decontamination of pipes and other equipment with contaminant residue. We believe that there are also opportunities for biocatalyst-enabled solutions to treat municipal wastewater streams.

Chemicals There are also significant market opportunities in the chemical industry for companies that can help reduce or eliminate petroleum dependency, as well as costly and wasteful manufacturing processes. For example, according to the EIA, in 2008, approximately 214 million barrels of petroleum were used in petrochemical feedstocks.

We believe that fermentable sugars produced from cellulosic biomass may serve as an alternate source of carbon for use in the manufacture of many chemicals. This potential market may provide an opportunity to leverage our funded work with Shell into a separate business in the non-fuels chemicals industry. Our license agreement with Shell permits us to use technology developed for Shell outside of the field of fuels and lubricants. In addition, our technology platform could be applied to develop biocatalysts for the conversion of sugar or other feedstocks, rather than petroleum-derived hydrocarbons, into commercially important chemicals.

5. HOW CODEXIS TECHNOLOGY WORKS

We are innovators in the directed evolution of enzymes and microbes to enable industrial biocatalytic reactions and fermentations via biocatalyst engineering, metabolic pathway engineering and fermentation microbe improvement.

Our approach to developing commercially viable biocatalytic processes begins by conceptually designing the most economically practical manufacturing process for a targeted product. We then develop optimized biocatalysts to enable that process design, using our directed evolution technology, including screening and validating biocatalysts under relevant conditions. Typical design criteria include stability in the desired reaction conditions, biocatalyst activity and productivity (yield), ease of product isolation, product purity and cost.

Alternative approaches to biocatalytic process development typically involve designing and engineering the biocatalytic processes around shortcomings of available biocatalysts, including, for example, biocatalyst immobilization (for stability and/or reuse), special equipment and costly product isolation and purification methods. We circumvent the need for these types of costly process design features by optimizing the biocatalyst for fitness in the desired process environment. As a result, we enable and develop cost-efficient processes that typically are relatively simple to run in conventional manufacturing equipment. This also allows for the efficient technical transfer of our process to our manufacturing partners.

The successful embodiment of our technology platform in commercial manufacturing processes requires well-integrated expertise in a number of technical disciplines. In addition to those directly involved in practicing our directed evolution technologies, such as molecular biology, enzymology, microbiology, cellular engineering, metabolic engineering, bioinformatics, biochemistry, and high throughput analytical chemistry, our process development projects also involve integrated expertise in organic chemistry, chemical process development, chemical engineering, fermentation process development, and fermentation engineering. Our tightly integrated, multi-disciplinary approach to biocatalyst and process development is a critical success factor for our company.

THE ENZYME OPTIMIZATION PROCESS

The enzyme optimization process starts by identifying genes that code for enzymes known to have the general type of catalytic reactivity for a desired chemical reaction. Typically, we identify gene sequences in published databases and then synthesize candidate genes having those sequences. Using a variety of biotechnology tools, we diversify these genes by introducing mutations, giving rise to changes in the enzymes for which they encode. The methods for diversifying these genes, and types of diversity being tested, often vary over the course of a biocatalyst optimization program. For finding initial diversity, methods typically include random mutagenesis and site-directed (included structure-guided) mutagenesis. We also test mutational variations that distinguish related enzymes among different organisms. Once we have identified potentially beneficial mutations, we test combinations of these mutations in libraries made using our proprietary gene recombination methodologies, gene shuffling and multiplexed gene SOEing.

With our proprietary gene shuffling methodology, we generate libraries of genes that have random combinations of the mutations we are testing. The pool of genes is used to transform host cells, which entails introducing the various genes, one each, into host cells. These cells are then segregated and grown into colonies. Cells from individual colonies are cultured in high throughput to produce the enzyme encoded by the shuffled gene in those cells. The enzymes are then screened in high throughput using test conditions relevant to the desired process. The screening results identify individual shuffled genes that produce improved enzymes having combinations of beneficial mutations and weed out enzymes having detrimental ones. Using different test conditions and/or different analytical methods, we can identify variant enzymes that exhibit various improved performance characteristics, such as stability, activity and selectivity, under conditions relevant to the desired chemical process.

In the next step in our optimization process, we use our proprietary software tool, ProSAR, to analyze protein sequence-activity relationships. We initially licensed ProSAR from Maxygen and further developed and customized ProSAR to address our specific needs. ProSAR aids in identifying specific gene and enzyme mutations that are beneficial, neutral or detrimental with respect to the desired performance characteristics. Earlier directed evolution methods did not separately evaluate individual mutations in libraries of variants which carry multiple mutations, where beneficial and detrimental performance characteristics may be mixed in an individual gene or enzyme. Capitalizing on the advent of inexpensive gene sequencing, we are able to determine which particular mutations are present in the genes and proteins we have screened.

Our ProSAR bioinformatics software relates the screening results to the mutations and ranks the individual mutations with regard to their degree of benefit or detriment, relative to whichever process parameter(s) the screening tested. Using that information, we can bias the pool of mutational diversity in the next iteration to further the accumulation of beneficial diversity and cancel out detrimental diversity in the individual genes in the resulting shuffled library. The ProSAR results also help us develop ideas about new diversity to test. ProSAR, combined with efficient gene synthesis and high quality library generation methods, has led to a significant increase in the efficiency and speed of enzyme improvement and optimization.

In another step of our optimization process, we take the best variants we have identified and prepare enough of each to test in the desired chemical process at laboratory scale, for in-process confirmation. This optimization routine is done iteratively, typically adding new diversity to the pool in each iteration. The gene that codes for the best performing enzyme in one iteration is used as the starting gene for the next iteration of shuffling and screening. As the enzymes improve over these iterations, the screening conditions are made increasingly more stringent. In this way, enzymes are rapidly optimized until all in-process performance requirements have been achieved and the economic objectives for the desired process have been met.

Multiplexed gene SOEing is our new proprietary methodology for rapidly generating gene variants. Using multiplexed gene SOEing, we rapidly generate collections of individual gene variants that have predetermined, as opposed to random, combinations of mutations we are testing. It is based on a biotechnology technique, which we refer to as SOEing, or Splicing by Overlap Extension, generally used

to make a hybrid, or spliced, gene from fragments of two genes and/or to introduce a specific mutation into a splice between fragments of one gene. We have automated the process to robotically make, in parallel, one hundred to several hundred variants, each with a predetermined combination of the mutations we are testing. The variants are introduced into host cells, and the encoded enzyme is produced and screened in high throughput, as described above.

Using multiplexed gene SOEing, we can test many mutations and combinations thereof in parallel, and because the mutation incorporation is controlled and predetermined before screening, as opposed to random incorporation and selection after screening, the resulting data set can be more optimal for ProSAR analysis.

We believe using multiplexed gene SOEing to quickly survey many mutations, followed by ProSAR-driven shuffling of beneficial mutations, is a particularly effective approach, providing rapid gains in enzyme performance.

ABOUT CODEX BIOCATALYST PANELS

Our Codex Biocatalyst Panels were initially developed to speed our own internal process for identifying enzymes with desired characteristics for further optimization. Each Codex Biocatalyst Panel is comprised of variants of one or more enzymes that catalyze one type of a generally useful chemical reaction. We assemble, on one or more microtiter sample plates, variants of a parent enzyme that we pre-optimize for stability in industrial chemical processes and for ready manufacturability. The variants are diversified to react to a variety of chemical structures that are susceptible to that type of chemical reaction.

Either we or our innovator pharmaceutical customers use the Codex Biocatalyst Panels to screen a new chemical structure against the assembled variants to rapidly identify variants that react with the new chemical structure. For some new structures, a variant on the panel could enable production of the desired product. We can also analyze the data from the panel screen using ProSAR to identify the mutations that are beneficial for the reaction of the new structure and further optimize the enzyme as needed using the enzyme optimization techniques described above. In cases where a customer wishes to screen a proprietary new chemical structure itself, we can produce a custom panel of new variants on a sample plate produced by multiplexed gene SOEing.

We may also use our Codex Biocatalyst Panels in our bioindustrial programs. In our biofuels research and development collaboration with Shell, we are developing a library of cellulases that have the potential to convert a wide variety of cellulosic biomass sources into fermentable sugars. The cellulosic biomass that we expect will be used to produce advanced biofuels is highly variable from region to region and can change over time. To optimize the local and seasonal conversion of cellulosic biomass to fermentable sugars, we expect to produce a Codex Biocatalyst Panel of cellulases that we or Shell can use to customize the biocatalysts that Shell uses at each advanced biofuel production facility. This technical innovation may ultimately make our sugar platform feedstock agnostic. Similarly, there is regional variation in coal. We may develop a Codex Biocatalyst Panel that we or our customers can use to tailor our carbon capture biocatalysts to the specific characteristics of the coal used in each energy facility that adopts our carbon capture technology.

THE GENE OPTIMIZATION PROCESS

For fermentation microbes, we enhance metabolic pathways by using gene optimization to improve the production and/or productivity of one or more enzymes in a series of in vivo reactions that make a desired product. We optimize the gene/enzyme using either in vitro or in vivo screening. For fermentation applications, the microbes containing the improved gene(s) are directly evaluated in laboratory scale fermenters.

The metabolic pathway may naturally exist in the microbe, but productivity and/or selectivity improvements are needed to economically produce more of the desired natural product and/or less of an undesired by-product. We can also introduce a new metabolic pathway to produce a desired product using our gene shuffling technology in combination with synthetic biology, a type of metabolic engineering in which new genes are introduced into a microbe.

We are using our gene/enzyme optimization methodologies in our biofuels program to optimize fermentation microbes, including optimization of:

- native and introduced (non-native) cellulase genes for increased productivity in our cellulase production microbes;
- an introduced (non-native) pathway in yeast for the conversion of xylose, a cellulose-derived sugar, to ethanol; and
- an introduced (non-native) pathway in a microbe for the production of our biohydrocarbon fuel molecule.

THE WHOLE GENOME SHUFFLING PROCESS

In addition to our gene optimization technology for enzymes, we have another complimentary technology in our platform for the optimization of fermentation microbes called Whole Genome Shuffling. Whole Genome Shuffling allows us to improve the performance of a fermentation microbe by shuffling unidentified mutations in unidentified genes across the genome. We start with a diversity of mutational variants of a fermentation organism, generated by conventional means such as random mutagenesis.

Our Whole Genome Shuffling involves introducing the entire genome of two or more such cells into a single cell, in which the genetic machinery of the combined cell recombines, or shuffles, the genomes. In one method, this is accomplished by protoplast fusion, in which the cell walls are removed to leave the cells' contents contained only by their cell membranes. The cell membranes of these protoplasts in the diverse population are induced to fuse together into fusants containing the genome of two or more of the parent cells. From these fusants, we regenerate normal cells, each with one copy of a hybridized genome.

Microbial colonies are then grown and screened for their performance in the fermentative production of the desired product. This process can be repeated, including with the introduction of new mutations, until the desired performance in the fermentation process is achieved. One of our collaborators is

operating a fermentation process for a generic pharmaceutical product using microbes we developed by Whole Genome Shuffling.

We are using our Whole Genome Shuffling technology in our biofuels program to optimize fermentation microbes, including optimization of:

- enzyme production hosts for increased production of cellulase enzymes;
- ethanol-producing yeasts for improved xylose utilization, ethanol productivity, and tolerance to higher ethanol concentrations; and
- our biohydrocarbon producing strain for increased productivity.

METABOLIC ENGINEERING AND SYNTHETIC BIOLOGY

In addition to our proprietary enzyme and microbe optimization technologies, we have built expert capabilities in a suite of new metabolic engineering technologies for the development and optimization of fermentation microbes. These technologies are generally applicable to our pathway and strain engineering programs. Genomics, transcriptomics, proteomics and metabolomics all provide more in-depth analyses of the metabolic functioning of fermentation microbes, and differences between variants, to guide further improvements. In many cases, these analyses help to identify enzymes that need to be modified (removed, increased, stabilized, or otherwise modified) in order to increase the overall productivity and performance of the strain.

Synthetic biology involves the design, synthesis and introduction of new genetic programming to organisms for new biological functions. This field has rapidly developed in recent years as DNA synthesis and sequencing costs have rapidly dropped. Using synthetic biology, we are taking advantage of the exploding publicly available gene and genome sequence information in our gene and metabolic pathway optimization projects. This information is being leveraged by our ProSAR software and multiplexed gene SOEing methodologies. For example, we use synthetic biology in our biofuels program to introduce non-native pathways for xylose utilization and for biohydrocarbon production and to optimize these pathways.