Impact of Government Biotechnology Regulations on Biofuel Development

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ABSTRACT

Commercial strategies using the advanced techniques of biotechnology for the production of biofuels may be affected by the biotechnology regulatory framework that exists in the United States and other countries. Use of genetic engineering to create improved microorganisms or enzymes for production of ethanol or other fuels might be subject to regulation by the U.S. Environmental Protection Agency. Projects involving the genetic engineering of plants and trees to improve the feedstocks used in biomass conversion may be subject to regulation by the U.S. Department of Agriculture and equivalent rules in other countries. This paper explains the history and the basis for biotechnology regulation in the United States and elsewhere, and surveys the regulatory programs that might be applicable to biofuels projects, as well as the procedural and data requirements such projects might face. With advance planning it should be possible to gain timely regulatory approval for biofuels projects.

Keywords: biofuels, genetic engineering, microorganisms, transgenic plants, regulation.

1 GENETIC ENGINEERING STRATEGIES FOR BIOFUELS

There are numerous strategies for the use of advanced biotechnologies to create improved biofuels products or processes, involving the creation of engineered or synthetic microorganisms for use in production of ethanol or biodiesel, or genetically engineered ("transgenic") plants as improved fuel feedstocks.

One strategy is to engineer microorganisms to overexpress desired enzymes, including enzymes not naturally found in the chosen microbial species (it is also possible to derive improved microbial strains by classical mutation and selection). Companies developing or using modified (or mutated) microorganisms for this purpose include Mascoma Corporation of Boston, Mass., Verenium Corporation of Cambridge, Mass., BioEnergy International LLC of Quincy, Mass., SunEthanol of Amherst, Mass., and HoosierGene of West Lafayette, Ind. Several companies are using engineered microbes to manufacture novel or improved industrial enzymes, for enhancing or accelerating biofuel production processes. An alternative strategy, pursued by companies like LS9, Inc. of San Carlos, Calif. and Amyris Biotechnologies of Emeryville, Calif., is to create novel or synthetic microorganisms optimized to produce biologically-based fuels resembling petroleum fuels.

Technical strategies involving transgenic plants might have as their goal the introduction of genetic changes into certain plant species to make them more readily useful for ethanol production. For example, one might introduce genes encoding key biodegradative enzymes into the plants, to enhance the conversion of cellulose into fermentable sugars. Among companies pursuing this approach are Agrivida, Inc. of Medford, Mass. and Farmacule Bioindustries Pty Ltd. of Brisbane, Australia.

2 GOVERNMENT REGULATION OF BIOTECHNOLOGY

2.1 The U.S. Regulatory Framework and its Historical Background

The products of biotechnology are regulated in the U.S. under a Coordinated Framework that was introduced in 1986 by the Reagan Administration, which decided that the products of biotechnology would be regulated under existing laws and in most cases under existing regulations, based on the intended end-use of each product, without need for new legislation. The term "Coordinated Framework" refers to the matrix of existing laws and regulations that have served to regulate the biotechnology industry since its publication in the Federal Register in June 1986 [1]. Most of the products of biotechnology have been drugs, biologics or diagnostics, and have been regulated by the U.S. Food and Drug Administration. However, many agricultural products, including transgenic plant varieties, have been regulated by the U.S. Department of Agriculture (USDA), and certain commercial products involving microorganisms are regulated by the U.S. Environmental Protection Agency (EPA) [1,2]. These two regulatory programs, put in place to oversee the environmental uses of biotechnology, are most likely to affect biofuels projects.

2.2 Biotechnology Regulation Outside the United States

Most of the major industrialized nations and regions of the world, including the European Union, Canada and Japan, have also developed biotechnology regulatory

frameworks, with particular emphasis (as in the U.S.) on assuring that proposed uses of engineered organisms in the open environment received appropriate scrutiny and risk assessment [1,3,4]. Although based on different legal frameworks, the scientific issues considered in risk assessments can be expected to be quite similar to those used by U.S. agencies. However, the uses of engineered plants and microorganisms has been controversial in the European Union, where there has been a moratorium on the use of transgenic plants in agriculture and in foods.

3 REGULATION OF GENETICALLY MODIFIED MICROORGANISMS FOR BIOFUELS PROJECTS

3.1 U.S. Environmental Protection Agency Regulation of Industrial Microorganisms

Under the U.S. regulatory framework, most of the genetically engineered microorganisms that have been used in commerce have been regulated by the EPA. Most of these have been microbial pesticides and have been regulated by EPA under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA), and most of EPA's biotech policies have been directed at products falling under this law. Genetically modified microorganisms used for nonpesticidal purposes that are not regulated by any other federal agency are potentially subject to EPA regulation under the Toxic Substance Control Act (TSCA). EPA's TSCA biotechnology policy (40 CFR Part 725 in the Code of Federal Regulations) covers certain microorganisms used in the environment and for industrial purposes and may also cover engineered microbes developed for biofuel production.

TSCA allows EPA to regulate the manufacture of "new" chemicals that are not already in commerce, for purposes not subject to regulation as a pesticide or under the food and drug laws. In the Coordinated Framework, EPA decided to use TSCA in this same "gap-filling" way, to capture those microorganisms that were not regulated by other federal agencies, and EPA issued final regulations under TSCA in 1997 (62 Federal Register 17910-17958) after a long, politically-charged rulemaking process [1,2]. Two main criteria will govern whether a microorganism is subject to TSCA regulations. First, its intended use must fall outside of other federal regulatory authority. Falling under TSCA jurisdiction to date have been nitrogen-fixing agricultural inoculants to promote plant growth, the manufacture of industrial enzymes or other bioproducts, proposed uses of microorganisms for bioremediation or waste treatment, and other industrial uses. Second, the microorganism must be considered to be "new". Under the regulations, new organisms are defined as "intergeneric", meaning that they include coding DNA sequences that have arisen from a taxonomic genus different than that of the host organism.

Under TSCA, manufacturers of chemicals new to commerce must file Pre-Manufacture Notices (PMNs) with EPA at least 90 days prior to the first intended commercial sale or use or importation and must submit all relevant health and safety data in their possession. The biotech rules instituted a similar procedure: new microorganisms used for commercial purposes subject to TSCA's jurisdiction require reporting 90 days in advance of the commercial activity, through Microbial Commercial Activity Notifications (MCANs) that are analogous to the PMNs filed for chemical entities. Certain outdoor research uses are also subject to regulation, but the rules provide several exemptions from MCAN reporting for specific organisms that qualify.

3.2 Impact of EPA Biotech Regulations on Biofuels Projects

The use of intergeneric microorganisms to produce biofuels or to manufacture enzymes for biofuel conversion would likely be subject to the TSCA biotechnology rule. However, only commercial uses will trigger the need for EPA reporting. TSCA is a commercial statute and so most research activities are exempt from reporting requirements. Use of engineered microorganisms in fermentation vessels for research purposes would be exempt from EPA reporting, provided that the fermentation conditions and the company's procedures are sufficient to minimize the potential release of the microbes into the open environment.

Commercial use of intergeneric microorganisms for biofuel production will likely require the filing of an MCAN prior to commencement, and would require the information shown in Table 1. EPA has 90 days to review each MCAN, and if the agency took no regulatory action within that period, the applicant would be free to begin commercial use of the microorganisms.

Regulation of Engineered Microorganisms under TSCA
• Description of the Recipient and New Microorganism.
• Genetic Construction of the New Microorganism.
• Phenotype and Ecological Characteristics of the New Microorganism.
• Production Process, Byproducts.
• Worker Exposure and Environmental Release.
• Health and Environmental Effects Data.

Table 1: Data to Include in MCAN Submissions to EPA under TSCA.

Since the adoption of the TSCA biotechnology rules in 1997, EPA has reviewed 18 MCANs, and aside from one that was withdrawn by the applicant, all these have been cleared for commercialization following EPA review [5]. Before adoption of the rules, EPA reviewed about 60 intergeneric microorganisms through the PMN program, about half of which were environmental introductions and half were for contained manufacturing [5]. Most of the

manufacturing applications have been for the production of industrial enzymes, and none appear to have been for microbial fermentation of biofuels. However, over the years, EPA has issued favorable rulings on the containment status of bioreactors for use with engineered microbes for bioremediation and industrial bioprocessing, and has informally considered the use of modified microorganisms for biofuel production.

4 REGULATION OF TRANSGENIC PLANTS FOR BIOFUELS PROJECTS

4.1 USDA Biotechnology Regulations

Under its biotechnology regulations (7 CFR Part 340 in the Code of Federal Regulations), USDA's Animal and Plant Health Inspection Service (APHIS) uses the Federal Plant Protection Act to regulate outdoor uses of transgenic plants. Technically, these regulations do not cover all genetically engineered plants, and instead cover only those plants engineered to contain sequences from certain microbial genera that contain species that are potential plant pests. In practice, because such "plant pest" sequences are used in most plant transformation procedures, the USDA regulations have covered the vast majority of transgenic plants that have been developed. Originally, permits were required for almost all proposed field uses of genetically engineered plants, and these permits required a detailed description of the modified plants, a description of the proposed field test, and an assessment of the potential environmental effects of the field test. These regulations were substantially relaxed in 1993 (58 Federal Register 17044-17059) and in 1997 (62 Federal Register 23945-23958), and today transgenic varieties of most common agricultural crops can be planted in the field simply upon 30 days advance notice to APHIS. Only less familiar uses of transgenic plants currently require the longer permitting process. In addition, the revised regulations provided a procedure under which applicants can petition to have specific transgenic plant varieties be "delisted" following several years of safe field tests, to proceed to commercial use and sale without the need for yearly permits.

These regulations have been extremely successful in fostering the growth of the U.S. agricultural biotechnology industry, and in allowing field testing and ultimately commercial use of transgenic plants to take place under an orderly, reasonable system. Through the end of 2007, APHIS had authorized over 13,000 field releases of transgenic plants under permits or notifications and has approved 73 delisting petitions for commercialization of engineered crop plants [6]. However, in 2003, USDA began requiring that most proposed outdoor uses of transgenic plants for industrial or pharmaceutical purposes be conducted through the permit process rather than the less burdensome notification procedure [7]. Most biofuels projects using transgenic plants would very likely fall under this requirement, which is described in more detail below. Furthermore, at this writing, USDA is in the midst of a comprehensive review of its regulatory program, and is soon expected to promulgate proposed rules that would revise the biotechnology regulations. The impact of such a revision is unclear, but it is likely to confirm and continue the current policy that industrial uses of transgenic plants would require project-by-project permits.

4.2 Impact of USDA Biotechnology Regulations on Biofuels Projects

USDA's policy requiring permits for industrial and pharmaceutical applications has arisen because it has felt that, as opposed to traditional uses of common crop plants for agricultural purposes, the agency has not yet developed familiarity and a level of scientific comfort with nontraditional uses of transgenic plants. These include production of pharmaceutical compounds or biologics, production of industrially-useful materials, and the uses of plants in hazardous waste clean-up (phytoremediation). Companies developing transgenic plants as potential feedstocks for biofuel production will likely need APHIS permits even for small-scale field testing.

Under the regulations, permit applications must be submitted 120 days before the proposed outdoor use of an engineered plant. Until a company gains significant experience with any given transgenic plant variety, it will be necessary to obtain a permit for each particular field site, as well as each time a new planting is made (although a single permit can cover multiple field sites, and sometimes multiple years). Each permit application must describe the plant, what heterologous DNA it contains and how it was constructed; it must identify and describe the proposed field sites; and it must address the scientific and procedural issues shown in Table 2. Special requirements may also apply for industrial uses of transgenic plants, particularly that the field test must be separated by a "substantial distance" from any sexually compatible crops, and that a 50-foot fallow zone must surround the plot.

Through the end of 2007, APHIS had issued 46 permits for field tests of plants expressing pharmaceutical or industrial products, although only one such permit appears to have been for a biofuel-related application [6]. This was a 2006 permit to Edenspace Systems Corp. for a field test of the "model" species tobacco expressing a microbial endoglucanase gene.

5 STRATEGIES FOR MANAGING REGULATION OF BIOFUEL PROJECTS

The biotechnology regulations should not be a significant obstacle to the use of engineered plants or microorganisms for biofuel production, as evidenced by the many agricultural, industrial and environmental projects that have received clearance under these rules [1,2,8,9]. The most important step in any regulatory plan is early

presubmission consultation with the agency. Applicants should contact the relevant agency well in advance of the anticipated starting date of the project, to discuss the data and other information the agency expects to see, and to begin the discussion on what issues are likely to be of concern to regulators. Agency officials will routinely agree to hold such presubmission meetings, which can be conducted under confidentiality, and they can often provide valuable rulings on the regulatory status of the project prior to formal submission.

Regulation of Transgenic Plants by USDA APHIS
• Stability of vector and introduced genes.
• Presence of infectious, pathogenic, toxic or deleterious functions encoded by introduced DNA.
• Reproduction and pollen/seed dispersal mechanisms.
• Ability to outcross with related species (particularly wild relatives).
• Potential weediness (ability to compete, survive and spread in the environment).
• Need for physical isolation from sexually compatible species.
• Standard Operating Procedures for planting, maintaining and monitoring plants.
• Post-termination scouting for volunteer plants.

Table 2: Issues in the Regulation of Transgenic Plants.

For projects involving engineered microorganisms, early consultation with EPA is especially desirable, to determine whether the intended commercial strain might qualify for an exemption from MCAN reporting, or whether use of the microbes in pilot plants would be considered as research uses not requiring MCAN reporting. If an exemption is not available for proposed commercial use, EPA staff will provide guidance as to the data that is required to be submitted with an MCAN. The company should plan on submitting the MCAN more than 90 days in advance of the anticipated commercial start date, since EPA rules allow the agency to suspend the review period if needed to request more information. Data required for an MCAN is likely to already be in hand or easily obtainable, and with proper planning, the need to submit an MCAN should not be a significant burden to most biofuels projects.

Permit applications for projects involving transgenic plants, including research field tests ,should be filed at least 4 months before the planned starting date, preferably allowing additional time in case the regulators request additional information. Consultation with USDA staff can help applicants develop detailed Standard Operating Procedures for the planting, growth and harvesting of the plants, as well as for the ongoing monitoring of the field test, including inspection and scouting procedures to ensure that "volunteer" transgenic plants do not begin growing on the site after termination. USDA procedures have become much more formalized in recent years, and with advance planning, biofuels companies should have no problem obtaining the needed approvals for field projects with transgenic plants.

6 CONCLUSIONS

The potential applicability of biotechnology regulations to those biofuels projects involving engineered plants and microorganisms might pose additional procedural burdens on applicants that would not be faced by companies using plants or microorganisms developed in more traditional ways. However, with careful advance planning, early agency consultation, and good communication and cooperation between company scientists, process engineers, legal and regulatory staff and management, it should be possible to successfully navigate biotechnology regulations so that these projects are not unduly delayed.

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