PMC-EF2a
(2.04.02)

U.S. DEPARTMENT OF ENERGY
EERE PROJECT MANAGEMENT CENTER
NEPA DETERMINATION

RECIPIENT: Consortium for Plant Biotechnology Research **STATE:** GA

PROJECT TITLE : Energy from Biomass Research and Technology Transfer Program

Funding Opportunity Announcement Number	Procurement Instrument Number	NEPA Control Number	CID Number
DE-FG36-02GO12026	DE-FG36-02GO12026	GFO-08-165f	GO12026

Based on my review of the information concerning the proposed action, as NEPA Compliance Officer (authorized under DOE Order 451.1A), I have made the following determination:

CX, EA, EIS APPENDIX AND NUMBER:
Description:

B3.6 Siting, construction (or modification), operation, and decommissioning of facilities for indoor bench-scale research projects and conventional laboratory operations (for example, preparation of chemical standards and sample analysis); small-scale research and development projects; and small-scale pilot projects (generally less than two years) conducted to verify a concept before demonstration actions. Construction (or modification) will be within or contiguous to an already developed area (where active utilities and currently used roads are readily accessible).

Rational for determination:

The Consortium for Plant Biotechnology Research, Inc. (CPBR) would demonstrate the feasibility of biomass energy and research technologies. The primary goals of CPBR's 2010 Energy from Biomass Research and Technology Transfer Program ("EBRTT") are: (1) to bring together industry, academe, and federal resources for research that would lead to the improved use of biomass and plant feedstocks for the production of fuels such as ethanol and renewable chemical feedstocks; (2) to facilitate technology transfer of the research results; and (3) to facilitate the commercialization of the results. The commercialization objectives are to improve the utilization of plants as energy sources; reduce the cost of renewable energy production; facilitate the replacement of petroleum by plant-based materials; create an energy supply that is safer in its effect on the environment, and contribute to U.S. energy independence. This a continuation of the proposal with CX 3.6 approved 7/31/2008, 8/19/2008, 9/17/2009, 11/25/2008, 3/5/2009, and 11/02/2009 for this project. No field trials of genetically modified plants would take place. The projects proposed are as follows:

1. Co-extraction of bio-oils and protein from biomass by University of Hawaii, Manoa. The work involves characterizing the efficiency of a novel co-solvent system, comprised of a polar covalent molecule (PCM) dissolved in a low melting point salt (i.e., ionic liquid or IL), to co-extract bio-oil and protein from microalgal biomass in terms of the metric lipid to biomass yield (w/w %). An added effort will be to characterize the extent to which the co-solvent treatment sufficiently disrupts the membrane carbohydrate polymers such that their enzymatic degradation to fermentable sugars is efficient. The extraction of bio-oil will be through the process of digesting biomass in various embodiments of IL-PCM co-solvent systems as described in a patent application (UH 0088523-003USO). Dried biomass will be suspended into IL-PCM co-solvent mixtures of varying polarity and subjected to mild heating under reflux at atmospheric pressure. Yields of bio-oils extracted, purity of the biooils, and the extent of protein extracted into the IL-PCM co-solvent will be measured.

2. Crude Glycerol as a Resource for Pyruvic Acid Production by the University of Georgia.
The overall objective of this 2-year project is to demonstrate the production of pyruvate from glycerol. The focus of the first year will be on developing bacterial strains with improved consumption of glycerol. The focus of the second year will be on designing and improving a fermentation process for the conversion of glycerol to pyruvate using the bacterial strains. This will involve laboratory culture of microorganisms and no work outside the laboratory.

This subaward has a pending institutional biosafety committee approval. However, that should not prevent DOE from allowing subawards to all other selectees. This one issue should be resolved soon and there are otherwise no significant impacts for this one project at the University of Georgia; however, this particular project cannot be awarded until the permit is obtained and a copy thereof sent to DOE.

3. Discovery of Novel Genes in Creeping Bentgrass by High Throughput Gene Silencing with Specialized Inverted Repeat cDNA Libraries by Iowa State University. This project requires approval by the Iowa State University Institutional Biosafety Committee (IBC). Approval has been granted under IBC ID #06-d-002-P dated January 12, 2010. This project does not involve the release into the environment (field planting) of a genetically engineered organism. Transgenic materials will be grown in contained environments (growth chambers and secured greenhouse).

4. Soy-Based Promoters for Crop Improvement by Ohio State University and South Dakota State University. The outcomes of this project are straightforward and funding will be applied to the overall goal of identifying and synthesizing soybean promoters. For this research, we will identify and characterize DNA components in 2 new families of soybean promoters and combine them in unique ways to generate novel promoters for regulation of genes of interest to the bioenergy community. Soybean represents the crop with the highest transgenic acreage of any crop with recognized utility as a bioenergy crop (for supplying oils suitable for biodiesel production). Providing novel, high expression soybean promoters to industry and university laboratories could contribute significantly to the biotechnological improvement of this important crop.

5. Improving Rice Photosynthesis and Yield by Montana State University. The funding would be applied to the production and testing of transgenic plants with modified levels of leaf starch. Transformation vectors containing genes controlling leaf starch biosynthesis will be transferred into rice cells using Agrobacterium. Once the transgenic plants are obtained, we will conduct experiments designed to test the effect of increased leaf starch upon whole plant growth.

6. Branch-chain fatty acid production in plants by the University of Kentucky. The purpose of this project is to develop renewable, environmentally friendly lubricants such as motor oils that can meet today's high performance automobile and other engine specifications. Plant oils can currently be used as automobile engine oils but only for short periods and do not meet automotive manufacturer's specifications due to the poor oxidative stability of the double bonds of the unsaturated fatty acids. Solutions to this problem include converting the double bonds commonly found in plant oils such as soybean oil into branch-chain groups. Such groups can have the adequate low temperature fluidity and lubricity + the needed very high oxidative stability. Therefore the overall goal of this research is to engineer plants to convert common unsaturated fatty acids that normally accumulate in seed oils (TAGs) into saturated branch-chain groups and selectively transfer these moieties into oil that can accumulate in plant tissues such as seeds. Plant oils containing high levels of branched chain fatty acids (BCFAs) will be developed by transferring the natural ability of some organisms to make such fatty acids and oils into oilseed crops that can be economically produced in the US. The specific objectives are: 1) Cloning and characterization of genes encoding branch-chain fatty acid biosynthetic enzymes from source organisms. 2) Cloning and characterization of enzymes involved in the accumulation of branch-chain fatty acids from source organisms. 3) Characterization of model plants and oilseeds engineered with enzymes involved in the formation and accumulation of branch-chain fatty acids. This will convert normal plant oil fatty acids into forms with superior industrial lubricant properties; high oxidative stability with good flow properties over a wide range of temperatures.

7. New genes for stress tolerance in bioenergy crop plants by Iowa State University. The goal of the project is to develop a RNA splicing assay to monitor environmental stress responses in corn and to determine what factors are responsible for the splicing reaction. The design phase of the project has already been completed in the development of this project. Most of the project will involve laboratory experimentation and data analysis. No construction, capital purchases or equipment installation/modification will be involved in this project.

8. Manipulation of ploidy levels in Miscanthus species by University of Illinois.
The overall objective of this project is to provide access to genetic variability in Miscanthus species for feedstock crop improvement by overcoming sterility and inter-specific hybridization barriers through the manipulation of Miscanthus genome ploidy levels. The specific objectives of this proposed project are to: Objective 1: Conduct chromosome doubling of triploid M. scanthus x giganteus accessions to generate hexaploid plants and of M. x giganteus' diploid parent accessions, M. sinensis and M. sacchariflorus, to create tetraploids. Evaluate growth rates, biomass accumulation, fertility and seed set of all Miscanthus chromosome doubled lines. Objective 2: Conduct controlled pollinations among chromosome-doubled hexaploid M. x giganteus lines, and between M. sinensis and M. sacchariflorus tetraploids to generate segregating hexaploid and tetraploid progeny. Objective 3: Using hexaploid M. x giganteus regenerate via anther culture, segregating triploid and sterile M. x giganteus plants and evaluate for biomass productivity.

Year1: Quantify nuclear DNA content of Miscanthus species accessions by flow cytometry. Initiate chromosome doubling of ten M. sinensis, five M. sacchariflorus and our three existing M. x giganteus accessions. Regenerate tetraploid and hexaploid M. sinensis, M. sacchariflorus, and M. x giganteus plantlets and test for DNA content and chromosome number. Grow out plants in the greenhouse. Initiate anther culture of hexaploid M. x giganteus to regenerate segregating triploids.

Year 2: Continue regeneration of triploid (from anther culture) and hexaploid M. x giganteus and tetraploid M. sinensis and M. sacchariflorus plants. Initiate greenhouse evaluation of biomass growth rates, cell wall composition, and pollen

viability of doubled lines. Initiate greenhouse hybridizations between different M. x giganteus hexaploids and M. sinensis and M. sacchariflorus tetraploids to attempt to recover fertile inter-specific hybrids. Continue to conduct hybridizations and greenhouse evaluation of all new polyploids. Due to time constraints associated with plant tissue culture regeneration and plant initiation of flowering to generate hybridized populations field evaluations of polyploids and populations from crosses will be not be field tested until after the end of this subaward.

All plant materials are native plants. NO recombinant plant materials are being used; hence no institutional biosafety permits are required.

9. Low Cost Conversion of Papermaking Sludge to Ethanol by North Carolina State University
This will be strictly laboratory work. The investigation will run several tests to evaluate different methods for separating carbohydrates from the inorganic components in paper sludge. The carbohydrate stream will then be evaluated based on the suitability for the production of films suitable for plastic film type applications as well as bioethanol. In addition, the project will investigate how the addition of waxes and nano-sized clays to the carbohydrate stream will affect the water resistivity. The project intends to identify an effective process that will utilize paper sludge to produce biofuels and biomaterials. All safety standards and regulations set in place by the university will be followed during this project.
10. Engineering Soybean Nematode Resistance by University of Illinois. The research is designed to enhance the genetic modification of soybean lines. There will be no attempt to introduce any of the transgenic plants into soybean field crops. The results will be used to confirm the hypothesis that modifying the shikimate pathway to develop a durable soybean line with resistance to the phytoparasitic nematodes will improve soybean yield. The governing body at most Universities for such R&D often is an institutional biosafety or biotechnology committee. Such an organization exists at University of Illinois and their approval has already been obtained and we can assume they will monitor such work regularly.

11. Practical Waterborne Agricultural Oil-Based Coatings by Iowa State University. This is strictly laboratory level R&D. Vegetable oil-based polymers will be combined with biocomposites additives such as distillers dried grains and solubles (DDGS), corn stover and lignin to create bioplastic composites. This project intends to investigate the processing requirements and properties of the bioplastic composites. The work for this project will be completed in a laboratory on Iowa State University campus. All standards and regulations set in place by the university will be followed during this project.

12. Sustainable Wood-Based Barrier Films for Liquid Packaging Applications by Iowa State University. This is strictly laboratory R&D work. The project will be grinding hardwood bleached kraft pulp into micro-fibrillated cellulose with and without pretreatment of various types of starches. In addition, layered paper sheets will be created using paper and microfibrillated cellulose (MFC) coating in combination with different additives added to the coating. This will investigate the optimum operating variables of the production of MFC and how the resulting MFC creates films of different properties for packaging and container products. Proposed tasks: 2.1 – Create layered paper sheets using a base thickness for the paper and varying thickness of the MFC coating. 2.2 – Add various additives to the MFC coating to improve strength, impart water resistance, and create barrier characteristics. 2.3 – Test the above materials to determine the effect of coating weight, and additives on imparting strength and barrier properties to the paper sheet.

13. Lignin-reduced and cellulose-expressing switchgrass by North Carolina State University. This is strictly laboratory work. The research is designed to genetically engineer switchgrass to decrease the lignin content within the cell structure. There will be 100 transgenic plants that will be developed in the laboratory and grown in a greenhouse. All transgenic material will be destroyed by autoclaving upon the conclusion of the project. The intent of the research is to enhance the biomass-to-biofuel conversion efficiency. The governing body at most universities for such R&D is an institutional biosafety or biotechnology committee. Such an organization exists at North Carolina State University and their approval has already been obtained and we can assume they will monitor such work regularly.

14. Molecular characterization of composition traits in sweet and energy sorghum germplasm for bioenergy production by Texas A&M University. This project will be conducted in both greenhouses and the laboratory. This project will involve collecting samples of different genotypes of sorghum grown in seven locations across the U.S., along with agronomic data to assess the relative affects of genotype and the environment have on the composition of the sorghum. This is intended to identify and localize genetic factors that influence composition for energy sorghum. The work will be located in research and extension centers throughout the state of Texas where there are established production practices in place for agricultural evaluation and testing. There is no transgenic material being evaluated in this study, as all the sorghum evaluated for this study was developed using conventional breeding approaches.

15. Cellulose Degrading Enzymes from Western Corn Rootworm Larvae by University of Nebraska, Lincoln. This is strictly lab work. This research will involve examining Western Corn Rootworm larvae and characterizing the midgut transcriptome to identify potential sources of cellulose degrading enzymes. Between 100-200 individual third instar Western Corn Rootworm larvae will be sampled. The governing body at most universities for such R&D often is an institutional biosafety or biotechnology committee. Such an organization exists at the University of Nebraska and their approval has already been obtained and we can assume they will monitor such work regularly.

16. Paper Mill Waste Bioconversion to Value-added Products by Kansas State University. This is strictly lab work. This research will involve examining the next generation of biomass enzymes to maximize the enzyme dosage for efficient sugar release and to study the effect of paper mill sludge ash on saccharification. Work will also include isolating microbial cultures for effective bioconversion of glucan and xylan components to L (+) lactic acid via simultaneous saccharification and fermentation, determining the commercial feasibility and scalability for enhanced L(+) lactic acid production, and developing fermentation processes for bioconversion of sugars derived from paper mill sludge to D(-) lactic acid using microbial cultures. The experiments are to develop the potential for manufacturing biobased chemicals with various applications from renewable paper mill sludge. All standards and regulations set in place by the university will be followed during this project.

Based on the information above, this project's impacts to the human and natural environment can be deemed less than significant and this project would qualify for Categorical Exclusions B3.6, and B5.1.

NEPA PROVISION

DOE has made a conditional NEPA determination for this award, and funding for certain tasks under this award is contingent upon the final NEPA determination.

Insert the following language in the award:

You are restricted from taking any action using federal funds, which would have an adverse affect on the environment or limit the choice of reasonable alternatives prior to DOE/NNSA providing either a NEPA clearance or a final NEPA decision regarding the project.

Prohibited actions include:
Subgrant award funding to University of Georgia for their Crude Glycerol as a Resource for Pyruvic Acid Production Project until a copy of a current institutional biosafety committee approval permit is provided to the DOE.
This restriction does not preclude you from:
Awarding funding to all other sub-grantees.
If you move forward with activities that are not authorized for federal funding by the DOE Contracting Officer in advance of the final NEPA decision, you are doing so at risk of not receiving federal funding and such costs may not be recognized as allowable cost share.

Note to Specialist :

None Given.

SIGNATURE OF THIS MEMORANDUM CONSTITUTES A RECORD OF THIS DECISION.

NEPA Compliance Officer Signature: _____ Date: 8/23/10
NEPA Compliance Officer

FIELD OFFICE MANAGER DETERMINATION

☐ Field Office Manager review required

NCO REQUESTS THE FIELD OFFICE MANAGER REVIEW FOR THE FOLLOWING REASON:

☐ Proposed action fits within a categorical exclusion but involves a high profile or controversial issue that warrants Field Office Manager's attention.
☐ Proposed action falls within an EA or EIS category and therefore requires Field Office Manager's review and determination.

BASED ON MY REVIEW I CONCUR WITH THE DETERMINATION OF THE NCO :

Field Office Manager's Signature: _____ Date: _____
Field Office Manager