

Program Announcement To DOE National Laboratories

LAB 00-18

Microbial Genome Program

The Office of Biological and Environmental Research (OBER) of the Office of Science (SC), U.S. Department of Energy (DOE), hereby announces its interest in receiving proposals in support of the Microbial Genome Program (MGP), focused on microbes of interest to the DOE, e.g. those involved in environmental processes, including waste remediation, carbon management, energy production and biotechnology. This announcement is focused on 1) whole genome functional analyses of genomic information from microorganisms; 2) bioinformatics tools for microbial genome annotation; 3) characterization of microbial genomic plasticity, e.g. lateral gene transfers and other forms of genomic information transfer; 4) novel technologies for comparative microbial genome sequencing that exploit previously sequenced microbial genomes; and 5) technologies to assess consortia and environmental diversity of hard-to-culture microbes. This announcement represents a significant departure from past MGP announcements in that the DOE will not solicit proposals to continue high throughput sequencing of microbial genomes. Rather, this is a shift in emphasis to exploiting already sequenced genomes to address DOE mission needs.

The Microbial Genome Program (MGP) supports key DOE business areas by providing microbial DNA sequence information that will further the understanding and application of microbiology relating to energy production, chemical and materials production, environmental carbon management, and environmental cleanup. The elucidation of microbial genome sequences is a natural outgrowth of past and current Biological and Environmental Research (BER) Programs, including DNA sequencing from the Human Genome Program, structural biology studies utilizing BER-supported facilities and synchrotrons located at DOE laboratories, and molecular microbiological research supported by BER environmental programs. The MGP benefits directly from capabilities at DOE national laboratories, DOE and National Institutes of Health Human Genome Centers, the National Center for Biotechnology Information (NCBI) at the National Institutes of Health (NIH), and the capabilities of universities and non-profits. The MGP represents a considerable interdisciplinary effort and will contribute to and draw from a wide variety of public and private programs. Over the last 5 years, sequencing of microorganisms that live in extreme environments (including the deep subsurface, geothermal environments, hypersaline environments, high-radiation environments, and toxic waste sites) has provided a considerable information base for scientific research related not only to DOE missions but also to other federal agency missions, and U.S. industry. Proposals are now being

sought in five complementary areas: whole-genome functional analyses, bioinformatics applied to microbial genome sequences, characterization of microbial genomic plasticity, novel microbial sequencing approaches, and the characterization of the diversity of microbial consortia and/or hard-to-culture microbes that mediate processes of relevance to the DOE. Each proposal must clearly state which area is being addressed; if a proposer wishes to address more than one area, the proposal must clearly describe the expected advantages of an integrated approach.

Candidate microorganisms for study can comprise archaea, bacteria, or communities made up of bacteria and/or archaea that mediate or catalyze metabolic events of energy or environmental importance. Preference will be given to those proposers using microbes for which complete or near-complete genomic sequencing information in the public domain exists. (See <http://www.ornl.gov/microbialgenomes/organisms.html> for a current list of microbes that have been and are being sequenced.) Priority will be given to studies on those microbes that can bioremediate metals and radionuclides, microbes that can degrade significant biopolymers such as celluloses and lignins or microbes that are involved in environmental carbon management, e.g. fix or sequester CO₂. Finally, microbes that participate in consortia with already-sequenced species are of interest. Strict pathogens or parasites will not be considered.

1) **Functional Analysis.** It is presently difficult, and in many instances impossible, to predict biological function from microbial genomic sequence data, even when the entire genome has been sequenced and published and is available for inspection. Better experimental and computational methods are needed to identify novel open reading frames and predict their functions at a whole-genome scale, particularly from completely sequenced microbial genomes. Accordingly, proposals are requested that will develop better ways to interpret sequence data from novel open reading frames, and even whole genomes, using both comparative genomic approaches as well as novel analyses. The DOE MGP is particularly interested in the use of sequence data for whole genome approaches to functional prediction, functional regulation, functional categorization (e.g. transporters, environmental sensors, redox enzymes, cytoskeletal components, DNA repair systems, metal reductases, biodegradative enzymes, etc.) as well as those approaches that identify and distinguish rare or unique ORFs that can be linked to restricted environmental niches or DOE-relevant bioremediation capacities. Identification of domains in gene sequences that mediate protein- protein interactions are also of great interest. Proposers should focus on microbes of mission interest to the DOE, as described above. It is estimated that between four and six awards for a total of

up to \$1 million could be available for this area in FY 2001, contingent upon the availability of appropriated funds.

2) **Bioinformatics.** It is estimated that by December 2000, completed genomic sequences of perhaps 50 archaea and bacteria will be publicly available, more than a third of them as a direct result of DOE Microbial Genome Program funding. In June 2000, a draft sequence for the entire human genome became available as well. For several microbes, complete sequences of close evolutionary relatives now or will soon exist. Computational comparative genomics can illuminate evolutionary pathways to complement traditional phenotype-based analyses, provide data for the prediction of gene function between organisms, and contribute to modeling pathways. The value of such comparative functional analysis is highlighted by the remarkable frequency of novel open reading frames in microbial genome sequences (up to half the genes in many cases) that currently lack any annotation. The evolutionary conservation of open reading frames and certain protein functions between microbes and more complex organisms (including human) emphasizes the value of microbial sequences for understanding the functions of uncharacterized microbial (and, potentially, human) genes. To this end, computational methods for interspecies genomic comparisons are an area of particular interest for this solicitation. Proposals are requested that propose ways in which microbial sequence data from all sources can be analyzed, compared, annotated, and used to predict the function of homologous genes in both prokaryotic and eukaryotic organisms. Thus, this announcement solicits proposals for research into:

- a) novel computational tools to increase the value of microbial genomic information, such as improved techniques for identifying distant sequence homologies, reconstructing phylogenetic trees, predicting gene function, or identifying and modeling gene expression networks, and
- b) algorithms and tools to extract longer stretches, and make more accurate base calls from current sequencing procedures in order to assist the closure process for microbial genomes.

Of special interest will be methods that use unique DOE resources in massively parallel, high-capacity supercomputers (machines in the multi-teraflop range). It is expected that computational tools developed under these awards will be widely distributed to the scientific community (e.g. via a WWW site) and some level of user support will be available. It is anticipated that between two and five awards for a total of up to \$1 million could be available for this area in FY 2001, contingent upon the availability of appropriated funds.

3) Characterization of Microbial Genomic Plasticity. Completed sequences for several microbes (e.g. *Thermotoga maritima*, (Nelson, K. et. al. (1999) *Nature* 399: 323-329) and *Deinococcus radiodurans* (White, O., et.al. *Science* (1999) 286:1571-1577) strongly suggest that entire blocks of genes have been laterally transferred during microbial evolution, even from sources in different biological kingdoms. How widespread this phenomenon may be, or any evolutionary constraints on it, is unknown. Proposals are solicited that would assess lateral gene exchanges, in terms of its frequency in different environmental niches, the mechanisms involved, as well as the circumstances in which it is observed. It is anticipated that between two and four awards totaling up to \$1 million could be available for this area in FY 2001, contingent upon the availability of appropriated funds.

4) Novel Approaches to Microbial Genomic Sequencing. Many microorganisms that are closely related by means of phylogenetic measures (e.g., 16S rRNA comparisons) display dramatic differences in phenotypic characteristics. Such differences can be chromosomal in origin, or they can be due to extrachromosomal genetic elements. The DOE MGP is interested in novel comparative sequencing approaches that exploit the completed sequence of one microorganism to efficiently determine the sequence of a related taxon or species. This element of this announcement could contribute to:

- a) new methods to accelerate genomic comparisons, without resequencing the entire genome of the related organism *de novo* (technologies up to the proof-of-principle stage are eligible for support). Technologies responsive to this element of this solicitation should be firmly grounded in already completed microbial sequencing projects; these may include subtractive hybridization approaches, or “DNA chips”, among others, but it is not the aim of this solicitation to support completely untested technologies;
- b) strategies to more efficiently identify specific sequence features associated with phenotypic differences; and
- c) techniques to characterize and quantify lateral gene transfer (especially any correlation with environmental selection).

A plan for making comparative sequence data publicly available by deposition into a community-accessible sequence database within three months of data acquisition must be included. A plan for efficient and timely annotation must be included in the Project Description. DOE expects that awardees will make all good faith efforts to publish in the open scientific literature the results of

their funded work, including the genome sequences of microbes sequenced under this announcement. (DOE data release requirements, a condition of any award, are available at: <http://www.sc.doe.gov/production/ober/EPR/data.html>). Proposers are encouraged to create process- and cost-effective partnerships that will maximize sequence data production and analysis, data dissemination, and progress towards understanding basic biological mechanisms that can further the development of biotechnology. It is anticipated that between two and four awards totaling up to \$1 million could be available for this area in FY 2001, contingent upon the availability of appropriated funds.

5) Consortia and Hard-to-Culture Microbes. Most of our current knowledge of microbiology is derived from individual species that either cause diseases or grow easily and readily as monocultures under laboratory conditions and are thus easy to study. The preponderance of species in the environment does neither and is thus largely unknown to science. Most are thought to grow as part of interdependent consortia in which one species supplies a nutrient necessary for the growth of another. Virtually nothing is known of the organization, membership, or functioning of these consortia, especially those involved in environmental processes in which DOE is interested. Technologies are sought that enable genomic analyses of microbial consortia as well as analyses of the genomic information content and diversity of those species that have proven refractory to laboratory culture but are plentiful in environments challenged with metal and radionuclide wastes, or involved in carbon sequestration. It is anticipated that between two and three awards totaling up to \$1 million could be available for this area in FY 2001, contingent upon the availability of appropriated funds.

Preproposals

Potential proposers are strongly encouraged to submit a brief preproposal that consists of two to three pages of narrative describing the research objectives and technical approach(s). Preproposals will be reviewed relative to the scope and research needs of the OBER Microbial Genome Program, as outlined above. The preproposal should identify, on the cover sheet, the title of the project, the institution, principal investigator name, telephone, fax, and e-mail address. A response to each preproposal discussing the potential programmatic relevance of a formal proposal will be communicated to the Principal Investigator within 14 to 21 days of receipt. Any renewal proposals must include a list of publications resulting from previous DOE Microbial Genome Program funding.

Program Funding

It is anticipated that up to \$5 million will be available for all MGP awards in Fiscal Year 2001; from twelve to as many as twenty-five awards are anticipated, contingent on availability of appropriated funds in FY 2001 and the size of the awards. Multiple year funding is expected, also contingent on availability of funds and progress of the research. Awards are expected to range from \$200,000 to \$1 million per year, total costs, with terms of one to three years.

DATES: Preproposals referencing Program Announcement 00-18 should be received by October 2, 2000.

Formal proposals in response to this Announcement should be received by 4:30 p.m., E.S.T., December 14, 2000, to be accepted for merit review and funding in FY 2001.

ADDRESSES: Preproposals referencing Program Announcement 00-18 should be sent to Dr. Daniel W. Drell, Office of Biological and Environmental Research, SC-72, Office of Science, U.S. Department of Energy, 19901 Germantown Road, Germantown, MD 20874-1290; e-mail is acceptable for submitting preproposals using the following address: joanne.corcoran@science.doe.gov.

Formal proposals referencing Program Announcement 00-18, should be forwarded to: U.S. Department of Energy, Office of Science, Office of Biological and Environmental Research, SC- 72, 19901 Germantown Road, Germantown, MD 20874-1290, ATTN: Program Announcement 00-18. This address must be used when submitting proposals by U.S. Postal Service Express Mail or any commercial mail delivery service, or when hand-carried by the proposer.

FOR FURTHER INFORMATION CONTACT: Dr. Daniel W. Drell, SC-72, Office of Biological and Environmental Research, Office of Science, U.S. Department of Energy, 19901 Germantown Road, Germantown, MD 20874-1290, telephone: (301) 903-4742, e-mail: daniel.drell@science.doe.gov.

Submission Information

The Project Description must be 20 pages or less, exclusive of attachments. It must contain an abstract or project summary on a separate page with the name of the proposer, mailing address, phone FAX and E-mail listed. The proposal must include letters of intent from collaborators (briefly describing the intended contribution of each to the research), and short curriculum vitae, consistent with NIH guidelines, for the proposer and any co-PIs.

DOE policy requires that potential proposers adhere to 10 CFR Part 745 "Protection of Human Subjects", or such later revision of those guidelines as may be published in the Federal Register.

Any recipient of an award from SC performing research involving recombinant DNA molecules and/or organisms and viruses containing recombinant DNA molecules shall comply with NIH "Guidelines for Research Involving Recombinant DNA Molecules," which is available via the world wide web at:

<http://www.niehs.nih.gov/odhsb/biosafe/nih/rdna-apr98.pdf>, (59 FR 34496, July 5, 1994,) or such later revision of those guidelines as may be published in the Federal Register.

Other useful web sites include:

MGP Home Page - <http://www.er.doe.gov/production/ober/microbial.html>

DOE Joint Genome Institute Microbial Web Page - http://www.jgi.doe.gov/JGI_microbial/html/

GenBank Home Page - <http://www.ncbi.nlm.nih.gov/>

Human Genome Home Page - <http://www.ornl.gov/hgmis>

The instructions and format described below should be followed. Reference Program Announcement LAB 00-18 on all submissions and inquiries about this program.

OFFICE OF SCIENCE
GUIDE FOR PREPARATION OF SCIENTIFIC/TECHNICAL PROPOSALS
TO BE SUBMITTED BY NATIONAL LABORATORIES

Proposals from National Laboratories submitted to the Office of Science (SC) as a result of this program announcement will follow the Department of Energy Field Work Proposal process with additional information requested to allow for scientific/technical merit review. The following guidelines for content and format are intended to facilitate an understanding of the requirements necessary for SC to conduct a merit review of a proposal. Please follow the guidelines carefully, as deviations could be cause for declination of a proposal without merit review.

1. Evaluation Criteria

Proposals will be subjected to formal merit review (peer review) and will be evaluated against the following criteria which are listed in descending order of importance:

Scientific and/or technical merit of the project

Appropriateness of the proposed method or approach

Competency of the personnel and adequacy of the proposed resources

Reasonableness and appropriateness of the proposed budget

The evaluation will include program policy factors such as the relevance of the proposed research to the terms of the announcement, the uniqueness of the proposer's capabilities, and demonstrated usefulness of the research for proposals in other DOE Program Offices as evidenced by a history of programmatic support directly related to the proposed work.

2. Summary of Proposal Contents

Field Work Proposal (FWP) Format (Reference DOE Order 5700.7C) (DOE ONLY)

Proposal Cover Page

Table of Contents

Abstract

Narrative

Literature Cited

Budget and Budget Explanation

Other support of investigators

Biographical Sketches

Description of facilities and resources

Appendix

2.1 Number of Copies to Submit

An original and seven copies of the formal proposal/FWP must be submitted.

3. Detailed Contents of the Proposal

Proposals must be readily legible, when photocopied, and must conform to the following three requirements: the height of the letters must be no smaller than 10 point with at least 2 points of spacing between lines (leading); the type density must average no more than 17 characters per inch; the margins must be at least one-half inch on all sides. Figures, charts, tables, figure legends, etc., may include type smaller than these requirements so long as they are still fully legible.

3.1 Field Work Proposal Format (Reference DOE Order 5700.7C) (DOE ONLY)

The Field Work Proposal (FWP) is to be prepared and submitted consistent with policies of the investigator's laboratory and the local DOE Operations Office. Additional information is also requested to allow for scientific/technical merit review.

Laboratories may submit proposals directly to the SC Program office listed above. A copy should also be provided to the appropriate DOE operations office.

3.2 Proposal Cover Page

The following proposal cover page information may be placed on plain paper. No form is required.

Title of proposed project
SC Program announcement title
Name of laboratory
Name of principal investigator (PI)
Position title of PI
Mailing address of PI
Telephone of PI
Fax number of PI
Electronic mail address of PI
Name of official signing for laboratory*
Title of official
Fax number of official
Telephone of official
Electronic mail address of official
Requested funding for each year; total request
Use of human subjects in proposed project:

If activities involving human subjects are not planned at any time during the proposed project period, state "No"; otherwise state "Yes", provide the IRB Approval date and Assurance of Compliance Number and include all necessary information with the proposal should human subjects be involved.

Use of vertebrate animals in proposed project:

If activities involving vertebrate animals are not planned at any time during this project, state "No"; otherwise state "Yes" and provide the IACUC Approval date and Animal Welfare Assurance number from NIH and include all necessary information with the proposal.

Signature of PI, date of signature
Signature of official, date of signature*

*The signature certifies that personnel and facilities are available as stated in the proposal, if the project is funded.

3.3 Table of Contents

Provide the initial page number for each of the sections of the proposal. Number pages consecutively at the bottom of each page throughout the proposal. Start each major section at the top of a new page. Do not use unnumbered pages and do not use suffices, such as 5a, 5b.

3.4 Abstract

Provide an abstract of no more than 250 words. Give the broad, long-term objectives and what the specific research proposed is intended to accomplish. State the hypotheses to be tested. Indicate how the proposed research addresses the SC scientific/technical area specifically described in this announcement.

3.5 Narrative

The narrative comprises the research plan for the project and is limited to 25 pages. It should contain the following subsections:

Background and Significance: Briefly sketch the background leading to the present proposal, critically evaluate existing knowledge, and specifically identify the gaps which the project is intended to fill. State concisely the importance of the research described in the proposal. Explain the relevance of the project to the research needs identified by the Office of Science. Include references to relevant published literature, both to work of the investigators and to work done by other researchers.

Preliminary Studies: Use this section to provide an account of any preliminary studies that may be pertinent to the proposal. Include any other information that will help to establish the experience and competence of the investigators to pursue the proposed project. References to appropriate publications and manuscripts submitted or accepted for publication may be included.

Research Design and Methods: Describe the research design and the procedures to be used to accomplish the specific aims of the project. Describe new techniques and methodologies and explain the advantages over existing techniques and

methodologies. As part of this section, provide a tentative sequence or timetable for the project.

Subcontract or Consortium Arrangements: If any portion of the project described under "Research Design and Methods" is to be done in collaboration with another institution, provide information on the institution and why it is to do the specific component of the project. Further information on any such arrangements is to be given in the sections "Budget and Budget Explanation", "Biographical Sketches", and "Description of Facilities and Resources".

3.6 Literature Cited

List all references cited in the narrative. Limit citations to current literature relevant to the proposed research. Information about each reference should be sufficient for it to be located by a reviewer of the proposal.

3.7 Budget and Budget Explanation

A detailed budget is required for the entire project period, which normally will be three years, and for each fiscal year. It is preferred that DOE's budget page, Form 4620.1 be used for providing budget information*. Modifications of categories are permissible to comply with institutional practices, for example with regard to overhead costs.

A written justification of each budget item is to follow the budget pages. For personnel this should take the form of a one-sentence statement of the role of the person in the project. Provide a detailed justification of the need for each item of permanent equipment. Explain each of the other direct costs in sufficient detail for reviewers to be able to judge the appropriateness of the amount requested.

Further instructions regarding the budget are given in section 4 of this guide.

* Form 4620.1 is available at web site:

<http://www.sc.doe.gov/production/grants/forms.html>

3.8 Other Support of Investigators

Other support is defined as all financial resources, whether Federal, non-Federal, commercial or institutional, available in direct support of an individual's research endeavors. Information on active and pending other support is required for all senior personnel, including investigators at collaborating institutions to be funded by a subcontract. For each item of other support, give the organization or agency, inclusive

dates of the project or proposed project, annual funding, and level of effort devoted to the project.

3.9 Biographical Sketches

This information is required for senior personnel at the laboratory submitting the proposal and at all subcontracting institutions. The biographical sketch is limited to a maximum of two pages for each investigator.

3.10 Description of Facilities and Resources

Describe briefly the facilities to be used for the conduct of the proposed research. Indicate the performance sites and describe pertinent capabilities, including support facilities (such as machine shops) that will be used during the project. List the most important equipment items already available for the project and their pertinent capabilities. Include this information for each subcontracting institution, if any.

3.11 Appendix

Include collated sets of all appendix materials with each copy of the proposal. Do not use the appendix to circumvent the page limitations of the proposal. Information should be included that may not be easily accessible to a reviewer.

Reviewers are not required to consider information in the Appendix, only that in the body of the proposal. Reviewers may not have time to read extensive appendix materials with the same care as they will read the proposal proper.

The appendix may contain the following items: up to five publications, manuscripts (accepted for publication), abstracts, patents, or other printed materials directly relevant to this project, but not generally available to the scientific community; and letters from investigators at other institutions stating their agreement to participate in the project (do not include letters of endorsement of the project).

4. Detailed Instructions for the Budget

(DOE Form 4620.1 "Budget Page" may be used)

4.1 Salaries and Wages

List the names of the principal investigator and other key personnel and the estimated number of person-months for which DOE funding is requested. Proposers should list the number of postdoctoral associates and other professional positions included in the proposal and indicate the number of full-time-equivalent (FTE) person-months and

rate of pay (hourly, monthly or annually). For graduate and undergraduate students and all other personnel categories such as secretarial, clerical, technical, etc., show the total number of people needed in each job title and total salaries needed. Salaries requested must be consistent with the institution's regular practices. The budget explanation should define concisely the role of each position in the overall project.

4.2 Equipment

DOE defines equipment as "an item of tangible personal property that has a useful life of more than two years and an acquisition cost of \$5000 or more." Special purpose equipment means equipment which is used only for research, scientific or other technical activities. Items of needed equipment should be individually listed by description and estimated cost, including tax, and adequately justified. Allowable items ordinarily will be limited to scientific equipment that is not already available for the conduct of the work. General purpose office equipment normally will not be considered eligible for support.

4.3 Domestic Travel

The type and extent of travel and its relation to the research should be specified. Funds may be requested for attendance at meetings and conferences, other travel associated with the work and subsistence. In order to qualify for support, attendance at meetings or conferences must enhance the investigator's capability to perform the research, plan extensions of it, or disseminate its results. Consultant's travel costs also may be requested.

4.4 Foreign Travel

Foreign travel is any travel outside Canada and the United States and its territories and possessions. Foreign travel may be approved only if it is directly related to project objectives.

4.5 Other Direct Costs

The budget should itemize other anticipated direct costs not included under the headings above, including materials and supplies, publication costs, computer services, and consultant services (which are discussed below). Other examples are: aircraft rental, space rental at research establishments away from the institution, minor building alterations, service charges, and fabrication of equipment or systems not available off-the-shelf. Reference books and periodicals may be charged to the project only if they are specifically related to the research.

a. Materials and Supplies

The budget should indicate in general terms the type of required expendable materials and supplies with their estimated costs. The breakdown should be more detailed when the cost is substantial.

b. Publication Costs/Page Charges

The budget may request funds for the costs of preparing and publishing the results of research, including costs of reports, reprints page charges, or other journal costs (except costs for prior or early publication), and necessary illustrations.

c. Consultant Services

Anticipated consultant services should be justified and information furnished on each individual's expertise, primary organizational affiliation, daily compensation rate and number of days expected service. Consultant's travel costs should be listed separately under travel in the budget.

d. Computer Services

The cost of computer services, including computer-based retrieval of scientific and technical information, may be requested. A justification based on the established computer service rates should be included.

e. Subcontracts

Subcontracts should be listed so that they can be properly evaluated. There should be an anticipated cost and an explanation of that cost for each subcontract. The total amount of each subcontract should also appear as a budget item.

4.6 Indirect Costs

Explain the basis for each overhead and indirect cost. Include the current rates.