

## SBIR/STTR STUDY AND CONTROL OF MICROBIAL BIOFILMS

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P.T.

National Heart, Lung, and Blood Institute  
National Institute of Dental and Craniofacial Research  
National Institute of Allergy and Infectious Diseases  
National Institute on Deafness and Other Communication Disorders  
National Institute of Arthritis and Musculoskeletal and Skin Diseases  
National Institute of General Medical Sciences  
National Institute of Diabetes and Digestive and Kidney Diseases  
National Institute of Child Health and Human Development  
National Institute of Neurological Disorders and Stroke  
National Institute of Nursing Research  
Office of Research on Women's Health

Application Receipt Dates:

Applications will be accepted for SBIR proposals three times a year with the following deadlines:  
December 15; April 15; August 15.

Applications will be accepted for STTR proposals three times a year with the following deadlines:  
December 1; April 1; August 1.

### PURPOSE

The National Heart, Lung, and Blood Institute (NHLBI), National Institute of Dental and Craniofacial Research (NIDCR), National Institute of Allergy and Infectious Diseases (NIAID), National Institute on Deafness and Other Communication Disorders (NIDCD), National Institute of Arthritis and Musculoskeletal and Skin Diseases (NIAMS), National Institute of General Medical Sciences (NIGMS), National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), National Institute of Child Health and Human Development (NICHD), National Institute of Neurological Disorders and Stroke (NINDS), National Institute of Nursing Research (NINR), and

Office of Research on Women's Health (ORWH) invite research grant applications to conduct studies on microbial biofilms leading to improved strategies and technologies to diagnose, prevent and treat biofilm-associated infectious diseases.

This program announcement must be read in conjunction with the "Omnibus Solicitation of the Public Health Service for Small Business Innovation Research Grant Applications (PHS 99-2)," and the "Omnibus Solicitation of the National Institutes of Health for Small Business Technology Transfer Grant Applications (PHS 99-3)." All of the instructions within the Omnibus Solicitations apply.

#### HEALTHY PEOPLE 2000

The Public Health Service (PHS) is committed to achieving the health promotion and disease prevention objectives of "Healthy People 2000," a PHS-led national activity for setting priority areas. This Program Announcement, SBIR/STTR Study and Control of Microbial Biofilms, is related to the priority areas of oral health, immunization and infectious diseases, unintentional injuries, diabetes and chronic disabling conditions, special population objectives, and heart disease and stroke. Potential applicants may obtain a copy of "Healthy People 2000" at <http://www.crisny.org/health/us/health7.html>

#### ELIGIBILITY REQUIREMENTS

Eligibility requirements for SBIR and STTR are described in the Omnibus Solicitation of the NIH for SBIR/STTR grant applications.

#### MECHANISM OF SUPPORT

A. FAST-TRACK APPLICATIONS. Applications may be submitted for the FAST-TRACK review option. Information on the FAST-TRACK process may be found at:

<http://www.nih.gov/grants/funding/sbir.htm>

B. INDIVIDUAL PHASE I APPLICATIONS. Phase I applications in response to this PA will be funded as Phase I SBIR Grants (R43) or STTR Grants (R41). Responsibility for the planning, direction, and execution of the proposed research will be solely that of the applicant. Applications for Phase I grants should be prepared following the directions for Phase I SBIR/STTR applications as described in the NIH Omnibus Solicitation. The NIH Omnibus SBIR Solicitation is available on the Internet at: <http://www.nih.gov/grants/funding/sbir1/toc.htm>

The National Institutes of Health (NIH) is employing features of the Modular Grant Application and Award procedures under its Small Business Innovation Research (SBIR) program. These features are explained in more detail below under "APPLICATION PROCEDURES."

The NIH OMNIBUS STTR Solicitation is available at:

<http://www.nih.gov/grants/funding/sttr1/toc.htm>

A limited number of hard copies of the NIH Omnibus SBIR and STTR Solicitations are available from:

PHS SBIR/STTR Solicitation Office

13685 Baltimore Avenue

Laurel, MD 20707-5096

Telephone: (301) 206-9385

FAX: (301) 206-9722

Email: [a2y@cu.nih.gov](mailto:a2y@cu.nih.gov)

#### C. INDIVIDUAL PHASE II APPLICATIONS

Phase II applications in response to this PA will be awarded as Phase II SBIR Grants (R44) or STTR Grants (R42). Phase II applications in response to this PA will only be accepted as competing continuations of previously funded NIH Phase I SBIR/STTR awards. The Phase II application must be a logical extension of Phase I research in the area of microbial biofilms.

Applications for Phase II awards should be prepared following the instructions for NIH Phase II SBIR/STTR applications. The Phase II SBIR instructions and application may be found on the Internet at: <http://www.nih.gov/grants/funding/sbir2/index.htm>

The Phase II STTR instructions and application may be found on the Internet at:

<http://www.nih.gov/grants/funding/sttr2/index.html>

Applications over \$500,000. Although the Phase II application has no official budgetary limit, applications requesting in excess of \$500,000 dollars direct costs in any single year of the grant period require prior approval before submission. Applicants who plan to submit a Phase II SBIR/STTR application requesting \$500,000 or more in any year are advised that it is important that they contact program staff listed under INQUIRIES as they begin to develop plans.

Applications requesting more than \$500,000 received without prior staff contact may be delayed in the review process or returned to the applicant without review (NIH GUIDE, Volume 22, Number 45, December 17, 1993).

## RESEARCH OBJECTIVES

### Background

A biofilm is an accumulation of microorganisms (bacteria, fungi, and/or protozoa, with associated bacteriophages and other viruses) embedded in a polysaccharide matrix and adherent to a solid biologic or non-biologic surface. Biofilms are medically important, accounting for over 80 percent of microbial infections in the body. Examples include infections of the: oral soft tissues, teeth and dental implants; middle ear; gastrointestinal tract; urogenital tract; airway/lung tissue, eye; urinary tract prostheses; peritoneal membrane and peritoneal dialysis catheters, indwelling catheters for hemodialysis and for chronic administration of chemotherapeutic agents (Hickman catheters); cardiac implants such as pacemakers, prosthetic heart valves, ventricular assist devices, and synthetic vascular grafts and stents; prostheses, internal fixation devices, and percutaneous sutures; and tracheal and ventilator tubing.

Several recent symposia and workshops sponsored by the American Society for Microbiology and the NIH have emphasized the unique features of bacteria and fungi growing as a biofilm rather than in free-floating, planktonic forms. In particular the microorganisms tend to be far more resistant to antimicrobial agents and to be particularly difficult for the host immune system to render an appropriate response.

The need for increased development of technologies for the study and control of microbial biofilms is based on many factors:

- Biofilms are remarkably difficult to treat with antimicrobials. The reasons for this are not clear. Antimicrobials may be readily inactivated or fail to penetrate into the biofilm. In addition, bacteria within biofilms have increased (up to 1000-fold higher) resistance to antimicrobial compounds, even though these same bacteria are sensitive to these agents if grown under planktonic conditions.
- Biofilms increase the opportunity for gene transfer between/among bacteria. This is important since bacteria resistant to antimicrobials or chemical biocides can transfer the genes for

resistance to neighboring susceptible bacteria. Gene transfer can convert a previous avirulent commensal organism into a highly virulent pathogen.

- Certain species of bacteria communicate with each other within the biofilm. As their density increases, the organisms secrete low molecular weight molecules that signal when the population has reached a critical threshold. This process, called quorum sensing, is responsible for the expression of virulence factors.

For example, *Pseudomonas aeruginosa* produces destructive proteinases when the number of these bacteria reach a high enough density in the airway biofilms of cystic fibrosis patients.

- Bacteria express new, and sometimes more virulent phenotypes when growing within a biofilm. Such phenotypes may not have been detected in the past because the organisms were grown on rich nutrient media under planktonic conditions. The growth conditions are quite different particularly in the depths of biofilms, where nutrients and oxygen are usually limited, and waste products from neighbors can be toxic. In short, bacteria found at the bottom of the biofilm look and act different than species located at the surface.

- Bacteria embedded within biofilms are resistant to both immunological and non-specific defense mechanisms of the body. Contact with a solid surface triggers the expression of a panel of bacterial enzymes which catalyze the formation of sticky polysaccharides that promote colonization and protection. The structure of biofilms is such that immune responses may be directed only at those antigens found on the outer surface of the biofilm, and antibodies and other serum or salivary proteins often fail to penetrate into the biofilm. In addition, phagocytes are unable to effectively engulf a bacterium growing within a complex polysaccharide matrix attached to a solid surface. This causes the phagocyte to release large amounts of pro-inflammatory enzymes and cytokines, leading to inflammation and destruction of nearby tissues.

The field of biofilm research has traditionally been hindered by an inability to study the biofilm in non-destructive, three dimensional ways. In addition, it has been difficult or impossible to assess gene expression and metabolism of the microbe at the single cell level within a biofilm. However, as a result of advances in laser technology, digital imaging, scanning electron microscopy, and new fluorescent probes, researchers can now build a three dimensional model of biofilms and identify the location in the biofilm where specific genes are being expressed.

Summary

This broad-based initiative on microbial biofilms is designed to support development of technologies and strategies for the prevention and treatment of microbial biofilm-associated diseases, and for advanced studies of microbial biofilms. In addition, this initiative is intended to capitalize on contemporary research in immunology, microbiology, bio-engineering and computer technology that might synergize with current biofilm research. Another potential aim of this initiative is to link clinical experts, such as nurses, physicians, respiratory therapists, and orthopedic technicians, with bioengineers and basic scientists to better identify the clinical problems associated with microbial biofilm-associated infection.

#### Research Objectives and Scope

Since microbial biofilms are a major problem affecting diverse organs and organ systems, several components of the NIH have joined in this Program Announcement. Therefore, examples of relevant research topics, which are listed below, may reflect interests of specific Institutes within the NIH. Importantly, the list should not be construed as complete or restrictive.

Applicants are encouraged to propose other topics that address the development of systems and technologies to advance the understanding of the formation of biofilms; to study their role in disease; and to develop the means to control them.

- o Development of improved imaging of biofilms in situ;
  
- o Development of improved clinically relevant in vitro and in vivo models of biofilms under specific in vivo conditions such as flow rate, nutrient content, and temperature;
  
- o Development of better probes (genetic, metabolic, and immunological) for real-time analysis;
  
- o Development of systems for improved studies of quorum sensing/signaling molecules;
  
- o Development of systems for the study of biofilm-specific gene expression; and the exchange of genetic material within biofilms;
  
- o Technologies for studies of organic contaminants on substrata, and their influence on biofilm structure;
  
- o Development of novel approaches to control pathogenic bacteria by, for example, devising strategies to favor growth of non-pathogenic microorganisms in biofilm communities;

- o Technologies and materials relevant to the interactions of biofilms with host tissues and artificial implants;
- o Development of materials and technologies to prevent pathogenic bacteria development for long term chronic users of devices; such as total hip replacements beyond ten years and home care use of tracheostomy tubes for mechanical ventilation;
- o Development or use of novel agents, materials, or coatings for preventing or treating infections related to cardiovascular and pulmonary devices, and musculoskeletal prostheses (artificial joints), internal fixation devices, percutaneous sutures, and engineered tissues;
- o Technologies for studies of pathogenic mechanisms of microbes growing in biofilms; and the mechanisms of resistance of biofilms to antimicrobial agents;
- o Systems to enhance the sensitivity of biofilms to antimicrobial agents;
- o Techniques or assays for studies of host immune responses, both innate and adaptive to biofilms;
- o Improved techniques for the study of the potential role of biofilms and host response in the development of systemic inflammatory response syndrome, septic shock, acute respiratory distress syndrome, and multiple organ dysfunction syndrome in injured, disabled or critically ill patients, or in model systems reflecting these clinical conditions;
- o Improved techniques for the studies of infectious lung disease in cystic fibrosis;
- o Development of treatments or products to inhibit the potential of diagnostic procedures such as bronchoalveolar lavage and bronchoscopy to disturb local biofilm flora and inoculate distant locations;
- o Development of mathematical models and computer simulations of biofilms; and
- o Development of the methodology for the prevention and control of biofilms from catheters, water unit lines, and other clinically important solid surfaces.

The focus of this Program Announcement is technology development. Support will not be provided for mechanistic studies of basic questions. Although testing of biological samples or procedures in whole organisms in the course of validating the technology is appropriate.

#### INCLUSION OF WOMEN AND MINORITIES IN RESEARCH INVOLVING HUMAN SUBJECTS

It is the policy of the NIH that women and members of minority groups and their subpopulations must be included in all NIH supported biomedical and behavioral research projects involving human subjects, unless a clear and compelling rationale and justification is provided that inclusion is inappropriate with respect to the health of the subjects or the purpose of the research. This policy results from the NIH Revitalization Act of 1993 (Section 492B of Public Law 103-43).

All investigators proposing research involving human subjects should read the "NIH Guidelines for Inclusion of Women and Minorities as Subjects in Clinical Research," which have been published in the Federal Register of March 28, 1994 (FR 59 14508-14513) and in the NIH Guide for Grants and Contracts, Vol. 23, No. 11, March 18, 1994, and is available at the following URL: <http://grants.nih.gov/grants/guide/notice-files/not94-100.html>

#### INCLUSION OF CHILDREN AS PARTICIPANTS IN RESEARCH INVOLVING HUMAN SUBJECTS

It is the policy of NIH that children (i.e., individuals under the age of 21) must be included in all human subjects research, conducted or supported by the NIH, unless there are scientific and ethical reasons not to include them. This policy applies to all initial (Type 1) applications submitted for receipt dates after October 1, 1998.

All investigators proposing research involving human subjects should read the "NIH Policy and Guidelines on the Inclusion of Children as Participants in Research Involving Human Subjects" that was published in the NIH Guide for Grants and Contracts, March 6, 1998, and is available at the following URL: <http://www.nih.gov/grants/guide/notice-files/not98-024.html>. Investigators may also obtain copies of these policies from the program staff listed under INQUIRIES. Program staff may also provide additional relevant information concerning this policy.

#### APPLICATION PROCEDURES

OMNIBUS SOLICITATIONS for both the SBIR and STTR programs are available electronically through the NIH, Office of Extramural Research "Small Business Funding Opportunities" web site

at <http://www.nih.gov/grants/funding/sbir.htm>. Hard copies, subject to availability, may be obtained from the PHS SBIR/STTR Solicitation Office, phone (301) 206-9385; FAX (301) 206-9722; Email [a2y@cu.nih.gov](mailto:a2y@cu.nih.gov). Helpful information for preparation of the application can be obtained: <http://grants.nih.gov/grants/funding/sbirgrantsmanship.pdf>

Applications are to be submitted on the grant application form PHS 6246-1 (1/98) (SBIR) and PHS 6246-3 (STTR) (1/98) located in the back pages of the OMNIBUS SOLICITATIONS, and will be accepted at the application deadlines. The title and number of this PA must be typed in Line 2 on the face page of the application.

Applications will be accepted for SBIR proposals three times a year with the following deadlines: December 15; April 15; August 15.

Applications will be accepted for STTR proposals three times a year with the following deadlines: December 1; April 1; August 1.

The OMNIBUS SOLICITATIONS give the general guidelines for levels of support and period of time for SBIR and STTR Phase I and II awards. (See NIH Guide, February 12, 1998: <http://www.nih.gov/grants/guide/notice-files/not98-014.html>). Because the length of time and cost of research may exceed that normally awarded for SBIR/STTR grants, larger budgets with longer periods of time may be requested if required to complete the proposed research.

#### MODULAR GRANT APPLICATION AND AWARD

The National Institutes of Health (NIH) is employing features of the Modular Grant Application and Award procedures under its Small Business Innovation Research (SBIR) program. See the "Modular Grant Application and Award" section of the OMNIBUS SOLICITATION, which can be found at <http://www.nih.gov/grants/funding/sbir1/modular.htm>.

SBIR Phase I grant applications requesting up to \$100,000 total costs (direct costs, indirect costs, and fixed fee) will request direct costs in a budget narrative format rather than being compiled from detailed and separate categories.

Applications requesting up to \$100,000 total costs (direct costs, indirect costs, and fixed fee) will request budgets using "Budget Justification" (form page 4 of PHS 6246-1) ONLY. Present the total amount requested for direct costs on line 7a of the Face Page of PHS 6246-1. Information, in narrative form, is to be provided on "Budget Justification" (form page 4) for Personnel, Fixed

Fee, and, when applicable, for Consultant Costs and Contractual Costs. No other budget information is to be submitted. Do not submit "Budget for Phase I Direct Costs Only" (form page 3 of PHS 6246-1). It is to be used as a "worksheet" only. Pages should be renumbered as necessary.

Applications requesting in excess of \$100,000 total costs (direct costs, indirect costs, and fixed fee) do not fall within the Modular Grant Application and Award procedures. Applications requesting in excess of \$100,000 should use "Budget for Phase I Direct Costs Only" (form page 3 of PHS 6246-1), and justify this request using "Budget Justification" (form page 4).

#### FAST-TRACK OPTION

The "Fast-Track" procedures are designed to expedite the decision and award of SBIR Phase II funding for scientifically meritorious applications for projects that have a high potential for commercialization. In order to apply for the FAST-TRACK option, applications for both Phase I and Phase II must be submitted together according to the instructions for Fast-Track applications as described in the OMNIBUS SOLICITATIONS (<http://grants.nih.gov/grants/funding/sbirsttr1/6method.htm#6g>).

SBIR applications are eligible for the Fast-Track review process upon meeting the following criteria:

The small business concern must submit a concise Product Development Plan (limited to ten pages) as a Product Development Plan Appendix to the Phase II application addressing each of the following areas:

- a. Company information, including size; specialization area(s); products with significant sales; and history of previous federal and non-federal funding, regulatory experience, and subsequent commercialization.
- b. Value of SBIR project, including lay description of key technology objectives, current competition, and advantages compared to competing products or services.
- c. Commercialization plans, milestones, target dates, market analyses of market size, and estimated market share after first year sales and after five years.
- d. Patent status or other protection of project intellectual property.

The Product Development Plan appendix should be labeled clearly.

Fast-Track SBIR applications for both Phase I and Phase II must be submitted together for concurrent initial peer review and evaluation. In order to identify the application as such, the words "Fast-Track" must be shown in item 2 on the face page of the Phase I application.

The Phase I application must specify clear, measurable goals (milestones) that should be achieved prior to initiating Phase II. Failure to provide clear, measurable goals may be sufficient reason for the scientific peer review group to exclude the Phase II application from Fast-Track review.

Applicants are ENCOURAGED to seek commitment(s) of funds and/or resources from an investor or partner organization for commercialization of the product(s) or service(s) resulting from the SBIR grant.

For all applications submit a signed, typewritten original of the application, including a cover letter, the checklist and two signed photocopies in one package to:

CENTER FOR SCIENTIFIC REVIEW  
NATIONAL INSTITUTES OF HEALTH  
6701 ROCKLEDGE DRIVE, ROOM 1040 - MSC 7710  
BETHESDA, MD 20892-7710  
BETHESDA, MD 20817 (for express/courier service)

## REVIEW CONSIDERATIONS

### Review Procedures

Application will be assigned on the basis of established NIH referral guidelines. When the subject of an application is of interest to more than one Institute, dual assignments will be made. Upon receipt, applications will be reviewed for completeness by the Center for Scientific Review (CSR). Incomplete applications will be returned to the applicant without further consideration.

Applications that are complete will be evaluated for scientific and technical merit by study sections of the CSR. As part of the initial merit review, all applications will receive a written critique and undergo a process in which only those applications deemed to have the highest

scientific merit, generally the top half of applications under review, will be discussed, assigned a priority score, and receive a second level review by the appropriate national advisory council or board.

## Review Criteria

Review criteria are described in the NIH Omnibus Solicitation and are as follows:

1. The soundness and technical merit of the proposed research. (Preliminary data are not required for Phase I proposals.
2. The qualifications of the proposed principal investigator, supporting staff, and consultants.
3. The scientific, technical, or technological innovation of the proposed research.
4. The potential of the proposed research for commercial application or societal impact.
5. The appropriateness of the budget requested.
6. The adequacy and suitability of the facilities and research environment.
7. Where applicable, the adequacy of assurances detailing the proposed means for (a) safeguarding human or animal subjects and/or (b) protecting against or minimizing any adverse effect on the environment.

For Fast-Track, Phase I applications should specify clear, measurable goals (milestones) that should be achieved prior to initiating Phase II. Failure to provide clear, measurable goals may be sufficient reason for the study section to judge the application non-competitive.

The initial review group will also examine: the appropriateness of the proposed project budget and duration; the adequacy of plans to include both genders and minorities and their subgroups as appropriate for the scientific goals of the research and plans for the recruitment and retention of subjects; the provisions for the protection of human and animal subjects; and the safety of the research environment. In addition, reviewers will be instructed to address the adequacy of plans for including children as appropriate for the scientific goals of the research, or justification for exclusion (see section on NIH POLICY AND GUIDELINES ON THE INCLUSION OF CHILDREN AS PARTICIPANTS IN RESEARCH INVOLVING HUMAN SUBJECTS).

## AWARD CRITERIA

Applications will compete for available funds with all other approved SBIR and STTR applications. Funding decisions for Phase I will be based on quality of the proposed project as determined by peer review, availability of funds, and program priority.

Fast-Track Phase II applications may be funded following submission of the Phase I progress report and other documents necessary for continuation. Phase II applications will be selected for funding based on the quality of the proposed project, the determination that Phase I goals were achieved, the project's potential for commercial success, and the availability of funds.

## INQUIRIES

Written, email, and telephone inquiries concerning this program announcement are strongly encouraged. NIH staff welcome the opportunity to clarify any issues regarding the application procedures or provide guidance to applicants regarding specific aspects of their proposals.

Direct inquiries regarding programmatic issues to:

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#### AUTHORITY AND REGULATIONS

This program is described in the Catalog of Federal Domestic Assistance No. 93.121 (NIDR), 93.856 (NIAID), and No. 93.173 (NIDCD), and No. 93.846 (NIAMS), No. 93.859 (NIGMS), No. 93.838 (NHLBI), and No. 93.849 (NIDDK). Awards are made under authorization of the Public Health Service Act, Title IV, Part A (Public Law 78-410, as amended by Public Law 99-158, 42 USC 241 and 285) and administered under PHS grants policies and Federal Regulations 42 CFR 52 and 45 CFR Part 74. This program is not subject to the intergovernmental review requirements of Executive Order 12372 or Health Systems Agency review.

The PHS strongly encourages all grant and contract recipients to provide a smoke-free workplace and promote the non-use of all tobacco products. In addition, Public Law 103-227, the Pro-Children Act of 1994, prohibits smoking in certain facilities (or in some cases, any portion of a facility) in which regular or routine education, library, day care, health care or early childhood

development services are provided to children. This is consistent with the PHS mission to protect and advance the physical and mental health of the American people.

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