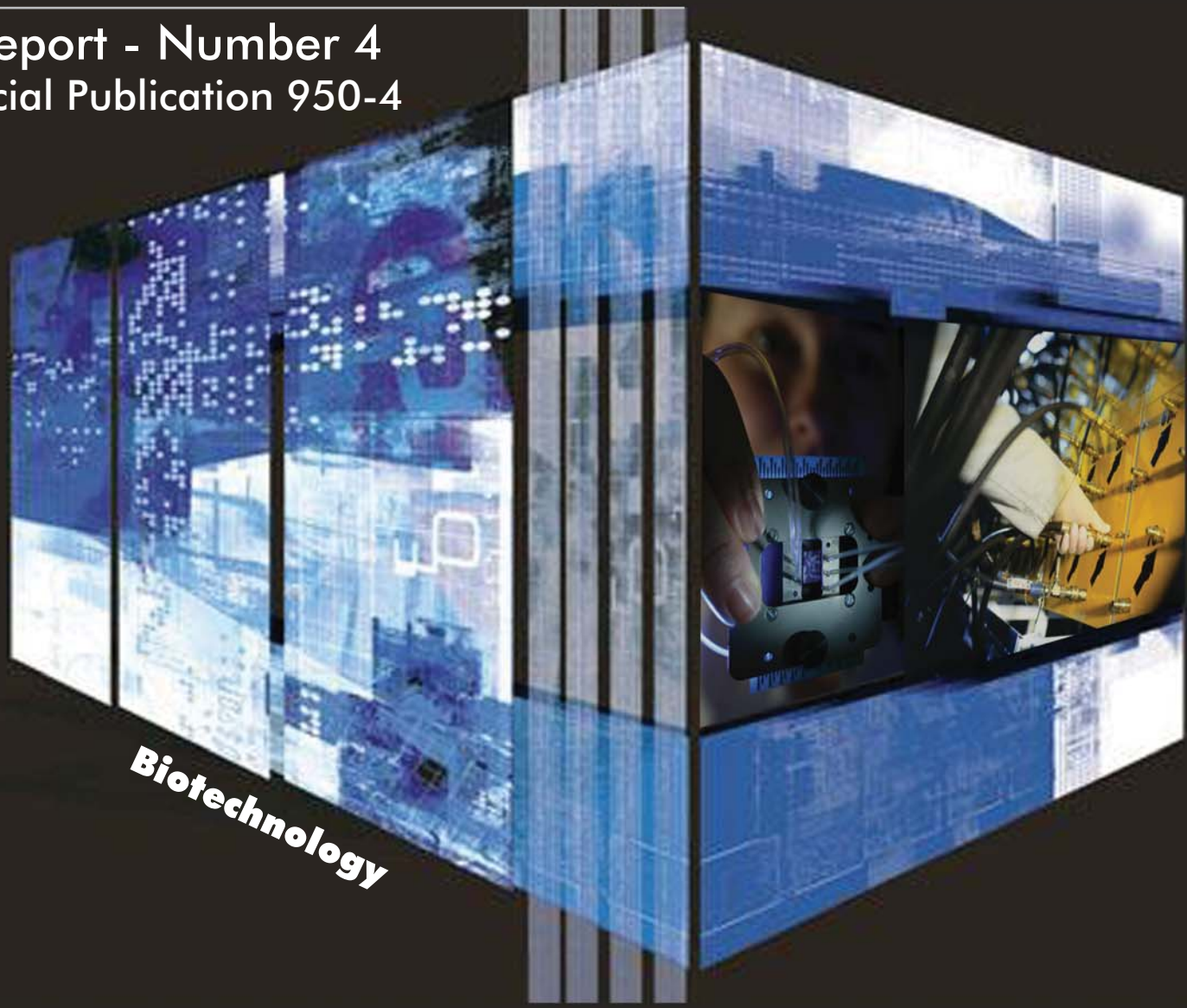
ADVANCED TECHNOLOGY PROGRAM

Performance of the Third 50 Completed ATP Projects

Status Report - Number 4

NIST Special Publication 950-4



September 2006

NIST
National Institute of Standards and Technology
Technology Administration
U.S. Department of Commerce

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TABLE OF CONTENTS

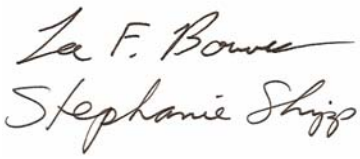
ACKNOWLEDGEMENTS

We are pleased to announce the completion of the next chapter in ATP's portfolio of status reports for completed projects. This compilation consists of the third batch of 50 "mini case studies" written to investigate the results and impacts of ATP's investment in innovative technologies. The goal of each status report is to provide the reader with a basic understanding of the technology, while also identifying any economic benefits that may have resulted from the ATP-funded project.

This process is a daunting one, requiring the efforts of many both inside and outside of ATP. The majority of the project was made possible by the former and current members of the status report team: Tony Colandrea, Stefanie Cox, Nashira Nicholson, Rick Rodman, Susan Stimpfle, and Virginia Wheaton. We would especially like to thank ATP Division Directors Michael Schen (Information Technology and Electronics Office) and Linda Beth Schilling (Chemistry and Life Sciences Office), and ATP Deputy Director Lorel Wisniewski, for their contributions to this project. Much appreciation also goes to the hard work of the others involved in preparing the status reports: project managers, reviewers, copy editors, and company representatives.

But these efforts aren't without rewards. As the portfolio of ATP status reports grows, we gain insight as to the role ATP plays in bridging the funding gap. We are confident this showcase of the third batch of 50 completed projects will help build on our understanding of ATP-funded innovations across many technology areas. We hope that you learn as much about the process of early-stage technology development, commercialization, and outcomes for the economy as we have in preparing these status reports.

Sincerely,



Lee Bowes, Economist
Stephanie Shipp, Director,
Economic Assessment Office
Advanced Technology Program

INTRODUCTION

Industry has proposed 6,924 projects to the ATP since 1990, of which 768, or 11 percent, have been selected by the ATP for funding. The number of participants for these funded projects totaled 1,511, with approximately an equal number of subcontractors. This study focuses on the third group of 50 projects that were completed and provides combined statistics for all 150 completed projects studied to date.

ATP: A Partnership with Industry

The ATP attracts challenging, visionary projects with the potential to develop the technological foundations of new and improved products, processes, and even industries. The ATP partners with industry on this research, fostering collaborative efforts and sharing costs to bring down high technical risks and accelerate technology development and application. These are projects that industry in many cases will not undertake without ATP support, or will not develop in a timely manner when timing is critical in the highly competitive global market. The program funds only research, not product development. The ATP is managed by the National Institute of Standards and Technology, an agency of the Commerce Department's Technology Administration.

ATP awards are made on the basis of a rigorous competitive review, which considers the scientific and technical merit of each proposal and its potential benefits to the U.S. economy. The ATP issues a proposal preparation kit that presents and explains the selection criteria to prospective applicants and provides guidance on preparing proposals.[1] U.S. businesses conceive, plan, propose, and lead the projects. Government scientists and engineers who are expert in the relevant technology fields review all proposals for their technical merit. Business, industry, and economic experts review the proposals to judge their potential to deliver broadly based economic benefits to the nation —including large benefits extending beyond the innovator (the award recipient).

The ATP delivers benefits to the nation along two pathways: 1) a direct path by which the U.S. award recipient or innovator directly pursues commercialization of the newly developed technologies; and 2) an indirect path which relies on knowledge transfer from the innovator to others who in turn may use the knowledge for economic benefit. Either path may yield spillover benefits. The ATP looks to the direct path as a way to accelerate application of the technology by U.S. businesses. It looks to the indirect path as a means of achieving additional benefits, or benefits even if the award recipient fails to continue. The ATP's two-path approach to realizing national benefits offers advantages: one path may provide an avenue for benefits when the other does not, and both paths together may yield larger, accelerated benefits as compared to having a single route to impact.

Project Evaluation

The ATP, like other federal programs, is required by law to report on its performance.[2] The ATP established its evaluation program soon after it began, even before evaluation was widely required by Congress. The Economic Assessment Office (EAO) of ATP plans and coordinates the evaluation of funded projects. It is assisted in this effort by leading university and consulting economists and others experienced in evaluation.

Performance is measured against the program's legislated mission. Emphasis is placed on attempting to measure benefits that accrue not only to the direct award recipients, but also to a broader population, i.e., spillover benefits. This emphasis reflects the fact the public funding covers part of the costs of these projects, and, therefore, a relevant question is how the broader public benefits from the expenditure.

This report constitutes one element of the EAO's multi-faceted evaluation plan: status reports. The purpose of status reports is to provide an interim assessment of the status of ATP-funded projects several years after they are completed. Although the ultimate success of the ATP depends on the long-run impacts of the entire portfolio of ATP projects, the performance-to-date of this partial portfolio provides some initial answers. This study contains an evaluation of 150 completed projects: the results of the 100 projects from the Status Report – Numbers 2 & 3, and the results and status reports of a third batch of 50 projects. These reports address the questions of what has the public investment of $321 million in the 150 projects produced several years after completion of the research, and what the outlook is for continued progress?

Study Approach

From the moment that ATP funded its first group of 11 projects in the 1990 competition, program administrators, the administration, Congress, technology policymakers, industry, and others in this country and abroad were keenly interested in the outcome. But technology development and commercialization are lengthy processes, and it takes time to produce results.

As more ATP-funded project are completed and move into the post-project period, sufficient time has elapsed for knowledge to be disseminated and progress to be made towards commercial goals. Thus, it is now possible to compile more complete aggregate portfolio statistics, and analyze these statistics with regard to implications for overall program success.

At the core of this study are 50 mini-case studies covering each of the completed projects. Each of these briefly tells the project story, recounting its goals and challenges, describing the innovators and their respective roles, and assessing progress to date and the future outlook. Photographs illustrate many of the projects.

Although the particulars vary for each project, certain types of data are systematically collected for all of them. Consistent with ATP's mission, the evaluation focuses on collecting data related to the following dimensions of performance:

- **Knowledge creation and dissemination**, which is assessed using the following criteria: recognition by other organizations of a project's technical accomplishments; numbers of patents filed and granted; citations of patents by others; publications and presentations; collaborative relationships; and knowledge embodied in and disseminated through new products and processes.
- **Commercialization progress**, which is gauged in terms of the attraction of additional capital for continued pursuit of project goals, including resources provided by collaborative partners; entry into the market with products and services; employment changes at the small companies leading projects and other indicators of their growth; awards bestowed by other organizations for business accomplishments of project leaders; and the analyst's assessment of future outlook for the technology based on all the other information.

The approach is to provide, in an overview chapter, the aggregate statistics of interest across a set of 150 projects, such as the total number of patents and the percentage of projects whose technologies have been commercialized. In addition, the aggregate statistics are combined to produce composite project metrics for overall performance. The composite performance scores allow one to see at a glance the robustness of a project's progress towards its goals. Underlying the simple scores is a wealth of data.

Sources of Information

Data for the projects were collected from many sources: ATP project records; telephone interviews with company representatives; interviews with ATP project managers; company websites; the U.S. Patent and Trademark Office; in-depth project studies conducted by other analysts; academic, trade and business literature; news reports; filings at the Securities and Exchange Commission; and business research services, such as Dun and Bradstreet, Hoover's Online, Industry Network, and CorpTech. Each one of the individual project write-ups was reviewed for accuracy by the project's lead company and ATP staff.

Study Limitations and Future Directions

Since developments continue to unfold for most of these projects, the output measures for the cases may have changed significantly since the data were collected. The cases provide a snapshot of progress several years after the completion of the ATP-funded projects.

Although undertaken at different calendar dates, the reports are written within about the same interval of time after ATP funding ended. Yet, different points in each technology's life cycle may be captured, depending on the technology area. Information technology projects, for example, may be expected to be further along than advanced materials and chemical projects. Examined at a later time, there may be less (or more) difference in the accomplishments among projects in different technology areas.

This study tracks outputs leading to knowledge dissemination but it does not assess the actual commercialization efforts by others who acquire the knowledge. The tracking of commercialization efforts is limited to the direct path of impact (i.e., commercialization by the award recipients or innovators).

"Completed" and "Terminated" Projects Defined

Projects do not necessarily finish in the order funded. For one thing, they have different lengths, ranging from approximately two years to no more than five years. For another, they are required to file a final report with the ATP and have financial and other paperwork completed before project closeout. The financial closeout is done through the National Institute for Standards and Technology (NIST) Grants Office, which notifies the ATP that it considers the project completed. This study assesses the first 150 projects the Grants Office declared "completed."

Not all ATP projects reach completion; some are stopped short and are classified as "terminated." Some of these were announced as award winners but never officially started. Other projects got off the ground but were closed for various reasons with a substantial amount of the technical work still unfinished. These terminated projects are assessed according to the principal reasons they stopped before completion. They are treated in Appendix B. While the terminated projects are generally regarded as unsuccessful, some produced potentially useful outputs.

Report Organization

The report has been divided into separate technology area "editions" in order to provide a smaller, more targeted compilation. However, the overview still provides a summary overview of the performance of the 150 completed projects as a group. It identifies some major outputs that appear useful as indicators of the degree of project success, and it uses these outputs in a prototype project performance rating system. A preview also notes some of the broad-based benefits that this portfolio of projects is producing and likely to produce. For additional background, the make-up of the portfolio of projects in terms of technologies, organizational structure, company size, and other features is provided.

The individual project reports, within the particular technology area, follow the overview. The reports highlight major accomplishments and the outlook for continued progress. A detailed account of the project under review is given, with attention to technical and commercial goals and achievements, information about technology diffusion, and views about the role played by ATP funding. A performance rating is assigned to each project based on a four-star scoring system. The rating depends on the accomplishments of the project in creating and disseminating new scientific and technical knowledge and in making progress toward generating commercial benefits, as well as the outlook for continued progress.

Three appendices provide supporting information. Appendix A provides a listing of technical and commercial achievements of each completed project. Appendix B provides a discussion of the terminated projects throughout ATP's existence. Appendix C provides a list of the first 150 completed projects and the respective composite performance ratings. The listed is sorted in descending order of performance rating, then by company name.

1. The current edition of the kit and other program materials may be obtained on ATP's website (www.atp.nist.gov), by e-mail (atp@nist.gov), by phone (1-800-ATP-Fund or 1-800-287-3863), or by mail (ATP, NIST, 100 Bureau Drive, Stop 4701, Gaithersburg, MD 20899-4701).
2. The Government Performance and Results Act (GPRA) is a legislative framework for requiring federal agencies to set strategic goals, measure performance, and report on the degree to which goals are met. An overview of the GPRA is provided in Appendix 1 of the General Accounting Office Executive Guide, Effectively Implementing the Government Performance and Results Act, GAO, Washington, D.C., GGD-96-118, 1996

Overview of Completed Projects

PART 1

Project Characteristics

This report provides an overview of the first 150 ATP-funded projects to reach completion. These projects reflect an investment of more than $621 million that was shared about equally by ATP and industry.

Of the initial 150 projects, 75 were led by small businesses that submitted single-company-applicant proposals to ATP. Eighty-seven percent involved collaborative relationships with other firms, universities, or both. Sixty-seven percent were funded in ATP's General Competitions.

In terms of classification by type, 25 percent of the projects were "Electronics, Computer Hardware, or Communications", while "Advanced Materials and Chemicals" accounted for 23 percent. "Manufacturing", "Information Technology", and "Biotechnology" each constituted about 17 percent of the remaining projects.

(The 150 completed status reports discussed in this chapter can be found on-line at http://www.atp.nist.gov/ under funded projects.)

Single Applicants and Joint Ventures

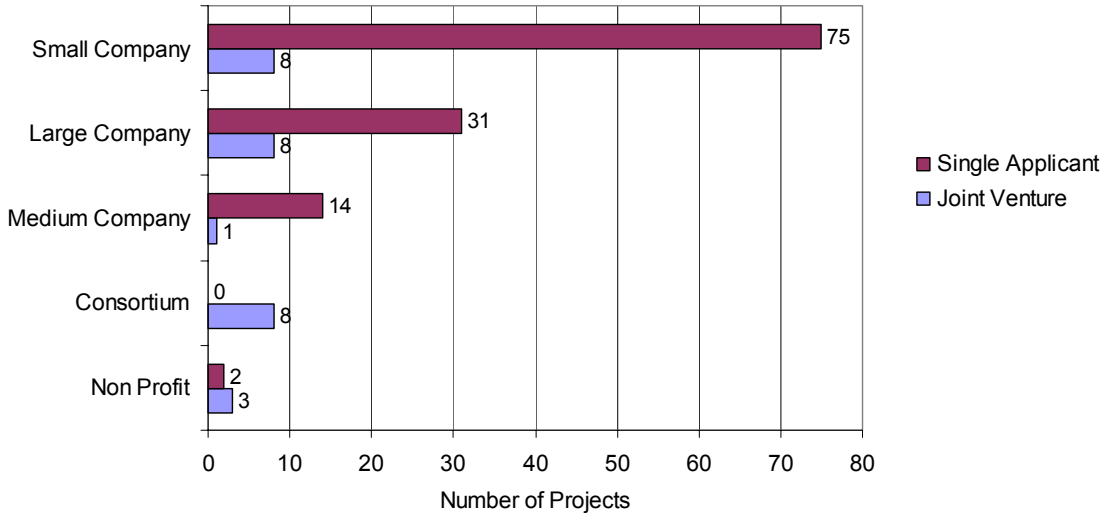
"Single-applicant projects," make up 81 percent of the first 150 ATP-funded projects; these projects were subject to an upper limit on ATP funding of $2 million and a time limit of 3 years. Nineteen percent of the 150 projects were joint ventures. Each of these projects had a minimum of two for-profit companies sharing research and costs for up to 5 years. Typically, the joint-venture membership included other for-profit companies, universities, and nonprofit laboratories. These projects, free of the funding constraint, tended to take on larger problems for longer periods of time.

Project Leaders

Figure 1-1 illustrates how project leadership of single-applicant and joint-venture projects was distributed among the various types of organizations. Small companies led most of the projects—75 of the 122 single-applicant projects and 8 of the 28 joint-venture projects. "Small" follows the Small Business Administration's definition and includes companies with fewer than 500 employees. Large companies—defined as Fortune 500 or

equivalent firms—led 31 of the single-applicant projects, or 25 percent, and eight of the joint ventures, or 29 percent. Medium-sized companies led only 14 single-applicant projects and one joint venture. Consortia led eight of the joint venture projects. Nonprofit institutions led two of the single-applicant projects[1], and three joint ventures.

Figure 1-1
Number of Single-Applicant and Joint-Venture Projects by Type of Leadership



Source: Advanced Technology Program First 150 Status Reports

A Variety of Technologies

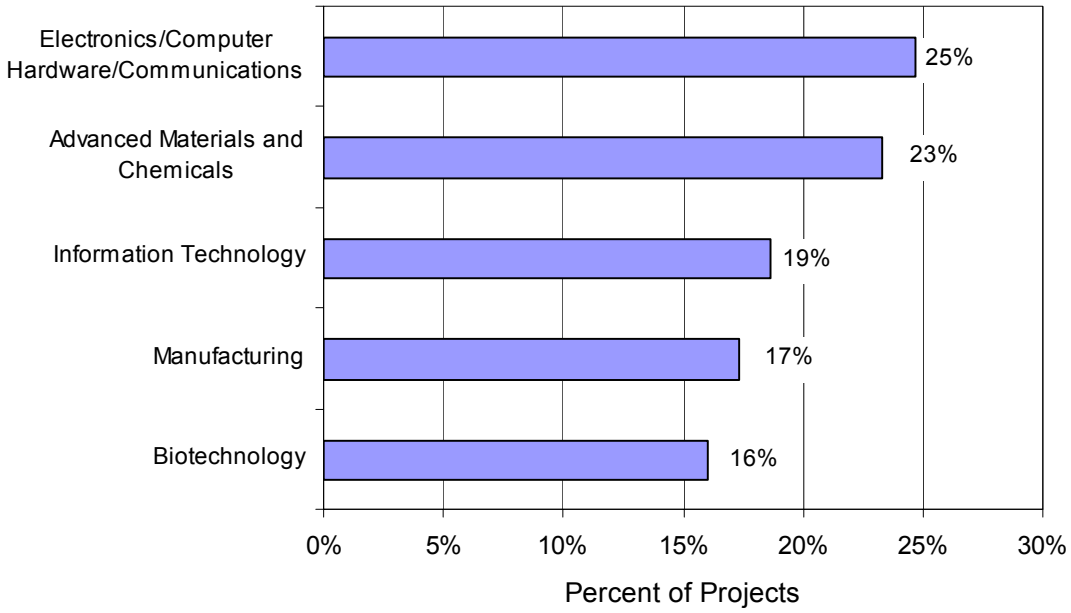
The 150 completed projects fall into the five technology areas used by ATP for classification purposes. Figure 1-2 shows the percentages of completed projects by technology area. The highest concentration, accounting for 25 percent of the total, is in "Electronics, Computer Hardware, or Communications." This category includes microelectromechanical technology, microelectronic fabrication technology, optics and photonics, and other electronics projects.

"Advanced Materials and Chemicals" account for 23 percent of the projects. "Information Technology," "Manufacturing," and "Biotechnology" account for, 19, 17 and 16 percent respectively of the 150 projects. The Manufacturing category includes areas such as energy conversion and energy generation and distribution, in addition to machine tools, materials handling, intelligent control, and other discrete manufacturing. The Advanced Materials and Chemicals category includes the subcategories of energy resources/petroleum, energy storage/fuel cell, battery, environmental technologies, separation technology, catalysis/biocatalysis, and other continuous manufacturing technologies, as well as metals and alloys, polymers, building/construction materials, and

[1] From the 1991 competition, when nonprofits were eligible to lead ATP projects.

other materials. The category of Biotechnology includes areas such as bioinformatics, diagnostic and therapeutic, and animal and plant biotechnology.

Figure 1-2
Distribution of Projects by Technology Area



Source: Advanced Technology Program First 150 Status Reports

The technology make-up of these 150 projects differs from that of the larger ATP portfolio of projects in part because the composition of ATP applicants and awardees over time changes. Of the first 150 completed projects, 67 percent come from ATP's General Competitions that were open to all technologies, while 33 percent come from ATP's focused program competitions, which were held from 1994 through 1998. These competitions funded technologies in selected areas of focus, such as in Motor Vehicle Manufacturing Technology and Digital Video in Information Networks.

It should be noted that while the five major technology areas are used to classify the projects, most of them are not easy to classify. Most ATP projects involve a mix of technologies and interdisciplinary know-how.

Collaborative Activity

Although only 19 percent of the 150 projects were joint ventures, 87 percent of all projects had collaborative arrangements. As shown in Table 1-1, 49 percent of the projects involved close research and development (R&D) ties with universities. Sixty-one percent reported collaborating on R&D with companies or other nonuniversity organizations. Slightly less than half the projects formed collaborative relationships with other organizations for commercial pursuit of their ATP-funded technologies. Thirty-five

percent of projects had collaborative relationships with both universities and nonuniversities for either R&D or commercial purposes.

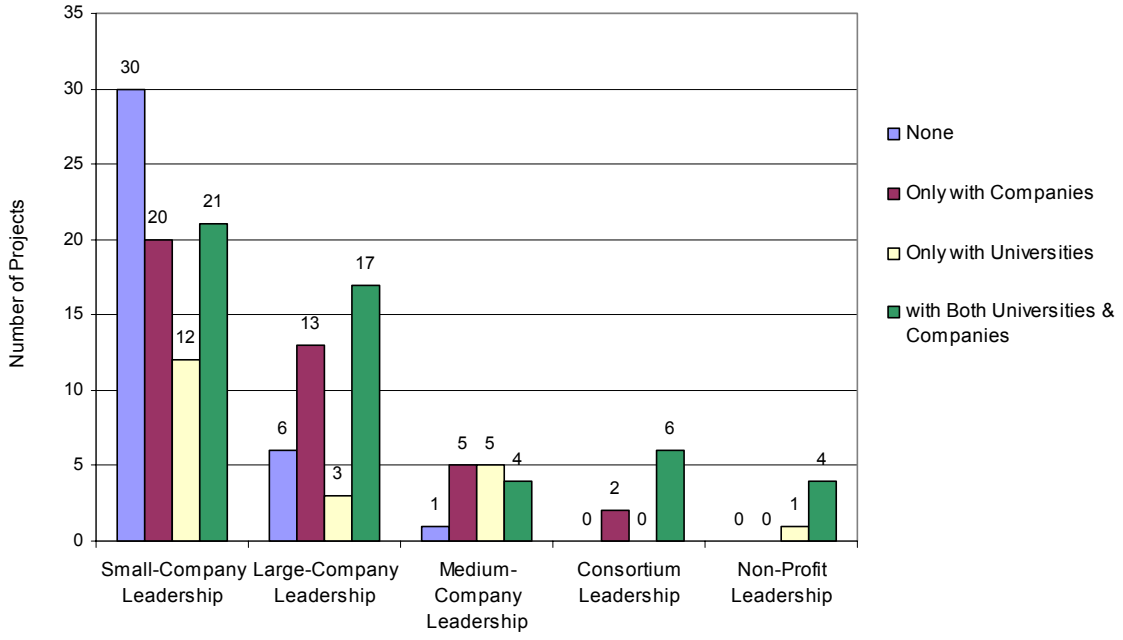
Table 1-1
Collaborative Activity

	Type of Collaboration	Percentage
A)	Collaborating on R&D with other companies or nonuniversity organizations	61%
B)	Close R&D ties with universities	49%
	Collaborating on R&D with other companies or nonuniversity organizations OR close R&D ties with universities (A or B)	75%
	Collaborating with both universities and non-university organizations (A and B)	35%
C)	Collaborating on commercialization with other organizations	46%
	Collaborating in one or more of the above ways	87%
Note: This assessment of collaborative relationships likely understates the numbers because it focused on the project's lead organization and probably missed some of the informal collaborative relationships of other participants.		

Source: Advanced Technology Program First 150 Status Reports

For more detail, Figure 1-3 illustrates the types of collaboration undertaken by projects with different forms of project leadership. It highlights the fact that under all forms of project leadership, projects were highly likely to involve collaboration with other companies. About 43 percent of the projects led by small and large companies involved university collaboration, while the share rose to 60 percent for projects led by medium-sized companies, and 75 percent for consortium-led projects.

Figure 1-3
Number of Projects with R&D Collaborations by Type of Collaboration and Type of Project Leadership



Source: Advanced Technology Program First 150 Status Reports

Costs of the Projects

As shown in Table 1-2, ATP and industry together invested in excess of $621 million on the 150 projects. They shared almost equally in project costs, with ATP providing a slightly larger share. ATP spent an average of $1.72 million per single-applicant project and an average of $3.97 million per joint-venture project. Across the 150 projects, the average total cost (ATP plus industry) per project was $4.14 million. Estimated benefits attributed to ATP from just a few of the 150 projects for which quantitative economic benefits have been provided exceed ATP's funding for all of the 150 projects. In addition, there is considerable evidence of large project benefits that have not yet been quantified.

Approximately 45 percent of single-applicant projects had total research costs under $3 million. These projects had an ATP share that ranged from a little more than $.5 million to $2 million. Slightly less than 50 percent had total research costs greater than $5 million, and one project had total research costs greater than $30 million. ATP's share of these costs were $2 million or more for 50 percent of the projects and were $5 million or higher for 36 percent. For one of the projects, ATP's share exceeded $10 million. Joint ventures, which made up only 19 percent of the total number of projects, accounted for 35 percent of total ATP funding.

Table 1-2
ATP Funding, Industry Cost Share, and Total Costs of 150 Completed Projects

	Single Applicant Projects	Joint Venture Projects	Total Projects
ATP Funding ($ Millions)	210.1	111.1	321.2
Industry Cost Share ($ Millions)	184.6	115.2	299.8
Total Project Costs ($ Millions)	394.7	226.3	621.0
ATP Share of Costs	53%	49%	52%
Industry Share of Costs	47%	51%	48%
Average Project Funding Provided by ATP ($ Millions)	1.72	3.97	2.14
Average Project Cost-Share Provided by Industry ($ Millions)	1.51	4.11	2.00
Average Project Funding Provided by Overall ($ Millions)	3.24	8.08	4.14

Source: Advanced Technology Program First 150 Status Reports

P A R T 2

Gains in Technical Knowledge

One of ATP's major goals is to build the nation's scientific and technical knowledge base. Each of the 150 completed ATP projects targeted a number of specific technical goals designed to achieve a new or better way of doing things. The knowledge created by each project is the source of its future economic benefit, both for the innovator and for others who acquire the knowledge. It is a good starting place for assessing completed projects.

(The 150 completed status reports discussed in this chapter can be found on-line at http://www.atp.nist.gov/ under funded projects.)

New Technologies and Knowledge Gains

Knowledge gains by the projects are diverse and encompass the five major technology areas. The technologies developed in the 150 projects are listed in column C in Tables A-1–A-5 in Appendix A. The set of tables provides the reader with a convenient, quick reference to the entire range of technologies. The entries are arranged alphabetically, by project lead company using the five technology areas. As was mentioned earlier, most of these projects are interdisciplinary, involving a mixture of technologies and generating knowledge in multiple fields.

Even those projects that were not fully successful in achieving all of their research goals, or those that have not been followed by strong progress in commercialization, have achieved knowledge gains. Moreover, some of the projects carried out by companies that have since ceased operations or stopped work in the technology area yielded knowledge, as indicated primarily by the presence of publications and patents. In these cases the direct market routes of diffusion of knowledge gains through commercialization by the innovators are likely lost. However, the indirect routes—whereby others acquire and use the knowledge—remain.

Of What Significance Are the Technical Advances?

Measuring the significance of technical advances is challenging. One factor that challenges measurement is the length of elapsed time that typically separates an R&D investment and its resulting long-term outcomes. In the interim period, various short-run metrics may serve as indicators that project results appear to be on track toward achieving long-term goals. One metric that has been used to signal the significance of a project's technical achievements is formal recognition in the form of an award from a third-party organization.

Thirty awards for technical accomplishments were made to participants for achievements related to ATP-funded projects. Participants in 19 of the 150 projects received awards for their technical achievements. Participants in seven of the projects received multiple technical awards. Table 2-1 lists the awards made to these projects by third-party organizations in recognition of their technical accomplishments.

Table 2-1
Outside Recognition of Technical Achievements of the First 150 Completed Projects

Project Awardee	Year	Awarding Organization	Award
American Superconductor	1996	*Industry Week* Magazine	Technology of the Year award
American Superconductor	1996	*R&D* Magazine	One of the 100 most important innovations of the year
Automotive Composites Consortium (a Partnership of DaimlerChrysler [formerly Chrysler], Ford and General Motors)	1999	*Popular Science* Magazine	Best of What's New for the Chevrolet Silverado composite truck box, "a breakthrough in the use of structural composites"
Cincinnati Lamb, UNOVA (Lamb Technicon)	1999	*Industry Week* Magazine	Top 25 Technology and Innovation Award
Communication Intelligence #1	1997	Arthritis Foundation	"Ease-of-Use Seal of Commendation" for the development of natural handwriting technology, for use by disabled people who have trouble with keyboard entry
DuPont	1993	*Microwave & Rf* Magazine	One of the Top Products of 1993, for high-temperature superconductivity component technology
Ebert Composites	1999	Civil Engineering Research Foundation	Charles Pankow Award for Innovation in Civil Engineering
Engineering Animation	1994	*Computerworld* Magazine	Smithsonian Award, for the use of information technology in the field of medicine
Engineering Animation	1995	Association of Medical Illustrators	Association of Medical Illustrators Award of Excellence in Animation
Engineering Animation	1995	International ANNIE Awards	Finalist, received together with Walt Disney, for best animations in the film industry
Engineering Animation	1996	*Industry Week* Magazine	One of the 25 Technologies of the Year, for interactive 3D visualization and dynamics software used for product development
GM Thermoplastic Engineering Design (Engineering Design with Thermoplastics)	2001	Internal GM R&D Award	Campbell Award for "Process Modeling and Performance Predictions of Injection-Molded Polymers"
GM Thermoplastic Engineering Design (Engineering Design with Thermoplastics)	2001	Society of Plastics Engineers	Best Paper Award from the Product Design and Development Division

Project Awardee	Year	Awarding Organization	Award
HelpMate Robotics	1996	*Discover* Magazine	One of 36 finalists for Technology of the Year, for the HelpMate robot used in hospitals
HelpMate Robotics	1997	Science Technology Foundation of Japan	Japan Prize, to CEO Joseph Engelberger, for "systems engineering for an artifactual environment"
Illinois Superconductor	1996	*Microwave & Rf* Magazine	One of the Top Products of 1996, for cellular phone site filters and superconducting ceramics
Illinois Superconductor	1997	American Ceramic Society	Corporate Technical Achievement Award
Integra Life Sciences**	1999	New Jersey Research and Development Council	Thomas Alvin Edison Award
Kopin Corporation	1998	*Electronic Products* Magazine	“Product of the Year” Award for expanding functionality of portable devices including PDAs, cell phones, and pagers
Kopin Corporation	1998	*IndustryWeek* Magazine	“25 Technologies of the Year” Award
Kopin Corporation	1999	*Photonics Spectra* Magazine	“25 Most Technically Innovative Products” Award for the CyberDisplay 320C
Kopin Corporation	2003	Consumer Electronics Show 2003	“Best Innovation" Award for the 44-inch LCoS HDTV
Molecular Simulations	1996	Computerworld Magazine	Finalist for Smithsonian Award, the 1996 Innovator Medal
NCMS	1994	Institute for Interconnecting & Packaging Electronics Circuits	Best Paper of Conference Awards
Perceptron (formerly Autospect, Inc.)	1998	International Body Engineering Conference	Best Paper Award
Strongwell Corporation	1998	Composite Fabricators Association Conference	Best of Show Award
The Dow Chemical Company	2004	Department of Commerce, NIST/Brookhaven	Gold Medal for Scientific/ Engineering Achievement for Dr. Daniel Fischer’s work on "a unique national measurement facility for soft X-ray absorption spectroscopy enabling breakthrough materials advances"
Xerox Palo Alto Research Center	2003	*JavaWorld*	Editors' Choice Award for the Most Innovative Java Product or Technology
X-Ray Optical Systems (XOS)	1995	*R&D* Magazine	R&D top 100
X-Ray Optical Systems (XOS)	1996	*Photonics Spectra* Magazine	Photonics Circle of Excellence Award

Source: Advanced Technology Program First 150 Status Reports

**The award went to Dr. Kohn of Rutgers University for his collaborative work with Integra on the project.

Examples of Projects with Knowledge Gains

Xerox Palo Alto Research Center: Xerox Palo Alto Research Center (PARC) expanded its research on modularity with a cost-shared award for $1.7 million from ATP's Component-Based Software Focus Program. The project began in 1995, and the researchers developed two prototype applications that extracted system-wide concerns into separate modules with their own code. They called this approach aspect-oriented programming (AOP).

As ATP funding ended, PARC began working with the Defense Advanced Research Projects Agency to create a general-purpose language and tool, which PARC patented and called AspectJ. This product:

- Is freely available through IBM's eclipse.org web site
- Has six trade books devoted to it
- Won the JavaWorld Editors' Choice Award for the Most Innovative Product or Technology in 2003
- Is used aggressively by IBM in developing new software products

AOP is well recognized in the computer industry and has eight patents associated with it. More than a dozen universities in North America and in the United Kingdom include it in their curricula. Although the average computer user does not know or care about aspects, programmers' use of AOP in designing web sites will bring speed, reliability, greater customization, and savings. End users receive better services, delivered more quickly, at a lower cost.

Orchid BioSciences (formerly Molecular Tool, Inc. Alpha Center): A small company, Molecular Tool, applied for and was awarded $1.9 million under the ATP Tools for DNA Diagnostics focused program in 1995, in order to compress most of the functions of SNP analysis that were being done in the 20-foot by 15-foot biotechnology laboratory onto a 1-square-inch glass chip..

Molecular Tool successfully developed a patented prototype SNP analysis tool in 1998 and gained the attention of the biotechnology industry. Orchid BioComputer (later renamed Orchid BioSciences) purchased Molecular Tool in 1998 to acquire the ATP-funded equipment and the company's project-related knowledge.

In 2000, Orchid BioSciences was performing DNA analyses using a single nucleotide polymorphism (SNP) analysis tool, which performed more than 800,000 DNA analyses per day. Orchid's SNP scoring tool, called SNPstream, analyzes up to 100,000 data points for increased accuracy. Furthermore, a typical result showed one in several billions statistical probability, increased from one in a million. SNP technology has had high-profile applications:

- Used to attempt to identify the remains of some New York City World Trade Center victims of 2001, which could not be identified by conventional DNA analysis due to sample degradation.

- Used in assisting major metropolitan police departments in forensics, including Los Angeles, Houston, and England's Scotland Yard. Also developed advanced forensic applications to identify individuals from unsolved crimes using degraded DNA samples for the Federal Bureau of Investigation. Orchid's express DNA service provides forensic DNA analyses in five business days compared with the standard four to five weeks.

- Used for the United Kingdom's scrapie genotyping program to help sheep farmers use selective breeding to eliminate the disease scrapie from their flocks. The company has genotyped over 1 million sheep to date.

The societal benefits of SNP analyses are growing. Typical DNA analysis cost has been reduced by approximately 70 percent, and the time it takes to perform DNA analysis has been reduced by approximately 75 percent, such that DNA analysis can now provide results in about a week (reduced from 4 weeks). Police departments are able to solve cold cases, because SNPstream is able to analyze DNA from degraded samples. It is hoped that pharmacogenetic applications (studying genetic variations related to the onset of disease, and pharmaceuticals) will improve medical treatment.

SciComp: ATP provided in cost-shared funds to $1.9 million to SciComp to develop a software synthesis technology that would simplify the process of mathematical modeling.

SciComp, Inc successfully incorporated simplified mathematical modeling (representing a mathematical device or process) into software for the derivative securities industry. Called SciFinance, this solution includes tools that can automate the pricing of complex derivative securities, organize libraries of pricing codes, and provide risk-management analysis.

As of 2004, SciFinance includes six financial products, four of which incorporate the ATP-funded synthesis technology and two that enhance the other products.

SciComp's software synthesis technology improved the productivity of mathematical modelers by tenfold. SciComp has been awarded two patents based on ATP-funded technology development, and the company has shared knowledge through nine published papers and made several presentations at conferences.

As of 2004, the volume of derivative securities trading has continued to grow, resulting in increased demand for software tools to assist in the pricing of complex derivative structures. SciComp is one of only a few companies that provide these tools.

PART 3

Dissemination of Knowledge

If knowledge from the projects is disseminated—either through products and processes commercialized by the innovators or through publications, patents, and other modes of knowledge transfer—it may benefit other producers in the economy and, subsequently, consumers. The resulting national benefits may go far beyond the returns to the innovating firms and the benefits to their customers.

(The 150 completed status reports discussed in this chapter can be found on-line at http://www.atp.nist.gov/ under funded projects.)

Multiple Ways of Disseminating Knowledge

New knowledge developed in a project can be diffused in a variety of ways. This section discusses two principal means: through patents filed and granted by the U.S. Patent and Trademark Office (USPTO) and cited by others, and through preparation of technical papers that are published or are presented at conferences. Collaborative activity among research and commercial partners, treated in Part 1, is another way by which knowledge is disseminated. Another way is through the observation and reverse engineering of the new goods or services produced directly by the innovators and their partners, discussed in Part 4. Among the other important ways—not explicitly covered here—in which knowledge developed in a project can be diffused are informal interactions among researchers, suppliers, customers, and others; movement of project staff to other organizations; distribution of nonproprietary project descriptions by government funding agencies; and project-related workshops and meetings.

Pathways of knowledge dissemination allow others to obtain the benefits of R&D without having to pay its full cost. When the technology is particularly enabling—in the sense of providing radically new ways of doing things, improving the technical bases for entire industry sectors, or being useful in many diverse areas of application—the spillover benefits to others are likely to be particularly large. The generation of spillover benefits, or positive externalities, from technological advancement is an important argument for public support of enabling technologies.

Balancing Intellectual Property Protection and Knowledge Dissemination

ATP encourages broad dissemination of knowledge produced in ATP-funded projects because it increases the number of potential users of the knowledge and, therefore, may increase national benefits. At the same time, ATP does not force innovating companies to compromise their ability and willingness to pursue early commercial applications of the technology by giving away all of their intellectual property. After all, these companies,

which contribute a substantial share of the costs, have agreed to tackle difficult research barriers and to take the technology to the marketplace as rapidly as possible.

Thus, it is not surprising that the amount of knowledge dissemination varies among the projects. Most of the projects pursue some forms of deliberate knowledge dissemination, such as publishing scientific papers, giving presentations, and forming collaborative relationships. Most projects also engage in considerable unintended knowledge dissemination; for example, as a company's scientists move and work among other companies and universities; as myriad formal and informal discussions occur; as others reverse-engineer their products; and through mergers and acquisitions of the innovating companies.

Public Disclosure of Patent Filing Information

When applying for a patent to protect intellectual property, an inventor must explicitly describe the invention. Because patent law requires that the invention is both novel and useful, the inventor must demonstrate that the invention is essentially different from any other invention and must describe how it can be used. When the USPTO grants a patent, the full application text describing how the invention may be used and how it is related to other technologies is put into the public record and becomes a medium through which knowledge is transferred to others. Hence, patents serve to disseminate knowledge.

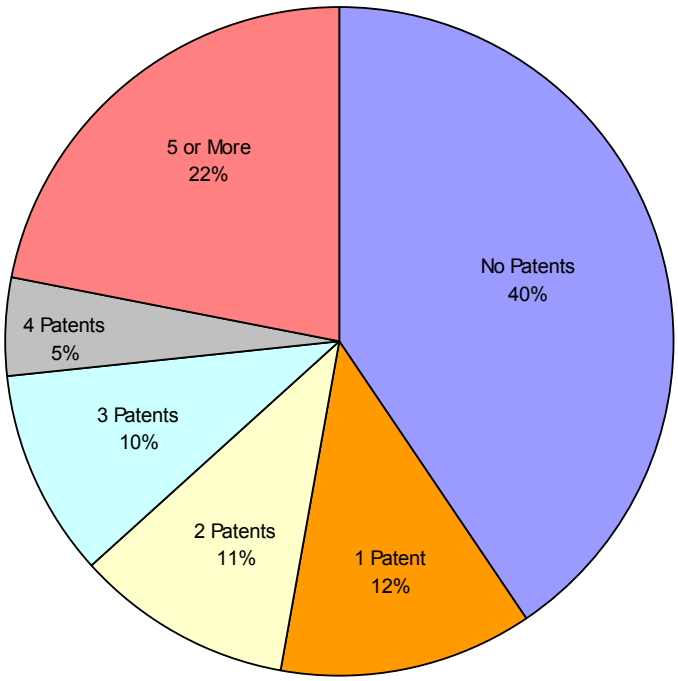
At the same time, patent data are not perfect signals of knowledge creation and dissemination. The decision to seek patent protection for intellectual property is influenced by many factors, including the ease with which others can copy the property's intellectual content and the difficulty of defending the patent position from infringement. Some companies may decide that patent protection is not worth its expense or that a strategy of trade secrets and speed-to-market is more effective. Conversely, patents may be filed as the basic ideas are forming, and trade secrets used in later stages. Furthermore, the importance of patents as a strategy varies among technology areas; for example, patents figure more strongly in electronics and manufacturing than in computer software. The absence of a patent does not mean that intellectual property was not created. But the presence of a patent is a signal that it was created. Despite the limitations, patent statistics serve as useful indicators of knowledge creation and dissemination, and they are widely used by researchers.

Of the 150 completed projects, 89 had filed 500 patents at the time the study data were collected.[2] Eighty-one of the projects had among them a total of 347 patents granted, or 70 percent of the total filed. Thirty-two of the projects had filed a total of 153 patents for which a final decision on granting was still pending.

[2] Patents filed and not yet granted are included here, in addition to those filed and granted, despite the fact that there is no public disclosure until patents are actually granted. The reason for including patents filed and not yet granted is to help offset the problem that there are substantial differences across industries in the lag time between patent filing and granting.

Figure 3-1 displays the distribution of the 150 projects by the number of patents filed, whether granted or not yet granted. More than half the projects have filed one or more patents. Participants in 12 percent of projects had filed a single patent, 26 percent had filed 2 to 4 patents each, and 22 percent had filed 5 or more patents. Forty percent of the projects had not filed a patent.

Figure 3-1
Distribution of Projects by Number of Patents Filed



Source: Advanced Technology Program First 150 Status Reports

Knowledge Disseminated by Patents as Revealed by Patent Trees

Each published patent contains a list of previous patents and scholarly papers that establish the prior art as it relates to the invention. The citations provide a way to track the spread of technical knowledge through patents granted to ATP-funded projects. By following the trail of the patent referenced, it is possible to construct what looks much like a horizontal genealogy tree.

Once the pool of ATP-related patents was identified, computerized tools made available by the USPTO were used to track subsequent patents that refer to each of the ATP-related patents as prior art and the links recorded.[3] The process is then repeated in turn for each of these patents, until the chain of references is complete. Next, the information is

[3] The references to prior patents contained in a published patent are based on information supplied by the applicant and on research by USPTO researchers. There is no way to distinguish between the two sources and no indication that one tends to dominate the other. (USPTO telephone interview with ATP staff, February 11, 2000.)

converted into a graphic format that illustrates the diffusion of knowledge along the path from ATP project patents in the tree.

With the passage of additional time, new branches may emerge as outgrowths of earlier patents. To the extent that later patents are dependent on the earlier ones, the patents in the tree represent developments in knowledge that would not have occurred, or at least not in the same timeframe, had ATP not stimulated the creation and dissemination of that platform knowledge.

Patent Tree Illustrating Knowledge Dissemination

The patent tree in Figure 3-2 shows citations of a patent that came out of an ATP-funded project led by **Texas Instruments, Inc.** during which the company developed a special insulating material, known as aerogel, to overcome problems with interconnect delays as a result of the continuing trend toward miniaturization. The company overcame impediments to aerogel processing early in the project, but in 1997, an industry competitor announced that it would begin using copper interconnect wiring in future integrated circuit designs. After the ATP-funded project Texas Instruments shifted focus away from aerogels for aluminum and began to develop copper interconnects.

The patent tree illustrates how an ATP-funded project whose direct path appears to have slowed or has come to a standstill nevertheless has the potential to remain influential along an indirect path of knowledge utilized and cited in subsequent patents. As the patent tree illustrates, a number of other companies are referencing the Texas Instrument patent, and the potential for beneficial impact from the research continues.

Figure 3-2
Patent Tree for Texas Instruments, Inc. - Patent 5,894,173
Project Impact After Innovator Reduced Activity

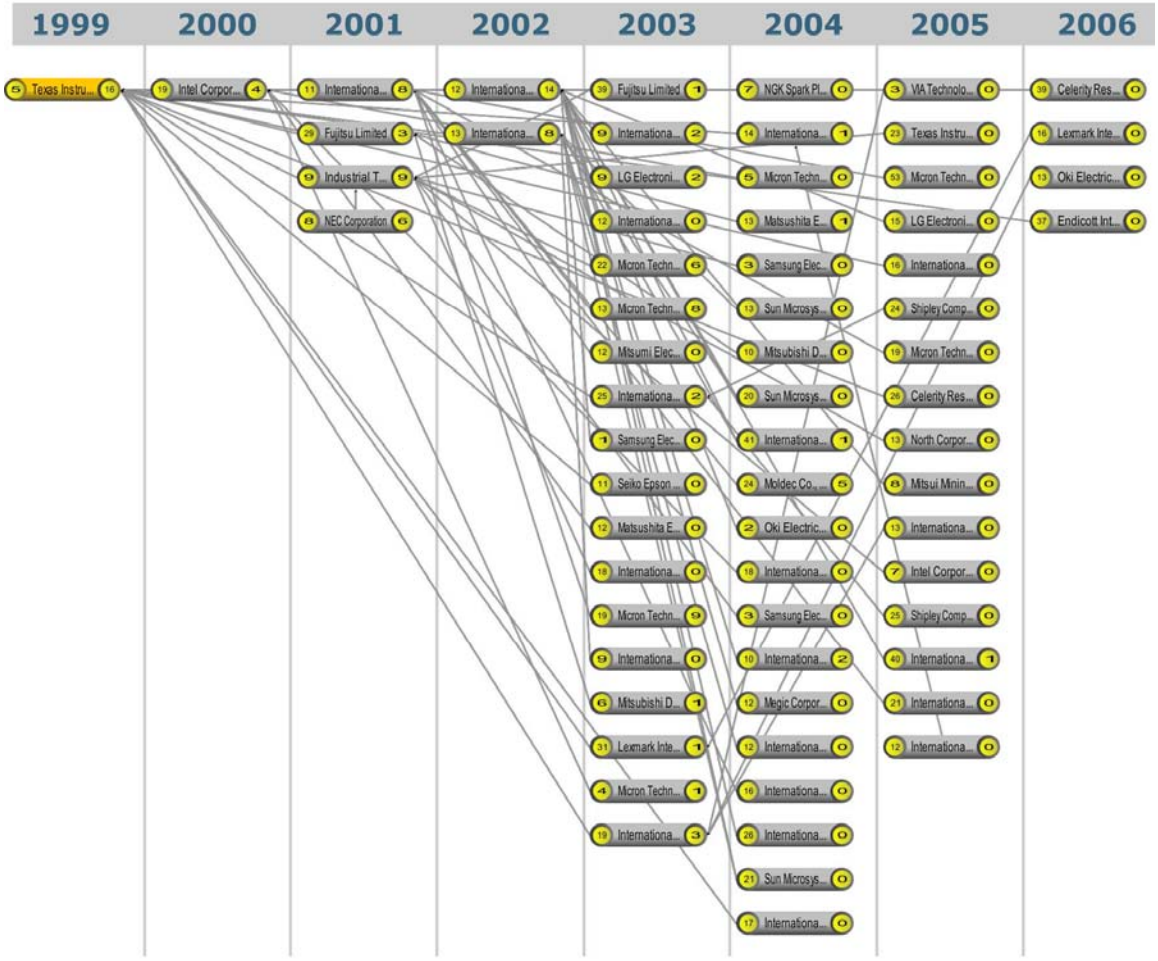


Figure 3-3
Patent Tree for Large Scale Biology Corporation - Patent 5,993,627
Project Impact Where Innovator Went Bankrupt

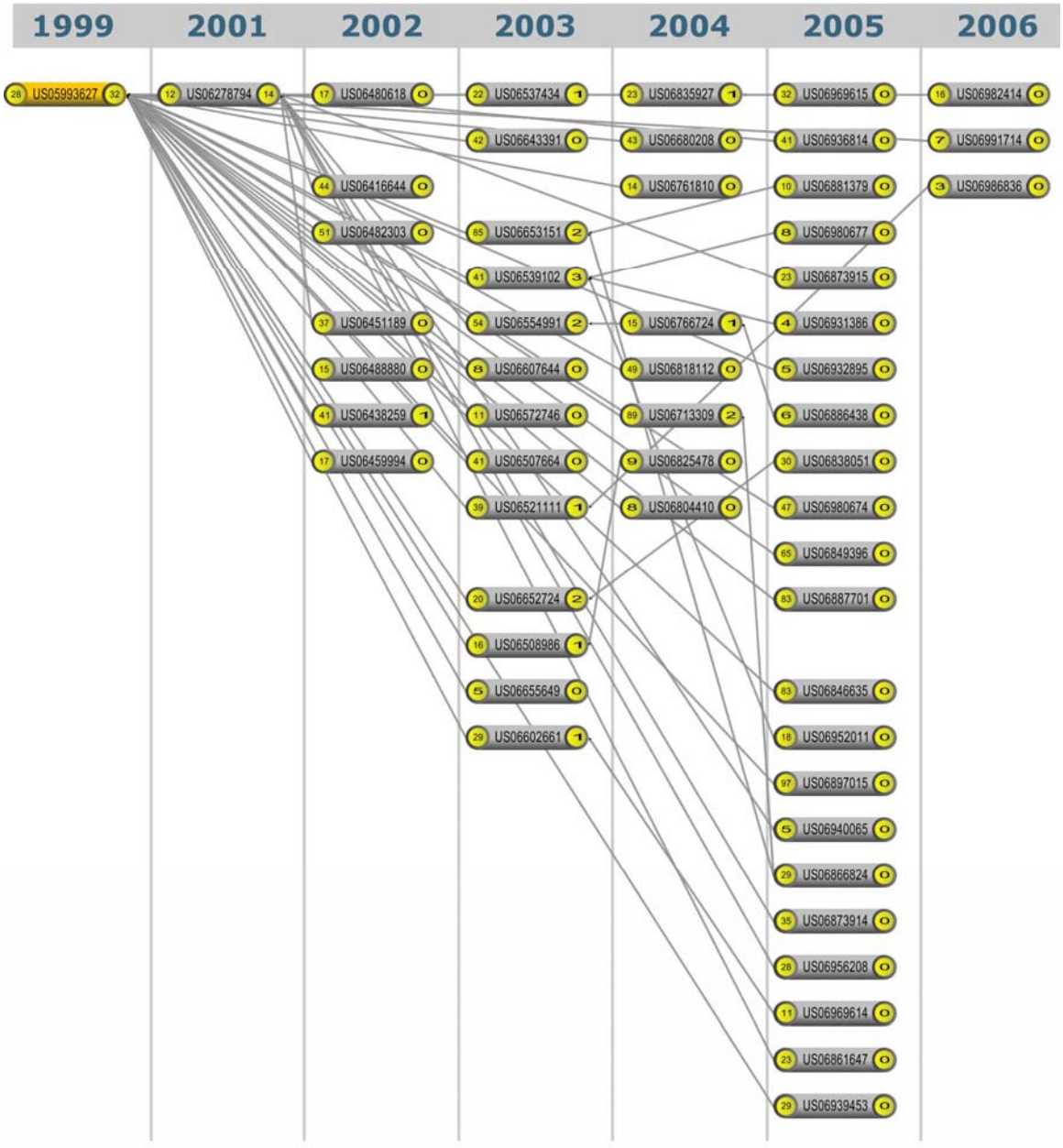


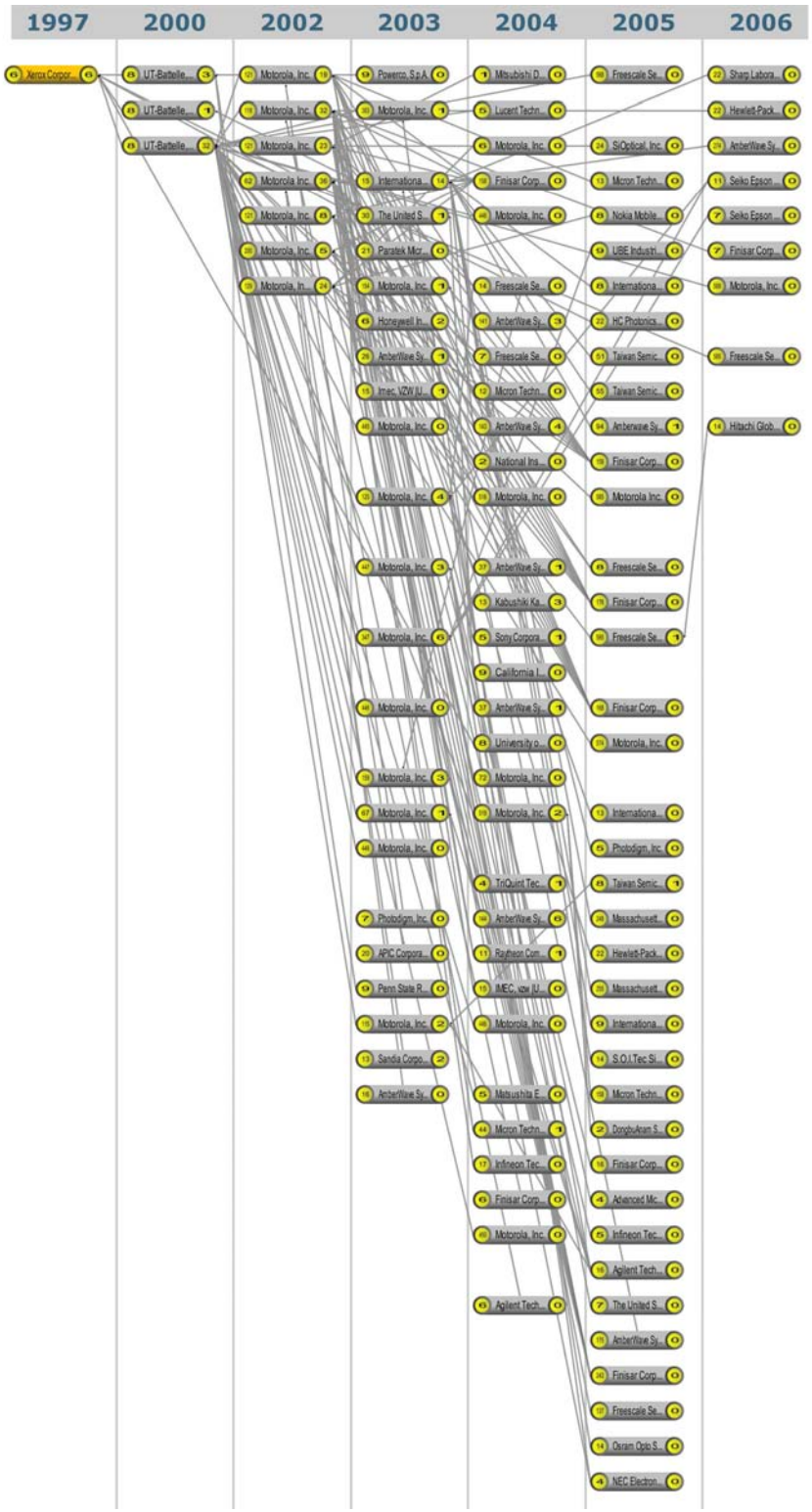
Figure 3-3 shows citations of a patent resulting from a project led by **Large Scale Biology Corporation**. Though the company went bankrupt, the patent tree illustrates how knowledge can outlive its creator and continue to be disseminated. An observer who equates business success of the innovator, one-to-one, with ATP project success may be mistaken, because the indirect path may nevertheless produce important benefits.

Patent Tree Illustrating Extensive Knowledge Flows

Figure 3-4 illustrates just how complex knowledge dissemination through patent citations can become. The path shown is for a patent resulting from an ATP-funded project led by **JDS Uniphase (formerly SDL, Inc.)** and **Xerox Corporation**. With the ATP award, the research team successfully developed high-performance, multibeam red laser diodes; two alternative methods for monolithic integrations of red, infrared, and blue emitters; and several valuable intermediary technologies. From these successes, the ATP-funded project built a strong U.S. technology base for multiple laser applications. Eighty-four inventions from this project have been commercialized into numerous products. This single Xerox patent resulted in approximately 110 citations.

For projects that have received a patent or patents, access to patent trees is available through the individual status reports on the NIST ATP website (http://statusreports-atp.nist.gov/basic_form.asp). Although representing only one aspect of knowledge dissemination, the patent trees extend awareness of the influence of the new knowledge.

Figure 3-4
Patent Tree for Xerox Corporation - Patent 5,963,447
Example of Extensive Knowledge Flows

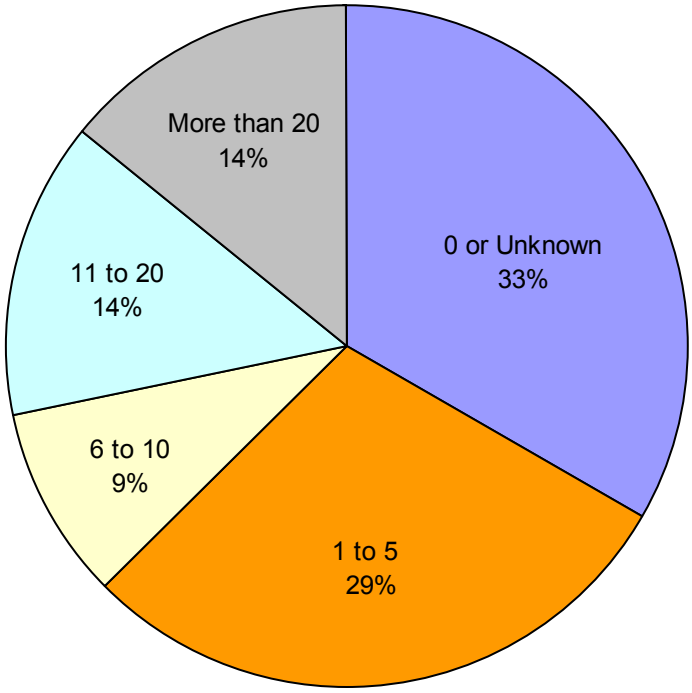


Knowledge Dissemination through Publications and Presentations

Participants in almost 66 percent of the 150 projects had published or had presented papers in technical and professional journals or in public forums. Participants in more than half of all projects had published, and the number of publications totaled at least 831 papers. Participants in nearly 47 percent of the projects had given project-related presentations, and the number of presentations totaled at least 739. Overall, publications and presentations for these 150 projects equaled or exceeded 1570.

Figure 3-5 gives the distribution of projects by their numbers of publications and presentations. Twenty-nine percent of the projects each had between one and five papers published or presented. Nine percent had between 6 and 10 papers published or presented, and another 14 percent had between 11 and 20. At the high end, 14 percent of projects each had more than 20 papers published or presented. Thirty-three percent had no known presentations or publications.

Figure 3-5
Distribution of Projects by Number of Publications and Presentations



Source: Advanced Technology Program First 150 Status Reports

Knowledge Dissemination through Other Means

Aside from publishing, presenting, and patenting, ATP-funded projects have a high rate of collaborative activities. Eighty-seven percent of the projects showed some type of collaboration (see Table 1-1). With so many partners, collaborators, and subcontractors involved, it would be difficult to secure the information. The involvement of so many participants in the projects provides rich avenues of further interaction, and those interactions in turn may increase knowledge flows through personal and professional contacts.

When the government enters into an agreement with an organization, certain information about the agreement is generally made public. Such is the case with ATP and company cost-sharing partnerships. Nonproprietary information has been disclosed to the public for each of the 768 projects funded by ATP in 44 competitions held from 1990 through September 2004 (project information is available on the ATP website[4]). Further, new nonproprietary project descriptions are added to the site as new awards are made. Evaluation reports, such as this one, are also available at ATP's website and provide information to the public.

[4] http://jazz.nist.gov/atpcf/prjbriefs/listmaker.cfm or http://atp.nist.gov (go to Funded Projects Database).

PART 4

Commercialization of the New Technology

New technical knowledge must be used if economic benefits are going to accrue to the nation. This generally means that a new product or process is introduced into the market by the innovating firm, its collaborators, or other companies that acquire the knowledge. In competitive markets, the producer is typically unable to capture all the benefits of a new product or process, and the consumer reaps part of the benefits. The higher up the supply chain the innovation occurs, the more value-added steps there are before final consumption, and the more intermediate firms in the supply chain may benefit, in addition to the final consumer.[5]

(The 150 completed status reports discussed in this chapter can be found on-line at http://www.atp.nist.gov/ under funded projects.)

Commercialization of Products and Processes—A Critical Step Toward National Benefits

When a product or service incorporating new technology reaches the marketplace, a buyer can learn a great deal about the technology. The mere functioning of a new product reveals some information. Intentional investigation, including reverse engineering, reveals even more. More than 60 percent of the 150 projects reviewed for this study had some commercial products or processes based on ATP-funded technology already on the market. Therefore, product use and examination are providing others with information about the new technologies.

Ninety-one of the projects had already spawned or expected to bring to market 222 new products or processes when the data for this report were collected. Companies in 18 additional projects expected to achieve their first commercialized results shortly[6], and

[5] For a detailed treatment of the relationship between spillover benefits (knowledge, market, and network spillovers) and commercialization, see Adam B. Jaffe, *Economic Analyses of Research Spillovers: Implications for the Advanced Technology Program*, GCR 96-708, (Gaithersburg, MD: National Institute of Standards and Technology, December 1996). He notes: "Market spillovers will not be realized unless the innovation is commercialized successfully. Market spillovers accrue to the customers that use the innovative product; they will not come to pass if a technically successful effort does not lead to successful commercialization" (p. 12). In commenting on spillovers that occur because new knowledge is disseminated to others outside the inventing firm, he observes: "Note that even in the case of knowledge spillovers, the social return is created by the commercial use of a new process or product, and the profits and consumer benefits thereby created" (p. 15).

[6] "Shortly" refers to the time when the question is asked. Since Status Reports are written about 5 years after ATP funding ends, the perspective is the same for all status reports. So, when a company answers that

companies in 17 projects that had already commercialized their technology expected to add new products and processes soon. Thus, 73 percent of the projects had spawned one or more products or processes in the market or were expected to do so shortly, for a total of 245 products or processes either on the market or expected shortly after the time the data were collected. Table 4-1 summarizes the commercialization results.

Table 4-1
Progress of Participating Companies in Commercializing the New Technologies

Degree of Progress	Number of Projects	Number of Products/Processes
Project has resulted in at least one Product/Process on the market AND additional Products/Processes are expected soon	17	63
Project has resulted in at least one Product/Process on the market, but no additional Products/Processes are expected soon	74	159
Project is expected to result in a Product/Process on the market soon, but no Product/Process is currently on the market.	18	23
Total Projects that have resulted in Products/Processes on the market OR are expecting to have Products/Processes on the Market soon.	109	245

Source: Advanced Technology Program First 150 Status Reports

A number of additional years have passed since the data for the first 150 projects were collected. Since that time, further developments have doubtless occurred with these projects, which have changed their commercialization results. This overview reports commercial progress of the first 150 projects, all at approximately comparable times following their completion.

A Quick Glance at the New Products

A variety of new products and processes resulted from the projects. For a convenient, quick reference, brief descriptions of the new products or processes for each project are listed in column D in Tables A-1–A-5 in Appendix A. For each new product or process, the new technology on which it is based is also listed in the tables, in column C.

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they expect a product or process on the market soon or shortly, they are referring to new product commercialization in the next 3 to 12 months.

Commercialization: A Critical Step, but Not the Final Word

Commercializing a technology is necessary to achieve economic benefit, but it does not ensure that the project is a full success from the perspective of either the company or ATP. Widespread diffusion of the technology may or may not ultimately follow the initial commercialization. Nevertheless, it is significant that these products and processes are actually on the market.

Rapidly Growing Companies

Rapid growth often signals that a small innovating company is on the path to taking its technology into the market, and one dimension of company growth typically is its employment gains.[7]

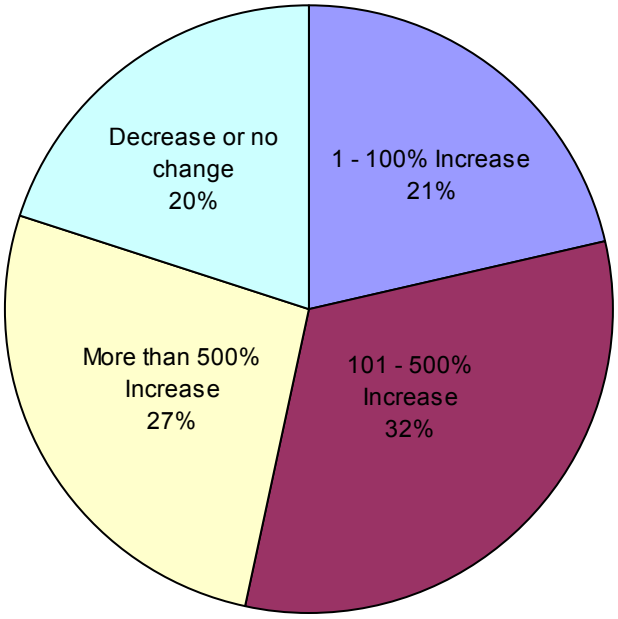
Figure 4-1 shows employment changes for the 75 small-company, single-applicant ATP award recipients.[8] Twenty-seven percent of these companies experienced job growth in excess of 500 percent from the beginning of the project until several years after the project had completed. Thirty-two percent —the largest share— experienced job growth in excess of 100 percent, ranging up to 500 percent. Mergers and acquisitions accounted for 20 percent, or nine of the 45 projects that experienced substantial job growth (substantial job growth being in excess of 100 percent).

Not all the small companies grew. A little more than one-quarter of them experienced no change or a decrease in staff. Several of the companies that were small when they applied to ATP grew so rapidly they moved out of the small-size category. As a group, of the 75 small single-applicant companies, 45 companies at least doubled in size; 14 of them grew more than 1,000 percent. ATP helped these companies develop advanced capabilities, which they subsequently leveraged into major business endeavors.

[7] Employment within the small companies is considered as an indicator of commercial progress. Assessing macroeconomic employment gains from the technological progress stimulated by the 150 projects is beyond the scope of this report.

[8] Employment changes in joint ventures, larger companies, and nonprofit organizations are less closely tied to the success of individual research projects, and, therefore, are not included in the employment data in Figure 4-1.

Figure 4-1
Employment Change at Small Companies that Received a Single-Applicant Award



Source: Advanced Technology Program First 150 Status Reports

The following examples illustrate the potential impact of ATP funding on the employment growth of funded companies.

Incyte Corporation grew from 4 to 215 employees due to the development of flexible techniques for manufacturing chem-jet-based microarrays. The technique synthesizes large arrays of specific DNA fragments suitable for medical diagnosis, microbial detection and DNA sequencing, and for creating supplies of detachable oligonucleotides for subsequent use. (Project number 94-05-0019)

Nanophase Technology increased employment from 2 employees at the start of the ATP project to 61 employees at the time the status report was written. The employment is a result of Nanophase's development of a technology that enabled a 25,000-fold increase in the development of nanoscale materials and a 20,000-fold reduction in cost. (Project number 91-01-0041)

Capital Attraction

Attraction of additional capital is another signal that a company is positioned to make further progress. Of the 150 projects, 104 had attracted additional capital to further pursue development of their technologies. Additional funding came variously from collaborative partners, venture capitalists, public offerings of stock, other governmental departments including state government programs, and other sources.

Members of the **Genosensor Consortium** attracted additional internal funding after successfully developing a technology for automated DNA sequence analysis during the ATP-funded project. (Project number 92-01-0044)

eMagin Corporation received a $3 million grant from the U.S. Air Force after successfully developing microdisplays that have been integrated into hundreds of medical, commercial, and military applications. (Project number 93-01-0154)

ABB Lummus attracted additional internal capital after the ATP project as a result of the company's successful development of a new, environmentally superior process to manufacture alkylate using solid-acid catalysts. (Project number 95-05-0034)

The Dow Chemical Company also attracted additional capital due to the methodologies developed during the ATP project to create a direct, economical, single-product oxidation process incorporating a silver-based catalyst for conversion of propylene to propylene oxide. (Project number 95-05-0002)

P A R T 5

Overall Project Performance

The individual performance of the 150 completed projects has varied, as, measured by the creation and dissemination of knowledge and the accelerated use of that knowledge for commercial purposes. Some of the award-recipient companies grew by leaps and bounds as they translated their knowledge gains from ATP-funded research into profitable and beneficial products, services, and production processes. Some continued to strive toward hard-to-achieve goals, while others showed little outward signs of further progress. A few that achieved impressive research accomplishments later failed in the commercialization phase. However, the achievements of the more successful projects, with their impressive new performance capabilities resulting in lower costs and higher quality products and processes, appear to have much more than compensated for the less successful projects. There is considerable evidence that the benefits attributable to ATP from the 150 completed projects substantially exceed their costs.

(The 150 completed status reports discussed in this chapter can be found on-line at http://www.atp.nist.gov/ under funded projects.)

Composite Performance Scores

During the intermediate period covered by this analysis—after project completion but before long-term benefits have had time to be realized—ATP uses a Composite Performance Rating System (CPRS) to help gain a sense of how projects in the portfolio have performed overall thus far against ATP's mission-driven multiple goals.[9] In this intermediate period of project life cycles, the focus is on progress toward the goals of 1) knowledge creation, 2) knowledge dissemination, and 3) commercialization. The CPRS uses a weighted composite of output data systematically collected for each of the 150 projects—some of which have been presented in aggregate form in the preceding sections of this overview—to assess overall performance of the portfolio of completed projects in this intermediate period.

The output data serve as indicator metrics of progress toward achieving goals. Examples of available indicator metrics signaling progress toward the creation and dissemination of knowledge are a) awards for technical excellence bestowed by third-party organizations,

[9] For an in-depth treatment of the CPRS, which was developed in prototype for ATP's use, see Rosalie Ruegg, *Bridging from Project Case Study to Portfolio Analysis in a Public R&D Program*, NIST GCR 03-851 (Gaithersburg, MD: National Institute of Standards and Technology, 2003).

b) patent filings, c) publications and presentations, d) knowledge dissemination from potential reverse engineering of new and improved products/processes on the market or expected soon, and e) collaborative activity. Available indicator metrics signaling progress toward commercialization of the new technology include a) attraction of additional capital, b) employment gains, c) project-related company awards for business success, d) moving products and processes into the market, and e) analysts' outlooks for future progress by the award-recipient companies.

Weights are assigned to the indicator data, which are combined to produce a composite numerical score that is then converted to a zero- to four-star rating for each project. A score of one star or less signals poor overall performance; two stars, moderate performance; three stars, strong performance; and four stars, outstanding performance. The distribution of CPRS scores computed for each project in a portfolio of projects is then examined, and the results taken as indicative of overall portfolio performance.

The resulting CPRS ratings provide an easy-to-grasp highlighting of portfolio performance in the intermediate period. They call out those projects that have exhibited outstanding or strong outward signs of progress towards long-run program goals during the years covered and those that have exhibited moderate or few signs of progress. However, the ratings are imperfect and should be viewed as only roughly indicative of overall performance.

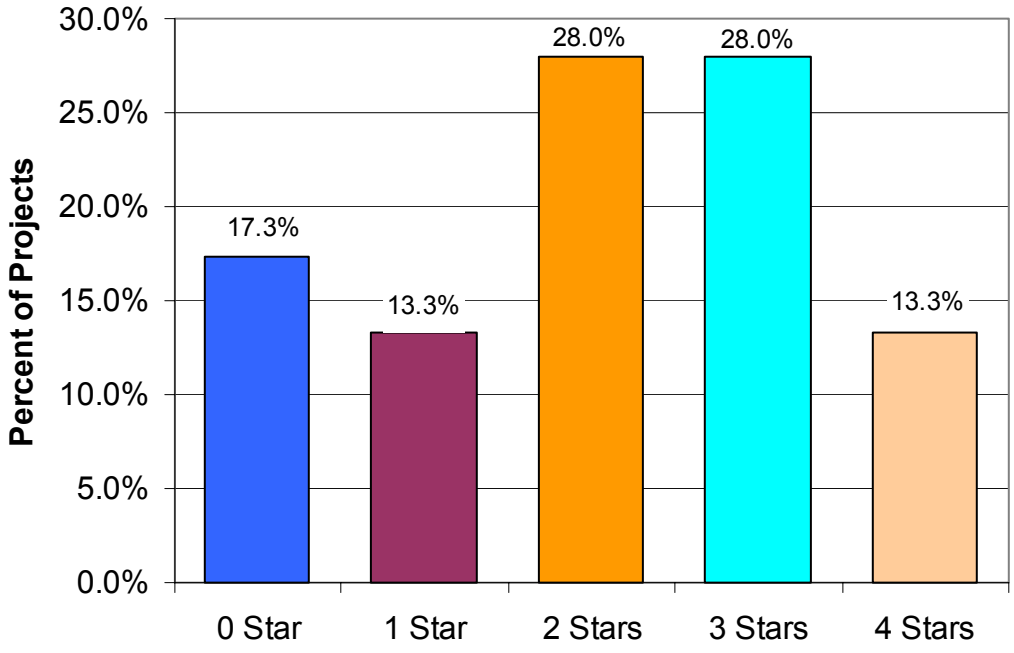
The performance metrics are consistent with the view of varying degrees of success—with knowledge creation and dissemination constituting partial success, and a continuation into commercialization constituting a fuller degree of success in terms of project progress. Some companies carried out their proposed research with a degree of success during the time of ATP funding, but then did not continue pursuit of their project's larger goals after ATP funding ended. At this stage of evaluation, ATP considers such projects only partial successes, because the direct path for achieving project goals is truncated. Such projects are not among the higher scorers in this report. It is possible, however, that developments along the indirect path (diffusion of knowledge from the project through publications, presentations, patents, and licensing) may nevertheless occur—particularly if a project produced effective knowledge transmitters, such as patents and publications. It is also possible that a company may work in secrecy for a long period of time with no visible outputs and then suddenly explode on the scene with a single output that will yield large societal benefits.

Limiting factors include the extent to which not all relevant effects are captured; moreover, the use of indicator metrics is constrained by data availability, the development of the weighting system is empirically driven rather than theoretically based, and the ratings do not directly measure national benefits. The degree of correlation between a project's performance score and its long-run societal benefits is impossible to know at this time. Projects with the same scores are not necessarily equal in their potential benefits. They are, however, somewhat comparable in terms of the robustness of their progress to date.

Scoring the First 150 Completed Projects

The distribution of CPRS scores for ATP's first 150 completed projects is shown in Figure 5-1. Combining the two and three-star categories shows 56 percent of projects performed at a moderate level. Thirteen percent of the projects performed at a high (four-star) level and approximately 30 percent of the projects scored one star or less, perhaps not surprising for companies taking on difficult goals.

Figure 5-1
Distribution of Projects by Star Rating



Source: Advanced Technology Program First 150 Status Reports

The 20 four-star projects overall include 16 single-applicant projects led by small companies and four joint ventures, two led by a consortium and two led by small companies. Leaders of these top-scoring projects are listed in Table 5-1.

Table 5-1
List of Four-star Projects

Aastrom Biosciences, Inc.	Nanophase Technologies Corporation
American Superconductor Corp.	National Center for Manufacturing Sciences (NCMS)
Automotive Composites Consortium (a Partnership of DaimlerChrysler [formerly Chrysler], Ford and General Motors)	Orchid BioSciences (formerly Molecular Tool, Inc. Alpha Center)
Cerner Corporation (formerly DataMedic - Clinical Information Advantages, Inc.)	SciComp, Inc.
ColorLink, Inc.	SDL, Inc. and Xerox Corporation
Cree Research, Inc.	Third Wave Technologies, Inc.
Engineering Animation, Inc.	Tissue Engineering, Inc.
Integra LifeSciences	Torrent Systems, Inc. (formerly Applied Parallel Technologies, Inc.)
Kopin Corporation	Xerox Palo Alto Research Center
Large Scale Biology Corporation (formerly Large Scale Proteomics Corporation)	X-Ray Optical Systems (XOS), Inc.

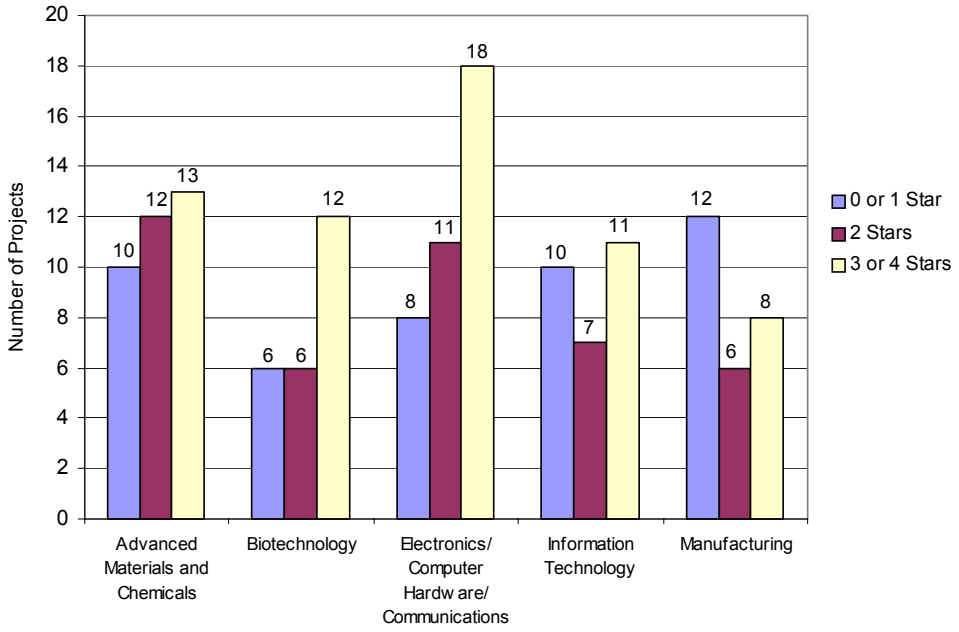
The three-star projects included 35 single-applicant projects and 7 joint-venture projects. Of the single-applicant projects, 25 were led by small companies, two by medium companies, and eight by large companies. Of the joint ventures, two were led by small companies, two by an industry consortium, two by a large company, and one by a nonprofit organization.

A few projects with low CPRS ratings had impressive technical achievements as indicated by the receipt of a third-party technical award, though most of the technical awards went to those with the highest overall ratings. In contrast, all of the awards for business acumen went to the projects with CPRS ratings of three or four stars

Performance by Technology Areas

Overall project performance in the intermediate period covered by the study varied by technology area, as illustrated in Figure 5-2. Of the 24 Biotechnology projects, 12 were three- or four-star projects. Of the 37 Electronics projects, half scored high. Of the 26 Manufacturing projects, close to third scored high, but 46 percent scored low. The 35 projects in the Advanced Materials and Chemical group were more evenly divided into high, low, and moderate scorers. The 28 Information Technology projects had 11 projects that were high-scoring projects, 7 moderate-scoring, and 10 low-scoring projects. Differences in life cycles among the technology areas may account for part of the performance differences, but the relatively small number of projects in each category does not support the drawing of robust conclusions about how projects in the different technology areas will perform.

Figure 5-2
Number of Composite Scores by Technology Area



Source: Advanced Technology Program First 150 Status Reports

Project Performance Translated into Economic and National Security Benefits

Photonics

ATP has provided cost-sharing funding to more than 120 photonics projects since 1991[10]. To access the economic benefits from a portion of these projects, the author adopted a cluster study approach to combine the methodological advantages of detailed case studies and of higher level overview studies. The following five projects were selected for analysis: Capillary Optics for X-Ray focusing and Collimating; MEMS-Based Infrared Micro-Sensor for Gas Detection; Infrared Cavity Ring-Down Spectroscopy; Optical Maximum Entropy Verification; and Integrated Micro-Optical Systems.

Findings from the study indicate that U.S. industry and consumers, and the nation, will enjoy at least $33 of benefits for every dollar of ATP's $7.47 million investment in the cluster of five projects. ATP technology translates into $1.90 already realized benefits generated for every dollar of ATP's investment in the five projects.

Component-Based Software (CBS)

Developing the capacity to build large software systems from assemblies of smaller, reusable, independent components is an important strategy to reduce software system

[10] Pelsoci, Thomas, M., *Photonics Technologies: Applications in Petroleum Refining, Building Controls, Emergency Medicine, and Industrial Materials Analysis*. NIST GCR 05-879 (Gaithersburg, MD: National Institute of Standards and Technology, September 2005).

costs, increase system reliability, and enable lower cost upgrades. Three projects included among the first 150 Status Reports were part of a portfolio of 24 projects that was included in an in-depth economic case study conducted by RTI.[11] These projects were led by **Reasoning Inc.**, **TopicalNet, Inc. (formerly Continuum Software)**, and **HyBrithms (formerly Hynomics Corp.)**.

Across the entire CBS portfolio, RTI's economic study estimated $840 million in net-present-value benefits and a benefit-to-cost ratio at 10.5, suggesting that the investment in the portfolio of projects as a whole was worthwhile. The net-benefits estimate is based on the cost of all 24 projects, but the benefits of only 8 were the subject of the detailed case study. In addition, the study found other benefits that were presented qualitatively, namely, enhancing the credibility of the mostly small software firms that were funded and assisting firms in strengthening their planning and management functions.

Reasoning Inc., TopicalNet Inc. (formerly Continuum Software), and Hynomics Corp. (formerly HyBrithms) had commercialization activities underway when RTI conducted its study. Their costs, but not their benefits, were included in RTI's aggregate portfolio net-benefit measure, because they were not among the eight projects selected by RTI for the portfolio benefits assessment. Thus, the RTI study results, at best, suggest that the three projects are part of a portfolio of projects found to be valuable. Of the three projects, two are rated as three-star performers, and one is a two-star performer.

It is also informative to look at how some of the other projects that were rated as top performers have progressed since the original data were compiled and the CPRS ratings calculated. Additional projects are profiled below.

Scalable Parallel Programming

One of the top-performing projects among the first 50 completed projects, originally profiled in Volume 1, was a project led by **Torrent Systems, Inc.** Although Torrent had fewer knowledge-dissemination outputs than the other top-performing projects, its exceptional commercialization efforts boosted it into the four-star group. The project developed a component software system that insulates programmers from the complexities of parallel programming while allowing them to use it productively in scalable applications. Torrent delivered this new capability in its software product, Orchestrate™. An early user of the new software, United Airlines, was able to increase its revenue by $100 million per year as a direct result of using Orchestrate™.[12]

When revisited in Status Reports, Volume 2, Torrent's technology was reported to be enabling e-businesses and other companies to process and analyze unlimited volumes of data. Torrent was listed in *Computerworld's* "100 Hot Emerging Companies" in 1998 and received a number of other awards recognizing both its software technology and business acumen.

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[11] White and Gallaher, November 2002.

[12] Information from Hoover's Online company search and Torrent's website, current August 31, 2000.

Since that time, Torrent, which had only two employees when it received its ATP award, has been acquired for a purchase price of $46 million by Ascential Software Corp., a global company with a market capitalization of $1.1 billion, headquartered in Westboro, Massachusetts.[13] According to Ascential's Chairman and CEO, Peter Gyenes, "Torrent's patented and proven parallel processing technology is a perfect complement to the rich feature set within our data integration solution, DataStage."[14] According to additional public statements by the company, Ascential has integrated Orchestrate™ into its DataStage XE product family, with the result that customers will be able to integrate data of virtually any volume and complexity, with infinite scalability, and turn growing amounts of data into valuable information assets.

United Airlines, first a Torrent customer and then an Ascential customer, is using Orchestrate™ and an IBM parallel-processing computer to design a system for managing airplane seat assignments. A statement by Bob Bongirno, managing director of applications development for United Airlines, which is posted at the Ascential Software Corp. website provides a user's perspective of the importance of the product:

> *"At United, we analyze 'astronomical' amounts of data every day through our Orion system to determine the optimum seat availability and price across tens of millions of passenger itineraries," he said. "For Orion and our other data-intensive applications, we demand a parallel processing technology that is robust and reliable enough to process massive data volumes on very large systems and will provide a state-of-the-art data integration foundation that helps us manage all our disparate data sources and accelerates the development of new applications. The combination of technologies from Torrent and Ascential holds great promise for meeting the data processing needs of customer-centric organizations like United."*

Thus the commercialization path has grown more complex for this ATP-funded technology as the technology has been combined with other software elements. At the same time, the impact potential of the technology appears strong. According to Doug Laney, META Group Vice President, the worldwide market for data integration was projected to grow from $900 million in 2001 to $1.3 billion in 2004,[15] and the technology platform funded in part by ATP appears well positioned to play a role in serving this growing market. Those projections were well-founded. Ascential grew rapidly in 2004, with a 46 percent increase in total revenue. In March 2005, Ascential agreed to be acquired by IBM for approximately $1.1 billion, strengthening IBM's fast-growing information integration business.[16] (Project number 94-06-0024)

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[13] Standard and Poor's stock report on Ascential Software Corp.

[14] Press Release, November 28, 2001, available on-line at www.ascentialsoftware.com, Press Center.

[15] Ibid.

[16] Company press release, "IBM to Acquire Ascential Software." March 14, 2005. (http://ibm.ascential.com/news/pr.html/view/1107)

High-Temperature Superconducting (HTS) Wire

The project led by **American Superconductor Corporation (AMSC)** is another of the top-rated 100 completed projects profiled originally in Status Report Volume 1. At the time Volume 1 was being written, the company was beginning to launch its commercialization effort. Since then, the company has reportedly continued making impressive advances, building the world's first high-volume HTS wire manufacturing plant with a capacity to manufacture 20,000 kilometers of wire per year when it is fully equipped. This new manufacturing capacity is said to give potential customers the ability to accelerate their schedules for launching commercial products incorporating HTS wire by making the product available to them in commercial quantities, at commercial prices.[17] AMSC's products and services listing now shows a vertically integrated portfolio that includes HTS wire, motors, generators, synchronous condensers, industrial power quality solutions, power conversion, and transmission grid solutions.

A press release issued October 1, 2003, announced that AMSC had received additional funding from the Department of Defense (DOD) and Department of Energy (DOE) to support further manufacturing scale-up for second-generation HTS wire. According to Dr. Paul Barnes, U.S. Air Force Superconductivity Team Leader, ensuring that the United States will have a reliable supply of the second-generation HTS wire is expected to be central to the development of many future military systems, including lightweight high-power generators and advanced weapon systems. According to James Daley, manager of the Superconductivity program at DOE, the technology is also expected to play an important future role in upgrading the nation's power grid.[18] (Project number 91-01-0146)

Visualization Software

As in the preceding examples, **Engineering Animation, Inc. (EAI)**, leader of another of the top-performing projects and originally profiled in Status Report Volume 1, continued to aggressively and successfully pursue applications of its award-winning imaging software capabilities developed in the ATP-funded project. Founded by two professors and two graduate students in 1990, EAI had 20 employees at the time ATP made the award. According to company officials, the ATP award allowed it to significantly extend its capabilities in computer visualization and computations dynamics and to form important collaborative relationships that enabled it to leverage the technology in many different directions. The company used its ATP-funded technology to improve the training of doctors as well as to guide medical procedures. Furthermore, patients reportedly had better outcomes when the visualization software was used during their surgical procedures.

In 1999, the company employed approximately 1,000 staff members and had sales of $71 million. At that time, EAI had extended and deployed its award-winning visualization

[17] Information provided by the company at its website, www.amsuper.com.

[18] Company press release, October 1, 2003.

capabilities to develop a virtual factory technology implemented at Ford Motor Company. This application of the software enabled faster design and analysis of factory models.

On October 23, 2000, EAI was acquired by Unigraphics Solutions Inc. for $178 million. Subsequently, through acquisition and merger, Unigraphics and another software services company, SDRC, became a combined subsidiary of Electronic Data Systems Corporation (EDS), the world's largest information technology outsourcing services company, which has a worldwide infrastructure and 138,000 employees.[19] Unigraphics and SDRC were combined to form EDS's fifth line of business, Product Lifecycle Management (PLM) Solutions. This union provided, through Unigraphics NX software, a unified approach to extended enterprise collaborations enabling the modeling and validation of products and their production processes digitally from initial concept to finished parts. Thus, EAI followed the business model for growth of merging with a much larger company.[20] An online search revealed that previously developed EAI products and books remain on the market. (Project number 91-01-0184)

Examples of strong projects from among the three and four -star group are described below. These, too, appear to be delivering important economic benefits.

Improving Software Efficiency through Reusable Components

An example is a four-star project led by **Xerox Parc** which is credited with developing aspect-oriented programming (AOP) and later developed products that incorporated its principles. After the ATP funded project ended Xerox developed AspectJ, an open-source language based on AOP. Aspect J extends Java; and is being further developed and used in IBM's software applications and by many others. Eight patents emerged from this ATP-funded project and more than 3,250 articles or books have been written about AOP. In June 2003, AspectJ won the JavaWorld Editors' Choice Award for the Most Innovative Product or Technology Using Java. (Project number 94-06-0036)

Miniature LCSs Enhance High-Definition Displays

Another four-star project with continued strong commercialization was led by **Kopin Corporation**. Kopin formed a joint venture with Philips, and together with their subcontractor, Massachusetts Institute of Technology facilitated a paradigm shift in high-definition display technology. During the ATP funded project, Kopin and Philips combined existing monochrome liquid crystal displays (LCDs), with color, signal processing, and high-definition technology. Independently, Philips successfully commercialized high-resolution projection HDTVs using the ATP-funded technology. Kopin also successfully applied the ATP-funded enabling technology in numerous applications including miniaturized display applications for use in viewfinders for camcorders and digital cameras, wearable computers, virtual reality games, and military

[19] Prior to the acquisition of Unigraphics, EDS was the major company stockholder. Information found at www.eds.com.

[20] Ibid.

applications. LCD projection display technology is a key product differentiator in U.S. electronics manufacturing. (Project number 94-01-0304)

Structural Composites for Large Automotive Parts
As a result of the ATP funded project the **Automotive Composites Consortium-ACC**, (A partnership of DaimlerChrysler [formerly Chrysler], Ford and General Motors) successfully produced a prototype box for a pickup truck that is stronger and more durable than steel, does not rust, is visually attractive, requires no bed liner, and improves fuel efficiency through its light weight (36 pounds, or 33 percent, lighter than steel). This pickup truck box gave the ACC member companies (General Motors [GM], Ford, and Chrysler, which later became DaimlerChrysler) the knowledge and tools to develop commercial products and to continue innovative research, based on this initial success. Applications of this successful ATP-funded technology include strong, lightweight components for aircraft, firefighter helmets, and marine motor covers. Project researchers shared their developments through one granted patent and several articles and presentations. As public acceptance of tough, durable composites increases, applications are expected to broaden. (Project number 92-01-0040)

To these examples, other promising technologies may be added—technologies that improve productivity, facilitate better weather forecasts, improve communications, enable new drug discovery, reduce energy costs, and improve health and safety.

What Difference Did ATP Make?

ATP aims to improve the international competitiveness of U.S. firms by funding projects that would not take place in the same timeframe, on the same scale, or with the same goals without ATP's support. A project may be successful in terms of achieving its goals, but if the same accomplishments would have occurred in the same timeframe without ATP, then the program has not had the intended effect. For this reason, evaluation studies of ATP—as well as other government programs—should apply the principle of "additionality" to correctly distinguish between benefits that would likely have occurred anyway and those benefits that are reasonably attributable to ATP.

In preparing the 150 individual mini-case studies, analysts asked project leaders about the role ATP funding played in their projects. Throughout the project selection process, beginning with the application, ATP presses the questions of why the project requires ATP funding, why funding is appropriate, what will happen if ATP funding is not provided, and how the expected outcome will differ with and without ATP involvement. During the evaluation process, these questions are again pursued retrospectively, i.e., what happened that was different as a result of ATP? Applied prospectively, the results are hypothetical. In evaluation studies, the results may be based on counterfactual survey and interview questions, such as those posed in the status report case studies. Evaluation studies have also used control group techniques, which provide more reliable evidence of the additional impacts of ATP.[21]

[21] See *Survey of Applicants 2002*, NIST GCR 05-876, (Gaithersburg, MD: National Institute of Standards and Technology, June 2005).

Forty-six percent of the respondents indicated their projects would not have happened at all without ATP funding. Indeed, some participants said their companies would have gone out of business had the ATP award not been made.

Thirty-eight percent of the respondents said they would have attempted the project at some later date or at a slower pace and that ATP funding enabled them to accelerate the technology. Table 5-1 shows the project time savings attributed to ATP for those projects that reported they would have proceeded without ATP funding. With ATP, the projects avoided delays ranging from six months to five years and more. The acceleration of some of the projects may seem short; however, the value of even a small acceleration can be substantial. Speed in developing and commercializing a technology can also mean increased global market share for U.S. producers.

Table 5-2
Effect of ATP Funding on Expected Timing of Research

Effect on Project			Number of Projects
Would not have conducted Project without ATP funding			69
Would have proceeded without ATP funding, but with a delay*:			57
	Length of Delay		
	6 months	*1*	
	12 months	*3*	
	18 months	*7*	
	21 months	*3*	
	24 months or more	*10*	
	More than 5 years	*11*	
	Delay, but time unspecified	*22*	
No Response			24
Total			150

Source: Advanced Technology Program First 150 Status Reports

*Another factor potentially influenced by ATP funding (the scope and scale of the project) was not explicitly covered.

**The Printed Wiring Board Joint Venture project had a split response: half the tasks would not have been done at all and half would have been delayed by at least a year. This result is recorded conservatively in Table 5-1 as a two-year delay.

A number of companies also reported other effects of their ATP awards. Some reported that receiving their award enhanced their ability to raise additional capital. Some reported that their award helped them form collaborative relationships for research and commercial activities. Others reported that receipt of their ATP award had enabled them to gain in international competitiveness.

What Constitutes Success and Failure for ATP?

Because individual project failure must be allowed and tolerated in a program that focuses on overcoming challenging technical barriers to innovation, it is essential to take a portfolio approach to assessing ATP. Moreover, success should be assessed against the legislated mission of the program.

Four general tests, and several additional specific tests—all derived from ATP's mission—if applied after sufficient passage of time, should reveal the extent to which ATP has successfully met its mission, as described below.

Test 1: Has the portfolio of ATP-funded projects overall produced large net social benefits for the nation?

Test 2: Has a substantial share of net social benefits accrued to citizens and organizations beyond ATP direct award recipients?

Test 3: Did ATP make a substantial positive difference in the size and timing of the benefits?

Test 4: Has the portfolio of ATP-funded projects enhanced United States' economic and technological competitiveness?

Additional specific tests of success include the following: Did the projects produce new scientific and technical knowledge? Did ATP increase collaboration? Were small businesses able to participate? Were manufacturing capabilities improved?

While the ultimate answers to these success "test questions" depend on the long-run impacts of the entire portfolio of ATP projects, the performance-to-date of the sub-portfolio of 150 projects provides emerging answers.

There is mounting evidence that the tests for program success are being met. First, there is strong evidence that social benefits of the portfolio are large and exceed program costs. Second, there are benefits extending well beyond those captured by the direct award recipients: there is substantial evidence of knowledge and market spillovers as others cite the project patents and use the products. Third, there is evidence that ATP has made a significant difference in the amount and timing of benefits, as well as having other beneficial impacts on the companies. Fourth, there is some evidence of improvements in the competitiveness of U.S. companies.

The performance ratings show that the majority of the projects continued to make progress in the several years after ATP funding ended. Moreover, the portfolio has been shown to contain a core group of highly active and productive projects that are successfully accomplishing their high-risk project goals.

ATP awarded a total of $621 million to the 150 completed projects. Questions of keen interest are what is the public investment producing in the way of benefits, and are the tests for program success being met? Estimated benefits attributed to ATP from just a few of the 150 projects for which quantitative economic benefits have been provided exceed ATP's funding for all of the 150 projects. In addition, there is considerable evidence of large project benefits that have not yet been quantified.

This completes the portfolio view of ATP. Appendix A that follows provides an overview of the 150 individual projects that make up the portfolio. Appendix B describes reasons that some ATP-funded projects did not proceed to completion. Appendix C lists the first 150 completed projects along with their CPRS star ratings.

Aphios Corporation

Marine Microorganisms and Saline Fermentation: A New Industrial Resource

In 1995, the discovery of new pharmaceuticals, such as antibiotics, from land-based genetic sources was beginning to dwindle. Scientists needed a new source to search for genetic material. Aphios, a small biotechnology company, had a plan to systematically collect and assess samples of microorganisms from a variety of marine sources at different ocean locations and depths (such as deep-water sediments, shallow mangrove swamps, and salt ponds) and to develop a technology platform for culturing, fractionating and screening the marine microorganisms in order to develop a library of potential therapeutics. Aphios applied for funding from the Advanced Technology Program (ATP) in 1995; their proposal included many recognized expert subcontractors, such as Bristol-Myers Squibb, Kelco division of Monsanto, CalBioMarine Technologies, Harbor Branch Oceanographic Institution, Massachusetts Institute of Technology, One Cell Systems, and Scripps Institution of Oceanography. The project was innovative and high risk because keeping a comprehensive library of marine microorganisms alive for future research posed significant challenges. Moreover, robust techniques were not readily available for efficiently culturing marine microorganisms in sea water-like media, reproduction of these organisms had never been done on a commercial scale, and little or no facilities existed to produce new medications from marine organisms.

ATP awarded cost-shared funding to Aphios for two years, beginning in 1995; the project was later extended to three years, because technical problems slowed progress. Aphios continued testing the marine samples after the project concluded, and by 2004, maintained a library of over 1,400 marine microorganisms from which they have derived over 20,000 partially purified fractions (partially purified portions of a microorganism) with the potential for making more than 56,000 partially purified fractions. While none of these have been commercialized as a therapeutic to date, research based on this project has identified several marine microorganisms that contain potential therapeutics for treating smallpox, influenza, HIV/AIDS, multiple-drug resistant bacteria, and dental plaque. Moreover, Aphios was awarded two patents for discoveries related to its ATP-funded research and has one patent pending, and they shared their knowledge in a variety of publications and presentations.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* *

Research and data for Status Report 95-01-0263 were collected during August – September 2004.

Marine Environments Provide Potential New Medications

In the early 20th century, the average American's lifespan was 49 years. All three leading causes of death were infectious diseases: pneumonia, tuberculosis, and gastrointestinal disease. This changed in 1929 with the discovery of penicillin, an antibiotic originally derived from mold that heralded the era of antibiotics. Scientists realized that microorganisms are a rich source of antibiotics and other clinically useful natural products, such as antimicrobial cleansers. Since 1929, 30,000 to 50,000 natural products have been discovered from terrestrial microorganisms. More than 8,000 of the

natural products discovered since 1929 exhibited or had antibacterial activity, which resulted in over 100 microbial products used as antibiotics, anti-cancer agents, and agrochemicals. These products, in conjunction with improved sanitation and the industrialization of medical and hospital care, helped to lengthen the American's average lifespan to 78 years by 1995. However, scientists were running out of new genetic and therapeutic discoveries from terrestrial sources and, therefore, needed to look elsewhere.

Because of the wide variety of marine microenvironments, oceans and seas contain a wealth of microorganisms that are vastly different from their terrestrial counterparts and provide a new source for scientists in isolating effective therapeutics. With their broad diversity of habitats (including polar, temperate, and tropical), marine environments possess a wide variety of salinities, temperatures, pressures, and conditions that foster high microbial diversity. However, out of the billions of species in these marine environments, only a few thousand had been isolated and described by the 1990s.

Aphios, a biopharmaceutical company, hoped to develop a marine microorganism library to be used in screening new drug leads.

A standard method used to discover new drugs is to isolate and screen novel chemical extracts produced by microorganisms, called secondary metabolites, together with parts of the microorganisms themselves, for biological activity against disease targets to identify active candidates (an initial response is called a hit. After identifying and dereplicating the hit, it may become a candidate). However, this screening process is time-consuming and costly. Macroorganisms, such as sponges and other invertebrates, are relatively easy to collect and evaluate, and some drug discoveries using this method were made in the early 1990s. Microorganisms (micro flora or micro fauna such as bacteria, protozoa, fungi, and algae) are more difficult to collect and analyze. If they could be collected and evaluated, they could provide an advantage over macroorganisms in large-scale processing because they are more amenable to reproduction through fermentation. Fermentation has long been used in the production of foods, beverages, enzymes, vitamins, industrial chemicals, pharmaceuticals, recombinant therapeutics, agrochemicals, and other valuable substances. Commercial fermentation relies on a microorganism demonstrating the ability to produce the large quantities (100,000 liter batches or more) of a desirable product necessary for high-volume sales.

Aphios Proposes to Culture and Catalog Microorganisms

Aphios, a biopharmaceutical company, hoped to develop a marine microorganism library to be used in screening new drug leads. In 1995, Aphios requested funding from ATP to support a two-year research project. The comprehensive screening project would involve collecting marine microorganism samples, isolating unique microorganisms, fermenting them to increase their quantities, preparing new extraction methods, developing and managing a library of microorganisms, scaling-up active extract production for further testing, and bioprocess engineering. Aphios' proposal to ATP included the following five subcontractors:

- Harbor Branch Oceanographic Institution, a nonprofit focused on understanding marine resources with one of the largest marine microorganism collections in the world, would provide microorganism isolates and new methods to maintain and grow samples.
- Bristol-Myers Squibb, a leading multinational pharmaceutical company, would provide high-throughput screening for therapeutic drug discovery.
- CalBioMarine Technologies would supply saline culture media development.
- One Cell Systems would develop new methods for screening and recovering microorganisms based on the company's existing proprietary technologies.
- The Massachusetts Institute of Technology (MIT) would develop saline fermentation media and scale-up technologies.

The proposed project would validate the usability of marine microorganisms for industry applications by:

- Developing new processes to enable the directed exploration and industrial utilization of marine microorganism capabilities by growing microorganisms in multiple saline fermentation media and selecting fractions of the microorganisms themselves, as well as of their secondary metabolites (products they generate)
- Screening large numbers of marine microorganisms for detection of commercially valuable substances
- Demonstrating that saline fermentation can be scaled up to support production of valuable substances

While the project's potential for developing new life-saving medications and sanitizing products was valuable, technical risks were high, including the following:

- It was challenging to keep marine organisms alive and grow them for preparation of extracts.
- No efficient techniques were available for fractionating active compounds from marine organisms.
- Industry lacked the basic tools to manipulate and control marine microorganisms with their unique genetic and biochemical resources.
- The conventional fermentation equipment was not designed to withstand or operate safely in saline conditions.

Industry needed substantial diverse collections of marine science samples to facilitate product discovery. ATP awarded funding for two years, beginning in August 1995; the project was later extended for a third year at no additional cost to ATP. If successful, the marine culture fermentation technology and knowledge base developed during this project would become the platform for rapid commercial expansion of marine microorganisms for therapeutic applications, such as the treatment of cancer. With an average 15-year drug discovery and testing phase, analysts predicted that the potential economic impact of a marine microorganism industrialization program could reach $20 billion by 2015.

Subcontractors Assist in Establishing a Library

In order to establish an extensive library of marine microorganisms, Aphios proposed a multi-step plan that drew on the resources and expertise of the company's expert subcontractors, as described below:

- Subcontractor Harbor Branch had an existing marine microorganism library; Aphios intended to begin with this library to secure 1,500 to 2,000 diverse marine microorganism cultures. Aphios would use new methods to isolate an additional 500 unique marine microorganisms using One Cell Systems' proprietary gel microdrop technology. Aphios would also establish an extraction laboratory and would develop extraction and sample preparation procedures to handle as many as 240 samples per week or 12,000 per year.
- Aphios intended to establish an expanded library that would consist of cryopreserved isolates and frozen extracts. They would isolate and characterize these microorganisms, develop saline culture media, and replicate the organisms in small-scale fermentation processes.
- Aphios would use Bristol-Myers Squibb's high-throughput screening methods and robotic equipment, which would involve developing new tools to classify, manipulate, and control marine microorganisms. This would enable them to optimize the manufacturing process (first isolating the microorganism, using the microorganism to generate desired products and then breaking it down to isolate the bioactive compounds with specific therapeutic value.
- Aphios would rely on saline fermentation media developed by CalBioMarine and scale-up technologies developed by MIT to demonstrate that marine microorganism culture (saline fermentation) could be scaled up to extremely large volumes and could enable the commercial-scale manufacturing of target marine molecules derived from the marine microorganisms.
- Ultimately Aphios would use samples from the library to develop hits for commercialization (new and unique compounds with biological activity toward disease targets) on the path to discovering

novel therapeutic products. However, Aphios understood that commercializing a product could take as long as 15 years.

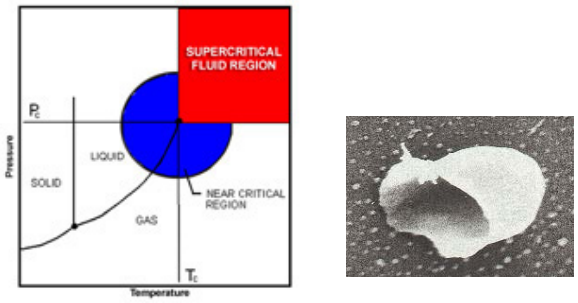
The First Year Shows Great Promise

Aphios' approach was to break down each microorganism and its secondary products into smaller segments. If the company tested the whole microorganism for bioactivity against a disease, it would not know which part of the microorganism was active, a process that may lead to false-positive or false-negative results. If the company purified each and every compound from the microorganism, the cost would be prohibitive. By partially purifying fractions (portions) of microorganisms, Aphios reached a cost-effective compromise.

Scientists were running out of new genetic and therapeutic discoveries from terrestrial sources and, therefore, needed to look elsewhere.

Aphios cultured the microorganisms by fermenting them in at least four different seawater-like media. Microorganisms digest nutrients and generate secondary metabolites (products). Some may be retained within the cell, and others may be excreted into fermentation broth. Aphios tested both the secondary products as well as the microorganisms themselves for bioactivity.

Aphios developed a proprietary method to partially purify the microorganisms. This method was called supercritical fluid fractionation, or SuperFluids CXF. The process utilizes a solvent-free system similar to that used in producing flavor extracts or decaffeinating coffee. The company enhanced the process in this ATP-funded project to purify extractions for screening and was awarded two patents for these advances. SuperFluids CXF involves breaking apart microorganisms that have initially shown bioactivity against a disease using supercritical fluids. These fluids (as shown by the pressure-temperature diagram below) are gases at ambient conditions. When brought to conditions above their critical points by raising temperature and pressure, these substances become fluids that simultaneously exhibit liquid-like properties (density and solvation) and gas-like properties (diffusivity and viscosity). SuperFluids CXF produces partially purified extracts and enhances the probability of clean "hits" by focusing on smaller portions of the microorganism. Supercritical fluid extraction disrupts the cells (as shown by the disrupted yeast cell below) and its bioactive constituents are extracted in different polarity-guided fractions. Processing a microorganism with SuperFluids CXF produces seven or more fractions.



Aphios' patented SuperFluids CXF technology penetrates the microorganism cells and swells them, while carefully controlling pressure and temperature (diagram, left). Many cells rupture at their weakest points as a result of decompression (part of the process), similar to a scuba diver experiencing the "bends" from rising too rapidly from deep water (shown by the sample disrupted yeast cell to the right). Some cells become more permeable, releasing their intracellular contents without rupturing. Aphios studied these portions of cells, or fractions, for bioactivity against disease, such as cancer and HIV.

Initial project results showed significant progress. By August 1996, Aphios had established a library of about 1,000 diverse marine microorganisms (from deep-water sediments, shallow mangrove swamps, salt ponds, sponges, algae, and corals) and had derived approximately 7,000 extracts. They had tested approximately 5,000 of the extracts for various bioactivities (anticancer, antiviral, antimicrobial). Out of the 1,000 cataloged microorganisms, 28 had yielded bioactive extracts, for an overall hit rate of 2.8 percent. The company expected this rate to increase as they completed more screening work on extracts from the organisms. Bristol-Myers Squibb indicated interest in pursuing six organisms further. Of these, two had already been refermented, and four were in the process of being scaled up (fermented again to grow larger quantities). Two organism samples had been refermented at a scale of 1 liter (20x the original fermentation). Preparations were underway to ferment one organism at a scale of 50 liters. Aphios had identified the organism using DNA sequencing and fatty acid analysis.

Technical Difficulties Slow Progress

During the first year, Aphios experienced some technical problems. Each marine sample contained many microorganisms, and the proposed cell isolation methodology from One Cell was not robust enough. Therefore, Aphios used traditional isolation techniques instead (researchers put the sample onto an agar plate and selected one culture to grow on a new plate).

Moreover, there was a concern about the consistent quality of microorganism samples from Harbor Branch, because approximately 20 percent of their samples were not active. Therefore, Aphios decided to create a brand new library from scratch during the second half of the two-year research period and received a one-year extension from ATP. Aphios then engaged Woods Hole and CalBioMarine Technologies to collect new samples using ocean-going ships and submarines that visited hydrothermal vents, shallow oceans, and tropical and hypersaline (super-salty) waters. During that time, Aphios registered 305 anti-HIV hits that required further testing. Their successes also included microorganisms that targeted tooth decay, influenza, and cancer.

In January 1998, Aphios decided to do an orderly reduction in personnel and commitments in preparation to voluntarily declare Chapter 11 bankruptcy. This strategy was elected in order to force a demand note and resolution of a lawsuit that had plagued the Company since 1995 and had been a significant impediment to attracting venture capital financing. When the ATP-funded project concluded in September 1998, internal and external funding was running short, and Aphios' future looked challenging. Aphios voluntarily declared Chapter 11 bankruptcy in January 1999 and exited from Chapter 11 on September 12, 2001.

Aphios Re-emerges to Test Marine Compounds

Relying on the key ATP-funded technology, Aphios survived by applying for Phase I Small Business Innovation and Research (SBIR) grants from the National Institutes of Health (NIH), selling products such as the anticancer drug paclitaxel to companies such as Quiral, Juiz de Fora, Brazil, and conducting contract research for pharmaceutical companies such as Novartis, Nyon, Switzerland. The company narrowed its focus and began discovering pharmaceutical compounds from its own library by performing its own strategic research, rather than seeking partner assistance. Aphios believed that its marine microorganism library contained many novel therapeutics, and it continued screening samples with promising results. Targets included Influenza A and B, tooth decay/plaque, multiple-drug resistant (MDR) bacteria, HIV/AIDS, cancer, smallpox, and Severe Acute Respiratory Syndrome (SARS). The ATP-funded technology allowed Aphios to pursue specialized research in numerous areas. Developments as of 2004 include the following:

- **Influenza**. Aphios continued Influenza A and B testing, which they had begun during the ATP-funded project. Researchers tested 2,000 fractions against Influenza A and B, identified 37 active hits, and followed up on purifying 5 fractions of the active compounds against Influenza A. Promising preclinical candidates included a fraction called CXF-Y2, which, at very low concentrations, has shown 100-percent inhibition of the influenza virus, and APP-310, which has high activity and low toxicity.

- **Tooth Decay and Plaque**. Research on tooth decay and plaque had also begun during the ATP-funded project in cooperation with the Gillette Company. Aphios screened the marine microorganism library (5,000 fractions) for sanitizing oral compounds. In 2000, Aphios received an SBIR grant from NIH to develop an anti-plaque compound from marine microorganisms. Aphios collaborated with MIT and Scripps, supported by the National Institute of Dental and Craniofacial Research. Aphios screened its marine molecules library for anti-plaque activity against microbes relevant to the oral cavity and identified bioactive components derived from a marine microorganism (APP-214). The company has scaled up manufacturing of the active ingredient Asterias. This compound is being further developed by medicinal chemistry to maximize effectiveness against microorganisms that affect the mouth. The goal is to develop a toothpaste or mouthwash with improved antimicrobial, anti-cavity, and antiplaque properties.

- **MDR bacteria**. Aphios screened 4,800 molecule fractions from 400 marine microorganisms and identified six isolates that fight MDR bacteria. These include methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *Enterococcus faecium* (VRE). One molecule fraction may also be active against *Bacillus anthracis* (anthrax).

- **HIV/AIDS.** Aphios also screened more than 10,000 microorganism fractions for anti-HIV activity. They identified 505 "hit" fractions, with a hit rate of 2.9 percent. Aphios is focusing on 170 of these results, including APP-069, a marine microorganism fraction with high activity and low toxicity. Aphios is conducting further testing and expects to enter clinical trials in the future.

- **Cancer**. Aphios screen several marine fractions against multiple cancer cell lines. Besides HIV-1, CXF-B13 is also a potential treatment for cancer. It is closely related genetically to a potent anticancer agent already in clinical trials in the United States.

- **Smallpox**. After the attacks of September 11, 2001, NIH researchers realized that the search for smallpox treatment was necessary, because the nation was not prepared to vaccinate the entire population should a bioterrorist attack occur. In 2003, NIH awarded Aphios an SBIR grant for a Phase I study to develop novel anti-smallpox therapeutics based on screening marine microorganism fractions. Aphios screened its microorganisms that appeared active against influenza and HIV. From these, researchers have identified seven active fractions from three unique microorganisms against Vaccinia variola, a less virulent form of smallpox. One example is APP-329, a high-activity, low-toxicity SuperFluids fraction.

- **SARS**. In 2003, Aphios began a three-year collaborative research agreement with NIH and the U.S. Army Medical Research Institute of Infectious Diseases to screen its library of unique marine microorganism fractions and natural therapeutics against SARS and other coronaviruses. Aphios has submitted a few samples for screening, but no hits have been discovered at this time.

New Pathogenic Viruses Increase the Need

The need for novel therapeutics, such as those from marine microorganisms, is greater than ever. In 2003, the U.S. Food and Drug Administration approved only 21 new chemical entities, which marks a steady decline since the peak of 56 in 1995. New pathogenic viruses create new major health threats, such as SARS and the "Bird Flu" of 2004. These threats are exacerbated by high-speed transportation, the ability of pathogens to mutate, and the potential for bioterrorism.

Aphios had established a library of about 1,000 diverse marine microorganisms and had derived approximately 7,000 extracts.

In an effort to meet the need for novel therapeutics, Aphios managed a library of over 1,400 unique marine microorganisms by September 2004. From these, 20,000 extracts had been prepared using Aphios' SuperFluids CXF fractionation methods. Researchers continue developing marine fractions that target HIV/AIDS, smallpox, Influenza A and B, MDR bacteria and oral plaque. Aphios anticipates commercializing one or more of these marine compounds within five years, attempting to capture part of the $315 billion annual pharmaceutical market.

Conclusion

Aphios cataloged 1,000 diverse marine microorganisms and derived approximately 7,000 extracts (partial microorganisms) in the first year of the ATP project; this initial library resides at Harbor Branch Oceanographic Institution. In the second and third year of the project, with the help of Woods Hole Oceanographic Institution and CalBioMarine Technologies, Aphios isolated an entirely new library of over 2,000 unique marine microorganisms from diverse sources. While the company experienced delays in developing its marine microorganism library, they have since refocused on research and continue to pursue the goals originally identified in its proposal to ATP: to develop potential novel therapeutics for the major diseases of modern society (such as HIV/AIDS, cancer, and influenza).

Aphios was awarded two patents for improvements it made to its existing extraction process during the project. By 2004, Aphios' library consisted of over 1,400 unique marine microorganisms and 20,000 derivative fractions (partially purified portions of a microorganism). The company continues to conduct research and development of marine extracts with the help of funding from other Federal agencies and is further developing the extracts through medicinal chemistry to maximize their effectiveness. Aphios expects to commercialize one or more marine compounds by 2009.

PROJECT HIGHLIGHTS
Aphios Corporation

Project Title: Marine Microorganisms and Saline Fermentation: A New Industrial Resource

Project: To develop a knowledge base and technology platform to tap into the valuable chemical diversity in the unexplored pharmaceutical, industrial, and environmental applications that may be possible through the enormous numbers and genetic and chemical diversity of marine microorganisms.

Duration: 8/1/1995 - 7/31/1998
ATP Number: 95-01-0263

Funding (in thousands):

ATP Final Cost	$2,000	77%
Participant Final Cost	600	23%
Total	$2,600	

Accomplishments: With ATP funding, Aphios researchers fractionated and tested marine microorganisms against a variety of diseases. In spite of several challenging technical obstacles, the company achieved most of its technical goals by 2004, isolating over 2,000 unique marine microorganisms and testing a significant fraction of these organisms against a variety of disease targets. Their accomplishments include the following:

- **Library**. Established a library of 2,000 unique marine microorganisms, of which 1,400 have been maintained. Derived 20,000 extracts (partially purified portions of a microorganism). Used proprietary SuperFluids CXF fractionation methods.
- **Influenza**. Tested 2,000 extracts of microorganisms against Influenza A and B, identified 37 active hits against Influenza, and were following up on purifying 5 fractions of the active compounds including APP-310, a SuperFluids fraction with high activity and low toxicity.
- **Tooth decay, plaque**. Screened 5,000 extracts from the marine microorganism library for oral compounds to prevent plaque and tooth decay. Identified bioactive components derived from a marine microorganism, APP-214, and scaled up manufacturing of the active ingredient Asterias. Aphios is developing this compound using medicinal chemistry to maximize antimicrobial, anti-cavity, and anti-plaque properties in toothpaste or mouthwash.
- **Multiple-Drug Resistant (MDR) bacteria**. Identified six isolates that are very effective against MDR bacteria: methicillin-resistant Staphylococcus aureus (MRSA) and vancomycin-resistant Enterococcus faecium (VRE) E. By screening 4,800 marine molecule fractions from 400 unique marine microorganisms, Aphios obtained a “hit” rate of 1.5 percent, including one molecule fraction that may also be active against Bacillus anthracis (anthrax).
- **HIV/AIDS**. Screened over 10,000 marine microorganism fractions for anti-HIV activity. Identified 305 “hit” fractions for a hit-rate of 2.9%. Aphios is currently prioritizing and confirming 170 of these results. High priority extract “hits,” include APP-069, a highly active and low toxicity marine microorganism SuperFluids fraction.
- **Cancer**. Screened several marine fractions against different cancer cell lines. Screening revealed symbiotic relationships between marine microorganisms isolated from a marine macroorganism with known anticancer bioactivity and identified one promising microorganism, CXF-B13, which is closely related to a potent anticancer agent already in clinical trials in the United States.
- **Smallpox**. Screened marine microorganism fractions that were active against influenza and HIV for smallpox. From these, researchers have identified seven active fractions from three unique microorganisms that are active against smallpox including APP-329, a highly active and low toxicity marine microorganism SuperFluids fraction against smallpox.
- **SARS**. Screened marine microorganisms fractions against SARS and other coronaviruses.

Aphios was awarded two patents and has one patent pending from this ATP-funded project:

- “Methods for fractionation of biologically-derived materials”
  (No. 5,854,064: filed July 31, 1996; granted December 29, 1998)
- “Method of fractionation of biologically-derived materials using critical fluids”
  (No. 6,569,640: filed October 13, 1998; granted May 27, 2003)

PROJECT HIGHLIGHTS
Aphios Corporation

Commercialization Status: An anti-plaque solution for toothpaste or mouthwash, which is being optimized through chemistry, is the product that is closest to commercialization. Novel therapeutics for MDR bacteria, influenza, HIV/AIDS, cancer, and smallpox are in preclinical drug discovery and development.

Outlook: The outlook for Aphios' marine microorganisms is good but uncertain. The technology is still high risk, and markets are still developing. If Aphios is able to commercialize one significant pharmaceutical product (such as AIDS treatment), it could save many lives and tap into an annual $315 billion market.

Composite Performance Score: * *

Number of Employees: 10 employees at project start, 12 as of September 2004.

Company:
Aphios Corporation
3-E Gill Street, New Boston Park
Woburn, MA 01801

Contact: Dr. Trevor Castor
Phone: (781) 932-6933

Subcontractors:

- Bristol-Myers Squibb
  Wallingford, CT and Princeton, NJ
- CalBioMarine Technologies (out of business, May 2004)
  Carlsbad, CA
- Harbor Branch Oceanographic Institution
  Fort Pierce, FL
- Massachusetts Institute of Technology
  Cambridge, MA
- One Cell Systems
  Cambridge, MA
- Scripps Institution of Oceanography
  La Jolla, California
- Woods Hole Oceanographic Institution
  Woods Hole, MA
- The Gillette Company
  Boston, MA
- Instituto Biomar
  Leon, Spain
- General Electric (formerly Betz-Dearborn)
  The Woodlands, Texas
- Global Pharma
  Lexington, MA

Publications: Aphios' marine microorganism research received public attention through of the following business and technical publications:

- "Aphios Developing Viral Inactivation, HIV Vaccines, Screening Marine Microorganisms." *Antiviral Agents Bulletin*, Vol. 11, no. 11, November 1998.
- "Patent Update: Aphios." *R&D Directions*, Vol. 5, no. 7, July 1999.
- Castor, T. P. "Rapid Discovery of Natural Therapeutics Using SuperFluids CXF Technology." *American Chemical Society*, 221: U613-U613, 299-IEC Part 1, April 2001.
- Hendrickson, Dyke. "Aphios Wins Grant to Put Influenza on the Run with Natural Agents." *Mass High Tech*, Vol. 21, no. 3, January 20, 2003.
- Hollmer, Mark. "Natural High: Aphios Profits by Taming Wild Compounds." *Boston Business Journal*, Vol. 23, no. 33, September 19, 2003.
- Castor, Trevor P. "Nature's Way." *Mass High Tech*, April 15, 2004.

Presentations: Aphios also disseminated knowledge through academic presentations:

- Castor, T.P., H.M. Chikarmane, G.T. Hong, P.B. Fernandes, S.A. Forenza, D.J. Hook, R. Lam, J. O'Sullivan, D. Mendola, B.J. Javor, S.A. Pomponi, K.E. Janda, J-F.P. Hamel, A. Ferrante, M.G. Haygood, and J.E. Thompson. "Diversity, Discovery and Development of Unique Biotherapeutics from Marine Microorganisms." American Society of Pharmacognosy 37, University of California, Santa Cruz, California, July, 1996.
- Castor, T.P. "Supercritical Fluid Extraction of Natural Products in Drug Discovery." Applications of Supercritical Fluids in Pharmaceutical Research, Saddle Brook, New Jersey, May 14, 1997.

- Castor, T.P. “Natural Products from Marine Organisms.” Marine Biotechnology Symposium, Commercial Applications of Marine Biotechnology 2, The Mount Desert island Biological Laboratory, Maine, July 16, 1997.

- Castor, T.P. “SuperFluids Fractionation of Diverse Marine Microorganisms for Drug Discovery.” Gordon Research Conference on Marine Natural Products, Ventura, California, February 22-27, 1998.

- Castor, T.P. “The Role of Molecular Diversity in New Drug Development.” Pharmaceutical Development for the Next Millennium, part of the Society of Nuclear Medicine 45, Toronto, Canada, June 1998.

- Castor, T.P. “SuperFluids Fractionation of Marine Organisms and Medicinal Plants for Drug Discovery.” Cambridge Healthtech Institute's High Throughput Screening for Drug Discovery 5, Dallas, Texas, June 1998.

- Castor, T.P. “Marine Microorganisms and Saline Fermentation: A New Industrial Resource.” IBC's Natural Products Drug Discovery & Development 4, Annapolis, Maryland, June 15-16, 1998.

- Castor, T.P. “Rapid Discovery of Natural Therapeutics Using SuperFluids CXF Technology.” ACS Meeting 221, San Diego, California, April 2001.

- Lallos, L.B., D. Patel, R. Student, L.C. Rosenberry, T.A. Tyler, and T.P. Castor. “Development of Novel Anti-Smallpox Therapeutics.” NERCE-BEID, Durham, New Hampshire, September 2004.

- Castor, T.P. “High Throughput Sample Preparation and Screening of Marine Microorganisms for Drug Discovery.” BIOPROSP - Symposium in Marine Bioprospecting, Tromso, Norway, October 2004.

- Lallos, L.B., D. Patel, R. Student, L.C. Rosenberry, T.A. Tyler, and T.P. Castor. “Development of Novel Anti-HIV Therapeutics,” Antiviral Research, HIV Dart, Montego Bay, Jamaica, December 2004.

Research and data for Status Report 95-01-0263 were collected during August – September 2004.

Cengent Therapeutics Inc. (formerly Moldyn Inc.)

Enhanced Molecular Dynamics Simulation for Drug Research

In the field of drug discovery, molecular dynamics (MD) simulation is used to understand protein structure, small molecule and protein interactions, and protein and protein interactions. While several commercial software packages started offering enhanced molecular dynamics simulation capabilities in the 1990s, the time required for a realistic simulation was too long, partly due to the slow speed of the computers and partly due to the way the biological system is treated in such simulation, which is atom-based. A complete ("all-atom") model simulation could be ideal for understanding atom-to-atom interaction in drug discovery, but could be a major bottleneck if feedback is needed quickly for biologists and chemists in an software/researcher-integrated drug discovery environment.

Moldyn Inc. was founded in 1993 by Photon Research Associates to research a way to make MD simulation via software less expensive and time-consuming by simplifying the MD simulation process. Moldyn proposed to replace the all-atom molecular model with a model that used groups of atoms that behave similarly to specific individual atoms in a disease process. Drug research using simulation processes is extremely laborious, time-intensive, and expensive; moreover, it carries high technical risk for any company attempting the process. Therefore, because Moldyn's own capital resources were limited due to the project's high risk, in 1994 the company applied for and received a three-year Advanced Technology Program (ATP) award. The proposed project to research MD simulation software to aid in finding new drug substances began in 1995. Moldyn's goal for MD simulation was to model approximate groups of atoms in a molecule instead of modeling exact maps of all the atoms of a molecule. If a breakthrough using MD simulation could be made, new drug research candidates could be discovered faster and more economically. If successful, MD simulation would replace the trial-and-error techniques prevalent in the drug research industry with a more precise and targeted identification method.

After two years, Moldyn had succeeded in developing the software and benchmarked it with other existing molecular dynamics tools. Its performance was shown to be superior. Moldyn then chose Molecular Simulations, Inc., to further develop it for incorporation with a suitable Graphical User Interface (GUI) and commercialization. Since Molecular Simulations already had competing products, the company spent very little in implementing a GUI but started offering the software as an independent module. Molecular Simulations then became a subcontractor and licensee, but stopped upgrading and selling the product. Subsequently, Cengent Therapeutics Inc. bought Moldyn and abandoned the software, as the business plan of Cengent did not include software product offerings.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* *

Research and data for Status Report 94-01-0137 were collected during May – July 2004.

Molecular Dynamics (MD) Simulation Software Could Speed New Drug Development

According to the Pharmaceutical Research and Manufacturers of America, it takes between 10 and 15 years and more than $800 million to bring a new drug to the U.S. market. This significant investment of time and money is due to the large number of target substances available for researchers to select from, as well as the

complexity of the research that is needed on these substances.

Molecular dynamics (MD) simulation is used to study the interactions between drug molecules and the active sites on a receptor protein. In drug research, a receptor protein is an organic molecule that needs to be altered to interrupt a disease process. The receptor molecule receives the drug target molecule at the "active site," located somewhere on the molecule. The target drug blocks the active site from further interaction with other proteins and stops the disease process, arresting the process at the molecular level. These target drugs must be extensively tested to ensure that they will bind with efficacy and selectivity at the active site on the receptor protein.

In the early 1990s, several commercial software packages offering MD simulation were available to help researchers study likely candidates for drug research, but these packages used all-atom molecular models. This meant that an entire molecule, such as a receptor protein, was modeled atom-by-atom by the software program until a complete mathematical model of the molecule was built (on the computer). The model was constructed according to three-dimensional coordinates for each atom in the molecule, which were entered by the user into the software program. Then candidate drug molecules were inserted near the active sites on the molecule to start the dynamics simulation. If the drug bound to the active site of the molecule, the drug was a candidate for more research.

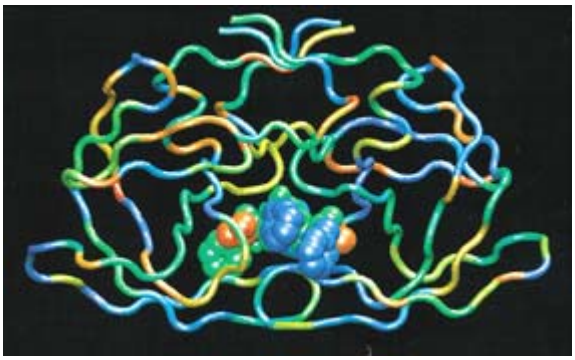


Figure 1. Example of a software-generated model of a drug/protein molecular interaction. This model represents 100,000 or more atoms.

There are thousands of proteins in the human body, and a protein molecule can consist of thousands of atoms. Therefore, many variables in the software must be controlled to ensure that the correct active site, which consists of atoms in a region of a molecule, gets targeted. Using software to model a large molecule consisting of 100,000 or more atoms could take months (see Figure 1).

Moldyn Proposes to Improve MD Simulation with Substructuring

To reduce the time required to model a molecule and to find its active sites, Moldyn, a company founded in 1993 by Photon Research Associates, wanted to modify the existing molecular modeling software. Moldyn initially received two Federal grants that they used to demonstrate the feasibility of MD simulation technology based on the generation of molecular clusters called "bodies". Then, in 1994, the company applied for and received a three-year ATP award to research ways to enhance the molecule-modeling software that was available at the time. Starting the research in 1995, the company attempted a breakthrough approach by modeling approximate groups of atoms in a molecule as flexible and rigid bodies instead of modeling all the molecule's atoms individually. Because Moldyn's MD simulation model assumes that approximate groups of atoms behave similarly to individual atoms, an atom group, called a substructure, could substitute for several exact maps of individual atoms in appropriate parts of the protein. If Moldyn's software enhancement was successful, molecules and drug interactions with molecules could be modeled more quickly, and new drug development candidates could be isolated faster, thus saving time and money.

Substructuring Poses Significant Technical Challenges

The implementation of Moldyn's proposed enhancements to the MD simulation software was formidable. The company wanted to modify a method that mapped the three-dimensional coordinates of every atom in a molecule. Molecules could consist of 100,000 atoms or more, so the software user must possess extensive knowledge regarding which nonactive substructures could have their degrees-of-freedom reduced without sacrificing accuracy in modeling the interaction of drug molecules to receptor protein active sites. Degrees-of-freedom are the atomic or molecular dynamic motions that move up, down, and across in a three-dimensional environment. The motions are related to the three-dimensional coordinates of each atom in a molecule. The Moldyn researchers sought to represent more efficiently the degrees-of-freedom of

molecules. Software similar to Moldyn's proposed technology was already being used in the aerospace industry to predict spacecraft movements in zero gravity environments.

Substructures in Moldyn's MD method can bend, twist, and fold, so that the resulting dynamic model of the molecule can have bends, twists, and folds just as would be realized in a traditional all-atom simulation. These dynamic degrees-of-freedom were an important variable, because reducing the degrees-of-freedom in regions of the protein that were unimportant to the dynamic motions of the active site was a critical time-saver in Moldyn's approach.

The company attempted a breakthrough approach by modeling approximate groups of atoms in a molecule as flexible and rigid bodies instead of modeling all the molecule's atoms individually.

Molecular Simulation Inc. (MSI), one of Moldyn's subcontractors, performed testing of the simulations. As the testing proceeded, the researchers had difficulty identifying which substructures needed their degrees-of-freedom reduced. Sometimes the location of the active sites was unknown. Substructures were generated according to the variables entered, so great care had to be exercised in substructuring. Researchers had already discovered earlier in the project that if they over-constrained parts of the molecule design by using poor substructuring, then extreme instabilities in the substructures resulted. Consequently, as a general rule, project researchers assumed that if the substructures remained stable, then the variables entered into the program were reasonable, and it was more likely that drug candidate discoveries would be made.

Moldyn Uses a Team of Code Developers and Testers

The software developers at Moldyn worked with the following team of eight subcontractors, who contributed to various project tasks:

- Karplus and Associates validated the software code.
- Ornet Consulting Group served as a primary software developer. The group had previous experience as the primary developer for an all-atom software package already on the market.
- H.R. Research developed and tested the mode structures (variables) in the Moldyn software. There were several modes that controlled how molecule and drug interactions were simulated.
- MSI provided alpha and beta testing of the software at each stage of its development. MSI also was the product licensee. They provided the graphic user interface package called "Insight" that computed and constructed three-dimensional molecular models on a computer screen.
- Haney Associates performed research at MSI. Haney also provided customer support for the companies that purchased the software.
- Vertex Pharmaceuticals, Bristol Myers Squibb Pharmaceutical Research Institute, and Northeastern University served as alpha and beta testers.

Software Is Licensed and Field Tested

In the fall of 1997, near the end of the project, Moldyn signed a licensing agreement with MSI to sell and distribute its commercial software. Moldyn's software was bundled with MSI's computational chemistry software, Insight. The field test (beta) version of the bundled software was released in November 1997.

Moldyn Faces Obstacles in the MD Simulation Market

After the project ended, Moldyn was ready to start commercialization. The product was launched in 1998, with a time-to-market that was two to three years ahead of the company's competitors. Moldyn's 1999 sales were $100,000, but many of the products were returned due to customer dissatisfaction. The software was difficult to use and many of the simulations were unstable and, therefore, unusable. In retrospect, project principal investigators realized that if testers from MSI and other project subcontractors did not have an extensive knowledge of chemistry and physics, they were not able to enter sufficiently complete variable sets to generate stable substructures for the simulations. When unstable substructures occurred, the results of the simulation were unusable and invalid. Subsequently, new sales dropped as MD simulation

techniques fell out of favor with many customers. This was probably also due to the introduction of new technologies, such as combinatorial chemistry techniques that use “lab on a chip” and other arrays to quickly test target drug substances. These new methods were more efficient than MD simulation because they did not require extensive user knowledge. The “lab on a chip” and combinatorial chemistry approaches did work.

At the time of the final report for the project, Moldyn was pursuing more funding to finance refinements in molecular modeling with Small Business Innovation Research proposals, as well as internal funding. However, based on the difficulties encountered, neither Moldyn nor its parent, Photon Research Associates, had the additional funds required to enhance the MD simulation software that they had developed during the ATP-funded project. However, they did share their project research through 3 publications and 13 presentations.

Moldyn’s molecular dunamics simulation software was launched in 1998, with a time-to-market that was two to three years ahead of the company’s competitors.

After the conclusion of the project, through a licensing agreement between Moldyn and Harvard University, Moldyn’s software was incorporated with Harvard's Chemistry at Harvard Macromolecular Mechanics (CHARMM) molecular modeling research software tool. CHARMM was originally developed in the early 1980s by project subcontractor Martin Karplus, and provided the molecular mechanics and dynamics framework for the Moldyn MD simulation software. As of 2004, CHARMM continues to be distributed by Harvard at no charge to academic and not-for-profit organizations for research purposes only. No intellectual property rights in either software package, the Moldyn software or CHARMM, were transferred in the Moldyn-Harvard licensing agreement.

In early 2000, Moldyn was acquired by Structural Bioinformatics Inc. (SBI), which subsequently changed its name to Cengent Therapeutics Incorporated in May 2003. According to the terms of the acquisition by SBI, the Moldyn software licensing agreement with Harvard was maintained.

Advent of New Technologies Causes Obsolescence of Molecular Dynamics Simulation

Moldyn’s software increased by a factor of 20 to 30 times the speed of simulating the all-atom model and achieved modeling accuracies of 10 to 20 percent or better in comparison to all-atom simulations. However, due to the program’s complexity and lack of predictability, consumers generally felt the program was undependable and not viable for commercial use. As of 2004, there was no customer interest in MD simulation software. The market for drug research had turned to high throughput screening techniques, including “lab on a chip” techniques that test for variables that flag new drug substances. Although it is very expensive for companies to set up high-throughput screening systems, as of 2004, all competitive drug research companies were using this technique.

Conclusion

Moldyn Inc. sought to improve molecular dynamics (MD) simulation software by modifying a fundamental task: substituting atom groups, called substructures, for many individual atoms in a molecule. The goal of the software was to virtually test the numerous potential drug substances to determine more quickly and at lower cost which ones were suitable as new commercial drug prospects. The most significant challenge in developing the software was to offer a faster but reasonably accurate molecular model that was superior to the all-atom model.

Although Moldyn made progress in several of the extremely complex tasks required to develop the software, the product failed. The primary reason for its failure was that the software users needed advanced knowledge in physics and chemistry, which the majority of the users lacked. Without this level of advanced expertise, the simulated models produced were unstable and therefore unusable. Consequently, the research community rejected the product. Despite the failure of the software, researchers learned much about the challenges of qualifying new drug agents for research. Moreover, they disseminated their knowledge through 3 publications and 13 presentations. As of 2004, MD simulation software is no longer used as a drug research technique; various high-throughput screening methods are now preferred.

PROJECT HIGHLIGHTS
Cengent Therapeutics Inc. (formerly Moldyn Inc.)

Project Title: Enhanced Molecular Dynamics Simulation for Drug Research (Enhanced Molecular Dynamics Simulation Technology for Biotechnology Applications)

Project: To develop a software that adapts a technology developed in the aerospace industry to simulations of biological molecule and drug interactions, for the purpose of qualifying drug research candidates in a more timely and efficient manner than by using trial-and-error techniques.

Duration: 2/15/1995 - 2/14/1998
ATP Number: 94-01-0137

Funding (in thousands):

ATP Final Cost	$1,988	60%
Participant Final Cost	1,341	40%
Total	$3,329	

Accomplishments: ATP funding enabled Moldyn to develop and test molecular dynamics (MD) simulation software. This type of software was of interest to the research community during a brief period in the mid-1990s. At that time, it provided some utility in helping experts better understand and characterize target proteins for drug research.

Commercialization Status: The MD simulation software was briefly commercialized through a license to Molecular Simulations Incorporated, but failed to gain sufficient sales and was discontinued. However, Moldyn's software was incorporated with Harvard's Chemistry at Harvard Macromolecular Mechanics (CHARMM) molecular modeling tool through a licensing agreement between Moldyn and Harvard Univesity.

Outlook: The outlook for this technology is poor. The software has been superceded by the use of high-throughput screening methods, which process vast numbers of possible new drug substances and select candidates that meet target variables.

Composite Performance Score: * *

Number of Employees: 7 at project start, 12 as of July 2004.

Company:
Cengent Therapeutics Inc. (formerly Moldyn Inc.)
10929 Technology Place
San Diego, CA 92127

Contact: Dr. Kal Ramnarayan
Phone: (858) 675-2400

Subcontractors:

- Bristol Myers Squibb Pharmaceutical Research Institute
  Princeton, NJ
- Haney Associates
  San Diego, CA
- H.R. Research
  Lawrenceville, NJ
- Karplus and Associates
  Cambridge, MA
- Molecular Simulation Inc.
  San Diego, CA
- Northeastern University
  Boston, MA
- Ornet Consulting Group
  Cambridge, MA
- Vertex Pharmaceuticals
  Cambridge, MA

Publications:

- Chin, D., K. Haney, H. Delak, C. Chun, and C. Padilla. "Nanosecond Simulations of the Unbinding Pathways of CBZ-Val-Phe-Phe-ValCBZ from the Active Site of HIV-1 Protease Using Multi-body Dynamics," *American Chemical Society Symposium Series on Rational Drug Design*, A. Parrill and R. Reddy, eds., 1998.
- Chin, D., K. Haney, H. Delak, H.M. Chun, and C.E. Padilla. "The Evaluation of Multi-Body Dynamics for Studying Ligand-Protein Interactions: Using MBO(N)D to Probe the Unbinding Pathways of Cbz-Val-Phe-Phe-Val-Cbz from the Active Site of HIV-1 Protease," *American Chemical Society Symposium Series 719, Rational Drug Design, Novel Methodology and Practical Applications*, A.L. Parrill, and M.R. Reddy, eds., p. 87-106, 2000.

- Chun, H., C. Padilla, H. Alper, D. Chin, M. Watanabe, V. Karlov, K. Soosaar, K. Blair, O. Becker, L. Caves, R. Nagle, M. Karplus, and D. Haney. "MBO(N)D: A Multi-body Method of Long-time Molecular Dynamics Simulations," *Journal of Computational Chemistry,* 21:3, p. 159-184, 2000.

Presentations:

- Thacher, T. "Advances in the Accuracy and Utility of Protein Ligand Models, IBC Rational Drug Design - Computational Approaches to Analyze Protein Ligand Interaction/Binding Affinity," Dec. 11-12, 1995.
- Chun, H. "Substructured Modeling Approach for Large Macromolecules - MBO(N)D," MSI Potential Energy Functions Consortium Meeting, Rhone-Poulenc Rorer, Collegeville, PA, June 24-25, 1996.
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Research and data for Status Report 94-01-0137 were collected during May – July 2004.

Dow AgroSciences LLC (formerly Mycogen Corporation)

Using Yeast Fermentation to Produce Cost-Effective and Biodegradable Lubricants

Squalene is a high-performing, biodegradable lubricant. In 1995, it was being used only in small-volume specialty applications such as watch lubricants, pharmaceuticals, and perfumes, because of its high price ($32 per pound). At the time, squalene was extracted from shark-liver oil. Cheaper lubricants could be produced from imported petroleum, but they are toxic and nonbiodegradable. Although shark-liver oil was a more environmentally friendly source of squalene for U.S. industrial use, the high cost of the product made large-scale manufacturing and use infeasible. Mycogen Corporation proposed to produce squalene from yeast fermentation, a technique that had never been tried before. In 1995, the company applied to the Advanced Technology Program (ATP) for cost-shared funding to investigate modifying the fermentation of several yeasts and to apply this more cost-efficient process to the production of squalene. The company was awarded ATP funds for a three-year project.

By the project's end in 1998, Mycogen had increased production of squalene from one yeast strain, but was unable to reach the company's target of producing squalene at a cost of $2 to $3 per pound. That same year, Dow AgroSciences acquired Mycogen and refocused the Mycogen division on plant genetics for agriculture, particularly on corn, sunflower, and canola crops. The yeast fermentation project ended, and the outlook for cost-effectively producing squalene from yeast fermentation is poor. The industry continues its research into developing and improving biodegradable lubricants such as vegetable oils, because analysts predict that the demand for these lubricants will reach $1 billion by 2010 (10 percent of the U.S. lubricants market), compared with $500 million in 2002.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)

No Stars

Research and data for Status Report 95-01-0148 were collected during November 2003.

Existing Lubricants Were Toxic or Expensive

In 1995, high-volume lubricant products were being produced from imported petroleum, which contributed to the U.S. trade deficit. In addition, petroleum-based products were not biodegradable and hence contributed to environmental pollution through both accidental spills and routine use in environmentally sensitive areas. Petroleum-based lubricants also required solvent-based removers for clean up, which created additional handling and disposal concerns. The annual market for lubricants, which included lubricant additives, hydraulic fluids, and lubricant base oils, was estimated at 50 million metric tons. The market for lubricant additives for diesel fuel was estimated at an additional 5 million metric tons.

Vegetable and animal fats and oils, which are biodegradable, were available for use as lubricants. They can be disposed of in landfill sites, and they are safer for workers and the environment, because they wash off in soap and water. However, these lubricants have drawbacks. They solidify at a relatively low temperature and have flash points that are too low for use in hot conditions (they break down or burn under normal hot engine conditions.

Isoprenes, such as squalene, are commercially important types of lipids (fats and waxes). They remain liquid at low temperatures and have high flash points (they do not freeze in cold weather and do not burn in hot engine conditions). However, at the time, squalene was being extracted from shark-liver oil and cost $32

per pound. Because of its high cost, it was used only in high-value specialty applications, such as watch lubricants, pharmaceuticals, and perfumes.

Scientists suggested that genetic engineering could alter yeasts to produce large quantities of the desired high-value isoprenes. Recent medical studies of cholesterol biosynthesis had resulted in advances in the understanding of isoprene biosynthesis. Researchers at Mycogen Corporation wanted to apply similar molecular biology techniques to manipulate yeast biosynthesis in order to maximize the yield of isoprenes (fats and waxes), particularly squalene.

ATP Funding Enables Innovation

Mycogen Corporation was a biogenetic technology company that developed and marketed improved crop varieties to increase agricultural production. Mycogen believed it could use plant biotechnology techniques to produce high-value squalene from yeast. The company proposed to use genetic engineering to alter the metabolic characteristics of an oleaginous (oily) yeast to increase the yeast's ability to produce isoprenes through biosynthesis. They proposed to ferment raw material from low-cost industrial waste streams, such as waste whey from dairy production or sugar cane residue. After developing the technical processes, Mycogen intended to design large-scale fermentation processes to produce large quantities of squalene. This kind of work had never before been attempted, but the potential for economic and environmental benefits was great. However, the research was innovative and risky, and Mycogen was unable to obtain funding. Therefore, in 1995 Mycogen applied to ATP for research funding and was awarded funds for a three-year project.

This project would combine genetic research and industrial microbiology to develop a production system for squalene and other lipids using oleaginous yeast fermentation. If successful, squalene could replace established petroleum-based products in hydraulic fluids and many other high-volume industrial applications.

Project researchers believed that increasing the availability and lowering the cost of isoprenes, especially squalene, would open larger market applications in areas such as lubricant base oils, hydraulic fluids, and lubricant additives. The new products would have high performance and biodegradability characteristics. The success of Mycogen's process depended on both the company's ability to construct a yeast organism that efficiently converts carbon sources into isoprenes and its ability to develop a cost-effective production system.

Mycogen Develops Challenging Milestone

The ATP-funded project aimed to modify the activity of four enzymes in oleaginous yeast to funnel carbon from fatty acid synthesis into the synthesis of squalene. Mycogen researchers selected a candidate yeast strain, *Yarrowia lipolytica,* and altered its genes to modify four enzymes they wished to manipulate: *ACCase, hydroxymethylglutaryl CoA reductase* (HMGR), *Squalene synthetase,* and *Squalene epoxidase*. The amount of *Squalene synthetase*, the immediate precursor to squalene, and HMGR needed to be increased, and *ACCase* and *Squalene epoxidase* needed to be decreased.

Squalene was used only in high-value specialty applications, such as watch lubricants, pharmaceuticals, and perfumes.

Initial success validated the research hypothesis. During the first year of the project, researchers were able to double squalene production levels from 0.45 to 0.9 percent of cell dry weight (cdw) compared with normal yeast production. Mycogen produced squalene levels that were 1.3 to 2.7 times higher than levels found in previous yeast strains. These results validated the concept that increasing the activity of HMGR was critical to producing isoprenes.

The researchers' plan, however, had lower than anticipated results. The researchers were unable to manipulate two of the four enzymes properly. *ACCase*, which controls the flow of carbon, was successfully inhibited, and HMGR, the rate-controlling enzyme, was successfully increased. However, *Squalene epoxidase* and *Squalene synthetase* were not manipulated successfully. Prototype production levels of squalene were too low, so Mycogen made no progress in developing viable commercial processes.

Research Makes Progress but Cannot Overcome Barriers

Mycogen abandoned yeast fermentation for squalene production. It became clear that the existing knowledge and technology for manipulating the genome of the *Yarrowia lipolytica* strain of yeast was too limited. Mycogen increased its total lipid production to 16 percent of cdw and its squalene production to 2 percent of cdw at a cost of $22 per pound. This cost was much too high (the target was $3 per pound). Researchers made a good effort in attempting to use yeast fermentation to produce squalene, but they could not overcome the technical barriers.

This project demonstrated that genetic manipulation could redirect carbon fatty acid synthesis to increase squalene synthesis compared with its normal presence in oleaginous yeast.

Although they could not successfully achieve the desired level of squalene production through fermentation techniques, Mycogen researchers made the following significant strides:

- Successfully transformed a yeast variety for the first time
- Developed a broader understanding of the metabolic pathways in yeast that lead to isoprene formation
- Used inexpensive carbon sources (e.g., cheese whey and sugar cane)
- Increased knowledge about using metabolic inhibitors for redirecting carbon synthesis in yeast

This project demonstrated that genetic manipulation could redirect carbon fatty acid synthesis to increase squalene synthesis compared with its normal presence in oleaginous yeast.

Dow AgroSciences Acquires Mycogen and Stops Research

The biotechnology industry was growing in the late 1990s, which changed Mycogen's research environment. Firms viewed consolidation as a way to increase visibility and capital. "It takes tremendous resources to commercialize these [biotech] products,"said Mike Sund, Vice President of Mycogen. Dow AgroSciences purchased Mycogen in 1998 (near the end of this ATP-funded project) and took over all of Mycogen's research projects. As a division of Dow AgroSciences, Mycogen refocused its research on the genetic manipulation of corn, sunflower, and canola crops. Yeast fermentation research ended after the ATP-funded project concluded in 1998.

Conclusion

Mycogen Corporation believed that oleaginous (oily) yeast varieties could be genetically altered to biosynthesize large quantities of isoprenes, a commercially important class of lipid. In particular, the company sought to produce squalene, a type of isoprene that is a biodegradable and nontoxic lubricant. In 1995, Mycogen was awarded cost-shared funding by ATP to genetically modify this yeast in order to cost-effectively produce squalene.

Oleaginous yeast fermentation trials demonstrated increases in production up to double the normal production of squalene. However, Mycogen was unable to manipulate two of the four desired enzymes, and the company was unable to reach its target of producing squalene at cost of $3 per pound. Dow AgroSciences acquired Mycogen in 1998 and ended the yeast fermentation research.

PROJECT HIGHLIGHTS
Dow AgroSciences, LLC (formerly Mycogen Corporation)

Project Title: Using Yeast Fermentation to Produce Cost-Effective and Biodegradable Lubricants (Oleaginous Yeast Fermentation as a Production Method for Squalene and Other Isoprenoids)

Project: To use modern genetic technologies to modify oleaginous yeast to stimulate the overproduction of isoprenes, a commercially important class of lipid (fats and waxes) and, in particular, squalene, an important biodegradable lubricant.

Duration: 9/30/95-9/29/98
ATP Number: 95-01-0148

Funding (in thousands):

ATP Final Cost	$797	88%
Participant Final Cost	112	12%
Total	$909	

Accomplishments: Although the project failed to accomplish its proposed goals, Mycogen Corporation made some strides in genetic research. The company demonstrated for the first time that yeast is transformable; they demonstrated that squalene could be hyper-produced in oleaginous yeast; and they gained a broader understanding of the metabolic pathways for isoprene formation in yeast. Mycogen was able to increase total lipid production to 16 percent of cell dry weight (cdw) and to increase squalene production from 0.45 to 2 percent of cdw (15 percent was needed) at a cost of $22 per pound. This was less than the market price of $32 per pound, but far short of the company's goal of $3 per pound.

Commercialization Status: Mycogen was acquired by Dow AgroSciences in 1998. The oleaginous yeast fermentation project was ended due to technical barriers with enzyme manipulation. Therefore, there will be no commercialization of products resulting from project research.

Outlook: The outlook for this project is poor. Mycogen's attempt to use biotechnology to manipulate oleaginous yeast to produce squalene was not cost competitive; therefore, research into oleaginous yeast fermentation has ended.

Composite Performance Score: No Stars

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Research and data for Status Report 95-01-0148 were collected during November 2003.

DuPont Qualicon (formerly DuPont FQMS Group)

Testing Food-Borne Bacteria Using DNA Analysis

As of 1994, food contamination was causing 10 million illnesses and nearly 4,000 deaths each year in the United States. Testing food samples for bacteria was costly and time consuming, requiring repetitive manual processes. Researchers at DuPont FQMS Group believed they could test food samples for contamination by using automated DNA analyses, a process that the health industry was using to analyze patient blood and tissue samples for disease. The company applied to the Advanced Technology Program (ATP) for cost-shared funding under a 1994 focused program, "Tools for DNA Diagnostics." The company's proposed project had high technical risk, because it required integrating many technologies, including miniaturization, micro-separation, contamination prevention, and computer analyses.

ATP awarded funding for a three-year research project beginning in 1995. DuPont FQMS (later called DuPont Qualicon) developed a functioning prototype to analyze DNA. The prototype used microfluidics (tiny amounts of fluid moving on a micro-scale), which reduced analysis time from 3 hours to 30 minutes, a sixfold improvement. The company received a patent for this work. Unfortunately, using a smaller sample increased the sample preparation time, which negated the time saved by the DNA analysis system. As a result, DuPont suspended further development of the technology until sample preparation methods were improved. No commercialization has resulted to date, and DuPont is now pursuing alternative bacteria-testing methods.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)

No Stars

Research and data for Status Report 94-05-0033 were collected during June – July 2004.

Food Sample Testing Was Slow and Costly

In the 1990s, DuPont FQMS Group was researching improved methods for testing bacterial contamination in foods. Although the company looked to the medical products industry for improved methods, testing for food-borne bacteria was more complicated than testing blood and tissue samples for disease. Bacteria such as *Salmonella* and *E. coli* involved a labor-intensive manual testing process that required skilled operators and provided low throughput (production). Moreover, samples required six days or more to generate results.

In a well-publicized 1993 case of food poisoning, four children died and hundreds of people became ill from eating hamburgers at a fast food chain that were infected with *E. coli*. As of 1994, as many as 4,000 Americans died and 10 million became ill each year from eating bacteria-infected food.[1] The annual number of premature deaths caused by food-borne pathogens exceeded deaths from accidental fire (3,300) and drowning (3,300).[2] More than 200 known diseases are transmitted through viruses, bacteria, parasites, toxins, and metals in the food supply.[3] Symptoms of food-borne illness range from mild gastroenteritis to life-threatening neurological problems, and liver and kidney failures. Infections in the U.S. population from the five major bacterial pathogens (*E. coli* O157:H7, *E. coli* non-O157 STEC, *Campylobacter*, *Listeria monocytogenes*, and *Salmonella*) cost $6.9 billion annually, according to a USDA Economic Research Service report in 2000. That estimate includes medical costs, productivity

[1] *Wall Street Journal*, CCXXVIII, no. 10, July 15, 1996.

[2] P. D. Frenzen, "Deaths Due to Unknown Foodborne Agents." *Emerging Infectious Disease* September 2004.

[3] Bryan, F.L. "Diseases Transmitted by Foods." Atlanta: Centers for Disease Control, 1982; P. S. Mead, L. Slutsker, V. Dietz, et al. "Food-Related Illness and Death in the United States," *Emerging Infectious Disease* 1999, 5:607-25.

losses from missed work, and the estimated value of premature death.[4] As a result of the increasing problem of food contamination, consumers began to demand increased testing for bacterial contamination in food, and public concern has remained steady since the mid-1990s.

The U.S. food market was substantial. For example, in the mid-1990s, the annual U.S. market for meat alone was $65.2 billion and was $28.5 billion for chickens and eggs. Although consumer demand for food safety testing was rising sharply, the technology used to test food samples for bacterial contamination had not improved significantly. The primary testing process, called amplification-based DNA technology, consisted of the following costly and time-consuming steps:

- First, the sample, such as 65 grams of hamburger meat combined with 650 milliliters of enrichment broth, was prepared. Preparing a small representative sample took 24 to 48 hours.
- After a representative sample was ready, technicians amplified the representative DNA sequences (making many copies), using a process called polymerase chain reaction (PCR). PCR can generate 100 billion copies of the DNA in a few hours.
- Finally, the DNA sequences were analyzed and reported. Existing testing methods to analyze the DNA relied on fluorescent detection, which pairs RNA molecules with genes that express them (called hybridization capture). Technicians measured the fluorescent signal at specific probe locations to determine the presence or absence of signals for specific bacteria.

Another method to analyze the DNA sequences was capillary electrophoretic separation. The DNA samples were placed in a capillary (a long, narrow tube) containing a gel medium called agar or agarose. An electric current flowed through the tube, forcing the DNA molecules to pass through the medium at different rates, depending on their size. Smaller fragments moved faster; larger ones moved slower. As a result, the DNA molecules separated into bands, so they could be identified by fragment size.

In 1994, commercial use of amplification-based DNA technology was limited due to cost. The only product approved for routine commercial use at the time was a kit for *Chlamydia* testing. Existing automated systems for detecting contaminants were based solely on some means of hybridization capture, or base pairing. They required skilled operators and had high material costs. As a result, DNA analysis systems' full potential was not being realized.

DuPont Proposes to Automate Sample Analysis Using Capillary Electrophoresis

Researchers in the medical field were relying increasingly on automated DNA analysis for disease testing of blood and tissue samples. Researchers at DuPont FQMS Group wanted to use similar automated techniques to test food samples for bacteria. DuPont had already invested $50 million to develop DNA-based diagnostic systems for microbial testing. The DuPont FQMS Group proposed to develop cost-effective, automated DNA diagnostics that would require minimal technician skills or specialized laboratory facilities. DuPont researchers believed that detection by capillary electrophoresis (analysis by fragment size) would be more versatile than hybridization (RNA base pairing). Capillary electrophoretic analysis would provide opportunities for automation, because it could detect the presence or absence of a specific DNA fragment by its length.

As of 1994, as many as 4,000 Americans died and 10 million became ill each year from eating bacteria-infected food.

DuPont planned to make a simple, automated system, affordable enough to be used in a wide range of laboratories, to support food processing and packaging plants. Their intent was to adapt the existing technology for routine use, so that technicians could operate it without receiving extensive training. Miniaturizing and then integrating the various processes proved to be technically risky; miniaturizing and integrating heightened the need for contamination prevention measures at the micro scale. Because the new system was to handle a wide range of food product samples, the design would be more complex than systems being

developed for the medical industry, which tested only tissue, blood, and urine samples.

In 1994, DuPont submitted a proposal to develop a functioning prototype food sampling system to ATP's "Tools for DNA Diagnostics" focused competition. DuPont's assessment showed that the proposed system would have a significant economic impact on the food processing industry as well as on agriculture, forensics, and medical sectors. If successful, consumers would benefit from improved safety of the food supply. The target markets were all food processors, commercial testing labs, and government regulators. In addition, the agriculture sector could test their crops before incorporating them into the food stream, thus eliminating contamination at the source. Forensic scientists could test more easily for deaths caused by food poisoning. Fewer cases of food contamination could save lives and reduce hospitalizations. Because of the risk associated with miniaturizing, internal funding was not available at DuPont. Therefore, ATP awarded DuPont a three-year, cost-shared award that began in 1995.

DuPont Conducts Parallel Research

Because food bacteria testing was a high priority for DuPont FQMS Group, the company explored multiple avenues simultaneously to maximize its success. In 1996, while the ATP-funded project was ongoing, DuPont released a commercial product for food bacteria testing called BAX, which had been developed through a separate research program. The BAX system screened for pathogenic microorganisms by amplifying specific DNA sequences and detecting the sequences by gel electrophoresis, a multiple-step process that is similar to capillary electrophoresis. DuPont hoped to enhance the benefits expected to result from the BAX product line with the results of this ATP-funded project.

PCR/Capillary Electrophoresis Achieved Some Success

DuPont FQMS researchers developed a functioning automated prototype, which combined PCR (to amplify DNA fragments) with capillary electrophoresis (to separate the fragments by size). In order to prevent contamination, researchers also developed a sealed plastic disposable unit to contain the samples. They were granted one patent for this integrated capillary electrophoresis system. The system delivered reagents to the sample in tablet form (reagent shelf life reached three years, which reduced cost of maintaining and storing reagents). The prototype used microfluidics (tiny amounts of fluid moving on a micro scale), which reduced analysis time from 3 hours to 30 minutes. However, DNA pattern results from sample testing were somewhat inconsistent and needed further development. In 1997 when the ATP-funded project ended, DuPont researchers believed that an integrated, automated system could offer many commercial advantages to the company's existing manual BAX system if the consistency of the pattern results could be improved.

DuPont planned to make a simple, automated system, affordable enough to be used in a wide range of laboratories, to support food processing and packaging plants.

Unfortunately, the miniaturized equipment required smaller samples. Sample preparation time for a typical 50-microliter sample (a microliter is one millionth of a liter) took 24 to 48 hours. However, this preparation time increased when preparing the smaller 1-microliter representative samples. Producing a smaller representative sample led to longer enrichment times or more complicated preparation procedures (for DNA extraction and clean up). As a result, the time saved by the automated system was lost.

DuPont Continues to Develop Bacteria Testing Methods

DuPont FQMS (later renamed DuPont Qualicon) could not justify continuing development of the automated system. Upon project conclusion in December 1997, researchers pledged to continue evaluating and refining the technology in the future. They did apply some of the basic PCR automation knowledge to their existing BAX system, but this project ended in 1998.

Sample preparation time has improved with technology. As of 2004, DuPont Qualicon is able to prepare food samples for DNA testing in about 8 hours (a threefold to sixfold reduction). As miniaturized electronics become more common, it is expected that DuPont will

reevaluate the advances it achieved in microfluidics during this project.

Conclusion

DuPont FQMS Group (later called DuPont Qualicon) believed that food-borne bacteria could be tested more quickly and less expensively by an automated system that combined polymerase chain reaction (PCR) and capillary electrophoresis. The research group sought to miniaturize and automate the testing process to meet strong consumer demand for improvements in the safety of the U.S. food supply. In 1995, ATP awarded cost-shared funding to DuPont to develop an automated system that would reduce testing time to improve food quality for consumers, as well as provide benefits to the agriculture and forensics industries.

DuPont built a functioning prototype that reduced testing time from 3 hours to approximately 30 minutes. The company was awarded one patent based on this technology, but additional steps were required in sample preparation that negated the time saved in analysis. DuPont Qualicon ended the research into this automated system in 1998, but the company did apply some of the automation knowledge gained in this project to its ongoing alternate food-borne pathogen-testing technologies.

PROJECT HIGHLIGHTS
DuPont Qualicon (formerly DuPont FQMS Group)

Project Title: Testing Food-Borne Bacteria Using DNA Analysis (Automated DNA Amplification and Fragment Size Analysis)

Project: To develop an automated, rapid DNA diagnostic system that can determine the presence or absence of specific microbial contamination as a means of quality control in the food industry.

Duration: 1/1/1995–12/31/1997
ATP Number: 94-05-0033

Funding (in thousands):**

ATP Final Cost	$1,966	75%
Participant Final Cost	$ 639	25%
Total	$2,605	

Accomplishments: With ATP funding, the DuPont FQMS Group (later called DuPont Qualicon) conducted a comprehensive study of food-borne bacteria testing. Because of the high risk of this project, the company would not have attempted this research without ATP support. The supporting technologies (such as sample preparation of minute volumes) required to commercialize the automated, miniaturized polymerase chain reaction (PCR) and capillary electrophoresis technology were not sufficiently developed during the project. However, the researchers made several advancements:

- Automated PCR technology
- Reduced analysis time from 3 hours to 30 minutes
- Miniaturized instrumentation and integrated PCR with capillary electrophoresis in one tool (the reduced sample size made sample preparation more complex, which negated the analysis time savings)
- Delivered reagents in the form of tablets, which had a three-year shelf life

DuPont researchers filed for and received one patent from this ATP-funded project:

- "Apparatus for integrated polymerase chain reaction and capillary electrophoresis" (No. 6,372,484: filed January 21, 2000; granted April 16, 2002)

Commercialization Status: DuPont Qualicon does not plan to commercialize automated capillary electrophoresis technology developed under this ATP award.

Outlook: The outlook for DNA testing of food-borne bacteria using an automated PCR/capillary electrophoresis system is poor. However, DuPont Qualicon has been successful in using fluorescence detection methods for its BAX system. Although the company does not plan to develop the proposed automated system, some of the miniaturization and automation knowledge gained during the ATP-funded research may be applied to future projects.

Composite Performance Score: No Stars

Focused Program: Tools for DNA Diagnostics, 1994

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** As of December 9, 1997, large single applicant firms are required to pay 60% of all ATP project costs. Prior to this date, single applicant firms, regardless of size, were required to pay indirect costs.

Research and data for Status Report 94-05-0033 were collected during June – July 2004.

Genosensor Consortium (c/o Houston Advanced Research Center)

Development of the Electronic DNA Chip

By the early 1990's, biological researchers were beginning to make great strides in DNA analysis and apply their findings to important areas, including human diagnostics, agriculture, and toxicology. However, these efforts proceeded slowly due to a lack of efficient, cost-effective sequencing tools. In response, a multidisciplinary group of organizations came together in 1992 and formed a consortium to develop an advanced form of DNA sequence analysis that would be simpler, faster, and less expensive than currently used methods. The new technology, in the form of a tabletop scientific instrument, would be capable of rapidly analyzing nucleic acid sequences contained in a DNA molecule analyzed on a microarray (a matrix containing many gene sequences that could be evaluated simultaneously) on a microfabricated chip. The consortium believed that this new technology, called genosensor technology, would revolutionize DNA sequence analysis and result in multibillion-dollar annual U.S. sales.

To obtain the resources needed to undertake the project, the Genosensor Consortium sought financial assistance. Private firms were unwilling to fund a project that could not prove a guaranteed return on investment, so the group submitted a proposal to the Advanced Technology Program (ATP) and was awarded cost-shared funding for a five-year project. By the end of the project in 1998, the Genosensor Consortium had produced two laboratory prototypes, using DNA microarrays, that demonstrated significantly higher throughput for DNA sequence analysis and lower costs than was available through current methods. Three consortium members have commercialized the new technology. The market is still emerging and they anticipate a profit in the future. The ATP-funded project resulted in numerous patents, publications, and presentations.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* * *

Research and data for Status Report 92-01-0044 were collected during September – December 2003.

New Tools for DNA Analysis Are Needed

By 1992, DNA analysis was viewed as the key to understanding complex diseases, drug effects and toxicology, mutations, and pathogens. However, traditional methods for sequencing DNA were slow, costly, and labor-intensive. Gel electrophoresis, for example, involved separating nucleic acids or proteins on the basis of size, electrical charge, and other physical properties. Although accurate, it was time-consuming and tedious. The DNA had to be prepared prior to sequencing and the fragments could take a long time to detect. Furthermore, only a few fragments could be analyzed at one time.

By the early 1990s, several new techniques had been developed that were faster and more efficient. One technique involved using fluorescent tags on DNA fragments so that hybridized sequences could be identified in real time. This method, which was also automated, resulted in increased throughput (for example, 400 base pairs of DNA could be sequenced in a day, with a throughput of approximately 10,000 nucleotides). However, the time required for the entire DNA sequencing process did not decrease because DNA samples still had to be prepared.

Genosensor Consortium to Develop Advanced DNA Sequence Analysis Technology

In 1992, a multidisciplinary group of organizations came together and formed a consortium to develop a faster, more economical technology for DNA sequencing. The new technology, called genosensor technology, would

be in the form of a small scientific instrument consisting of a microfabricated chip containing DNA probes (an array), as well as associated sensor and computer technology. It would be able to instantaneously analyze the sequence of nucleic acids in a DNA molecule.

In the early 1990's, the use of DNA arrays was a promising solution for decreasing the time, effort, and expense in DNA analysis. With a DNA array, which includes a large number and high density of probes and the use of fluorescent dye labeling to detect base-pairing or hybridization, many gene expressions can be observed simultaneously. This was significant because, although every cell contains the same genetic material, the pattern of genes that are expressed in that cell characterizes its state of health or disease.

The Genosensor Consortium included Baylor College of Medicine; Beckman Instruments, Inc.; Genosys Biotechnologies, Inc.; Houston Advanced Research Center (HARC); Laboratories for Genetic Services, Inc. (LGS); Lincoln Laboratory, Massachusetts Institute of Technology (LL/MIT); MicroFab Technologies, Inc. Triplex Pharmaceutical Corporation. Genometrix, Inc. later joined the group.

Genosensor Consortium Anticipates Broad-Based Benefits

The Genosensor Consortium believed that the new technology would provide important benefits in areas such as human diagnostics, agriculture, and toxicology.

In the early 1990's, the use of DNA arrays was a promising solution for decreasing the time, effort, and expense in DNA analysis.

In human diagnostics, the genosensor could be used to quickly detect mutations within genes associated with diseases, such as cystic fibrosis, Alzheimer's, and cancer. Treatment could then be provided to a patient early enough to prevent the onset of a serious medical condition. The information could also be used to rapidly screen individuals who carry recessive genetic diseases and to provide them with genetic counseling when appropriate.

In agriculture, information obtained from genome sequencing could be used to rapidly develop new techniques for genetic engineering in crops and livestock, resulting in strains more resistant to disease and harsh climates and crops with a greater yield or higher nutritional value.

In toxicology, genosensors could be used to quickly detect genetic mutations in an individual that are caused by chronic exposure to radiation or chemical agents in the environment. With this knowledge, affected individuals could obtain treatment earlier, and employers could take steps to reduce chemical or radioactive hazards in the workplace, thereby decreasing work-related healthcare costs and disability claims.

New Technology Poses High Risk

The combination of small physical scale and efficiency of microelectronics and data processing in the consortium's proposed genosensor would increase the throughput of DNA sequencing by a hundredfold. This would result in a significant reduction in the cost of sequencing. However, developing the genosensor technology would also involve a high level of risk. It was a new technology in which the following six physical sciences and biotechnologies, rarely paired in the past, would be brought together in new ways:

- Chemistry of oligonucleotide synthesis
- Chemistry and biophysics of DNA hybridization
- Microluminescence and microelectronic detection
- Microlithography of solid supports
- Micro-robotics and microfluidics
- Parallel computer processing

Due to the risks, as well as the resources required for this multidisciplinary project, the consortium sought financial support. Private firms were unwilling to fund a project that could not prove an immediate return on investment, so the Genosensor Consortium submitted a proposal to ATP. In 1992, it was awarded $9.2 million in cost-shared funding.

Genosensor Technology Is Successfully Developed

The Genosensor Consortium organized the ATP project into five tasks:

- *Genosensor Development.* This would involve creating two types of DNA chips: a permittivity DNA chip and an optical or fluorescence DNA chip. (Permittivity is the measure of the ability of a material to resist the formation of an electric field within it.) In a permittivity DNA chip, hybridization is detected through electrical permittivity changes that occur at the array surface upon binding with DNA targets. In an optical or fluorescence DNA chip, hybridization is detected through an optical or fluorescent change that occurs at the array surface upon binding with DNA targets.
- *Instrumentation Development*. This would involve constructing the necessary electronic interface to connect the permittivity and optical genosensors with the computer workstation required for DNA sequence recognition.
- *Computer Development*. This would involve creating the software and hardware needed to associate the two-dimensional hybridization pattern on the genosensor array to the correct analysis of the sequence of nitrogenous bases that make up the target DNA molecule.
- *System Integration*. The group would assemble the deliverables from the genosensor, instrumentation, and computer technology development programs into demonstrable laboratory prototypes for DNA sequence analysis.
- *Applications Development*. The group would use the prototype systems to diagnose Lesch Nyhan and cystic fibrosis diseases.

HARC would play a central role in the development of the technology. It would fabricate different types of arrays using technology and techniques that had been established at LL/MIT. It would then conduct hybridization analyses with Baylor to determine the best substrate material to use for both the permittivity and optical DNA chips, while Triplex developed the DNA probes. HARC would then conduct sensitivity studies for the instrumentation to quantitatively determine the required signal amplification and noise rejection levels; and, with LL/MIT, would take measurements from both hybridized and non-hybridized DNA attached on the DNA chip prototypes to determine the specification requirements for the instrumentation boards. With Beckman Instruments and Baylor, HARC would develop fluorescent dye chemistry that could be used as an optical marker for detecting a hybridization event on the array.

Researchers believed that microarray technology for DNA analysis could be used for preventative medicine.

Much of the computer development also would take place at HARC: algorithms would be developed on a computer workstation at the research center; PC boards to host special purpose chip processors to increase computational speed would be assembled; and, optimal array studies would be performed.

Finally, the system prototypes would be integrated at HARC with guidance from Beckman Instruments. The mutation detection applications would be developed at HARC and at LGS.

By the end of the ATP-funded project, the consortium had accomplished the following:

- Invented several new electrical and optical methods for detecting hybridization on DNA microarrays, based on the application of microelectronics principles.
- Achieved highly resolved (50-mm) fluorescent images of DNA microarrays without the use of lenses. This enhanced by tenfold the sensitivity and the ability to scan large arrays of DNA probes very quickly (nine microarrays per second). This technology was unique and resulted in an issued patent.
- Developed permittivity-based detection. This approach may lead to the development of integrated DNA microarray devices that can directly detect hybridization through electrical permittivity changes that occur at the array surface upon binding DNA targets. This technology was also unique and was patented.
- Developed designs for an integrated DNA microarray device to electrically enhance the rate of DNA hybridization. These designs were patented.

- Developed a conceptual framework for achieving the desired sample throughput criteria required for pharmaceutical and diagnostic applications, whereby the solid support underlying the array would be carefully modified. This would result in a significant increase in the rate of target binding to the microarray, under low salt conditions. The consortium also developed various chemistry modifications. The modifications resulted in at least a tenfold improvement in the rate of hybridization.

In 1998, the consortium demonstrated two prototype systems. The first system, developed by Beckman Coulter, Inc. (Beckman Instruments had acquired Coulter Corporation in 1997), had fully automated hybridization and detection. The second system, developed by Genometrix, had at least a tenfold speed improvement over conventional microarray imagers. Both prototype systems represented advancements in the miniaturization of pharmaceutical and diagnostic platforms.

The ATP-Funded Project Benefits Participants

The ATP-funded project was beneficial to all of the consortium members. Genometrix, a provider of genomic services and information, was incorporated in 1993. In 1994, it received $300,000 for start-up costs from a venture capitalist. The company attributed its ability to attract this financing to the ATP award. From 1994 to 1998, Genometrix grew from 3 part-time to 40 full-time employees. That year, the company also had more than 20 partners, including LL/MIT and Motorola. The company believed that the ATP award played an important role in its growth.

Genosys Biotechnologies, a supplier of custom synthetic DNA and gene arrays, also benefited from the ATP-funded project. The company found that its participation proved valuable in helping it evaluate the most effective methods to produce and analyze gene arrays for new product and custom service development.

For Beckman Coulter, a leading provider of instrument systems and complementary products that simplify and automate laboratory processes, the ATP-funded project influenced its decision to further develop nucleic acid diagnostics technology.

Genosensor Technology Is Commercialized

Genometrix, Genosys Biotechnologies (now Sigma Genosys), and Beckman Coulter commercialized the genosensor technology.

Genometrix

Genometrix planned to market a wide range of products and services, which it would offer to genomic, pharmaceutical, and diagnostic customers. First, the company planned to offer genotype database services. Its database would use a panel of genetic markers to predict the effect of a drug, in terms of toxicity and efficacy, based on an individual's genetic profile. Pharmaceutical companies could then subscribe to the database and use it to screen patients for clinical trials, which would significantly increase the success rate of the trials and reduce the development time of a new drug.

Genometrix's next goal was to become a leading supplier of cost-effective, high-throughput, microarray-based products and genotype information services for the research and development of genomics-based pharmaceuticals. Finally, the company wanted to become a leading supplier of microarray-based diagnostic instrumentation. The instrumentation would be used to rapidly diagnose patient illness and to select optimum molecular medicines for treatment.

By March 2000, Genometrix was selling DNA chip microarrays for the high-throughput analysis of genetic variation, gene expression profiling, and other data analysis and storage services. The company was also constructing a DNA repository that it planned to license to customers, such as Schering-Plough and the National Cancer Institute, for use in genotyping analysis. In June, Michael Hogan, the chief scientific officer of Genometrix and a professor at Baylor College of Medicine, collaborated with two colleagues to test DNA microarray technology for screening DNA samples from patients with lung cancer to determine if their genes are different from those of healthy people. The group planned to eventually use these data to determine the probability that an individual would develop lung cancer.

DNA microarray technology was becoming successful by mid-2000. However, the technology was also creating its own problems. According to an article in the

June 2000 issue of the journal OncoLog, the speed and efficiency of the technology generated more data points than could be evaluated by conventional data analysis methods. To solve this problem, information specialists at Genometrix and the M.D. Anderson Cancer Center at the University of Texas began to develop new statistical techniques to handle the large volume of data.

By October 2000, Genometrix was providing sample analysis and database services for genotyping and gene expression applications to Schering-Plough Research Institute and similar organizations for research, discovery, and clinical trial applications. In January 2001, Genometrix collaborated with Vistagen, Inc. to combine in vitro stem cell biology with microarrays to discover surrogate markers for toxicology. The markers would be used in the development of pharmaceuticals.

In March 2001, the company released the VistaLogic Information System, an integrated, PC-based bioinformatics system for genomic researchers. Genometrix was also making plans to use its DNA chip microarrays for protein analysis and had partnered with Motorola to develop DNA clinical systems. The company also joined with GE Medical Systems to research the use of molecular imaging techniques with genetic probes to develop technologies for detecting and quantifying risk levels for diseases such as breast and prostate cancers.

Three years after the ATP-funded project ended, Genometrix had not only met, but had exceeded most of its original goals for commercialization. However, in spite of its technical successes, the company was also experiencing financial difficulties. In 2001, a year after announcing that it would make an $84 million initial public offering, it withdrew the filing due to concerns about market conditions and the general decline in the biotechnology industry. In 2002, Genometrix sold its intellectual property rights to the array technology and its tangible assets to High Throughput Genomics Inc. a genomics firm in Tucson, Arizona. Shortly thereafter, the company filed for bankruptcy and went out of business.

Genosys Biotechnologies (now Sigma Genosys)

In 1998, Genosys Biotechnologies was acquired by Sigma Aldrich and its name changed to Sigma Genosys. After the ATP-funded project, Sigma Genosys collaborated with HARC to evaluate custom oligonucleotide-based arrays (an oligonucleotide is a linear sequence of up to 20 nucleotides, the basic building blocks of nucleic acids) for gene expression profiling (the analysis of gene expression patterns). In 1999, the company introduced Panorama Gene Arrays for gene expression profiling. In 2003, Sigma Genosys also sold human cancer oligoarrays. These arrays represent 2,886 genes and have demonstrated greater specificity, sensitivity, and accuracy than similar oligo tests.

In 2003, the company was one of the largest suppliers of custom oligos for a variety of uses, including DNA primers (segments of DNA that are complementary to a given DNA sequence), antisense therapeutics (a means of treating many diseases that are difficult to control, such as cancer and viral infections, by blocking the messenger RNA transcripts used to produce disease-causing proteins), and DNA probes.

Beckman Coulter

At the end of the ATP-funded project in 1998, Beckman Coulter was very optimistic about genosensor technology and its potential for growth. The company anticipated that it would apply genosensors and related nucleic acid technologies to clinical diagnostics. It also envisioned that, in the future, nucleic acid diagnostics would not only replace current diagnostic methods, but could lead to the development of far more advanced diagnostic methods.

That year, Beckman Coulter signed several collaborative agreements to share intellectual property with Affymetrix, a biomedical company that develops state-of-the-art technology for acquiring, analyzing, and managing complex genetic information for use in biomedical research. In 2002, Beckman Coulter was granted an option to license technology for analyzing single nucleotide polymorphisms (SNPs), markers of genetic diversity, from Orchid BioSciences, Inc. a company with expertise in SNPs and a leader in genetic diversity. This technology, SNP-IT, which was developed in another ATP-funded project, is used to search for specific SNPs on strands of DNA to discover potential interaction sites for drugs. In 2003, Beckman Coulter was also leveraging Orchid's work on an image analyzer. According to Jim Osborne, Vice President of Advanced Technology at Beckman Coulter, the

company gained expertise in the ATP project that led to its relationship with Orchid.

In July 2003, Beckman Coulter formed a limited liability corporation with Affymetrix to further array commercialization efforts, such as the Miniature Integrated Nucleic Acid Diagnostic (MIND) device developed by Affymetrix in another ATP-funded project. The MIND device can rapidly and accurately diagnose a wide variety of diseases by extracting DNA from a small sample of blood and analyzing it through DNA probe-array hybridization.

In 2003, Beckman Coulter has also started to commercialize arrays. The company has continued to work with consortium members LL/MIT and Baylor and is seeking additional alliances.

Conclusion

With ATP's assistance, the Genosensor Consortium accomplished its goal. It successfully developed an advanced form of DNA sequence analysis, an electronic chip, using novel microarrays. The new chip can analyze DNA faster, more efficiently, and at less cost than previous methods.

Since 1997, three of the consortium members have commercialized the technology. Genometrix was the first member to sell microarray-based products and services; by 2000, these included microarrays for high-throughput analysis of genetic variation and for gene expression profiling, data analysis, sample analysis, and database and storage services. That year, however, due to the economic downturn, which had a significant effect on the biotechnology industry, the company was acquired by High Throughput Genomics.

Sigma Genosys (formerly Genosys Biotechnologies) has also commercialized the new technology; by 2003, its products included Panorama Gene Arrays for gene expression profiling and human cancer oligoarrays. In 2003, Beckman Coulter also began to commercialize arrays.

The market for microarray-based products and services has continued to expand. As of 2004, there were many companies in the biomedical, pharmaceutical, and agriculture industries that had commercialized arrays.

During the ATP-funded project, members of the Genosensor Consortium received one patent, published numerous papers, and gave a number of presentations on their research.

PROJECT HIGHLIGHTS
Genosensor Consortium (c/o Houston Advanced Research Center)

Project Title: Development of the Electronic DNA Chip (Genosensor Technology Development)

Project: To develop a microfabricated chip that incorporates synthetic DNA probes, together with necessary sensor and computer technology, for an automated, low-cost DNA sequencer.

Duration: 8/18/1993-7/31/1998
ATP Number: 92-01-0044

Funding (in thousands):

ATP Final Cost	$9,234	50%
Participant Final Cost	$9,366	50%
Total	$18,600	

Accomplishments: ATP funding enabled the Genosensor Consortium to develop an electronic chip for DNA sequence analysis using miniaturization, microelectronics, and data processing. The new chip was simpler to use, more efficient, and less expensive than other current means of performing DNA sequence analysis.

By the end of the ATP-funded project, the consortium had accomplished the following:

- Invented several new electrical and optical methods for detecting hybridization on DNA microarrays, based on the application of microelectronics principles.
- Achieved highly resolved (50-mm) fluorescent images of DNA microarrays without the use of lenses. This enhanced by tenfold the sensitivity and the ability to scan large arrays of DNA probes very quickly (nine microarrays per second).
- Developed permittivity-based detection. This approach may lead to the development of integrated DNA microarray devices that can directly detect hybridization through electrical permittivity changes that occur at the array surface upon binding DNA targets.
- Developed a conceptual framework for achieving the desired sample throughput criteria required for pharmaceutical and diagnostic applications, whereby the solid support underlying the array would be carefully modified. This would result in a significant increase in the rate of target binding to the microarray, under low salt conditions. The consortium also developed various chemistry modifications. The modifications resulted in at least a tenfold improvement in the rate of hybridization.
- Developed designs for an integrated DNA microarray device to electrically enhance the rate of DNA hybridization.

The Genosensor Consortium members filed and were granted the following patent:

- "Multi-site detection apparatus " (No. 5,532,128: filed December 12, 1994; granted July 2, 1996)

Commercialization Status: Three of the consortium members have commercialized products resulting from their project research. For example, Genometrix began to sell microarray-based products and services in 1997, during the ATP project. The company earned revenue of $2 million for these products and services that year. By 2000, the company specialized in DNA chip microarrays for genetic variation and gene expression profiling and offered services for data analysis and storage. It also provided sample analysis and database services for genotyping and gene expression research; its customers included organizations such as the Schering Plough Research Institute. In 1999, Sigma Genosys began to sell Panorama Gene Arrays, which profile gene expression in human cytokines, B. subtilis, and E. coli. In 2003, the company sold human cancer oligoarrays. In 2003, Beckman Coulter started to commercialize arrays.

Outlook: Since 1997, the market for microarray-based products and services has continued to grow. In 2003, there were a number of companies that had commercialized arrays for differential expression. These companies have not made a substantial profit yet, but they anticipate an increasing demand for their products in the future.

Composite Performance Score: ***

Consortium Members:

- Houston Advanced Research Center (HARC)
- Baylor College of Medicine
- Genometrix, Inc.
- Lincoln Laboratory, Massachusetts Institute of Technology (LL/MIT)
- Sigma Genosys (formerly Genosys Biotechnologies, Inc.)
- Beckman Coulter (formerly Beckman Instruments, Inc.)
- Dynacare/Dynagene (formerly Laboratories for Genetic Services, Inc.)
- MicroFab Technologies, Inc.
- Antigenics (formerly Triplex Pharmaceutical Corporation)

PROJECT HIGHLIGHTS
Genosensor Consortium (c/o Houston Advanced Research Center)

Company:
Beckman Coulter, Inc.
Corporate Staff
200 S. Kraemer Blvd.
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Brea, CA 92822-8000
(Beckman Coulter is one of the companies commercializing the technology)

Contact: Jim Osborne
Phone: (714) 773-8427

The group also shared its project research in many publications and presentations.

Publications:

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Presentations:

- Eggers, M.E., W.J. Balch, L.G. Mendoza, R. Gangadharan, A.K. Mallik, M.G. McMahon, M.E. Hogan, D. Xaio, T.R. Powdrill, Bonnie Iverson, G.E. Fox, R.C. Willson, K.I. Maillard, J.L. Siefert, and N. Singh. "Advanced Approach to Simultaneous Monitoring of Multiple Bacteria in Space," 27th International Conference on Environmental Systems, Lake Tahoe, NV, July 14-17, 1997.
- Matson, R.S. "Recent Developments in Ras Oncogene Screening," Biochip Array Analysis 3rd Annual Conference, International Business Communications, San Diego, CA, March 5-6, 1997.
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Genosensor Consortium (c/o Houston Advanced Research Center)

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- Matson, R.S., P.J. Coassin, J.B. Rampal, and R.W. Staub. “Probe-Target Interactions on Disposable, Plastic Film Based Array Panels,” Genetic Screening & Diagnosis of Human Disease Conference, Cambridge Healthtech Institute, San Francisco, CA, March 8-10, 1995.

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Research and data for Status Report 92-01-0044 were collected during September – December 2003.

Incyte Corporation (formerly Combion, Inc.)

Using Chem-Jet Techniques to "Print" High-Density Microarrays

In 1994, the biotechnology industry predicted that DNA analysis could be used to diagnose disease and to customize treatments. Existing analysis methods could only analyze one or two genes, and the methods were slow, costly, and labor intensive, with potential for error or contamination. Researchers needed new automated DNA analysis techniques in order to increase the number of genes that could be studied in a single experiment, while at the same time increasing speed and lowering cost. Combion, Inc. was unable to find investors to fund this unproven technology and applied to the Advanced Technology Program (ATP) for support in 1994 as part of the focused program, "Tools for DNA Diagnostics." ATP awarded cost-shared funding for a three-year project that began in 1995.

Combion successfully produced microarrays and received five patents for this technology, which enables fast, flexible, low-cost analyses. Incyte Pharmaceuticals purchased Combion in 1996 in order to acquire this microarray production expertise. Incyte, which changed its name to Incyte Genomics in 2000, shifted its focus to bioinformatics, gathering genetic data based on microarray analyses. The company licensed its microarray technology to Agilent Technologies in 2001. In 2002, Incyte changed its name to Incyte Corporation. As of 2004, Agilent is the leader in chem-jet microarray manufacturing and counts Incyte among its customers. Analysts project continued strong growth in the microarray market. For example, in 2004, industry analysts Frost & Sullivan estimated that global DNA microarray markets generated $596 million in 2003 and will reach $937 million by 2010, growing at approximately 6.7 percent annually. Promising research based on these microarray analyses is focused on discovering new drugs to treat cancer, diabetes, AIDS, and other life-threatening diseases.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* * *

Research and data for Status Report 94-05-0019 were collected during March – April 2004.

Focused Effort on Protein Research Technique Proposed

In 1994, biological research was leading to an unprecedented understanding of genetic material, as well as ways to manipulate the material. Researchers relied on techniques for synthesizing and sequencing DNA in order to study diagnostics, therapeutics, environmental monitoring, agriculture, forensics, and toxicology. Researchers wanted to identify gene sequences ("normal" genes or their mutations) and to measure their expression level (called "abundance"). Although most cells contain a person's identical genes, not all of the genes are used in each cell (different genes are "turned on" or "expressed" in a kidney cell compared with a heart or lung cell, and normal expression levels are altered by disease). The pattern of genes expressed in a cell characterizes its current state (such as its relative health or its response to medication). Scientists wanted to identify which genes each type of cell expresses in order to understand how cells achieve specialization, such as in the kidneys or liver. DNA analysis had the potential to identify complex genetic diseases, measure drug effects and toxicology, detect mutations, analyze pathogens, and measure changes over time. The genes that are expressed differently in two different tissues may indicate the cause of disease.

Efforts to gain an understanding of gene expression were hampered by the lack of practical, cost-effective tools to synthesize and detect oligonucleotides (which are subunits of DNA). Traditional methods in molecular biology worked on a "one-gene-in-one-experiment"

basis, which limited production speed and made it impossible to see the "whole picture" of gene function in context. These methods were costly in terms of labor, facility, and reagent requirements. They also required a long waiting period to obtain results. Furthermore, concerns had been raised about the reliability of the results, because multiple manual operations can lead to errors and contamination. Researchers were attempting to develop ways to monitor the whole genome on a single microscope slide, so that they could gain a better understanding of the simultaneous interactions among thousands of genes.

In 1994, biological research was leading to an unprecedented understanding of genetic material, as well as ways to manipulate the material.

Researchers wanted to place DNA molecules representing many genes on a single microscope slide, or chip, called a DNA microarray. Microarray technology would allow them to simultaneously detect many gene expressions. They were striving to develop methods such as spotting and photolithography to lay down DNA sequences to create an orderly arrangement of probes for base-pairing, or hybridization. (Base-pairing refers to RNA molecules that pair with the genes that express them. This indicates which genes are turned on in the cells.) Researchers purified messenger RNA (working copies of genes within cells that indicate which genes are expressed) from specific types of cells, such as liver or pancreas cells, and then "labeled" them by attaching a fluorescent dye that makes them detectable. Researchers added these RNA molecules to the DNA molecules on the microarray. After incubation, the unpaired, or nonhybridized, samples were washed away. Researchers then measured the fluorescent signal at specific probe locations.

Microarray Could Allow Many Genes to Be Analyzed Simultaneously

Research at the California Institute of Technology (CalTech) had demonstrated the feasibility of creating a microarray using a novel liquid jet or chem-jet printing approach to "print" small amounts of reagents in a particular order onto a microscope slide in order to synthesize oligonucleotides, or DNA segments, in situ (right on the slide). Similar to desktop inkjet printers, this non-contact printing process would not be affected by how the fluid wets a substrate and would not be contaminated by the substrate. Using computer-assisted design methods, researchers generated the sequence of reagents applied ("printed") to the microscope slide to create the unique DNA molecule.

Combion, Inc. was a start-up biotechnology company of four scientists who were developing commercial products based on microarrays. In 1994, Combion requested funding from ATP to pursue using the CalTech chem-jet methods to design and build automated microarrays right on a microscope slide. As a subcontractor, CalTech would contribute scientists to the research effort. ATP awarded cost-shared funding for three years beginning in 1995 as part of the focused program, "Tools for DNA Diagnostics."

Combion and CalTech researchers believed that microarray technology for DNA analysis could be used for preventative medicine. Applications of the technology would include designing drugs to treat disease causes rather than symptoms, targeting at-risk populations, providing genetic testing, and customizing drug treatments for diseases such as HIV/AIDS. Together, the disciplines of biology, chemistry, and information technology could increase the speed of the drug-discovery pipeline. Diagnostic products would reduce the cost and improve the quality of healthcare by allowing faster, less expensive, and more definitive diagnoses to be carried out initially in clinical labs and, ultimately, in the physician's office.

Combion and CalTech researchers' technical plan for this research involved developing the following four areas:

- **DNA sequencing by hybridization (SBH)**. Combion would develop sensor methods that are sensitive only to completely matched DNA-RNA hybrids, or pairs. The purpose was to understand genes and what they do.
- **DNA hybridization-based diagnostics**. Combion would acquire a genetic database and would compare DNA sequences with it to determine DNA hybridization mismatches (mismatches would indicate genetic mutations or pathogen sequences that are targets for disease). Combion would also develop appropriate analysis software.

- **Microorganism detection by hybridization pattern recognition**. Combion would simultaneously detect multiple pathogens based on complex pattern recognition algorithms for larger arrays, relying on deconvolution. (Deconvolution is a mathematical technique that removes extraneous variations in order to extrapolate the shape of objects hidden in images.)
- **Addressable oligonucleotide libraries**. Combion would detach and extract single oligonucleotide species, as well as subsets or mixtures of the members of the array, to build libraries for various medical research applications.

ATP-Funded Project Changes Focus to Meet Market Demand

By 1996, while the ATP-funded project was ongoing, the DNA diagnostic market changed dramatically. Increasing competition was eroding profit margins. As a result, Combion focused on understanding genes and what genes do (SBH), requiring high throughput (large numbers of analyses), rather than on the array of services the company originally planned. Combion and CalTech made a business decision to stop pursuing the other areas of research, although they were still technically viable (hybridization-based diagnostics, microorganism detection by hybridization pattern recognition, and oligonucleotide libraries). The company hired six additional researchers and gained the attention of a public company, Incyte Pharmaceuticals. Incyte purchased Combion in 1996 to obtain microarray expertise and continued this ATP-funded development, with an eye toward customers in the pharmaceutical industry and hospitals.

Incyte researchers made the strategic assessment to specialize in hybridization assays for diagnostic and gene-expression monitoring. The team pursued SBH and successfully developed gene expression microarrays (GEMs) using a glass-slide matrix and fluorescence detection methods, together with the company's existing GEMTools software to interpret results. The software identifies over- or under-expressed genes in diseased cells in order to measure the presence or change in the progress of disease. Each Incyte chem-jet microarray contained up to 10,000 genes by 1998, at project close. The company received five patents for this work.

In 1998, Incyte continued performing microarray analyses, but temporarily stopped SBH development due to the high cost. Competitors had developed other microarray technologies, including photolithography and pin-based deposition, so Incyte purchased the pin-based deposition technology, because it was less expensive. The chem-jet method developed with ATP funding opened the door to high-throughput custom microarrays, but Incyte put this specific technology on hold for a time. The company relied on the foundational knowledge and software tools developed during the project to build its microarray analysis business.

Combion and CalTech researchers believed that microarray technology for DNA analysis could be used for preventative medicine.

Incyte and CalTech made a little progress with the other three research areas. Researchers reduced analysis time for DNA hybridization-based diagnostics from 36 hours at project start to 24 hours. Incyte's GEMTools software measured images to detect differences in a cell related to disease state, disease progression, or genetic variation. At the end of the project, they were able to analyze 10,000 probes at a time. The technology allowed them to improve the efficiency of custom-made arrays. The technology changed their manufacturing process. However, company researchers still had major technical barriers to overcome, and competitors were pursuing commercialization of their products. Therefore, the company abandoned DNA diagnostics based on pattern recognition and addressable oligonucleotide libraries, although these application areas were still technically feasible.

Incyte and CalTech researchers published two articles and gave more than 20 presentations at a variety of industry and trade conferences to share their knowledge.

Incyte Changes Focus beyond Microarrays

Two years after the conclusion of this project, biotechnology made headlines in 2000 when the completed human genome sequence was announced. Several companies announced that they had finished deciphering the entire human genetic code. This achievement, combined with other research, generated more than a billion pieces of DNA data; moreover, the

wealth of data was growing each day. Incyte's in-house microarray manufacturers and researchers built on their ATP-project knowledge and continued using pin-based deposition microarrays. From these analyses they built databases of genetic information. In response to the focus on the human genome, Incyte Pharmaceuticals changed its name to Incyte Genomics in 2000.

By 2001, Incyte shifted its focus to bioinformatics (medical databases, licenses, and tools) based on its microarray analyses. The company sold access to its databases by subscription, which supported researchers from universities and pharmaceutical companies in exploring complex illnesses such as diabetes, Alzheimer's, and cardiovascular disease. The company changed its name to Incyte Corporation in 2002. Incyte ultimately wanted to develop a drug pipeline of its own. Incyte hoped to identify a promising drug target and to work with a pharmaceutical company willing to take the drug through the expensive and lengthy Food and Drug Administration clinical trials. Incyte held patents for 600 full-length human genes and the proteins they encode, which was more than any other firm. These patents may be the key to a major drug discovery. Analysts predicted that the big growth would be in the drug discovery arena. As of 2004, Incyte was focusing on drug discovery (both internal programs and in collaboration with pharmaceutical companies) with clinical and pre-clinical trials targeting the following diseases:

- HIV (Phase II clinical trials)
- Rheumatoid arthritis, multiple sclerosis, neuropathic pain, atherosclerosis (Phase I clinical trials)
- Cancer (discovery phase)
- Diabetes (preclinical trials)

ATP-Funded Microarray Technology Is Licensed

In 2001, Incyte finalized agreements with Agilent Technologies to use Incyte's pin-based microarrays in order to identify critical genes involved in various biological processes and, especially, to identify potential new targets for drugs. Agilent developed highly specific complementary DNA- and oligonucleotide-based microarrays for detecting and monitoring gene expression (complementary DNA, or cDNA, is a portion of the DNA that specifies a protein; it is complementary to a messenger RNA [mRNA] molecule). The agreements provided Agilent with one of the most comprehensive microarray product lines available, according to Roy A. Whitfield, Chief Executive Officer of Incyte.

Agilent also licensed Incyte's key chem-jet-based microarray and gene expression patents, which strengthened Agilent's position as a major manufacturer of printed DNA microarrays. These patents include the ATP-funded inventions to synthesize oligonucleotides right on the slide and added to Agilent's existing chem-jet intellectual property. Incyte later purchased ready-made chem-jet microarrays from Agilent for use in Incyte's research into drug targets.

The patents that resulted from the ATP-funded research provide Agilent more flexibility in the manufacturing process. As of April 2004, the company was developing a new production process that takes advantage of that increased flexibility. For example, the process will allow technicians to deliver the same reagent to multiple layers of sequences on a single spot. The company is moving toward more multiplexing (putting multiple arrays on a single slide at higher densities). Researchers typically have samples from seven different organs to analyze (for example, heart, lung, liver, kidney) and currently process each tissue independently. Agilent plans to introduce a single slide with eight different tissue arrays that can be processed independently.

Agilent anticipates moving toward microarrays that have more features and more DNA arrays. Currently they print up to 44,000 arrays per slide and expect this to increase.

In 2004, Frost & Sullivan industry analysts estimated that global DNA microarray markets generated $596 million in 2003 and will reach $937 million by 2010, growing at approximately 6.7 percent annually.

Conclusion

In 1994, the biotechnology industry sought ways to conduct high-throughput DNA analyses. With ATP support, Combion successfully developed patented chem-jet-based microarray fabrication methods and gained the attention of Incyte Pharmaceuticals. Incyte purchased Combion in 1996 and continued the ATP-funded research. Incyte changed microarray production methods in 1998 and put this technology on hold due to high development costs. In 2001, Incyte (its name had been changed to Incyte Genomics) sold licenses for the

ATP-funded technologies to Agilent Technologies, a company that markets comprehensive chem-jet-based microarray products and accessories to pharmaceutical and research organizations. Incyte (renamed to Incyte Corporation in 2002) uses the arrays for research that seeks to discover new drug targets and to improve drug effectiveness for HIV, rheumatoid arthritis, multiple sclerosis, neuropathic pain, atherosclerosis, cancer, and diabetes. As of 2004, Agilent is relying on this ATP-funded project's advances to develop enhanced microarrays, providing more data to researchers in their search for high-value medications. The global DNA microarray industry is forecasted to reach $937 million by 2010.

PROJECT HIGHLIGHTS
Incyte Corporation (formerly Combion, Inc.)

Project Title: Using Chem-jet Techniques to "Print" High-Density Microarrays (DNA Diagnostic Systems Based on Novel Chem-jet Techniques)

Project: To develop a method similar to ink-jet printing for synthesizing large arrays of specific DNA fragments suitable for medical diagnosis, microbial detection, and DNA sequencing and for creating supplies of detachable oligonucleotides (subunits of DNA) for subsequent use.

Duration: 2/1/1995-1/31/1998
ATP Number: 94-05-0019

Funding (in thousands):

ATP Final Cost	$1,415	64.5%
Participant Final Cost	780	35.5%
Total	$2,195	

Accomplishments: With ATP funding, Combion successfully developed flexible techniques for manufacturing chem-jet-based microarrays. These techniques allowed Combion to "print" many DNA arrays directly on the microscope slide, or chip. The company (purchased by Incyte in 1996) also accomplished the following:

Combion received the following five patents for technologies related to the ATP-funded project, which it licensed to Agilent Technologies in 2001. These technologies provided increased flexibility in producing microarrays and provided the foundation for later related patents filed by Agilent.

- "Jet droplet device"
  (No. 5,958,342: filed May 17, 1996; granted September 28, 1999)
- "Apparatus for the chemical synthesis of molecular arrays"
  (No. 5,981,733: filed September 16, 1996; granted November 9, 1999)
- "Methods and solvent vehicles for reagent delivery in oligonucleotide synthesis using automated pulse jetting devices"
  (No. 5,874,554: filed December 13, 1996; granted February 23, 1999)
- "Methods for performing multiple sequential reactions on a matrix"
  (No. 5,847,105: filed August 13, 1997; granted December 8, 1998)
- "Jet droplet device"
  (No. 6,001,309: filed May 15, 1998; granted December 14, 1999)

Commercialization Status: Microarray expertise and knowledge gained in this project formed the foundation for Incyte's highly successful bioinformatics business, which operated from 1999 to 2001 (selling subscriptions to databases of DNA information). Although Incyte put the specific chem-jet microarray manufacturing techniques developed in this project on hold from approximately 1998 to 2004, the company licensed the technology to Agilent in 2001. As of 2004, Agilent was about to commercialize the ATP-funded technology in conjunction with their numerous other patented chem-jet technologies. Agilent is the leading manufacturer of chem-jet-based microarrays. The company expects to benefit from the ATP-funded technology in two areas:

- The patented ATP-funded technology, which is a new manufacturing process with greater flexibility, applies a reagent to multiple layers of sequences on a single spot.
- Agilent expects to add more microarray features in the future. As of 2004, the company offers up to 44,000 DNA spots per slide. They are also moving toward multiplexing (they will put up to 8 different tissue samples on a single slide at high densities). Researchers typically analyze seven different sample organ tissues (such as heart, lung, kidney, liver) to test for disease.

Outlook: The outlook for DNA microarray manufacturing is good but clouded. The tendency toward consolidation is forcing out smaller companies, and price competition is strong. Global sales were $249 million in 2001 and $596 million in 2003. More importantly, this market serves the DNA diagnostic and pharmaceutical market, which holds great promise for discovering new and improved treatments for many diseases. Incyte's drug targets as of 2004 included HIV, rheumatoid arthritis, multiple sclerosis, neuropathic pain, atherosclerosis, cancer, and diabetes.

Composite Performance Score: ***

PROJECT HIGHLIGHTS
Incyte Corporation (formerly Combion, Inc.)

Number of Employees: 4 at project start, 10 upon sale of Combion to Incyte in 1996, 676 as of December 1997, 1,322 as of December 2000, 215 as of March 2004.

Focused Program: Tools for DNA Diagnostics, 1994

Company:
Incyte Corporation
Experimental Station
Rt. 141 & Henry Clay Rd.
Building E336/226
Wilmington, DE 19880

Contact: John Keller
Phone: (302) 498-6796

Subcontractor:
California Institute of Technology
Pasadena, CA

Publications:
Incyte published its results in the following articles:

- Schena, M., R. A. Heller, T. P. Theriault, K. Konrad, E. Lachenmeier, and R. W. Davis. "Microarrays: Biotechnology's Discovery Platform for Functional Genomics." *Trends in Biotechnology* 16, pp. 301-306, 1998.
- Theriault, T.P., S. W. Winder, and R. C. Gamble. "Application of Ink-Jet Printing Technology to the Manufacture of Molecular Arrays." *DNA Microarrays: A Practical Approach.* Mark Shena, Ed. Oxford University Press, pp. 101-119, 1999.

Presentations:
Incyte provided the following presentations at a variety of industry and trade conferences:

- "DNA Diagnostic Systems Based on Novel Chem-Jet Techniques." Biochip Array Technologies: Fabrication and Application, International Business Communications, Washington, DC, May 1995.
- "Synthesis of Oligonucleotide Arrays Using Chem-Jet Technology." Biochip Array Technologies, International Business Communications, Marina Del Rey, CA, March 1996.
- "Incyte Microarray Technologies: Chem-jet Synthesis and Gene-jet Gridding." Biochip Arrays and Integrated Devices for Clinical Diagnostics, International Business Communications, San Diego, CA, March 1997
- "Synthesis of Oligonucleotide Arrays Using Chem-jet Technology: Diagnostic System Design." NIST Public Meeting, Gaithersburg, MD, September 1997.
- "Gene Identification and Expression Level Monitoring by DNA Microarrays." GASLINI Course in Cancer Genetics, Sestri Levante, Italy, September 1997.
- "Microarray Technology for Transcript Imaging." Advances in DNA Diagnostics, National Managed Health Care Congress (NMHCC), Washington, DC, October 1997.
- "GeneJet Gridding Technology for DNA Microarrays." Microfabrication Technology for Biomedical Innovations, Cambridge Healthtech Institute, San Jose, CA, October 1997.
- "Incyte Ink-Jet Microarray Technologies: CombiJet Synthesis; GeneJet Gridding." NIST Public Meeting, Gaithersburg, MD, March 1998.
- "Microarray Technology for Enhanced Drug Discovery." Discovery 98: Emerging Technologies for Drug Discovery, NMHCC, San Diego, CA, May 1998.

Research and data for Status Report 94-05-0019 were collected during March – April 2004.

JDS Uniphase (formerly Uniphase Corporation)

The BioLaser: A Compact Blue Laser for DNA Diagnostics

In 1994, scientists and researchers were attempting to capitalize on the building successes of the "genetic revolution" and move DNA analysis instruments into more laboratories in the United States and around the globe. At the time, U.S.-based companies, such as Perkin-Elmer's Applied Biosystems Division, already had DNA analysis instruments on the market. However, these instruments were expensive and required large power supplies and significant levels of maintenance by factory-trained technicians. The price, life-cycle costs, and infrastructure requirements kept DNA analysis out-of-reach for many laboratories in the United States and for many more laboratories worldwide.

Uniphase Corporation and Perkin-Elmer responded to the Advanced Technology Program's (ATP) 1994 focused competition, "Tools for DNA Analysis," with a joint venture proposal to focus industry attention and resources on a problem with existing DNA analysis tools: the size and power requirements of internal lasers. Lasers were typical components of the automated DNA analysis machines. They were used to signal computers that a particular DNA sequence of interest was present in a sample. Uniphase and Perkin-Elmer proposed a research program that would result in the BioLaser, a solid-state, blue-light laser hoped to be half the cost, 10 times smaller, and 250 times more efficient than existing lasers used in DNA analysis machinery.

At the close of the ATP-funded research project in late 1997, technical barriers prevented Uniphase and Perkin-Elmer from developing the BioLaser at a cost that was commercially viable. However, two ancillary products were created, one for the bioscience industry and one for the digital photo-finishing industry.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* *

Research and data for Status Report 94-05-0004 were collected during September - October 2003.

Lasers for DNA Detection Require Improvements

Machines used in 1994 for automated DNA analysis, such as Perkin-Elmer/Applied Biosystems Division's Model 373, were large and expensive. They also consumed substantial electrical power and required costly maintenance, which raised the total life-cycle costs well above the machine's initial $100,000 price. Moreover, the laser used to identify target DNA sequences did not work with the existing DNA analysis machines. The argon-based lasers were expensive; they constituted one of the most costly components of the entire DNA analysis system. Also, the large size of both the laser head (the portion of the laser that creates the light needed for the machine to function) and the power supply was a problem. These argon-based lasers also wore out quickly, requiring additional expenses for field visits by a trained service representative to replace the laser.

Uniphase and Perkin-Elmer Propose to Address the Laser Problem

Uniphase Corporation and the Applied Biosystems Division of the Perkin-Elmer Corporation jointly proposed to develop the BioLaser, a compact, inexpensive source of blue laser light for instruments intended for DNA analysis. The BioLaser would address industry concerns about DNA analysis machine power, size, initial price, and life-cycle costs.

In 1994, laser-based DNA analysis instruments relied on fluorescent dyes that linked to specific genetic regions of interest for analysis. Lasers were used to excite the dyes so that a computer-linked optical device could identify the dyes and analyze the genetic regions that were bound to the dyes.

The lasers used in 1994 were argon-based, blue-light lasers. The argon gas made the lasers bulky, expensive, and relatively inefficient compared with the envisioned laser that could move DNA-based analysis toward commercial acceptance. The physical limitations of the argon-based lasers also presented substantial roadblocks to developing diagnostic instruments with the low cost, small size, and convenience required for rapid, widespread domestic and international adoption. Uniphase's proposed solid-state laser (as opposed to the argon-based lasers), which would be manufactured using novel methods adapted from other industries, could be smaller, more efficient, longer lived, and half the cost of argon-based lasers.

Technical Barriers to BioLaser Development

Uniphase's proposed solid-state laser would work by directing infrared laser light originating in a semiconductor diode laser through a waveguide (material that amplifies and alters the laser light to a useful frequency) made of nonlinear optical materials (materials used to significantly increase the frequency of the laser light). These optical materials would double the diode's original light to a specific frequency for blue light. Reliably doubling this particular light frequency with a waveguide represented the most significant technical challenge that Uniphase and Perkin-Elmer sought to overcome. Other technical challenges identified by the companies included the following:

- Reliably fabricating the waveguides
- Maintaining the laser light within a narrow frequency band at high power
- Coupling the infrared light to the waveguide material with minimal loss of intensity and power

Project Success Would Benefit the U.S. Economy

In 1994, a typical instrument for the acquisition of genetic information was the Perkin-Elmer/Applied Biosystems Model 373 genetic detection and analysis system. The Model 373 used an expensive argon laser-based fluorescence detection system. Uniphase and Perkin-Elmer proposed to greatly reduce the cost and to increase the efficiency of the laser excitation and detection systems. They envisioned that their successful BioLaser would be half as expensive, 10 times smaller, 250 times more efficient, and 5 times longer lived than lasers used in existing instruments.

The laser used to identify target DNA sequences did not work with the existing DNA analysis machines.

If Uniphase and Perkin-Elmer had succeeded in creating their proposed BioLaser, industry experts projected that the market for automated DNA research projects could have grown as more laboratories in the United States and overseas could have purchased the lower cost, easier-to-maintain machines.

ATP Funding Essential to Gaining Industry Attention

ATP's 1994 "Tools for DNA Analysis" focused competition invited applications for ATP funding from industry participants with proposals to address roadblocks in the DNA analysis industry. Uniphase and Perkin-Elmer responded to that competition.

Their successful BioLaser would be half as expensive, 10 times smaller, 250 times more efficient, and 5 times longer lived.

A partnership of industry powerhouses like these two companies was not common in 1994. The DNA analysis industry was not yet mature, and trade associations and other sources of funding for pre-competitive research did not have enough capital to galvanize the industry to solve specific technical problems. ATP funding facilitated parallel research at multiple companies by

numerous scientists and helped DNA analysis research. ATP awarded Uniphase and Perkin-Elmer $1.36 million to conduct initial research into the BioLaser.

Research Team Well Equipped to Tackle Technical Challenges

Affymetrix and Molecular Dynamics joined Uniphase and Perkin-Elmer as informal collaborators on the project. The Uniphase and Perkin-Elmer team also brought in New Focus, Inc. as a subcontractor who would develop specialty antireflection coatings for the photonic (light-directing) components of the BioLaser.

The team assembled by Uniphase and Perkin-Elmer had substantial experience in lasers and bioscience. The team aggregated this expertise and focused on solving the technical challenges posed by solid-state, blue-light lasers in DNA analysis applications. Highlights of the companies' expertise include the following:

Uniphase. This San Jose, California public company had significant experience with optical technologies. Uniphase also maintained a growing position in the marketplace for optical technologies for biomedical applications. Through a series of mergers and acquisitions, Uniphase later became JDS Uniphase.

Applied Biosystems Division of Perkin-Elmer. This division of the Wellesley, Massachusetts public company was focused on the following markets: basic research; commercial research (pharmaceutical and biotechnology); and standardized testing, including forensic human identification, HIV genotyping, and food testing. This division spun off into a separate California-based public company and was later acquired by the Connecticut-based Applera Corporation.

Affymetrix. This Santa Clara, California public company offers an expanding portfolio of integrated products and services, including its integrated GeneChip platform, to address growing markets focused on understanding the relationship between genes and human health. The company's customers include pharmaceutical, biotechnology, agrichemical, diagnostics, and consumer products companies, as well as academic, government, and other nonprofit research institutes. Affymetrix completed an initial public offering in 1996 during this ATP-funded project.

Molecular Dynamics. Molecular Dynamics, a private company based in Cambridge, Massachusetts, focused on genetic sequencing and discovery. The company's flagship products were a series of DNA microarrays that consisted of thousands of DNA samples deposited in small spots on the surface of a slide. Molecular Dynamics worked with Uniphase and Perkin-Elmer to improve the method of analyzing these DNA microarrays using solid-state, blue-light lasers. The company was acquired in 1998 by what later became Amersham Biosciences.

New Focus, Inc. New Focus is a Sunnyvale, California private company that is a leading supplier of photonics research tools. Their product lines include tunable lasers, high-performance photodetectors and receivers, laser modulators, mechanical positioners, and optical components. The proposed blue laser for biotechnology applications required state-of-the-art antireflective coatings for the laser diode, waveguide frequency doubler, and fiber to eliminate distortion. Because New Focus maintained a fully equipped optical coating facility within a short drive from Uniphase, New Focus was hired as a subcontractor to handle all antireflective coating work for the project. New Focus completed an initial public offering in 2000.

Technical Barriers Remain, Yet Products Resulted

All members of the joint venture worked diligently during the three-year project to find a way to manage blue-light, solid-state lasers. The team constructed a prototype laser in the laboratory for testing and further development. During the project, however, technical barriers proved insurmountable, and innovations in DNA analysis methodologies made the BioLaser concept obsolete for DNA analysis applications.

Technical barriers proved insurmountable, and innovations in DNA analysis methodologies made the BioLaser concept obsolete.

The most significant technical barrier Uniphase and Perkin-Elmer faced was in developing new fluorophore

labels for light generated by anything other than argon ions in the traditional gas-based laser. (Fluorophore labels create fluorescent light when excited by a laser to signal a DNA match that can then be read by a computer.) Because the fluorophore labels bind directly to the DNA that the diagnostic machine is analyzing, the labels must be compatible with the enzymes used to cut the DNA; must be chemically stable so as not to interfere with the method used to separate the DNA; and must be able to absorb large amounts of laser-generated light before fluorescing.

Before the end of the ATP-funded project, the joint venture participants were able to generate fluorophore labels for blue-light, solid-state lasers. The labels and laser modifications, however, were still expensive. The expected overall cost reduction from the prototype BioLaser generated during the course of the ATP-funded project would have been only 9 percent; a 50-percent reduction was required for commercial acceptance.

After the close of the project, Uniphase continued research into diode-based, red-light lasers as a possible solution for a DNA analysis machine. However, in the late 1990s, shortly after the end of the project, a new method of DNA separation and analysis was introduced, which eliminated the need for relatively high-powered lasers such as the blue and red lasers that Uniphase and Perkin-Elmer sought to create. The new method, polymerase chain reaction (PCR), also used laser-induced fluorescence. With PCR, the fluorescence would occur on DNA probes. Higher powered lasers, however, would destroy the reactions on the DNA probes and ruin the PCR output.

Unrelated to the automated DNA analysis marketplace, Uniphase created two new products flowing from innovations discovered during the ATP-funded project. The first, a Blue Laser Module, was a stripped-down, inexpensive blue laser for tabletop applications within the biotechnology industry. The product represented an adaptation of an existing technique to meet cost pressures. It entered the marketplace in 1999 and sales ranged between $50,000 and $500,000 annually from 1999 to 2002. The second product was a specialized, low-noise blue laser for digital photo-finishing. This new product enhanced digital photo-finishing capabilities above and beyond other commercially available techniques. Uniphase introduced the product in 2000 and earned between $250,000 and $1 million annually from 2000 to 2002. The product made photo-finishing easier and cheaper than was previously possible.

Conclusion

In 1994, a joint venture led by Uniphase and Perkin-Elmer's Applied Biosystems Division sought to reduce the size, cost, and power requirements for the lasers used in DNA analysis. One goal of this ATP-funded project was to make DNA analysis machines accessible to more laboratories in the United States and abroad. The joint venture partners worked diligently for three years, but the expected cost savings on lasers for DNA analysis were not realized. However, the technical developments led JDS Uniphase to unanticipated new products, as well as a patent application, and led Perkin-Elmer to publish an article about the technology.

PROJECT HIGHLIGHTS
JDS Uniphase (formerly Uniphase Corporation)

Project Title: The BioLaser: A Compact Blue Laser for DNA Diagnostics

Project: To develop a compact, efficient, and cheaper source of blue light for fluorescence-based diagnostic instruments and techniques for physicians and biomedical researchers.

Duration: 1/1/1995-12/31/1997
ATP Number: 94-05-0004

Funding (in thousands):

ATP Final Cost	$1,361	50%
Participant Final Cost	1,369	50%
Total	$2,729	

Accomplishments: Although the Uniphase/Perkin-Elmer team achieved partial technical success, they did not succeed in reducing the cost of lasers for DNA analysis applications enough to earn commercial acceptance. The companies did not pursue their research after the close of the ATP-funded project because a new DNA identification technology entered the field, the polymerase chain reaction. However, the following ancillary new products or product improvements resulted from the ATP-funded project:

- The Blue Laser Module, an inexpensive blue-light laser for tabletop applications in the bioscience industry (a new product for Uniphase)
- The MicroBlue SLM, a specialized laser for the digital photo-finishing marketplace (a new product for Uniphase)

The ATP-funded project also resulted in one patent application by Uniphase, as well as the following publication by Perkin-Elmer:

- O'Neill, Michael D. "Sequencers Benefit from Solid State Detectors." Laser Focus World, October 1995: p. 135.

Commercialization Status: The commercialization status is positive for the two ancillary products developed by JDS Uniphase. The Blue Laser Module reached the market in 1999 and has achieved sales as high as $500,000 per year. The MicroBlue SLM was first marketed in 2000 and generated $1 million in annual sales.

Outlook: The outlook for the two ancillary products flowing from the ATP-funded research is good. However, the outlook is poor for the blue-light, solid-state BioLaser that was the initial focus of the project.

Composite Performance Score: * *

Focused Program: Tools for DNA Analysis, 1994

Company:
JDS Uniphase Corporation
1768 Automation Parkway
San Jose, CA 95131

Contact: Dr. Kevin Kalkhoven
Phone: (408) 434-1800

Applied Biosystems Division (formerly a division of Perkin-Elmer)
850 Lincoln Centre Drive
Foster City, CA 94404

Subcontractors:

- Affymetrix
  Santa Clara, CA
- Molecular Dynamics
  Cambridge, MA
- New Focus, Inc.
  Sunnyvale, CA

Research and data for Status Report 94-05-0004 were collected during September-October 2003.

Large Scale Biology Corporation (formerly Large Scale Proteomics Corporation)

Protein Discovery and Analysis Standardized through Improved Gel Media

In 1994, the U.S. biotechnology industry knew far more about the DNA molecule containing the body's genetic code than about the proteins resulting from specific genetic codes. Knowledge of protein structure and function remains critically important: proteins catalyze and control chemical reactions within the body and manifest most of the impacts of aging, disease, drugs, toxic agents, and radiation. Scientists struggled to understand the structure and function of many proteins, because the primary technique for separating proteins from other biological materials was slow and costly. This technique, known as electrophoresis, had not advanced substantially since its development in 1975. Electrophoresis required that small, individual gel media house the protein samples. The gels were then bombarded with, among other things, electric current to separate the proteins. This separation took 20 hours or more per individual gel and produced widely inconsistent results. In 1994, Large Scale Proteomics Corporation (LSPC) submitted a proposal to the Advanced Technology Program (ATP) to standardize gels into larger sized, more consistent media for faster protein separation. If successful, the speed of protein analysis would increase dramatically, reducing cost and facilitating the discovery of novel therapeutics for a wide variety of ailments.

After three years of focused effort enabled by ATP funding from 1995 to 1998, LSPC scientists succeeded in decreasing the variability in gel results to less than 15-percent variation in protein position. This represented a nearly 100-percent improvement from what was possible before the ATP-funded project began. They achieved a throughput (cycle speed) of 100 analyses per cycle. Company scientists also believed they could develop an innovative, high-risk, integrated protein analysis system if they were able to obtain additional funds. In 1997, LSPC received a second ATP award and ultimately developed ProGEx, a novel low-cost, high-throughput, integrated platform for protein discovery and analysis. The company was acquired by Large Scale Biology Corporation (LSBC) in 1999. In 2001, with the help of their ProGEx system, LSBC created the first draft of the Human Protein Index, a database that contains more than 115,000 proteins that work within the human body.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* * * *

Research and data for Status Report 94-01-0284 were collected during October 2003 – March 2004.

Focused Effort on Protein Research Technique Proposed

Proteins carry out the molecular functions that keep the body alive, such as producing energy, breaking down waste, and fighting infection. They are key to the life of healthy cells, and they are central to disease and treatment as well. Most human diseases involve protein abnormalities such as overproduction, underproduction, or a failure to function, and most drugs target proteins in an attempt to correct the abnormalities that cause disease. Understanding proteins is essential to understanding biological processes and new methods of intervention. The study of proteins is called proteomics. Combinations and relative quantities of certain proteins indicate relative health or illness. Protein markers are proteins that, when present in body fluids such as blood or urine, may be used to make an early diagnosis of disease or track its progress or response to treatment.

In 1994, many industry experts believed that the promise for a proteomics revolution in biomedical science equaled or surpassed the possibilities enabled by the genetic revolution. Economic forecasts predicted a multi-billion-dollar domestic protein sequencing and analysis market, and hundreds of billions more in revenues should the protein-based revolution lead to the creation of new therapeutics and medicines. Although the pipeline for drug development can be longer than a decade, if protein diagnostics enabled only one therapeutic, the benefits in re-captured productivity from fewer sick days and from sales alone would provide a hundredfold return on the initial investment in standardizing the process used for protein separation, known as two-dimensional (2-D) electrophoresis.

The 2-D electrophoresis process was a powerful but variable and labor-intensive laboratory bench-level research tool (as opposed to a tool that required extensive analysis in a sophisticated, specialized laboratory). Large Scale Proteomics Corporation (LSPC) developed a research plan to transform 2-D electrophoresis into an economical, standardized, automated, routine analytical system. This system would be based on using innovative, low-cost gel media through which proteins could be quickly and inexpensively sequenced along the two dimensions of size and pH (degree of acidity or baseness of a gel). The research plan attempted to eliminate the technical barriers that had prevented widespread quantitative use of 2-D electrophoresis to answer major questions in the protein research and customized therapeutics industries.

Successful Project Could Have Vast, Positive Effects on the Nation's Health

If the LSPC-proposed system worked as planned, it would give scientists the chance to map portions of an individual's various protein expression sequences and to study protein function so that they could analyze the impact of medications on the individual patient. Used in a systematic way, a successful project would give scientists an in-depth understanding of protein structure and function. They could learn which proteins are present in unusually large or small quantities in diseased tissues, a step toward finding the molecular roots of a given disease.

The research project could also determine conclusively whether a 2-D electrophoresis system was capable of providing important data on the effects of a broad range of pharmaceuticals in vivo. The results could be used to establish "normal" values for the population as a baseline for future analysis of the many proteins synthesized within the body. The result could be a database that reveals novel patterns of relationships between genes and proteins and between proteins and physical manifestations of disease, with potential importance to drug discovery. Existing genetic sequence data did not indicate where and when different genes may be activated or changed (and, therefore, where and when certain diseases would manifest themselves), so the protein function database could help uncover those triggers. Careful analysis of the data could lead to customized medications, fewer incidents of rejection following organ transplants, and lower incidence of drug-resistant viruses.

Government Support Necessary for Protein Discovery Tool

Because no company in the industry had improved substantially upon 2-D electrophoresis since its development in 1975, sources of private funding for LSPC's efforts were difficult to find. Moreover, the company had identified numerous technical risks associated with building an economical, automated 2-D electrophoresis. The most daunting risk was in producing high-quality, consistent 2-D patterns necessary for identifying particular proteins. Gels have a similar consistency to gelatin, and the soft structure was not conducive to exact results because it was easy for a protein to lose its direction during separation. Therefore, passing proteins through the substance did not always result in the same proteins migrating to the same place in every gel.

Economic forecasts predicted a multi-billion-dollar domestic protein sequencing and analysis market.

If successful, the research project would drive the total cost per electrophoresis gel as low as possible. The lower cost would increase the use of 2-D

electrophoresis in taking a "snapshot" of the proteins at work, facilitating data comparisons and increasing the chance that new knowledge or therapeutics would result to benefit society. However, by developing and selling the low-cost gels, LSPC could not capture even a small fraction of the overall return on their investment into creating standardized 2-D electrophoresis units. The near-term required rates of return to compensate for the technical risk made using internal capital nearly impossible, because the limitations would place funding for 2-D electrophoresis research behind numerous other corporate priorities. The potential for return was in providing protein analyses (proteomics) for pharmaceutical companies and research hospitals on a fee-for-service basis in the future.

Although LSPC was able to commit $1.4 million in internal funds, they turned to ATP for the additional financial support necessary for their research program. The company submitted a proposal during ATP's 1994 general competition and received an award of $1.9 million to augment their internal funding.

Project Eventually Leads to Protein Research Platform

LSPC proposed the following six-step research program for the ATP-funded project:

1. Produce a one-dimensional (1-D) electrophoresis gel that was larger than the industry standard and was available to sequence more proteins than what the industry could handle in 1995. But the larger size was not the only goal. LSPC also wanted to significantly reduce positional variation (the degree of change in the separated protein's final position from gel to gel) and improve readability. With added size and accuracy, the 1-D gel could sequence more proteins more reliably than ever before.
2. Determine ideal pH ranges for analyzing cellular protein standards.
3. Create a new, simple, standard way to manufacture gels that are capable of analyzing 500 protein segments at once. This manufacturing approach would not require research scientists to spend time and energy mixing, pouring, and setting the gel; moreover, it would open doors for the creation of a ready-to-use gel.
4. Reduce the cost of producing a 2-D slab gel by 90 percent while maintaining low positional deviation.
5. Generate test samples that exemplify the effects of commercially important drugs *in vivo*.
6. Evaluate the positional and quantitative reproducibility of the combined 1-D and 2-D system and determine whether the system can detect and characterize drug effects.

LSPC successfully developed larger gels that were easier to read and handle (step 1), and they optimized and standardized the gels' pH levels (step 2).

The researchers developed an automated serial production line, but had difficulty in molding and reproducing gels. They were able to achieve a throughput of 100 analyses per cycle (step 3). Although the new larger gels held their shape better, scientists were still trying to drill precise holes into the gels in order to hold the sample proteins. The holes and the path of the migrating proteins still could not be predicted with enough reliability (step 4) to develop an efficient system for detecting and characterizing proteins.

In addition, scientists analyzed the effects of 50 pharmaceuticals on the protein composition of rodent liver (step 5) and built a demonstration database using proprietary data analysis and visualization software to characterize drug mechanisms (step 6). More research and advanced testing would be necessary after the close of the ATP-funded project in 1998 before a commercial product would be available.

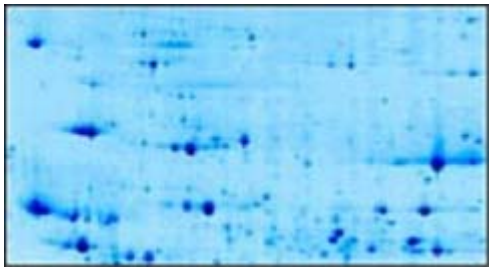
ATP-Funded Project Generates Significant Knowledge for the Industry

Even though the company did not create a gel electrophoresis-based system for characterizing drug effects, the project did achieve other promising technical outcomes. LSPC scientists succeeded in holding positional deviations to under 15 percent (which was more than twice as accurate as what was achieved prior to the ATP award). The company also designed, built, and tested a new gel-manufacturing process and received three patents; more were filed and awarded flowing from subsequent, related research into this type of technology.

Although the positional deviation was still too high to meet their goals without additional research, LSPC scientists overcame the deviation and produced a high-

quality machine for protein analysis. This created a change in the business model that would focus less on selling gel media and more on conducting the protein analysis in-house, using a machine that compensated for the weaknesses inherent in the gel media. The company searched for additional financing in order to continue their research into separating proteins through slab-sized electrophoresis gels. The company found the capital through a combination of internal funds, outside investors, and additional government awards. Moreover, LSPC won a separate ATP award to make an integrated system that can separate and analyze proteins in an economical and expeditious manner.

Through these additional investments, LSPC (which was acquired by Large Scale Biology Corporation [LSBC] in 1999) created a protein discovery platform called ProGEx (see illustration), which combines 2-D gel electrophoresis with high-throughput mass spectrometry to enable new and more effective disease diagnostics, therapeutics, and vaccines. By 2001, the ProGEx platform had reduced the cost of protein discovery machines to 50 percent of their 1990s cost. Moreover, the knowledge generated by the ProGEx platform allowed LSBC to complete the first version of the Human Protein Index, a database that contains more than 115,000 proteins from 157 normal human tissues.



A digitized gel image. The ProGEx system uses 2-D gel electrophoresis to identify proteins by electrical charge and molecular weight, which are indicated by their position on the gel. One image is equivalent to thousands of specific protein tests done at once. Software algorithms interpret the results.

In 2000, LSBC conducted an initial public offering and then expanded its research activities into proteomics. The company identified more than 70 protein markers associated with chronic diseases such as osteoporosis, obesity, diabetes, analyzing up to 1,000 gels per week (up to 1 million protein analyses per week). The company also began to form strategic partnerships to develop new products and research tools.

During an interview with ATP staff, LSBC Vice President of Operations, Ms. Connie Seniff, reflected on the ATP-funded project and the focused resources it brought to bear. She estimated that the technology had advanced three to four years faster than would have been possible without ATP support.

During and after the ATP-funded project, LSBC has sought to disseminate knowledge of its technical platforms whenever possible. Articles on the technology developed during the project appeared in business journals as well as in the Journal of the National Cancer Institute and the International Journal of Electrophoresis, in addition to numerous academic articles published by company scientists. Company representatives also gave more than 40 presentations during the ATP-funded project. LSBC created a new subsidiary company, Eclipse Diagnostics, in December 2003, which is focused on promising proprietary technology for potentially discovering biomarkers for disease from blood tests based on mass spectrometry analyses. Its databases are populated with results from the company's earlier automated 2-D gel electrophoresis. In preliminary research collaborations with the National Cancer Institute, Eclipse Diagnostics analyzed a group of blood samples and accurately diagnosed 100 percent of ovarian cancer patients from the samples. If successful, the commercial implications for biomarker discoveries in clinical trials and future therapeutic treatments are huge.

The knowledge generated by the ProGEx platform allowed LSBC to complete the first version of the Human Protein Index, a database that contains more than 115,000 proteins from 157 normal human tissues.

The company closed its 2-D gel fee-for-service proteomics laboratory in February 2004 due to a downturn in business. The market shifted toward pure mass spectrometry analyses, the next generation in protein analysis, and LSBC relocated its staff to

California. Nevertheless, LSBC continues its work to advance the proteomics revolution, based, in significant part, upon knowledge gained from ATP-funded research.

Conclusion

With ATP's award paving the way, Large Scale Proteomics Corporation (LSPC) began an effort to standardize gels to be used in protein discovery and analysis in order to develop an efficient system to detect and characterize drug effects on proteins. While the gels could not be fully standardized, the knowledge generated from the ATP-funded project later led to a protein analysis and discovery platform that has vastly increased the biotechnology industry's level of knowledge about protein structure and function. LSPC, acquired by Large Scale Biology Corporation (LSBC) in 1999, published its findings and ultimately produced the Human Protein Index, a database that contains more than 115,000 proteins from 157 normal human tissues. The greatest potential lies in discovering biomarkers for disease from blood tests. In 2003, LSBC created a new subsidiary company, Eclipse Diagnostics, to focus on biomarkers. The company closed its two-dimensional (2-D) gel proteomics laboratory in 2004 and shifted toward pure mass spectrometry analyses, the next generation in protein analysis. The outlook for protein analyses continues to be strong.

PROJECT HIGHLIGHTS
Large Scale Biology Corporation (formerly Large Scale Proteomics Corporation)

Project Title: Protein Discovery and Analysis Standardized through Improved Gel Media (Standardization of 2-D Protein Analysis Using Manufacturable Gel Media)

Project: To develop economical production methods for reproducible media used in two-dimensional (2-D) electrophoresis and then use the system to detect and characterize protein-level effects of pharmaceuticals in animal test systems.

Duration: 1/16/95-1/15/98
ATP Number: 94-01-0284

Funding (in thousands):

ATP Final Cost	$1,902	47%
Participant Final Cost	1,445	53%
Total	$3,347	

Accomplishments: Although Large Scale Proteomics Corporation (LSPC) did not succeed in revolutionizing protein discovery through their initial research, the knowledge the company gained enabled them to set up a plan for future research into gel electrophoresis media. The company attracted significant additional funding for continued research into protein discovery. Eventually, they developed the ProGEx product line for protein identification and research. The company also completed the first version of the Human Protein Index by identifying more than 115,000 proteins from 157 medically relevant human tissues.

LSPC received three patents for technology developed during this ATP-funded project.

- "Automated system for two-dimensional electrophoresis" (No. 5,993,627: filed June 24, 1997, granted November 30, 1999).
- "Automated system for two-dimensional electrophoresis" (No. 6,136,173: filed June 24, 1999, granted October 24, 2000).
- "Automated system for two-dimensional electrophoresis" (No. 6,123,821: filed September 27, 1999, granted September 26, 2000)

The company's foundational knowledge gained during this project led to later additional patents that were filed covering related technology. These research areas would not have been possible without the early ATP support.

The company's core technology (including the technology flowing from the ATP-funded research) also received media coverage in the following publications:

- *Start-Up* (covered twice in this entrepreneurial newspaper that deals with emerging medical venture opportunities)
- *Science*
- *Proteomics*
- *Genetics Engineering News*
- *Venture Capital Health Care*
- *High Tech Separations News*
- *Drug & Market Development*
- *Spectrum* (a newsletter on pharmaceutical industry dynamics)
- *BioWorld*

In addition, during the ATP-funded project, LSPC scientists and company officers gave more than 40 formal presentations. The presentations were given at conferences in the United States and in Europe, such as the Conference on Toxicological Mechanisms, the Society of Toxicologic Pathology, the Division of Animal Clinical Chemistry, International Electrophoresis, Informatics and Genome Research, Functional Genomics, the Association of Biomolecular Resources, and the American Association for Clinical Chemistry. Topics included molecular toxicology, mechanistic toxicology, functional genomics, drug discovery, pharmaceuticals, biotechnology, agricultural biotechnology, and clinical chemistry.

Commercialization Status: The 2-D gel and ProGEx line of protein analysis tools has been upgraded and improved over the years. Large Scale Biology Corporation (LSBC), which acquired LSPC in 1999, still sells research products and databases created through use of technology flowing from the knowledge acquired during this ATP-funded project. The company performs up to one million mass spectrometry analyses of proteins per week. In 2003, LSBC created a new subsidiary company, Eclipse Diagnostics, which focuses on discovering biomarkers for disease from blood tests.

Outlook: The outlook for the technology flowing from the ATP-funded research continues to be strong. LSBC uses the protein research tools, and the industry (e.g., pharmaceutical companies and research hospitals) uses the databases created by the protein research tools.

PROJECT HIGHLIGHTS
Large Scale Biology Corporation (formerly Large Scale Proteomics Corporation)

Composite Performance Score: * * * *

Number of Employees: 17 as of December 1997 (Large Scale Proteomics Corporation), 114 as of March 2004 (Large Scale Biology Corporation).

Company:
Large Scale Biology Corporation
3333 Vaca Valley Parkway
Vacaville, CA 95688

Contact: Ronald J. Artale
Phone: (707) 469-2320

Publications and Presentations:

Academic publications related to this project include the following:

- Anderson, N.L., R. Esquer-Blasco, J.-P. Hofmann, L. Meheus, J. Raymackers, S. Steiner, F. Witzmann, and N.G. Anderson. “An updated two-dimensional gel database of rat liver proteins useful in gene regulation and drug effects studies.” Electrophoresis 16: 1977-1981, 1995.

- Anderson, N.L., J. Taylor, J.-P. Hofmann, R. Esquer-Blasco, S. Swift, and N.G. Anderson. “Simultaneous measurement of hundreds of liver proteins: Application in assessment of liver function.” Toxicologic Pathology 24: 72-76, 1996.

- Weinstein, J.N., T.G. Myers, P.M. O'Connor, S.H. Friend, A.J. Fornace, Jr., K.W. Kohn, T. Fojo, S.E. Bates, L.V. Rubinstein, N.L. Anderson, J.K. Buolamwini, W.W. van Osdol, A.P. Monks, D.A. Scudiero, E.A. Sausville, D.W. Zaharevitz, B. Bunow, V.N. Viswanadhan, G.S. Johnson, R.E. Wittes, and K.D. Paull. “An information-intensive approach to the molecular pharmacology of cancer.” Science 275: 343-349, 1997.

- Myers, T.G., N.L. Anderson, M. Waltham, G. Li, J.K. Bulamwini, D.A. Scudiero, K.D. Paull, E.A. Sausville, and J.N. Weinstein. “A protein expression database for the molecular pharmacology of cancer.” Electrophoresis 18: 647-653, 1997.

- Anderson, L. and J. Seilhamer. “A comparison of selected mRNA and protein abundances in human liver.” Electrophoresis 18: 533-537, 1997.

- Li, G., M. Waltham, N.L. Anderson, E. Unsworth, A. Treston, and J.N. Weinstein. “Rapid mass spectrometric identification of proteins from two-dimensional polyacrylamide gels after in-gel proteolytic digestion.” Electrophoresis 18: 391-402, 1997.

- Anderson, N. G., and N. L. Anderson. “Proteome and Proteomics: New technologies, new concepts, and new words.” Electrophoresis 19: 1853-1861, 1998.

- Doherty, N.S., B.H. Littman, K. Reilly, A.C. Swindell, J.M. Buss, and N.L. Anderson. “Analysis of changes in acute-phase plasma proteins in an acute inflammatory response and in rheumatoid arthritis using two-dimensional gel electrophoresis.” Electrophoresis 19: 355-363, 1998.

The company made 44 presentations over the years 1995 – 1998.

Research and data for Status Report 94-01-0284 were collected during October 2003 – March 2004.

Medical Analysis Systems (formerly Navix, Inc.)

Rapid, Low-Cost DNA Diagnostic Technology

In 1995, DNA diagnostics required significant manpower, typically from the highly paid research-grade scientists, which made DNA diagnostic testing prohibitively expensive for clinical diagnostic facilities. Moreover, the capital equipment costs required to add DNA diagnostic capabilities to existing, protein-based laboratory diagnostic methods increased the already daunting capital burden. Navix, Inc., with its subcontractor Profile Diagnostics, Inc., submitted a proposal to the Advanced Technology Program's (ATP) 1995 focused program "Tools for DNA Diagnostics." They proposed to eliminate much of the complexity and to reduce the amount of new equipment needed by a clinical diagnostics laboratory. If successful, Navix could reduce the number of steps required for DNA diagnostics by containing all materials and reactions in a single reaction chamber. This reaction chamber could reduce the cost to administer each test from several hundred dollars to approximately $1 per test.

ATP awarded Navix cost-shared funds to pursue a focused research program. The company succeeded in creating a technique to identify disease-causing sequences of DNA from sample strands, and, in 1999, they received a patent for their efforts. However, this covered only part of the research and development plan. Business problems and technical failures prevented Navix from designing a system that could identify disease-causing strands in a single reaction chamber so that ordinary laboratory machines could process and interpret the results. After the close of the ATP-funded project in 1998, Navix merged with Medical Analysis Systems. A new market study showed that a competitor had entered the marketplace with a better DNA diagnostic product. Therefore, Medical Analysis Systems did not pursue further research into the technology. The company went out of business in 2003 due to business reasons unrelated to the ATP-funded project.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
No Stars

Research and data for Status Report 95-08-0017 were collected during August - October 2003.

U.S. Market Share Threatened

In 1995, the U.S. in vitro diagnostic test manufacturers enjoyed a significant market share of the estimated $17 billion laboratory-use-only annual world market for DNA-based research. Trends throughout both the U.S. healthcare market and the global diagnostic market, however, showed the movement away from "for research only" products and toward more actual DNA-based in vitro diagnostic work. Moving from laboratory work to in vitro work required significantly lower capital and per-test costs. In addition, the global health problem of drug-resistant pathogens placed pressure on healthcare providers to be certain of the nature of the disease before they prescribed treatments. All of these factors put added pressure on the industry to transform DNA diagnostics from a prohibitively expensive research tool to an inexpensive diagnostic technique that could be made available to healthcare providers.

To assist that transition, ATP initiated a focused program in 1995 to fund innovations in "Tools for DNA Diagnostics." Navix, Inc. responded to that focused program with a proposal to create a DNA diagnostics tool that would attempt to eliminate sample preparation steps. The preparation steps represented the most expensive and time-consuming component of DNA diagnostic research.

New Method Could Reduce Costs and Meet Demand

Navix proposed to develop a single-step, homogenous, quantitative assay to detect specific DNA sequences using a process the company called Self-Detected Target-Cycling Reaction (SD-TCR). The proposed SD-TCR process would use a novel protein-assisted strand replacement reaction that incorporated into the assay a protein enzyme coupled to one strand of a DNA probe. This probe corresponded to the DNA sequence correlated with the disease of interest. When double-stranded DNA from a patient was introduced, the probe (covered with an enzyme that actively sought out target double-stranded DNA) would find and replace any areas in the patient's DNA that correlated with disease. This binding released the strand with the enzyme that, in turn, activated a second enzyme that would show as an amplified, colored diagnostic result. In short, if the color appeared, a mutation (or other sequence of interest) was present.

Factors put added pressure on the industry to transform DNA diagnostics from a prohibitively expensive research tool to an inexpensive diagnostic technique.

If successful, the SD-TCR process would dramatically reduce the number of required steps for typical DNA diagnostics to just one. Instead of the costly sample preparation steps, the Navix technology would not require any modification or manipulation of the sample DNA other than simple DNA isolation and its addition to the reaction chamber. After being added to the reaction chamber, the sample DNA would then be allowed to react at controlled temperatures for about an hour. If successful, the SD-TCR process would reduce the time required for DNA diagnostics from days to hours. Moreover, with the addition of a reaction chamber to the system, the cost of conducting a full test analysis would be dramatically reduced from hundreds of dollars per test to less than $1.

Navix Faces Technical Challenges

Navix's technical challenge was to coordinate the three distinct chemical reactions necessary for the SD-TCR technology into a single step. The reactions (DNA identification, amplification, and signal generation) were all possible when they occurred separately, but they needed to be integrated into a working system. Navix entered into a subcontractor agreement with Profile Diagnostics, Inc. to pursue integrating the three reactions. Both Navix and Profile Diagnostics had years of experience in optimizing stand-alone DNA identification, amplification, and signal generation reactions. Together, the team sought to create new DNA diagnostic protocols that would not require new automated instruments. Instead, they asked the question, What can an existing diagnostic laboratory accomplish, and how can we enable one-step DNA diagnostics with those materials?

Navix scientists believed that the materials in a DNA diagnostics laboratory could be integrated through two technically risky and complex steps. First, the scientists would have to compartmentalize and optimize the DNA identification and detection functions into different areas within the reaction chamber without risking contamination. Second, they would have to integrate the three different reactions so that each stage would be compatible with the others and there would be no possibility of premature signal color development that would lead to incorrect results. Navix proposed that they could tightly integrate these reactions in order to achieve a single-step, homogeneous DNA assay.

SD-TCR Process Could Bring DNA Diagnostic Testing into the Clinical Laboratory

Navix expected that its SD-TCR process would allow the rapid identification of those DNA sequences whose presence is correlated with the potential for genetic diseases, likelihood of future cancers, or other genetic sequences that do not cause disease, but might predispose individuals to illness. Navix scientists were aware that most of the costs for DNA diagnostics came from the use of separate equipment for identification, amplification, and signal generation. Therefore, the scientists proposed to conduct the reaction in one container.

The proposed reaction vessel would have all the necessary components to conduct the analysis so that, once sample double-stranded DNA was added to the reaction chamber, no further manual or automated additions or manipulations would be required. If any of the DNA sequences associated with disease were present, a cascade of chemical reactions would occur,

causing specific reactions that would lead to color-coded indicators that laboratory equipment could read and analyze. Moreover, they proposed a system that would be compatible with the clinical detection computer systems on the market in the mid-1990s so that diagnostic laboratories would not incur high costs to incorporate the Navix system into their diagnostics offerings.

Navix Seeks Funding for Concentrated, Focused Research

At the time of Navix's 1995 proposal, no other companies were working on projects similar to the SD-TCR chamber. ATP funding became necessary when Navix's internal corporate resources proved inadequate to fund such high-risk research. Without ATP's funding, the research would have taken significantly longer, because Navix and Profile Diagnostics would not have been able to devote eight scientists to concurrent, full-time research. ATP awarded Navix $1.97 million to conduct parallel, focused research with Profile Diagnostics over a two-year period.

Technical and Business Problems Forestall Success

Navix and Profile Diagnostics hoped their ATP-funded project would achieve the following milestones:

- A patent for a DNA amplification process within one year of ATP funding
- A patent for a DNA detection process within one year of ATP funding
- Clinical trials within 15 months of ATP funding
- FDA application submission within two years of ATP funding
- First sales within three years of ATP funding

Navix and Profile Diagnostics scientists succeeded in developing a two-stage reaction for DNA identification and amplification. During stage one, the target nucleic acid groups from the sample double-stranded DNA created bonds with probes in the reaction chamber containing nucleic acids indicative of disease. This released another series of molecules into the assay. During stage two of the reaction, specific sites within the reaction chamber bound with the molecules released from the first stage of the reaction. If future technology developed as expected, the enzyme on the released DNA strand would activate a second enzyme to catalyze a chemical reaction to convert a colorless substrate into a colored product that existing laboratory equipment could analyze. This technology led to Navix receiving a U.S. patent in 1999.

The SD-TCR process would dramatically reduce the number of required steps for typical DNA diagnostics to just one.

Technical problems with the stability of the processes within the reaction chamber as well as business issues delayed the project. By the end of the second year of the project, Navix had filed amplification and detection patents. The U.S. Patent and Trademark Office awarded the amplification patent. Navix then provided an additional $1 million in funds to continue the research. ATP extended the project by an additional five months to allow for further research.

At the end of February 1998, Navix's ATP-funded project concluded; however, the company had fallen short of their goals. Later in 1998, Navix merged with Medical Analysis Systems. After analyzing the market following the merger, Medical Analysis Systems decided against further research or efforts to commercialize the ATP-funded technology because other products had beaten Medical Analysis Systems to market.

By early 2003, Medical Analysis Systems closed due to business reasons unrelated to the ATP-funded project. The company could not remain solvent with the products it offered. The company earned no revenues from the ATP-funded project and commercialized no technology.

Conclusion

Navix and subcontractor Profile Diagnostics submitted a proposal to create a one-step DNA diagnostic reaction chamber that would reduce the per-test cost from several hundred dollars to less than $1. After achieving initial technical success, business and subsequent technical issues prevented Navix from reaching its ultimate goal of commercializing a one-step DNA diagnostic system. After a 1998 merger with Medical Analysis Systems, the company halted further research into its Self-Detected Target-Cycling Reaction process. Five years later, the company went out of business.

Navix scientists believed that the materials in a DNA diagnostics laboratory could be integrated through two technically risky and complex steps. First, the scientists would have to compartmentalize and optimize the DNA identification and detection functions into different areas within the reaction chamber without risking contamination. Second, they would have to integrate the three different reactions so that each stage would be compatible with the others and there would be no possibility of premature signal color development that would lead to incorrect results. Navix proposed that they could tightly integrate these reactions in order to achieve a single-step, homogeneous DNA assay.

PROJECT HIGHLIGHTS
Medical Analysis Systems (formerly Navix, Inc.)

Project Title: Rapid, Low-Cost DNA Diagnostic Technology (DNA Diagnostics Using Self-Detected Target-Cycling Reaction [SD-TCR])

Project: To develop a rapid, low-cost DNA diagnostic system that detects DNA sequences associated with disease and automatically triggers complementary cascade reactions for DNA amplification and signal generation using current clinical laboratory instruments.

Duration: 9/1/1995-2/28/1998
ATP Number: 95-08-0017

Funding (in thousands):

ATP Final Cost	$1,973	69%
Participant Final Cost	895	31%
Total	$2,868	

Accomplishments: Although no products resulted and the company did not completely meet their technical goals, Navix and subcontractor Profile Diagnostics pursued aggressive parallel research and created a two-step process to identify areas of DNA that correlate with disease. The effort led to two patent applications, one of which was granted in 1999:

- "Homogeneous diagnostic assay method utilizing simultaneous target and signal amplification" (No. 5,858,665: filed July 25, 1996, granted January 12, 1999)

Commercialization Status: Navix did not commercialize any products from its ATP-funded research. Business issues delayed research long enough for another competitor to beat Navix to the market. After Navix's merger with Medical Analysis Systems in 1998, the new company decided against pursuing further research or commercializing the reaction chamber product. The new company went out of business in 2003.

Outlook: Because Medical Analysis Systems failed, and other products on the market have surpassed the company's patented technology, the outlook for this technology is poor.

Composite Performance Score: No Stars

Number of Employees: 8 at project start; 0 as of September 2003 (the company has gone out of business).

Focused Program: Tools for DNA Diagnostics, 1995

Company:
Medical Analysis Systems (formerly Navix, Inc.)
(Medical Analysis Systems is no longer in business)

Contact: No available contact

Research and data for Status Report 95-08-0017 were collected during August - October 2003.

Monsanto Company (formerly Agracetus, Inc.)

Biopolymers to Give Cotton Fibers Synthetic-Like Qualities

In 1994, researchers in the U.S. cotton industry believed that genetic engineering was the key to improving cotton fibers. For more than 100 years, U.S. scientists have used breeding techniques and biological innovations to improve the quality of the domestic cotton crop. By the early 1990s, however, advances in chemistry and manufacturing techniques enabled synthetic fibers to surpass cotton in characteristics such as strength, wrinkle resistance, and ability to bind with dyes. Key players in the U.S. cotton industry knew that, in order to compete with largely foreign-made synthetic fabrics, they would have to make significant research advances within a short period of time. The potential to lose cotton sales to synthetic fabrics was projected at more than $200 million annually until at least 2005, given normal market growth.

One major focus of this research was to incorporate biopolymers into cotton plants. As of 1994, there were no viable methods for inserting these biologically derived substances into the cotton in a way that would allow the fiber to take on synthetic characteristics while retaining its natural ability to feel cool in the summer and warm in the winter. If biopolymers could be inserted successfully, however, cotton plants would gain the favorable qualities of synthetic materials without losing the qualities of pure cotton. With cost-shared funding from the Advanced Technology Program (ATP), Agracetus, Inc., pursued an aggressive research project from 1995 to 1998 to incorporate biopolymers into cotton fiber. By the end of the ATP-funded project, the research had advanced the state of knowledge of biopolymers several years ahead of where it would have been otherwise. Despite the added knowledge, Agracetus researchers were unable to reach a high enough concentration of the biopolymer to develop a competitive fiber.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)

No Stars

Research and data for Status Report 94-01-0074 were collected during December 2002 - January 2003.

Cotton Losing Its Competitive Advantage

Though U.S.-grown cotton was the premier natural textile fiber as of 1994, the industry faced substantial challenges from synthetic fabrics and commodity pricing; the cotton industry simply could not earn a profit because of price competition and substitute goods. The synthetic fabric industry, through innovative chemistry and manufacturing processes used primarily in laboratories and plants overseas, created man-made fibers that had many qualities that made them superior to cotton. This forced the cotton industry to compete by trying to reduce costs. Inefficiencies in production, however, made cotton more expensive than necessary. For example, textile producers were forced to blend polyester into their fabrics solely to increase fabric strength. Furthermore, they had to use far more cotton than otherwise would be necessary in order to compensate for shrinkage. These processes generated additional costs that reduced the U.S. cotton industry's profit.

If less favorable properties such as shrinkage could be cost-effectively reduced, however, cotton could regain its competitive advantage. For example, in fleece products alone, 25 percent less cotton per garment could be used. Consequently, the industry spent significant time and money researching ways to

incorporate synthetic fabric properties into cotton plants. As of 1994, however, there were no promising methods of eliminating cotton's unfavorable characteristics.

Synthetic Properties Could Be Introduced Into Pure Cotton

In 1994, the Middleton, Wisconsin-based agricultural products company, Agracetus, showed through initial research that it was theoretically possible to genetically engineer a set of traits into cotton plants that could revolutionize cotton production, capture additional global market share, and lift prices above commodity levels. If the company could translate the biotechnical theory into practice within a prototype cotton plant, the result could be a commercial crop with greater fiber strength, reduced shrinkage, enhanced wrinkle-resistance, better thermal properties (such as an ability to breathe in the summer and retain heat in the winter), and superior dye binding.

Agracetus focused on the genetic engineering steps necessary to modify cotton fiber.

If successful, Agracetus envisioned a new generation of cotton created exclusively by U.S. companies that would increase export opportunities for cotton farmers, mills, and textile manufacturers. Initial projections showed potential sales of more than $1 billion between 2000 and 2005, assuming technical success and rapid market penetration.

Agracetus Seeks Funding for Genetic Engineering Research

Agracetus' strategic plan required that the company devote nearly all its available resources to cotton fiber products in order to develop a first-generation product known as Fiber #8802. The company had enough resources to fund the necessary studies on gene identification in cotton fibers and plant transformation through gene insertion. Ideally, Agracetus executives wanted Fiber #8802 to have additional properties. Specifically, the fiber should have the correct number and types of genes to improve the cotton's dyeability, dimensional stability (reduced incidences of wrinkling and shrinking), heat-withstanding ability, and stain-repelling properties. All of these qualities were necessary for the commercial acceptance of a premium product.

The technical risk involved in incorporating these characteristics into Fiber #8802, however, was too high for Agracetus to undertake. Company researchers felt strongly that research was necessary in order to quickly advance the cotton industry to the point where it could capture additional global market share. The researchers received management's approval to search for additional funding to enable this research. Agracetus approached trade groups within the U.S. cotton industry for resources, but because of the depressed profitability of the industry, funds were not available for a project with such a high technical risk.

After failing to obtain funds from other sources, Agracetus submitted a proposal to ATP and was awarded $1 million to carry out their genetic engineering research. This research was essential to get the leaders in the industry to focus on the technical roadblocks to biopolymer development that had hindered commercialization for years.

Ambitious Research Goals Require Collaboration

During the ATP-funded project, Agracetus focused on the genetic engineering steps necessary to modify cotton fiber. Internal research completed prior to the start of this project suggested that two bacterial genes were necessary for cotton to form polyester-like compounds such as poly-3-hydroxybuteric acid (PHB). The primary technical goal of the project was to get PHB levels in cotton up to the point where the fiber took on the favorable qualities of synthetics while retaining the superior thermal characteristics of pure cotton.

Agracetus envisioned a new generation of cotton created exclusively by U.S. companies.

In order to test whether PHB levels resulted in the appropriate mix of cotton and synthetic traits, Agracetus relied on tests from the Cotton Incorporated Research Center in Raleigh, North Carolina; the Starlab Specialty

Testing and Research Laboratory in Knoxville, Tennessee; and the International Textile Center of Texas Tech University in Lubbock, Texas.

Biopolymer Concentration in Prototype too Low for Commercialization

By the end of the ATP-funded project in 1998, Agracetus had produced a prototype plant and had it tested by the three research laboratories. Agracetus achieved a major technical milestone by developing a prototype plant with elevated levels of PHB. Initial testing showed that, while the fiber had many of the necessary properties for commercial applications, a major problem remained. In order to achieve the desired synthetic-like properties within the cotton fibers, the biopolymer added to the cotton plant would have to be increased five- to tenfold. Research conducted during the course of the ATP project showed that this increase would be extraordinarily difficult to achieve because when the PHB levels increased that significantly, other favorable traits were "crowded out," reducing the plant's commercial viability.

Given the continued risk and relatively small likelihood of success, Agracetus executives decided not to conduct additional research and development after the end of the project. Although no products resulted from this project, Agracetus' ATP-funded research accelerated the pace of research in biopolymers at a crucial time when the industry faced significant pressure from overseas competitors.

Agracetus was acquired by Monsanto Company in 1996. During the following year, corporate priorities shifted, and no further research or development of Agracetus' proposed technology has occurred since the end of the grant.

Agracetus Shares Project Knowledge to Help U.S. Cotton Industry

Agracetus researchers took a number of steps to publicize the results of their ATP-funded research in order to assist the U.S. cotton industry. Agracetus scientists wrote articles on the technology that appeared in professional journals, company representatives delivered presentations at conferences, and the popular press reported on the technology several times. Even though Agracetus is not pursuing further commercialization, sequencing the cotton genome is the subject of a number of domestic and international research projects.

Conclusion

In 1995, with the help of ATP funding, Agracetus started an aggressive research plan to determine if it was possible to incorporate the most beneficial properties of synthetic fabric into natural cotton through genetic engineering. The results of a successful project could have increased U.S. market share and annual sales by more than $200 million. By 1998, when the project ended, Agracetus researchers were able to use genetic engineering to introduce a biopolymer into cotton plants. The biopolymer concentration, however, would have to be increased 5 to 10 times the achieved level in order to be commercially viable. However, the company disseminated its knowledge from the project throughout the industry through professional publications, the popular press, and company presentations.

PROJECT HIGHLIGHTS
Monsanto Company (formerly Agracetus, Inc.)

Project Title: Biopolymers to Give Cotton Fibers Synthetic-Like Qualities (Transgenic Cotton Fiber with Polyester Qualities Via Biopolymer Genes)

Project: To develop a genetically engineered version of cotton that outperforms standard cotton fibers in such properties as dyeability, strength, resistance to wrinkling, and shrinkage.

Duration: 2/1/1995-1/31/1998
ATP Number: 94-01-0074

Funding (in thousands):**

ATP Final Cost	$1,019	87%
Participant Final Cost	149	13%
Total	$1,168	

Accomplishments: Through its aggressive research plan, Agracetus advanced the industry's knowledge of biopolymers in cotton by two years. In addition, the company produced a prototype plant with elevated levels of poly-3-hydroxybuteric acid (PHB). Although the PHB concentration was not high enough for commercialization, simply raising the PHB level at all represented a technical achievement.

Agracetus disseminated its project knowledge through a variety of media. Company employees published papers in 10 professional journals and publications. Agracetus representatives gave presentations at five conferences in the United States and around the world. The popular press, which included *Knight Ridder*, *Associated Press, The Wall Street Journal*, and *USA Today*, also reported on Agracetus' project more than 20 times.

Commercialization Status: Due to the difficulty in attaining high enough PHB levels in the cotton fibers without "crowding out" the fibers' favorable traits, no commercialization efforts resulted from this ATP-funded research.

Outlook: Because no products will result from this project unless another company begins research after the cotton genome is sequenced, the outlook for the technology is poor.

Composite Performance Score: No Stars

Company:
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** As of December 9, 1997, large single applicant firms are required to pay 60% of all ATP project costs. Prior to this date, single applicant firms, regardless of size, were required to pay indirect costs.

Research and data for Status Report 94-01-0074 were collected during December 2002 - January 2003.

Orchid BioSciences (formerly Molecular Tool, Inc.)

Integrated Microfabricated Devices for DNA Typing

Genetic analysis is the study of DNA to determine information relating to identity or disease. Studying the subtle changes in genes when a cell becomes diseased offers opportunities to search for new molecular targets for drugs. Genetic analysis promises to provide more accurate diagnoses and customized drug therapies. In 1994, genetic analysis was cumbersome and expensive, with each test costing approximately $100. Molecular Tool, Inc. was an innovative start-up company with a desire to improve genetic analytical testing for human health. The company had already developed practical applications for thoroughbred horse genetics and wanted to apply improved techniques for human genetic analysis. Molecular Tool intended to automate and simplify analyses by creating a miniature "lab on a chip." If successful, they could reduce cost and space requirements from a manually operated 20-foot x 15-foot laboratory, down to an automated 1-square-inch glass chip. Industry analysts predicted that affordable genetic analyses could be performed on desktop systems, but technical risks included placing microscopic samples on the chip, taking images of the samples, and analyzing those images. Molecular Tool applied for funding from the Advanced Technology Program (ATP) under the "Tools for DNA Diagnostics" focused program in 1994.

ATP awarded cost-shared funding for a three-year project, beginning in 1995. Molecular Tool successfully developed a patented prototype single nucleotide polymorphism (SNP) analysis tool in 1998. (SNPs, or "snips" are useful genetic markers or places in the genetic code that detect minute variations in the DNA sequence.) Orchid BioComputer (later renamed Orchid BioSciences) purchased Molecular Tool that year and acquired the ATP-funded lab on a chip technology. Following further development, Orchid reduced the cost of a typical DNA analysis by approximately 70 percent and increased accuracy to 1 in several billions statistical probability from 1 in a million. The company continued enhancing the technology and commercialized its SNPstream Genotyping System, as well as providing SNP analyses on a fee-for-service basis by 2001. Orchid acquired three identity genomics testing competitors and became a leading provider for the forensic and paternity DNA testing markets. In 2002, the company decided to focus on its DNA analysis services business and sold its SNP genotyping business, which included an exclusive license to Orchid's SNP analysis technology, to Beckman Coulter, a leading provider of tools for clinical laboratories. Beckman continues to market and improve the SNPstream Genotyping System, based on ATP-funded technology, while Orchid focuses its efforts on fee-for-service genetic analyses. Orchid had sales in excess of $62 million in 2004. The global DNA diagnostics industry is expected to grow from $1 billion in 2003 to $6 billion by 2010.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* * * *

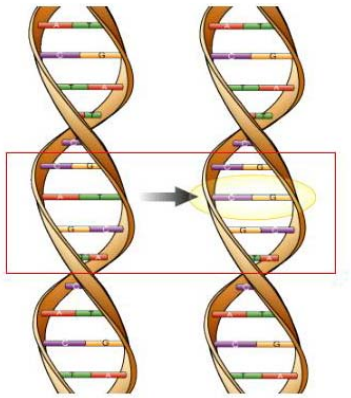
Research and data for Status Report 94-05-0034 were collected during June – August 2004.

Genetic Analysis Is Cumbersome and Costly

In the early 1990s, Molecular Tool, Inc. was a small Maryland biotechnology company of highly skilled microbiologists that wanted to develop advanced genetic applications. Located near a racetrack, Molecular Tool received its first commission to perform advanced genetic testing for thoroughbred racehorses. The company had developed a proprietary genotyping biochemistry, which it called Genetic Bit Analysis (GBA)

to identify and analyze variations in the individual bases of DNA. Molecular Tool used GBA to verify horse parentage and, in 1994, generated more than 500,000 individual horse genotypes.

In DNA sequencing, the scientist looks at a long string of DNA and identifies fragments that determine identity or information relating to disease. The most common type of DNA sequence variation is the single nucleotide polymorphism (SNP, or “snip”), a place in the genetic code where DNA differs from one person to the next by a single letter (see illustration below). The human genome contains more than 2 million SNPs. The genetic code is specified by the four nucleotide "letters": A (adenine), C (cytosine), T (thymine), and G (guanine). A SNP occurs when a single nucleotide (A, T, C, or G) in the genome sequence is mismatched. The analyst unzips the DNA sample and looks at one side. For example, in the illustration below, the left side of the first highlighted DNA sequence shows C**A**GT. The second SNP variation shows C**C**GT. For a variation to be considered a SNP, it must occur in at least 1 percent of the population. For example, a place in the genome where 93 percent of the population has a T and the remaining 7 percent has an A is a polymorphism. SNP scoring (analysis) is simpler than complete DNA sequencing.



Example of a SNP variation. An individual DNA sequence varies from the general population by one nucleotide. Small changes in DNA sequence such as these can affect drug response and disease susceptibility. Reproduced with permission from BioTeach http://bioteach.ubc.ca.

Certain SNPs may predispose some people to a particular disease, and this may explain why some respond better to certain drugs. When a SNP occurs, the gene’s function may change; for example, it may cause the individual to develop a bacterial resistance to antibiotics. SNPs can identify differences in drug metabolism between individuals; they can pinpoint which patients may not respond to a drug or may suffer an adverse drug response. Furthermore, SNPs can also be used in genetic diagnosis to uniquely identify individuals; for example, to link a defendant to the crime scene in a criminal case. Analysts predicted that genetic analyses could some day be performed by affordable desktop systems in clinics and physicians’ offices. These analyses would make more accurate customized diagnoses and even predict disease to facilitate preventive medicine.

Technology available in 1994 required several hundred to several thousand test-tube tests to decode essential elements among the 3 billion bits of information that make up the human genetic code. Performing 1,000 genetic tests to uniquely identify one human sample (or to test for disease) required at least two lab technicians, a 20-foot by 15-foot laboratory, several machines to perform rote tasks, and 12 hours of time, at a cost of $100 or more per test.

Molecular Tool sought opportunities to advance DNA testing for human healthcare by optimizing and dramatically miniaturizing existing tools. The company aimed to identify the top 1,000 human SNPs that contribute to the lack of efficacy in drugs, adverse side effects, and predisposition to disease.

Molecular Tool Proposes to Miniaturize Biotechnology

Reducing the size of the lab components to fit onto a chip would lower cost through reduced chemical use, increased automation, and greater efficiency. Molecular Tool’s goal was to compress most of the functions of SNP analysis that were being done in the 20-foot by 15-foot biotechnology laboratory onto a 1-square-inch glass chip. In the same way that a computer chip manipulates electrons through microscopic wires, biotechnology chips would manipulate fluids along microscopic channels in order to search the DNA strands for SNP variations. A subcontractor, Naval Research Laboratories, would assist with the microfluidics technology. They would develop stamping methods to create patterned DNA surfaces and a film-patterning approach to validate Molecular Tool’s optical-scanning system.

Molecular Tool submitted a proposal to ATP in 1994 under the focused program, “Tools for DNA

Diagnostics," and was awarded cost-shared funding for a three-year project beginning in 1995. If successful, Molecular Tool hoped that the miniaturized tool would make comprehensive DNA diagnostics routinely available at a low cost. The new tool would improve genotype processing power more than a hundredfold; moreover, dramatic reductions in cost would make DNA testing more accessible. This, in turn, could support preventive medicine by providing valuable information to people who are genetically predisposed to certain diseases. Preventive disease management could reduce healthcare costs over time.

Molecular Tool faced significant technical risks. Liquids behave very differently in minute quantities; activities such as heating, mixing, and separating fluids would be unpredictable. Challenges included developing the necessary techniques for micromachining and for handling fluids on a microscopic scale. One of the key challenges was to find ways to move tiny amounts of fluid through channels etched on the glass chip. Although primitive versions of analytical devices had been constructed on microchips, integrated microinstrumentation was a new field. Multiple processing steps had never been integrated on a single chip. In addition, when moving strands of DNA, another challenge occurs: the oligonucleotides (short single strands of DNA) might not behave as expected due to chemical sensitivity. Molecular Tool focused their research on two principal objectives: first, to develop a microanalytic device to perform SNP analysis on single genetic bits; and second, to demonstrate the feasibility of creating a microdevice capable of analyzing 100 SNPs.

Miniaturized and Automated Instruments to Speed Processing

Molecular Tool encountered several technical challenges in developing their strategy of miniaturization and integrated automation, in which they would adapt a proven genotyping biochemistry to micromachined instruments and microfluidic processing. Their technical challenges included the following:

- Reduce labor requirements by achieving higher throughput (larger sample quantities)
- Reduce the cost of biochemicals by processing smaller samples and reducing reagent consumption
- Reduce processing complexity by miniaturizing and combining equipment

A carefully prepared sample, such as drops of blood, would be put on a glass chip, where channels etched on the chip would direct the liquid in a manner similar to circuits directing electricity in computers. As the blood passed through different routes, reagent chemicals would extract genetic material and, through a chemical process known as amplification, would make many copies of the material so multiple genetic tests could be performed. Various reagent chemicals applied to the chip would react with the genetic material in the sample. A laser would then be used to detect which chemicals had been absorbed, and advanced software would be used to interpret the chip data results. From this analysis, scientists could determine the genetic "fingerprint" of the sample. In another application of the technology, a doctor could determine if the patient carried genes linked to a variety of illnesses.

Molecular Tool's goal was to compress most of the functions of single nucleotide polymorphisms analysis that were being done in the 20-foot by 15-foot biotechnology laboratory onto a 1-square-inch glass chip.

Molecular Tool's ultimate goal was to reduce the size of genotyping tools 1,000-fold over their existing tools. Moving tiny amounts of sample material short distances in minute channels would result in deep reductions in reagent consumption.

Molecular Tool's proposed system had three main components:

- Micromachined analytical instruments in glass (the chip) would be built with photolithographic methods (photographic process used to transfer circuit patterns) on a sub-millimeter scale
- Electro-osmotic pumping (moving liquid through a semi-permeable membrane) would precisely measure nanoliter volumes of sample material (a nanoliter is one-billionth of a liter), mix reagents, and allow the material to interact with sensitive fluorescence detection systems

- Fluorescent dye would attach to specific cells, and researchers would measure the fluorescent signal at specific probe locations to detect the genes

Molecular Tool's plan was to reduce the size of sample testing sectors, or wells, from 1 square centimeter down to less than 1 square millimeter. After developing an early prototype, the company would integrate the tools, automate them, and increase throughput. Ultimately, their goal was to shrink the entire genotyping lab to the size of a 1-square-inch microchip.

ATP-Funded Project Achieves All Technical Goals

At the conclusion of the project in 1998, Molecular Tool had successfully converted existing SNP analysis technologies into a miniaturized format to create a high-throughput and economical DNA analysis of disease states and forensic identification (DNA fingerprinting). The company fabricated DNA microarrays, "printing" an orderly arrangement of DNA fragments that represented the genes onto the glass slide for analysis. Each DNA fragment representing a gene was assigned a specific location on the array. DNA or RNA in the overlaid sample attach (through a process called hybridization) to a complementary spot on the array; that is, Gene-A will stick to a spot composed of a Gene-A fragment. Molecular Tool developed high-resolution optics in order to see patterns in tiny sizes. They scanned the fluorescent-labeled (hybridized) microarrays, looking for SNP variations, and relied on robotics to increase analysis speed. Each glass chip had an array with 384 wells, which was an increase from 96 wells at the start of the project. Molecular Tool achieved 100 percent of their technical goals and developed a working prototype tool to identify and validate SNPs, sometimes referred to as SNP scoring. They were awarded five patents for their advances and published their findings in academic journals. ATP funding was critical to developing this SNP scoring technology.

Technical Success Gains Commercial Attention

In 1998, Molecular Tool was purchased by Orchid BioComputer, located in Princeton, NJ. The strategy underlying the acquisition was the combination of Orchid's high-throughput microfluidics technology with Molecular Tool's proprietary SNP analytic chemistry and GBA capabilities to create even faster, more flexible tools to analyze correlations between SNPs and specific genotypes, diseases, and therapeutics. Commenting on the Molecular Tool acquisition in 1998, Dale Pfost, Ph.D., Orchid's then-CEO, said, "The genetic variations expressed in SNPs are the foundation of pharmacogenomics and pharmacogenetics. The pharmaceutical industry has expressed great interest in targeting subgroups of patients based on their individual genetic differences to improve therapy through the use of existing drugs and the development of new drugs that have greater efficiency and fewer side effects than those available today. Companies can use the [SNP] platform as a rapid and cost-effective way to assess disease correlation and … responses to different drugs."

Orchid's efforts to combine Molecular Tool's analysis technology with its microfluidics platform enabled Orchid to focus on developing automated high-throughput instrument systems, including the automation of sample preparation. Orchid later added a microscope with a digital camera to the SNP analysis tools to more easily read results.

Orchid Commercializes SNP Technology

In 1999, a group of pharmaceutical and biotechnology companies formed a consortium, called The SNP Consortium Ltd. Together the companies invested $45 million, with the goal to produce a map of the human genome. At the same time, Orchid opened a new SNP genotyping facility to house labs devoted to microfluidic chips, chemistry, and optics. The consortium contracted with Orchid to use this facility to identify up to 300,000 SNPs and to "map" at least 150,000, or determine their location within the human genome; the consortium would provide this information to the public free of charge. Furthermore, the consortium wanted Orchid to test identified genetic markers. Orchid used its SNP genotyping tools and GBA software developed during this project to perform the genotyping assays.

Medical Groups Use SNP Technology to Study Disease and Treatment

Also in 1999, Orchid began scoring SNPs for medical applications. The company joined with the University of Washington's School of Medicine to perform high-

throughput SNP genotyping to study genetic variability, its relationship to the onset of disease, and pharmaceuticals. Moreover, Orchid's genotyping was being used at the Mayo Clinic to determine whether patients under- or over-metabolize drugs so that drug dosage could be more carefully tailored to the patient.

SNPstream Product Enters the Market

In cooperation with Luminex Corp., Orchid used the ATP-funded technology to develop SNPstream, an affordable, rapid-throughput system for performing SNP scoring. The system combined Luminex's LabMAP system with Orchid's proprietary line of SNPware reagent kits. In 1999, the SNPstream system was able to score thousands of SNPs per day.

By the end of 2000, Orchid could score a million SNPs per day, with the goal of finding and patenting clinically important relationships between SNPs and pharmaceuticals. The ultimate goal was to improve drug treatment options to fight disease. The company offered a successful initial public offering that year and changed its name to Orchid BioSciences.

"The genetic variations expressed in SNPs are the foundation of pharmacogenomics and pharmacogenetics."

In 2001, Orchid entered the market with its SNPstream ultra-high throughput, array-based genotyping system (see illustration), which was based on the ATP-funded technology. The system was capable of handling more than 800,000 genotypes per day. SNPstream analyzes up to 100,000 data points for increased accuracy (in 1994, Molecular Tool had been testing only 1,000 data points). Furthermore, a typical result showed 1 in several billions statistical probability, increased from 1 in a million. Total cost for the processing of sample of DNA had been reduced by approximately 70 percent, and labor involved in processing had been reduced by approximately 75 percent. The entire process of DNA testing which previously took up to 4 weeks, could now be completed in about a week. At the same time, the company began to conduct genetic analysis on a fee-for-service basis for the biotechnology and pharmaceutical companies, as well as academic laboratories.

A technician reads results from the automated, scalable SNPstream genotyping system, which includes a robotic analysis instrument and computer readout.

Orchid Expands and Licenses Its Technology

Orchid's success with SNP technology allowed the company to expand and license its technology to other laboratories, which could adapt the technology to their instrument platform. By 2001, Orchid acquired three more companies that provided compatible identity genomics testing: GeneScreen, Inc., Lifecodes Corp., and Cellmark Diagnostics (a subsidiary of AstraZeneca, which had pioneered the introduction of DNA testing for paternity and forensic analyses in the United Kingdom). Orchid granted several SNP-related non-exclusive licenses to use its proprietary SNP technology to Applied Biosystems, Amersham Pharmacia Biotech, and Quest Diagnostics, as well as an exclusive license to PerkinElmer to use Orchid's SNP technology to perform fluorescence-based analyses.

Orchid Refocuses on DNA Services

By the end of 2002, four years after the ATP-funded project ended, Orchid was concentrating on its highly successful DNA services business and had moved away from product development, customization, and sales. Beckman Coulter, a leading provider of instrument systems for life sciences and clinical laboratories, purchased the instrumentation piece of Orchid's business, to continue to develop and offer SNP scoring systems and consumables to the research market. Beckman began producing and selling reagent kits, software, and systems globally. Beckman, by virtue of its exclusive license from Orchid, incorporated Orchid's proprietary SNP-identification technology

for performing SNP analyses on DNA sequencers, microarray plates, and flow cytometers. "Our collaborative efforts with Orchid enhance our offerings to important segments of the marketplace and are a key strategic element in our genomics program," said George Bers, President of Life Science Research for Beckman Coulter.

Orchid continues to be a leading provider of identity genetics services for the forensic and paternity DNA testing markets. Their work includes parentage testing to aid child support enforcement agencies, criminal casework analysis, DNA testing for convicted offender DNA databases, casework for law enforcement laboratories, the identification of victims of accidents, and consulting services for attorneys. Orchid's SNP technology was used in collaboration with the New York Office of the Medical Examiner in an attempt to identify the large number of World Trade Center victims for whom conventional identification methods had failed. Orchid has forensic contracts with major metropolitan police departments, such as Los Angeles and Houston, as well as with London's Scotland Yard. In 2003, the Federal Bureau of Investigation awarded Orchid two contracts to develop SNP technology for advanced forensic applications to identify individuals using degraded DNA samples. Orchid also offers a premium "DNA Express Service" to help law enforcement agencies analyze backlogs of DNA evidence from unsolved crimes. DNA Express provides forensic DNA analyses in five business days compared with the standard four to five weeks.

Orchid continues to develop new applications for uses of its SNP technology, as it has to conduct global disease testing programs. For example, in 2002, Orchid was awarded a contract by the Department of Environment and Rural Affairs in the United Kingdom to help selectively breed disease out of the sheep population. The company processes samples for the United Kingdom's pioneering sheep genotyping program to help farmers use selective breeding to eliminate the disease scrapie from their flocks. Scrapie is a progressive brain disease in sheep that causes itching so intense that the animals scrape off their wool. In 2003, Orchid analyzed nearly 500,000 samples for scrapie susceptibility and has genotyped more than 1.4 million sheep to date. Orchid's contracts in forensic, paternity, and animal testing have led to financial success for the company. Identity genetics accounted for 97 percent of Orchid's 2003 revenues, or $49 million. Total sales grew 23.5 percent in 2004 to $62.5 million.

Genomics Is Growing Globally

Genomics is still an emerging technology growth area. Relying on the original ATP-funded technology concepts, Orchid BioSciences and Beckman Coulter are well positioned for continuing expansion in the diagnostic tool industry. Global molecular diagnostics spending has risen from $78 million in 1994 to $1 billion in 2003, and it is expected to reach $6 billion by 2010. Genomics research spending is expected to increase from $4.5 billion in 2000 to $7 billion in 2010 (Andrew Broderick, Genomics, SRI Business Intelligence, 2003, p. 41, 48).

Conclusion

As a result of funding provided by ATP, Molecular Tool, Inc. successfully developed a prototype lab-on-a-chip single nucleotide polymorphism (SNP) analysis tool for DNA testing. They were able to shrink the space requirement for DNA analyses from a 15- x 20-foot laboratory to a 1-square-inch chip. In addition, they achieved comparable reductions in labor, time, and chemical reagent use. The company was awarded five patents for its technology advances and published its findings. When Orchid BioSciences bought the company and its ATP-funded SNP analysis system in 1998, it continued development. Following project conclusion in 1998, Orchid built a large SNP scoring laboratory in 1999 and began analyzing large numbers of samples. The company began marketing its SNPstream analysis system in 2001. In 2002, Orchid sold the instrumentation portion of this business to Beckman Coulter, a leading instrument manufacturer in the biotechnology industry. As of 2004, Beckman continues developing and marketing SNPstream, based on ATP-funded technology, which is capable of performing more than 800,000 genotypes per day. Orchid, which provides DNA analyses to multiple markets, continues to find new uses for its SNP detection technology, and most recently earned $62.5 million in sales in 2004.

PROJECT HIGHLIGHTS
Orchid BioSciences (formerly Molecular Tool, Inc.)

Project Title: Integrated Microfabricated Devices for DNA Typing

Project: To scale the size of the company's state-of-the-art Genetic Bit Analysis (GBA) technology down by a factor of 1,000, developing the necessary techniques for micromachining and for handling fluids on a microscopic scale to make a simple, compact DNA typing instrument.

Duration: 2/15/1995 – 2/14/1998
ATP Number: 94-05-0034

Funding (in thousands):

ATP Final Cost	$1,940	74%
Participant Final Cost	$ 684	26%
Total	$2,624	

Accomplishments: With ATP funding, Molecular Tool succeeded in developing a miniaturized DNA analysis instrument that reduced the cost and required laboratory space. The project met 100 percent of its goals. Accomplishments include:

- Molecular Tool developed a functioning prototype single nucleotide polymorphism (SNP) detection and analysis tool.
- The prototype device contained 384 reaction sectors, or wells, which was an increase from 96 wells at the start of the project in 1994.
- Orchid BioSciences purchased Molecular Tool in 1998 and integrated advanced microfluidics and optics, including a digital camera to improve analysis.
- Orchid's SNP analysis instrument, called SNPstream, performs more than 800,000 DNA analyses per day.

In addition, Orchid developed commercial applications for SNP technology:

- The SNP technology was used in identifying the remains of some New York City World Trade Center victims, which could not be identified by conventional DNA analysis due to sample degradation.
- Orchid has contracts with major metropolitan police departments for forensics, including Los Angeles, Houston, and London's Scotland Yard. They also developed advanced forensic applications to identify individuals from unsolved crimes using degraded DNA samples for the Federal Bureau of Investigation. Their DNA Express service provides forensic DNA analyses in five business days compared with the standard four to five weeks.
- Orchid collaborated with the University of Washington's School of Medicine to study genetic variations, the onset of disease, and pharmaceuticals.
- Orchid collaborated with the Mayo Clinic to tailor drug dosage based on whether patients under- or over-metabolize drugs. This is just the beginning of such improvements in medical treatment based on pharmacogenetics.
- Orchid analyzed SNPs for the United Kingdom's scrapie genotyping program to help sheep farmers use selective breeding to eliminate the disease scrapie from their flocks. The company has genotyped nearly 1 million sheep to date.

Molecular Tool filed six patent applications from this ATP-funded project, of which the following five were awarded:

- "Covalent attachment of nucleic acid molecules onto solid-phases via disulfide bonds"
  (No. 5,837,860: filed March 5, 1997; granted November 17, 1998)
- "Attachment of unmodified nucleic acids to silanized solid phase surfaces"
  (No. 5,919,626: filed June 6, 1997; granted July 6, 1999)
- "Covalent attachment of nucleic acid molecules onto solid-phases via disulfide bonds"
  (No. 6,030,782: filed November 21, 1997; granted February 29, 2000)
- "De novo or 'universal' sequencing array"
  (No. 6,322,968: filed November 21, 1997; granted November 27, 2001)
- "Covalent attachment of unmodified nucleic acids to silanized solid phase surfaces"
  (No. 6,136,962: filed June 23, 1998; granted October 24, 2000)

PROJECT HIGHLIGHTS
Orchid BioSciences (formerly Molecular Tool, Inc.)

Commercialization Status: After acquiring Molecular Tool in 1998, Orchid BioSciences developed a thriving DNA analysis service business based on its SNPstream genotyping system and commercialized it in 2001. Beckman Coulter purchased the instrumentation from Orchid in 2002 and continues to sell the system. Orchid is a leading product and service provider of DNA forensic and paternity services, based on the ATP-funded SNPstream system:

- **Product**: SNPstream Ultra High Throughput (UHT) is an automated array-based genotyping tool. It entered the market through Orchid BioSciences in 2001. The business pertaining to the production of the SNPstream instruments and consumables was sold to Beckman Coulter in December 2002. Beckman also has an exclusive license from Orchid for the use of SNP identification technology for use with the system. As of 2004, Beckman continues to develop and enhance the system, marketing to research and clinical laboratories.

- **Service**: Orchid BioSciences provides genetic analyses using SNPstream UHT on a fee-for-service basis (for biotech companies, pharmaceutical companies, and criminal justice agencies). Orchid's facility was providing up to 1 million SNP scores per day by the end of 2000 on a fee-for-service basis. Sales exceeded $1 million the first year and exceeded $62 million in 2004.

Outlook: SNP scoring plays a key part in the evolving DNA diagnostics industry. The outlook for integrated microfabricated devices and services related to DNA typing is excellent. Uses include paternity testing, forensic testing, and pharmacogenetic testing. The global market reached $1 billion in 2003 and is projected to reach $6 billion by 2010. It is hoped that further testing will ultimately lead to customized drug treatments based on patients' unique DNA.

Composite Performance Score: * * * *

Number of Employees: 20 employees at project start, 60 as of February 1998 (at project end, Molecular Tool); 430 as of December 2003 (Orchid BioSciences).

Focused Program: Tools for DNA Diagnostics, 1994

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Subcontractor:
Naval Research Laboratory
Washington, D.C.

Publications: Molecular Tool and Orchid BioSciences shared knowledge through the following publications:

- Head, S. R., P. Parikh, Y.-H. Rogers, W. R. Bishai, P. Goelet, and M. T. Boyce-Jacino. "Nested Genetic Bit Analysis (N-GBA) for Mutation Detection in the p53 Tumor Suppressor Gene." *Nucleic Acids Research*, Vol. 25, No. 24: 5065-71, 1997.
- Head, S. R., P. Parikh, Y.-H. Rogers, W. R. Bishai, P. Goelet, and M. T. Boyce-Jacino. "Solid-Phase Sequence Scanning for Drug Resistance Detection in Tuberculosis." *Molecular and Cellular Probes*, 13: 81-87, 1999.
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- Pohl, M.G., L. Picoult-Newberg, R. Patch, and M. Boyce-Jacino. "Primer Design for High Throughput Primer Extension." *American Journal of Human Genetics* 67 (4) 2000: 273, Suppl. 2.
- Donaldson, M.A., J. Lathrop, C. A. Gelfand, N. Nouri, T. Ryder, and M. Boyce-Jacino. "The SNPcode 100 Genotyping Kit: A Generic System for SNP Genotyping on High Density Arrays." *American Journal of Human Genetics* 67 (4): 273-273, Suppl. 2, 2000.
- Swenson, R. E.; Q. Xu, C. Gelfand, M. Donaldson, and M. Boyce-Jacino. "Polymerase Signaling Assay for DNA Sequencing and DNA Polymorphism Analysis." *American Journal of Human Genetics* 67 (4): 273-273, Suppl. 2, 2000.

PROJECT HIGHLIGHTS
Orchid BioSciences (formerly Molecular Tool, Inc.)

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Research and data for Status Report 94-05-0034 were collected during June – August 2004.

Valentis, Inc. (formerly Progenitor, Inc., a subsidiary of Interneuron Pharmaceuticals)

Use of Gene Therapy to Treat Cardiovascular Disease

In 1994, Progenitor, Inc., received an award from the Advanced Technology Program (ATP) to develop transplantable endothelial cells from precursor stem cells, which could be genetically engineered or otherwise modified for specific medical purposes. The company believed that these endothelial-like cells could contribute to the development of new blood vessels and could be used to treat complications that often followed angioplasty. Early in their research, the company discovered that many of the endothelial cells that it was isolating expressed the Del-1 marker, an essential gene that regulates angiogenesis, the process by which new blood vessels are formed in adults. This discovery prompted the company to change its focus to this essential gene.

By the end of the project in 1998, Progenitor had developed an understanding of how the gene regulates angiogenesis and can be used to treat ischemia; it did not, however, complete the necessary research to commercialize the gene. Shortly thereafter, Progenitor went out of business, but in April 1999, Valentis, a biopharmaceutical company, acquired the rights to the specific gene and has continued to develop it for the treatment of heart disease and peripheral arterial disease. In July 2003, the company initiated Phase II of a clinical trial for the Del-1 gene.

COMPOSITE PERFORMANCE SCORE
(based on a four star rating)
* *

Research and data for Status Report 94-01-0301 were collected during March - July 2003.

Dysfunctional Endothelial Cells Thought to Lead to Vascular Disease

Endothelial cells regulate many critical physiological processes. These processes include local control of blood vessel tone, modulation of the immune response, and new blood vessel development. The malfunctioning of these cells is thought to play an important role in the onset of diseases involving blood vessels, blood flow, and arteries, such as hypertension, atherosclerosis (hardening of the arteries), and ischemia (heart attack).

In the 1990's, angioplasty (a procedure for alleviating blockage of an artery in which a balloon-tipped catheter is introduced into the artery at the point of obstruction and inflated to push the vessel open) and the replacement of vessels by grafting were the most advanced means of managing coronary heart disease; however, these procedures were limited in their effectiveness and often required follow-up procedures. Angioplasty had a relatively high incidence of restenosis, which was thought to be caused, in part, by the loss of functional endothelial cells during the procedure. Artificial devices were used to keep the blood vessels open, but the lack of efficient endothelial cell activity resulted in blood clotting or coagulation on the surface, often causing vascular obstruction and limiting the long-term usefulness of the prostheses.

Progenitor Proposes Cell Therapy Based on Endothelial Precursors

Progenitor had previously been successful in isolating cell lines with endothelial characteristics from the murine (mouse) yolk sac. It had also isolated a human yolk sac line with endothelial-like properties. The company wanted to develop the technology to use the yolk sac-derived endothelial cell lines to treat vascular disease.

The yolk sac is the first site for endothelial and blood cell development in mammals. Progenitor believed that the endothelial cell lines derived from the yolk sac might be accepted as tissue grafts more easily than other types of cell lines and could potentially contribute to the normal development of blood vessels. This would be useful in treating ischemia and restenosis following angioplasty. The cells could also reduce occurrences of restenosis by rapidly repairing the endothelial lining of blood vessels damaged by angioplasty. Cells from human yolk sacs could be obtained as by-products (i.e., exempted tissue donations) from normal full-term births.

Progenitor realized that developing this technology was a high-risk endeavor. Cellular therapy was a new field that was rapidly changing. However, if successful, Progenitor anticipated $400 million in sales in the U.S. with a potential increase to $1 billion per year if it could capture the angioplasty markets in Japan and Europe. (In 1995, there were approximately 40,000 angioplasties performed each year in Japan and 400,000 performed in Europe.) The company also believed that use of the new technology would result in an overall reduction in direct health care costs by $2 billion through more successful angioplasties and a corresponding decrease in repeat angioplasties or the alternative procedure, coronary artery bypass graft surgery, due to restenosis.

The company also believed that use of the new technology would result in an overall reduction in direct health care costs by $2 billion.

In order to bring the concept of cell therapy based on endothelial precursors to the regulatory review stage in time to remain competitive with other emerging technologies, Progenitor needed financial assistance. After seeking funding from other sources, the company submitted a proposal to ATP and received $2 million for a three-year project.

Progenitor Anticipates Broad-Based Benefits

Progenitor believed that the successful commercialization of its technology for using yolk sac-derived endothelial cell lines to treat vascular disease would positively impact U.S. economic growth and productivity in health care delivery. As there is no cure for cardiovascular diseases, a significant amount of U.S. government funds for health care is spent on therapy for this family of diseases, particularly atherosclerosis. Implementation of the new technology would also produce benefits to the health care industry. Commercializing the cell-based therapy would involve many different core technologies to facilitate production, distribution, and administration, including technologies for the large-scale production of cells under regulated quality practices, and technologies to characterize, store, and manipulate the cells in different applications.

Progenitor Changes Focus to Del-1 Gene

Progenitor's overall technical goal for the ATP-funded project was to isolate a human cell line with endothelial properties equivalent to the murine cell line and to find techniques to ensure that the grafted cell line would not be rejected when transplanted so that it could develop the cell line's potential for treating vascular disease, such as ischemia and restenosis.

During the ATP project, Progenitor discovered that a high percentage of the human yolk sac-derived endothelial cells that it was isolating strongly expressed the Del-1 marker, an essential gene that regulates angiogenesis. This discovery, which was made by Vanderbilt University at the time of the ATP project, had important implications because angiogenesis has therapeutic applications in several fields, including ischemia and restenosis.

At the same time, Progenitor was becoming aware of the many regulatory and safety requirements it would have to meet before commercializing a product based on its original plan. Meeting these requirements would be more time consuming and costly than the young company had anticipated. Alternatively, commercializing Del-1 products would involve meeting fewer requirements. Thus, it would cost less and involve less business risk for Progenitor. It would also result in approximately the same broad-based economic benefits as commercializing yolk sac-derived endothelial cell line products. After discussions with ATP, Progenitor decided to change its project goals and focus on developing the Del-1 marker. The company's new objectives were:

- To understand how Del-1 regulates angiogenesis

- To develop therapies to modulate Del-1 so that tumor angiogenesis could be blocked and therapeutic angiogenesis could be initiated to treat ischemia

Progenitor made considerable progress in meeting these revised goals. By the end of the ATP project, the company had accomplished the following:

- Observed that Del-1 is associated with angiogenesis in both experimental tumors and in most clinical tumors that it examined;
- Observed that Del-1 interacts with a protein that is critical to tumor growth;
- Found that Del-1 exists in several protein forms, one of which is likely to be an antagonist of Del-1's normal function (i.e., it could impede the development of new blood vessels). Progenitor thought that this could then be the basis of a cancer therapy;
- Found that the normal, full-length protein form of Del-1 is highly angiogenic and, thus, could potentially be useful for treating ischemia

Progenitor was assisted in its research by the following subcontractors: Biosupport Inc.; University of Wisconsin at Madison; University of Colorado; Ohio University; Cell Genesys; Microbiological Associates; Southern Research Institute; University of California, San Diego; Stanford University; Vanderbilt University; and, the National Jewish Cancer Center.

Valentis Continues Research of Del-1 Gene

In addition to making significant progress toward reaching its goal of developing the Del-1 marker, by the end of the ATP project in 1998, Progenitor had also published four papers, given four presentations, and filed for two patents. However, the company was also experiencing financial difficulties. A year before, Progenitor had filed an initial public offering and had purchased Mercator Genetics. As a result of the purchase, the company increased its research activities which led to a significant financial loss. By December 1998, about half a year after the ATP project concluded, the company closed.

In April 1999, Valentis, Inc., a leading company in the field of biopharmaceutical delivery, acquired the rights and intellectual property related to the Del-1 gene and continued the research initiated by Progenitor during the ATP project. By December 2000, Valentis was developing plans to conduct clinical studies with Del-1 gene medicine in patients with peripheral arterial disease (PAD) and in patients with coronary artery disease.

In 2003, Valentis completed the Phase I clinical trial of Del-1 for the treatment of PAD and initiated a Phase II clinical trial. Valentis presented the data from the Phase I clinical trial at the 6th Annual Meeting of the American Society of Gene Therapy in Washington, DC. It also published a paper entitled "Neovascularization of Ischemic Tissues by Gene Delivery of the Extracellular Matrix Protein Del-1" in the Journal of Clinical Investigations. Data for the Phase II clinical trial are expected in the third quarter of 2004.

Conclusion

With ATP's assistance, Progenitor continued its research on cell lines with endothelial characteristics, which it had successfully isolated in the past, for the treatment of heart disease. During the course of the project, however, the company observed that the Del-1 gene, which regulates angiogenesis, had a high presence in the endothelial cells that it was isolating. This was a critical discovery, as angiogenesis can be used in the treatment of ischemia and restenosis. Progenitor also realized that commercializing Del-1 products would entail meeting fewer regulatory and safety requirements. This prompted the company to change its focus to developing the Del-1 gene.

By the end of the project in 1998, Progenitor had made significant progress in understanding how Del-1 regulates angiogenesis and could be used to treat ischemia. The company, however, did not complete the research necessary to commercialize the gene. In December 1998, Progenitor went out of business due to financial difficulties. Four months later, in April 1999, Valentis acquired the rights and intellectual property related to the Del-1 gene and has continued the research initiated by Progenitor in the ATP-funded project. In 2003, the company completed a Phase I clinical trial of Del-1 for the treatment of peripheral arterial disease. It also initiated a Phase II clinical trial, which is expected to be completed in 2004.

PROJECT HIGHLIGHTS
Valentis, Inc. (formerly Progenitor, Inc., a subsidiary of Interneuron Pharmaceuticals)

Project Title: Use of Gene Therapy to Treat Cardiovascular Disease (Application of Gene Therapy to Treatment of Cardiovascular Diseases)

Project: To develop a supply of transplantable endothelial cells that might provide the basis of cell-based therapies for vascular disorders, antirejection and anticlot coatings, and "mini-organs" for in-body delivery of therapeutic agents.

Duration: 6/1/1995–5/31/1998
ATP Number: 94-01-0301

Funding (in thousands):

ATP Final Cost	$1,996	71%
Participant Final Cost	799	29%
Total	$2,795	

Accomplishments: ATP funding enabled Progenitor to conduct research on the Del-1 marker. During the project, the company gained an understanding of how the gene regulates angiogenesis and can be used to treat ischemia.

Progenitor filed for a total of 9 patents, 2 of which were granted:

- "Nucleic acid encoding developmentally-regulated endothelial cell locus-1" (No. 5,874,562: filed June 7, 1995, granted February 23, 1999)
- "Developmentally-regulated endothelial cell locus-1" (No. 5,877,281: filed June 5, 1996, granted March 2, 1999)

Commercialization Status: Since acquiring the rights to the Del-1 gene in 1999, Valentis, Inc. has continued to conduct the research on Del-1 gene medicine that was initiated by Progenitor during the ATP project. In 2003, the company completed a Phase I clinical trial and initiated a Phase II clinical trial for Del-1 angiogenesis product for the treatment of peripheral arterial disease. Valentis presented data from the Phase I clinical trial at the 6th Annual Meeting of the American Society of Gene Therapy in Washington, DC. The Phase II clinical trial is expected to be completed in 2004.

Outlook: Valentis anticipates commercializing the Del-1 gene medicine in the future for a large market.

Composite Performance Score: * *

Company:
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Contact: John Reddington
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Subcontractors:

- Biosupport Inc.
  Redmond, WA
- University of Wisconsin
  Madison, WI
- University of Colorado
  Boulder, CO
- Ohio University
  Athens, OH
- Southern Research Institute
  Menlo Park, CA
- Microbiological Associates
  Rockville, MD
- Cell Genesys
  Foster City, CA

Publications and Presentations:

Since 1995, Progenitor has published the following papers:

- Wei, Yanzhang, Thomas Quertermous, and Thomas E. Wagner. "Directed Endothelial Differentiation of Cultured Embryonic Yolk Sac Cells In Vivo Provides a Novel Cell-Based System for Gene Therapy." Stem Cells. 13:541-547. (1995).

- Hidai, Chiaki, Thomas Zupancic, Kalyani Penta, Adel Mikhail, Masatoshi Kawana, Elena E. Quertermous, Masafumi Fukagawa, Y. Aoka, Yasuhisa Matsui, Doros Platika, Brigid L.M. Hogan, Ralph Snodgrass, and Thomas Quertermous. "Cloning and Characterization of Developmental Endothelial Locus-1." Genes and Development. Vol. 12, No. 1, p. 21-33. (January 1998).

- Sturtz, F., L. Cioffi, S. Wittmer, M. Sonk, A. Shafer, Y. Li, N. Leeper, J. Smith-Gbur, J. Shulok, and D. Platika. "Tetracycline-Regulated Expression Vector Tightly Regulate In Vitro Gene Expression of Secreted Proteins." GENE. Vol. 221, No. 2, p. 279-285. (October 1998).

- Cioffi, L., F. Sturtz, S. Wittmer, B. Barut, J. Smith-Gbur, V. Moore, T. Zupancic, B. Gilligan, R. Auerbach, F. Gomez, F. Chauvin, M. Antczak, D. Platika, and H.R. Snodgrass. "A Novel Endothelial Cell-Based Gene Therapy Platform for the In Vivo Delivery of Apolipoprotein E." Gene Therapy. Vol. 6, No. 6, p. 1153-1159. (June 1999).

Since 1996, Progenitor has made the following presentations:

- Sturtz, F., L. Cioffi, B. Barut, S. Wittmer, J. Smith-Gbur, E. Beck, R. Snodgrass, and D. Platika. Poster. "Miniorgans: Genetically Engineered Yolk Sac Cells Secreting an Externally Regulated Gene Product." KEYSTONE Meeting on Tissue Engineering. Taos, New Mexico. (January 23-29, 1996).

- Cioffi, L., F. Sturtz, S. Wittmer, V. Moore, J. Smith-Gbur, E. Beck, T. Zupancic, B. Barut, D. Platika, and R. Snodgrass. "Gene Therapy for Atherosclerosis: Modulation of Cholesterol Levels by APOE Secreting Endothelial Cells." Joint Meeting of the ASBMB, ASIP, and AAI. (June 1996).

- Zupancic, T.J., C. Hidai, K. Penta, A. Mikhail, T. Schweitzer, V. McGaughy, J. Smith-Gbur, T. Quertermous, D. Platika, and H.R. Snodgrass. "Cloning and Characterization of Del-1, a Novel Developmentally Regulated Gene Involved in Formation of the Cardio-Vascular System." Joint Meeting of the ASBMB, ASIP, and AAI. (June 1996).

- Snodgrass, R. "Del-1 in Angiogenesis." IBC Angiogenesis Conference. (July 1997).

Since acquiring the rights to the Del- 1 gene in April 1999, Valentis has published the following paper:

- Zhong, J., B. Eliceri, D. Stupack, K. Penta, G. Sakamoto, T. Quertermous, M. Coleman, N. Boudreau, and J.A. Varner. "Neovascularization of Ischemic Tissues by Gene Delivery of the Extracellular Matrix Protein Del-1." Journal of Clinical Investigations. Vol. 112, No. 1, p. 30-41 (July 2003).

Research and data for Status Report 94-01-0301 were collected during March - July 2003.

APPENDIX A

Development of New Knowledge and Early Commercial Products and Processes, 3rd 50 of Status Reports

Table A-1: Advanced Materials and Chemicals; Table A-2: Biotechnology; Table A-3: Electronics, Computer Hardware, or Communications; Table A-4: Information Technology; Table A-5: Manufacturing

Table A-1. Advanced Materials and Chemicals

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
ABB Lummus Global, Inc. (formerly ABB Lummus Crest)	95-05-0034	Developed a new, environmentally superior process to manufacture alkylate, an ideal unleaded gasoline additive, using solid-acid catalysts	As of 2005, the joint venture partners were seeking commercial opportunities to build new solid-acid alkylation plants
Advanced Refractory Tech	95-01-0131	Developed a diamond-like nanocomposite (DLN) coating technology. The company established improved manufacturing techniques for DLN films and developed several applications, such as electrosurgical blades and flat panel displays	A number of products with DLN coatings are currently being sold. These include components that are used in manufacturing CDs, DVDs, polyethylene terephthalate juice bottles, and metal cans and components used in semiconductor cluster tools
Air Products and Chemicals, Inc.	93-01-0041	Developed ceramic-steel seals and processes to remove contaminants from oxygen	The company is continuing its research and development (R&D) into their prototype air-separation unit for producing high-purity oxygen so that future commercialization may be possible. However, the company does not intend to pursue commercialization initiatives until a 30-percent decrease in production cost is achieved
Automotive Composites Consortium (a Partnership of DaimlerChrysler [formerly Chrysler], Ford and General Motors)	94-02-0027	Developed a composites-manufacturing process called Structural Reaction Injection Molding (SRIM) for f producing large automobile structural parts, such as the box of pickup trucks	Commercialized the access door and tail cone for the Air Force C-17 cargo plane by Boeing, firefighter helmet shells by Lion Apparel, the inner tailgate sections for the GM Cadillac Escalade EXT hybrid SUV beginning in 2001, the load floor sections for the "Stow 'n Go" system to fold down second-and third-row seats in the Chrysler

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
			Town & Country LX and Dodge Grand Caravan SCT beginning in 2005, the midgate (a door that folds down to extend cargo space) for the GM Chevrolet Avalanche beginning in 2001, the motor covers for marine applications by SeaRay (Marine division of Brunswick Corp), and the pickup truck box and tailgate assembly for the 2001 to 2004 GM Chevrolet Silverado. Boeing's 787 "Dreamliner" uses SRIM composites for structural parts, increasing fuel efficiency by 3 percent. Overall fuel savings is 20 percent compared with the 747. First commercial flight is scheduled for 2008
Bosch (formerly Allied Signal)	95-07-0020	Developed a synergy between design and casting processes that resulted in the following accomplishments: elimination of porosity problem (zero rejects for porosity); reduction from one large and three small defects per part to two small defects per part; acceleration of research by two years ahead of where it otherwise would have been through parallel research efforts; and reduction of defects in a specific type of valve body design by up to 85 percent	The technical challenges of this project were too numerous and difficult to overcome. As a result, AlliedSignal created no new products for brakes using the technology developed under the ATP-funded project. The Top Die Casting Company produced some components using the new processes, such as air brake valves and brackets. Stahl Specialty Company used one step of the aluminum manufacturing process to assist in aluminum filtration. That process had a small impact on several of the company's product lines
BP Amoco	93-01-0234	Developed a process using silver nitrate as a facilitating agent in high-efficiency contactors and had developed a promising new complexing agent that would potentially cost less than silver nitrate when used for facilitated transport	Although the process was technically sound, the company was experiencing costly operating problems. Amoco was unable to demonstrate the economic feasibility of using this new technology for olefin-paraffin separations and therefore did not commercialize the technology
Catalytica Energy Systems (formerly Catalytica, Inc.)	94-01-0190	Developed catalysts with enhanced activity and selectivity for use in the chemical and petroleum-refining industries	Developed a Multiple Stream Mixer/Reactor (MMR) which may prove to be a very valuable tool for the emerging nanotechnology sector, producing nanoparticles for many industries. The company expected to sell its first major

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
			production MMR system in 2005 or 2006
Crucible Materials Corporation, Crucible Compaction Metals Division	94-01-0287	Developed alloys with high levels of nitrogen that demonstrated the potential to produce high-strength, corrosion-resistant stainless steel	Commercialized high-nitrogen alloys that could improve the performance of stainless steel (SS100)
GM Thermoplastic Engineering Design (Engineering Design with Thermoplastics)	92-01-0040	Developed models and generated data for "virtual design" in order to improve the design and development of thermoplastic automotive parts. The project team linked two commercial software tools, Moldflow (formerly C-MOLD) and ABAQUS, with new failure theories for plastics in order to integrate mold design with parts performance	Commercialized virtual design tools that have shortened development time and have improved the performance of thermoplastic parts, which has benefited many manufacturers (for example, Delphi's thermoplastic radiator tank and many other parts; GM's injection-molded plastic intake manifold and other engine components; GE Plastics' improved raw material, which is used in business equipment, optical media, and telecommunications devices). The project resulted in the International Organization for Standardization (ISO) issuing a new standard (ISO 94-5)
Honeywell (formerly Allied Signal)	93-01-0104	Developed powder injection molding used in the ceramic industry for chinaware, spark plugs, oxygen sensor components, and oxygen sensor insulators	Commercialized ceramic powder injection molding technology that is being used in chinaware, spark plugs, oxygen sensors, ball bearings, manufacturing components (for example, stamping punches and guide rollers), engine and machine components (for example, nozzles, seals, shafts, valves, and heating units), and bio ceramics (for example, artificial bones for human replacement surgery)
Honeywell (formerly Allied Signal)	95-07-0003	Developed "aqueous injection molding" (AIM) process improvements for ceramic splitter vanes	Commercialized ceramic splitter vanes in 1998. They had plans to commercialize other small, complex, high-volume parts like blades and nozzles

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
IBM T.J. Watson Research Center	93-01-0149	Developed a conducting polymer of acid-doped polyaniline (PANI) with thermal stability greater than 250 degrees C from 150 degrees C, increasing processability and solubility, and increasing conductivity by 2.5 orders of magnitude	Commercialized a water-soluble version of PANI that was licensed to Monsanto Chemical Corporation in 1997, and IBM is pursuing further licensing opportunities
PCC Structurals	95-07-0011	Developed a casting technology that combines the superalloy processing capabilities of investment casting with the economic advantages of sand casting and achieves part sizes sufficient to produce exhaust frames for industrial gas turbine engines	PCC did not commercialize the new casting technology. They did develop prototypes of a new casting technology that will allow manufacturers to produces large structural superalloy components for industrial equipment industries, such as the Industrial Gas Turbine industry
Praxair, Inc.	94-01-0111	Developed new materials highly selective for oxygen, including IC-2, IA-1, IA-2, and IA-3, which have the potential of meeting all characteristics of a successful material with further development	The O2-selective materials developed during this ATP-funded project have not been commercialized. However, as of 2003, Praxair has continued work on their development through a project with the Department of Energy with hopes to commercialize in the future
The Dow Chemical Company	95-05-0002	Developed a direct, economical, single-product oxidation process incorporating a silver-based catalyst for conversion of propylene to propylene oxide	Dow researchers expect that they might complete a process to develop a direct oxidation propylene sometime between 2006 and 2014. A successful process will reduce energy consumption, cost, and waste in the manufacturing of many types of plastics, lubricants, coatings, surfactants (detergents), and composite materials
Wyman-Gordon	95-07-0026	Developed an incremental forging process to produce near-net shape forgings for industrial gas turbines using a lower-tonnage press than was previously possible	Wyman-Gordon has incorporated the incremental forging process into its business operations

Table A-2. Biotechnology

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
Aphios Corporation	95-01-0263	Developed a knowledge base and technology platform to tap into the pharmaceutically, industrially, and environmentally valuable chemical diversity that remains unexplored in enormous numbers of marine microorganisms	An anti-plaque solution for toothpaste or mouthwash, which is being optimized through chemistry, is the nearest product to commercialization. Novel therapeutics for multiple-disease-resistant (MDR) bacteria, influenza, HIV/AIDS, cancer, and smallpox are also undergoing trials in preclinical drug discovery and development
Cengent Therapeutics Inc. (formerly Moldyn Inc.)	94-01-0137	Developed a software that adapts a technology developed in the aerospace industry to simulations of biological molecule and drug interactions, for the purpose of qualifying drug research candidates in a more timely and efficient manner than by using trial-and-error techniques	The MD simulation software was briefly commercialized through a license to Molecular Simulations Incorporated, but failed to gain sufficient sales and was discontinued. However, Moldyn's software was incorporated with Harvard's Chemistry at Harvard Macromolecular Mechanics (CHARMM) molecular modeling tool through a licensing agreement between Moldyn and Harvard University
Dow AgroSciences LLC (formerly Mycogen Corporation)	95-01-0148	The company made strides in genetic research and demonstrated for the first time that yeast is transformable. They demonstrated that squalene could be hyper-produced in oleaginous yeast; and they gained a broader understanding of the metabolic pathways for isoprene formation in yeast	No commercialization occurred because the oleaginous yeast fermentation project was ended due to technical barriers with enzyme manipulation
DuPont Qualicon (formerly DuPont FQMS Group)	94-05-0033	Developed a functioning automated, rapid DNA diagnostic prototype system that reduced analysis time from 3 hours to 30 minutes. The system can determine the presence or absence of specific microbial contamination as a means of quality control in the food industry. However, DNA pattern results from sample testing were somewhat inconsistent and needed further development	Additional steps were required in sample preparation that negated the time saved in analysis. DuPont Qualicon ended the research into this automated system in 1998, but the company did apply some of the automation knowledge gained in this project to its ongoing alternate food-borne pathogen-testing technologies

Genosensor Consortium (c/o Houston Advanced Research Center)	92-01-0044	Developed a technology for automated DNA sequence analysis	Provided sample analysis and database services for genotyping and gene expression research to organizations such as the Schering Plough Research Institute. In 1999, consortium member Sigma Genosys began to sell Panorama Gene Arrays, which profile gene expression in human cytokines, B. subtilis, and E. coli. In 2003, Sigma Genosys sold human cancer oligoarrays. In 2003, consortium member Beckman Coulter started to commercialize arrays
Incyte Corporation (formerly Combion, Inc.)	94-05-0019	Developed a method akin to ink-jet printing for synthesizing large arrays of specific DNA fragments suitable for medical diagnosis, microbial detection and DNA sequencing, and for creating supplies of detachable oligonucleotides for subsequent use	Microarray expertise and knowledge gained in this project formed the foundation for Incyte's highly successful bioinformatics business, which operated from 1999 to 2001 (selling subscriptions to databases of DNA information). Although Incyte put the specific chem-jet microarray manufacturing techniques developed in this project on hold from approximately 1998 to 2004, the company licensed the technology to Agilent in 2001. As of 2004, Agilent was about to commercialize the ATP-funded technology in conjunction with their numerous other patented chem-jet technologies
JDS Uniphase (formerly The Uniphase Corporation)	94-05-0004	Although the attempt to develop a compact, efficient, and cheaper source of blue light for fluorescence-based diagnostic instruments and techniques for physicians and biomedical researchers was unsuccessful, the project led to the development of two unanticipated products	Commercialized the Blue Laser Module, a stripped-down, inexpensive blue laser for tabletop applications within the biotechnology industry, that reached the market in 1999 and has achieved sales as high as $500,000 per year. They also sold the MicroBlue SLM, a specialized, low-noise blue laser for digital photo-finishing, that was first marketed in 2000 and generated $1 million in annual sales
Large Scale Biology Corporation (formerly Large Scale Proteomics Corporation)	94-01-0284	Developed the ProGEx product line for protein identification and research. The company also completed the first version of the Human Protein Index by identifying more than 115,000 proteins from 157 medically relevant human tissues	The 2-D gel and ProGEx line of protein analysis tools has been upgraded and improved over the years. Large Scale Biology Corporation (LSBC), which acquired LSPC in 1999, still sells research products and databases created through use of technology flowing from the knowledge acquired during this ATP-funded

			project. The company performs up to one million mass spectrometry analyses of proteins per week
Medical Analysis Systems (formerly NAVIX)	95-08-0017	Developed a two-stage reaction for DNA identification and amplification. The process identifies areas of DNA that correlate with disease	Navix did not commercialize any products from its ATP-funded research. Business issues delayed research long enough for another competitor to beat Navix to the market
Monsanto (formerly Agrecetus)	94-01-0074	Developed a prototype plant with elevated levels of poly-3-hydroxybuteric acid (PHB). Although the PHB concentration was not high enough for commercialization, simply raising the PHB level at all represented a technical achievement	Due to the difficulty in attaining high enough PHB levels in the cotton fibers without "crowding out" the fibers' favorable traits, no commercialization efforts resulted from this ATP-funded research.
Orchid BioSciences (formerly Molecular Tool, Inc. Alpha Center)	94-05-0034	Developed techniques for micromachining and for handling fluids on a microscopic scale to make a simple, compact DNA typing instrument	Developed the SNPstream Ultra High Through-Put (UHT), automated array-based genotyping tool. Entered the market through Orchid BioSciences in 2001. Product, intellectual property, and research and development were sold to Beckman Coulter in December 2002. As of 2004, Beckman continues to develop and enhance the system, marketing to research and clinical laboratories. Orchid BioSciences provides genetic analyses using SNPstream UHT on a fee-for-service basis (for biotech companies, pharmaceutical companies, and criminal justice agencies). Orchid's facility was providing up to 1 million SNP scores per day by the end of 2000 on a fee-for-service basis
Valentis, Inc. (formerly Progenitor, Inc.; a subsidiary of Internueron Pharmaceuticals)	94-01-0301	Developed an understanding of how the Del-1 gene regulates angiogenesis and can be used to treat ischemia	In 2003, the company completed a Phase I clinical trial and initiated a Phase II clinical trial for Del-1 angiogenesis product for the treatment of peripheral arterial disease

Table A-3. Electronics, Computer Hardware, or Communications

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
eMagin Corporation (formerly FED Corporation)	93-01-0154	Developed manufacturing techniques for large-scale, flat-panel displays based on arrays of field emitters, a sort of “flat CRT”	Commercialized two microdisplays, SVGA 3D and SVGA+ rev2. The microdisplays are integrated into hundreds of medical, commercial, and military applications. For example, firefighters see through thick smoke by looking through a thermal-imaging camera lens to find victims, even under a blanket. They can also use the lens to find the source of a fire quickly and put it out. Researchers and doctors are using the display to enhance vision for magnetic resonance imaging (MRI), endoscopic surgery, and eye surgery
INSIC (formerly NSIC) - Short Wavelength	90-01-0231	Developed optical recording standards to improve upon traditional magnetic recording	NSIC members did not commercialize optical recording devices because remaining technical obstacles would have required significant further development of the frequency-doubling technology; and by the end of the project, competition was looming from direct-lasing green and blue diode lasers
Kopin Corporation	94-01-0304	Developed liquid crystal projection display technology capable of producing high-quality, high-resolution images for high-definition TV	Commercialized the CyberDisplay 320 Monochrome, the CyberDisplay 320 Color, the CyberDisplay 640 Color, the CyberDisplay 1280 Monochrome 60" diagonal projection HDTV, the CyberDisplay 1280 Monochrome 55" diagonal projection HDTV, the CyberDisplay 1280 Monochrome 46" diagonal projection HDTV, and CyberDisplay 1280 Monochrome 43" diagonal projection HDTV
Planar Systems, Inc. (American Display Consortium)	93-01-0054	Developed a group of patterning technologies necessary to manufacture color flat-panel displays, including large-area photo exposure tools, large-area masks, wet and dry etching tools, printing tools, panel alignment methods and a final inspection tool	Subcontractor, Photronics (now Infinite Graphics, Inc. [IGI]), commercialized customized large-area photo masks for use in high-end printer circuits, calibration plates, x-ray systems, and flat-panel displays. Photonics also developed two processes: mask cleaning & laser pattern generator

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
			Subcontractor, Plasma-Therm successfully commercialized dry etching processes in its Clusterlock 7000 for 6-inch wafers. The company was sold to a Swiss company, Oerlikon-Buehrle in 1999. Planar used the Plasma-Therm etcher to produce AMEL microdisplays until 2002. Subcontractor, YieldUp (now FSI) developed drying tools for wet-etched substrates. This is now used for flat-panel displays and primarily computer chip manufacturing. Also used in hard disk drive cleaning and photomask cleaning. Currently, the ATP-funded component is key in seven larger processing systems:ZETA Spray Cleaning System, ANTARES CX Advanced Cleaning System, EXCALIBUR Vapor HF Etching System, MERCURY Spray Cleaning System, YieldUP 4000 Immersion Etch System, YieldUP 2000 Rinse Dry Module, and YieldUP 2100 STG Rinse Dry Integration Module
SDL, Inc. and Xerox Corporation	91-01-0176	Demonstrated the first integration of multiple-wavelength laser diodes on a single semiconductor device. In the course of this work, the team established several intermediary technologies and accomplished important research in the field of gallium nitride (GaN)-based blue laser diodes. Demonstrated technologies include two alternative methods for monolithic integrations of red, infrared, and blue emitters; red laser diodes with powers of up to 120 mW single mode; lasers in the 700- to 755-nm range; green and blue lasers with frequency doubling; and the lasing of blue GaN diodes at room temperature	After the ATP-funded project, SDL commercialized several laser products that were based on technologies developed in the course of the project: a single-mode laser using facet passivation technology; a single-mode laser for PDT applications; a dual-spot single-mode laser for data storage, printing, displays, and alignment; a multi-mode laser; fiber coupled laser bars for solid state laser pumps, medical systems and displays; and a DBR laser for frequency doubling, interferometry, atomic clocks, and spectroscopy
Superconductor Technologies Inc. (formerly Conductus)	91-01-0134	Developed a prototype superconducting DSP switch	Commercialization of the technology developed and tested during this ATP-funded project was not pursued due to a lack of interest in the technology on the

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
			part of the semiconductor and communications industries
Texas Instruments Inc.	94-01-0221	Developed a special insulating material, known as aerogel, to be integrated adjacent to on-chip interconnects in order to overcome problems with interconnect delay as a result of the continuing trend toward miniaturization. Texas Instruments and NanoPore developed the world's first fully automated manufacturing process to dry an aerogel quickly	The company overcame impediments to aerogel processing early in the project, but in 1997, an industry competitor announced that it would begin using copper interconnect wiring in future integrated circuit designs. Texas Instruments then shifted focus away from aerogels for aluminum and began to develop copper interconnects. Before shifting focus, however, Texas Instruments transferred its aluminum circuit aerogel knowledge to NanoPore, which later sold the rights to continue development of the product to Honeywell. Honeywell's development efforts resulted in a product that they marketed briefly in 2002 to companies for use in manufacturing semiconductors. However, Honeywell withdrew the product in 2004 after it did not fulfill its potential as a new and innovative insulator

Table A-4. Information Technology

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
Accenture (formerly Andersen Consulting Center for Strategic Research)	94-06-0012	Developed a prototype technology for reusable software components based on software architecture considerations, including formal languages to express semantics, a graphical user interface programming environment, automated techniques for assuring that the separate components are logically compatible and properly combined, and automated systems to generate executable systems	No product was commercialized as the technology focus of the industry changed shortly after the project concluded
Cerner Corporation	94-04-0008	Developed information tools to automate, validate and distribute clinical practice guidelines for mass use	Used general concepts from the ATP-funded project to execute guidelines in its Cerner Millennium product. With Cerner Millennium, clinicians are electronically alerted about potential patient safety and regulatory issues through evidence-based medical information
Cerner Corporation (formerly DataMedic - Clinical Information Advantages, Inc.)	94-04-0038	Developed a knowledge-base-driven automated coding system in the form of a software component, CHARTnote which uses MEDencode, a technology that automatically gathers, codifies, and records specific detailed information about a patient	The software is currently incorporated into and sold with approximately 7 CHARTstation products, manufactured by VitalWorks. It is also sold separately and with other products. Products include GIstation, EMstation, EYEstation, RADstation, and other areas including internal medicine and family practice, renal dialysis, and rehabilitative medicine
InStream	94-04-0018	Developed the first behavioral healthcare (BHC) Web portal for claims processing	The software product was briefly commercialized in 1998, but was quickly overtaken by competing products after a lack of funding prevented InStream from providing the necessary upgrades and market penetration to reach positive cash flow
Lucent Technologies (formerly AT&T Bell Laboratories)	94-06-0011	Developed and successfully demonstrated their software (Symphony) to develop an easy-to-use, graphics-user interface (GUI) software assembly system for nonprogrammers that handles the complexity of building reliable,	No commercialization resulted from this project because of AT&T's corporate restructuring in 1996. Lucent decided to discontinue its development of the reusable software component product

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
		custom-designed software by using libraries of reusable, software components	
SciComp, Inc.	94-06-0003	Developed a component software and a software synthesis technology for creating mathematical models in the field of scientific computing	As of 2004, SciComp offered three software tools in the SciFinance solution that incorporate the ATP-funded software synthesis technology; SciFinance also includes two additional products that enhance SciPDE and SciMC. SciComp experienced greater demand for these products as the market
Titan Systems (formerly Intermetrics)	94-04-0040	Developed a script language and a related suite of software tools to facilitate the process of developing customized home healthcare workstations for homebound or limited-mobility, chronically ill patients	A product was not commercialized. The intellectual property was acquired by HealthVision, which chose not to further develop it
Xerox Palo Alto Research Center	94-06-0036	Developed a new programming technique called aspect-oriented programming (AOP). They also developed two prototype applications of specialized computer languages	AspectJ, an open-source language that extends Java, is now used in a significant percentage of IBM's new products and is an open-source platform. PARC transferred AspectJ to the open-source eclipse.org project in December 2002

Table A-5. Manufacturing

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
Abrasive Technology Aerospace, Inc.	95-02-0053	Developed an integrated CAD/CAM approach to applying superabrasive coatings to complex surfaces of electroplated superabrasive grinding wheels	In 2000, Abrasive Technology began to market and sell electroplated superabrasive grinding wheels using the CAD/CAM technology it developed during the ATP-funded project, and still continues to do so. The company has used the new technology to produce grinding wheels for a variety of industries, including automotive and aerospace
Cincinnati Lamb, UNOVA (Lamb Technicon)	95-02-0019	Developed an experimental prototype of a flexible line boring station with intelligent tooling and controls	The BOA technology was not commercialized because auto manufacturers found less expensive machine tools to meet their specifications
General Electric Corporation R&D	95-07-0018	Developed an intelligent process for applying thermal barrier coatings to critical components in turbine engines for power plants in order to raise firing temperatures and increase fuel efficiency	GE successfully produced an improved gas turbine engine for its new H-System combined-cycle power plant, which can achieve 60-percent energy efficiency. The high-performance thermal barrier coatings developed in part using technology from this project were essential to the design of this model. GE also applied the knowledge to upgrade existing F-System plants, which achieved 56-percent efficiency. Other companies have used the process on marine aircraft and heavy diesel engines, as well as other applications
IBM Corporation	94-03-0012	Developed an automated tool kit that could be used by vendors to develop, maintain, and join interoperating families of enterprise resource planning (ERP) and manufacturing execution system (MES) applications	IBM did not commercialize its new automated tool kit. Instead, it commercialized a service based on its new Framework for Adaptive Interoperability of Manufacturing Enterprises (FAIME) technology, enterprise application integration (EAI) services
Montronix	95-02-0020	Developed a diagnostic system that can monitor the vital signs of machining operations in real time to provide a trouble-shooting aid for process engineers who are increasingly challenged to efficiently machine smaller	The developed monitoring system later evolved into a standard Montronix product line called Spectra. A key accomplishment of this project was providing free Internet-based simulated machine-tool modeling

A. Awardee Name	B. Project Number	C. Technology Developed	D. Products or Processes Commercialized or Expected to be Commercialized Soon
		volumes of a wider variety of parts	(http://mtamri.me.uiuc.edu/testbeds/testbed.intro.html). The web-based simulation is still in use by government, research and industry
United Technologies Research Center	95-06-0011	Developed a prototype handheld device to detect refrigerant leaks during manufacture of components containing refrigerant	No commercialization occurred. All three companies cited cost of development, lack of funding, competition, and uncertain market demand as contributing factors to discontinuing research into this technology. The markets for the laser emitter for the handheld unit were also limited
York International	95-06-0004	Developed a prototype heat exchanger that was 25 percent smaller and had the same heat transfer capability as the standard size. Furthermore, York developed a method and a tool that they still use in their ongoing research and development. They also demonstrated that oval-tube geometry is 10 percent more efficient for heat transfer than round tubes	Using the methods developed during this project, York developed a new commercialized plate fin, called HiQ. York uses the fin in its ECO2 rooftop heating/cooling units. Its proprietary enhancements yield approximately twice the heat transfer when compared to a standard fin. Due to the prohibitive manufacturing capital cost, York has postponed commercializing oval-tube coil technology

APPENDIX B

Reasons for Terminating ATP Projects

At the end of an ATP competition, projects are selected for award and the winners are announced. Most of these projects proceed through their multi-year research plans to completion. Some are not carried through to completion for a variety of reasons. These projects are collectively called "terminated projects."

Between 1990 and September 2004, there were 768 ATP awards issued, of which 84[22] projects ended before completion. Below is a percentage distribution by category of the reasons for termination.

Change in goals

- 54 percent ended because of changes in the strategic goals of the companies, changes in the business climate or markets, changes in company ownership, or other business-related facts.

Lack of technical progress

- 12 percent ended because of lack of technical progress, which sometimes occurs at go/no-go decision points recommended by the participant(s).

Project no longer meets ATP criteria

- 11 percent ended because changes in scope, membership, performance, or other factors meant that the project no longer met ATP's technical and/or economic criteria.

Lack of agreement among joint venture members

- 2 percent ended because the joint venture members could not reach an agreement on some issues.

Financial distress

- 11 percent ended due to the financial distress of a key participant.

Early success

- 5 percent ended due to early success of the project!

Although projects may end early, it is not necessarily an indication of total failure. Projects that ended early produced important knowledge gains; involved integrated planning for research, development, and business activities that may have some benefit to participating companies; and entailed substantive cross-disciplinary contact among scientists and other researchers, cross-talk among technical and business staff, and negotiations among executives at different companies.

[22] Included in this figure are four projects that were cancelled before the project began, comprising approximately 5 percent of the total.

These characteristics still benefit the economy by stretching the thinking and horizons of participants in the process. Companies may learn about new opportunities and apply integrated planning of research and business activities to other projects. In summary, terminated projects may have some positive impact even though they incur costs.

APPENDIX C

Composite Performance Rating System (CPRS) Star Ratings—First 150 Completed Projects

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
91-01-0243	Aastrom Biosciences, Inc.	1st 50	****
91-01-0146	American Superconductor Corp.	1st 50	****
94-02-0027	Automotive Composites Consortium (a Partnership of DaimlerChrysler [formerly Chrysler], Ford and General Motors)	3rd 50	****
94-04-0038	Cerner Corporation (formerly DataMedic - Clinical Information Advantages, Inc.)	3rd 50	****
96-01-0263	ColorLink, Inc.	2nd 50	****
91-01-0256	Cree Research, Inc.	1st 50	****
91-01-0184	Engineering Animation, Inc.	1st 50	****
93-01-0085	Integra LifeSciences	1st 50	****
94-01-0304	Kopin Corporation	3rd 50	****
94-01-0284	Large Scale Biology Corporation (formerly Large Scale Proteomics Corporation)	3rd 50	****
91-01-0041	Nanophase Technologies Corporation	2nd 50	****
90-01-0154	National center for Manufacturing Sciences (NCMS)	1st 50	****
94-05-0034	Orchid BioSciences (formerly Molecular Tool, Inc. Alpha Center)	3rd 50	****
94-06-0003	SciComp, Inc.	3rd 50	****
91-01-0176	SDL, Inc. and Xerox Corporation	3rd 50	****
94-05-0012	Third Wave Technologies, Inc.	2nd 50	****
92-01-0133	Tissue Engineering, Inc.	1st 50	****
94-06-0024	Torrent Systems, Inc. (formerly Applied Parallel Technologies, Inc.)	1st 50	****

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
94-06-0036	Xerox Palo Alto Research Center	3rd 50	****
91-01-0112	X-Ray Optical Systems (XOS), Inc.	2nd 50	****
95-05-0034	ABB Lummus Global, Inc. (formerly ABB Lummus Crest)	3rd 50	***
95-01-0131	Advanced Refractory Tech	3rd 50	***
93-01-0113	Amersham Pharmacia Biotech (formerly U.S. Biochemical Corporation)	1st 50	***
91-01-0177	Auto Body Consortium	1st 50	***
94-01-0115	Calimetrics, Inc.	1st 50	***
93-01-0055	Caterpillar Corporation	2nd 50	***
95-01-0022	Corning Tropel (formerly Tropel Corporation)	2nd 50	***
92-01-0136	Cynosure, Inc.	1st 50	***
95-02-0055	Dana Corporation	2nd 50	***
92-01-0115	Diamond Semiconductor Group, LLC	1st 50	***
98-02-0034	Digital Optics Corporation	2nd 50	***
94-01-0402	Displaytech, Inc.	2nd 50	***
90-01-0064	E.I. Du Pont de Nemours & Company	1st 50	***
94-02-0025	Ebert Composites Corporation	2nd 50	***
95-11-0012	EDO Specialty Plastics (formerly Specialty Plastics Inc.)	2nd 50	***
93-01-0154	eMagin Corporation (formerly FED Corporation)	3rd 50	***
91-01-0178	Ford Motor Company	2nd 50	***
95-07-0018	General Electric Corporation R&D	3rd 50	***
92-01-0044	Genosensor Consortium (c/o Houston Advanced Research Center)	3rd 50	***
92-01-0040	GM Thermoplastic Engineering Design (Engineering Design with Thermoplastics)	3rd 50	***

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
92-01-0116	Honeywell (formerly Allied Signal Inc.)	2nd 50	***
95-09-0052	Hynomics (formerly Hybrithms Corporations, formerly Sagent Corporation)	2nd 50	***
94-05-0018	Hyseq, Inc.	2nd 50	***
93-01-0149	IBM T.J. Watson Research Center	3rd 50	***
92-01-0017	Illinois Superconductor Corporation	1st 50	***
94-05-0019	Incyte Corporation (formerly Combion, Inc.)	3rd 50	***
91-01-0016	Information Storage Industry Consortium (INSIC, formerly NSIC)	2nd 50	***
91-01-0262	Kopin Corporation	1st 50	***
93-01-0101	Kurzweil Applied Intelligence, Inc	2nd 50	***
93-01-0183	MicroFab Technologies, Inc.	2nd 50	***
91-01-0224	Molecular Simulations, Inc. (formerly Biosym Technologies, Inc.)	1st 50	***
96-01-0172	Nanogen, Inc.	2nd 50	***
90-01-0166	Nonvolatile Electronics, Inc.	1st 50	***
93-01-0071	Perceptron, Inc.	1st 50	***
93-01-0205	Physical Optics Corporation (POC)	2nd 50	***
94-04-0024	PPD Informatics (formerly Belmont Research, Inc.)	2nd 50	***
92-01-0123	Sage and 3M Corporation	1st 50	***
94-02-0010	Strongwell Corporation	2nd 50	***
94-04-0046	Surgency (formerly Benchmarking Partners)	2nd 50	***
95-05-0002	The Dow Chemical Company	3rd 50	***
97-01-0087	TopicalNet (formerly Continuum Software, Inc.)	2nd 50	***
93-01-0124	Vitesse Semiconductor Corporation	1st 50	***

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
94-04-0027	3M Company, Health Information Systems	2nd 50	**
95-02-0053	Abrasive Technology Aerospace, Inc.	3rd 50	**
91-01-0187	AlliedSignal, Inc.	1st 50	**
90-01-0060	American Display Consortium	1st 50	**
95-01-0263	Aphios Corporation	3rd 50	**
91-01-0142	AstroPower, Inc.	1st 50	**
93-01-0250	BioTraces, Inc.	1st 50	**
95-07-0020	Bosch (formerly Allied Signal)	3rd 50	**
93-01-0234	BP Amoco	3rd 50	**
94-01-0190	Catalytica Energy Systems (formerly Catalytica, Inc.)	3rd 50	**
94-01-0137	Cengent Therapeutics Inc. (formerly Moldyn Inc.)	3rd 50	**
95-02-0019	Cincinnati Lamb, UNOVA (Lamb Technicon)	3rd 50	**
90-01-0210	Communication Intelligence Corporation	1st 50	**
93-01-0211	Communication Intelligence Corporation	1st 50	**
94-01-0287	Crucible Materials Corporation, Crucible Compancton Metals Division	3rd 50	**
93-01-0091	Elsicon (formerly Alliant Techsystems, Inc.)	2nd 50	**
92-01-0022	FSI International, Inc.	1st 50	**
92-01-0074	Geltech Incorporated	1st 50	**
94-01-0147	Genzyme Corporation (formerly GelTex Pharmaceuticals, Inc.)	2nd 50	**
91-01-0034	HelpMate Robotics, Inc. (formerly Transitions Research Corporation)	1st 50	**
93-01-0104	Honeywell (formerly Allied Signal)	3rd 50	**
95-07-0003	Honeywell (formerly Allied Signal)	3rd 50	**

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
94-05-0004	JDS Uniphase (formerly The Uniphase Corporation)	3rd 50	**
94-01-0133	Laser Power Corporation	2nd 50	**
90-01-0212	Light Age, Inc.	1st 50	**
90-01-0121	Lucent Technologies Inc.	1st 50	**
93-01-0191	M&M Precision Systems Corporation	2nd 50	**
92-01-0053	Mathematical Technologies Inc.	1st 50	**
91-01-0088	Michigan Molecular Institute	1st 50	**
93-01-0027	Micron Optics, Inc.	2nd 50	**
95-02-0020	Montronix	3rd 50	**
95-08-0009	Nanogen, Inc.	2nd 50	**
94-01-0357	Norton Diamond Film	2nd 50	**
93-01-0054	Planar Systems, Inc. (American Display Consortium)	3rd 50	**
94-06-0026	Reasoning Systems, Inc.	2nd 50	**
90-01-0232	Saginaw Machine Systems, Inc.	1st 50	**
91-01-0263	Spire Corporation	1st 50	**
94-01-0221	Texas Instruments Inc.	3rd 50	**
94-06-0034	TopicalNet, Inc. (formerly Continuum Software, Inc.)	2nd 50	**
94-01-0063	Union Switch and Signal, Inc.	1st 50	**
94-01-0301	Valentis, Inc. (formerly Progenitor, Inc.; a subsidiary of Interneuron Pharmaceuticals)	3rd 50	**
91-01-0261	Westinghouse Plasma Corp.	1st 50	**
93-01-0041	Air Products and Chemicals, Inc.	3rd 50	*
94-04-0017	American Healthware Systems	2nd 50	*

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
94-02-0040	Budd Company, Design Center	2nd 50	*
92-01-0132	GE Corporate Research and Development	2nd 50	*
91-01-0069	Honeywell, Inc., Technology Center	2nd 50	*
94-03-0012	IBM Corporation	3rd 50	*
92-01-0034	Ingersoll Milling Machine Company	2nd 50	*
90-01-0231	INSIC (formerly NSIC) - Short Wavelength	3rd 50	*
94-04-0037	KOOP Foundation, Inc.	2nd 50	*
95-10-0067	KOOP Foundation, Inc.	2nd 50	*
91-01-0258	Microelectronics Center of NC	1st 50	*
95-02-0005	Perceptron (formerly Autospect, Inc.)	2nd 50	*
93-01-0045	Philips Laboratories	2nd 50	*
94-01-0111	Praxair, Inc.	3rd 50	*
91-01-0267	PreAmp Consortium	1st 50	*
91-01-0134	Superconductor Technologies Inc (formerly Conductus)	3rd 50	*
93-01-0109	Thomas Electronics, Inc.	1st 50	*
95-06-0011	United Technologies Research Center	3rd 50	*
95-07-0026	Wyman-Gordon	3rd 50	*
95-06-0004	York International	3rd 50	*
94-06-0012	Accenture (formerly Andersen Consulting Center for Strategic Research)	3rd 50	-
94-04-0025	Accenture (formerly Andersen Consulting)	2nd 50	-
92-01-0055	Accuwave Corporation	1st 50	-
91-01-0135	Aphios Corporation	1st 50	-

Project Number	Project Identifier (Title/Lead Organization)	Data Set	Overall Project Success
91-01-0025	Armstrong World Industries, Inc.	1st 50	-
92-01-0007	Calmac Manufacturing Corporation	2nd 50	-
94-04-0008	Cerner Corporation	3rd 50	-
95-01-0148	Dow AgroSciences (Mycogen Corporation)	3rd 50	-
94-05-0033	Dupont Qualicon (formerly Dupont FQMS Group)	3rd 50	-
92-01-0109	Eagle-Picher Research Laboratory	2nd 50	-
92-01-0122	ETOM Technologies, Inc. (formerly Optex Communications, Inc.)	1st 50	-
95-05-0031	General Electric Company	2nd 50	-
90-01-0126	Hampshire Instruments, Inc.	1st 50	-
91-01-0017	IBM Corporation	1st 50	-
92-01-0103	IBM Corporation	1st 50	-
94-04-0018	InStream	3rd 50	-
94-06-0011	Lucent Technologies (formerly AT&T Bell Laboratories)	3rd 50	-
91-01-0057	MediaBin (formerly Iterated Systems Incorporated)	2nd 50	-
95-08-0017	Medical Analysis Systems (formerly NAVIX)	3rd 50	-
94-01-0074	Monsanto(formerly Agrecetus)	3rd 50	-
92-01-0124	NetOptix Corporation (formerly Galileo Corporation)	1st 50	-
95-07-0011	PCC Structurals	3rd 50	-
95-07-0006	Praxair Surface Technologies, Inc.	2nd 50	-
92-01-0035	Sheffield Automation (formerly Giddings & Lewis)	2nd 50	-
91-01-0071	Thermo Trilogy Corporation	1st 50	-
94-04-0040	Titan Systems (formerly Intermetrics)	3rd 50	-