Genetic Footprinting Making Its Mark

As molecular biologists know, the popular methods to identify gene functionality are labor-intensive. They normally require isolating, storing and characterizing each created mutant individually, an arduous process when you are characterizing numerous mutants. Due to an original procedure invented by Dr. Patrick Brown, an associate professor and HHMI investigator in Stanford's biochemistry department, and Dr. Victoria Smith, a former post-doctoral fellow at Stanford now working at Genentech, this process may have just gotten easier.

Genetic footprinting is a fast, high-throughput method for analyzing gene functionality in microorganisms and for high-resolution genetic dissection of any cloned gene. The critical difference between this and other methods is that genetic footprinting allows mutations to be made, and their consequences for gene function determined, many thousands at a time rather than one at a time.

"Instead of making each mutant and analyzing it independently, you make the mutants and analyze them in parallel," said Rachel Crowley, the graduate student currently working with Brown on genetic footprinting applications. "It has the potential to give you a lot more information."

"We're not crippled by our assumptions," added Smith. "With this method one doesn't have to pick out the certain gene you think might code for something." Instead, all of the possibilities are examined.

Two applications

The genetic footprinting strategy can be applied in two distinct ways to attack two different experimental problems. The first procedure was developed by Smith and Brown to determine the functions of all of the thousands of genes in the genome of a microorganism. The entire genome is subjected to the genetic footprinting process.

Using this method, Smith and Brown examined the biological roles of virtually every gene on one specific chromosome of Brewer's yeast (Sci-Continued on page 2

OTL's Homepage Gets a Facelift and Adds Some Features

With all the places to visit on the Internet these days, just going online and doing a search can be daunting. However, OTL would like to persuade readers to visit its revamped web site at www.stanford.edu/group/OTL, which will help all those associated with or interested in OTL to understand and explore our office further.

At the homepage, users can select from the choices listed in Fig. 1. OTL's homepage naturally contains the basics, including descriptions of the office and staff, frequently asked questions, and directions to the office.

Trusty old Brainstorm is also now located on the web site. Back issues from the Winter of 1993 to Summer of 1997 will soon be accessible.

One of the most recent web site options is the "Featured Technology" category. "Featured Technology" highlights hot new technologies looking for a home. OTL will normally feature a new

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A Sampling of Licenses Granted by OTL in the Last Quarter

<table>
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<tr>
<th>Docket(s)</th>
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The procedure
Both applications have similar procedural steps. The first involves insertion mutagenesis: intro-
duction of a specific DNA into random sites in the targeted gene of interest, or into the entire genome of the microorganism. The result is a library of mutant DNA molecules in the first appli-
cation noted above and a population of cells har-
boring different insertion mutations in their gen-
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The mutagenized DNA molecules or mutant cells from above are then subject to one or more selective conditions, a process called functional selec-
tion. Functional selection determines whether any of the mutations affect the function of the gene or fitness of the cell.

An example of functional selection for genetic footprinting is the analysis of the entire genome of a microorganism is the growth of cells in a medium lacking a nutrient required for cell survival. Cells that contain intact versions of the genes which synthesize the missing nutrient will survive and grow. In contrast, cells containing disrupted ver-
sions of these genes will fail to grow. The next step is to identify the genes which had a role in cell fitness during the selection.

Polymerase Chain Reaction (PCR) is used to identify where the mutations were made. Specific PCR products corresponding to the positions of the mutations in the targeted gene after selection (se-
lected population) and the positions of mutations in each gene before selection (unselected popula-
tion) are compared by gel electrophoresis.

Gel electrophoresis shows a number of "bands," each band representing a position where the gene se-
quence was disrupted by a mutation. The posi-
tion and intensity of the bands indicates the loca-
tion of each mutation and its abundance in the popu-
lation, respectively.

The "footprint" shows itself as any missing bands on the gel. The presence of a band for the unselected wild-type after selection indicates that the gene was not selected.

Future directions
Besides being highly sensitive and accurate, the genetic footprinting procedure is simple to implement in most well-equipped molecular genetic laboratories.

Nifty Fifty Festivities for OTL

O TL's Big 50 festivities, held September 3, 1997, in Alumni Grove at Stanford, was a rousing time complete with music, a video, terrific food and new and old friends. The Big 50 was in celebration of OTL achieving a year-end income of over $50 million (see page 4 for the figures).

Gary Leuenberger, owner of Enchantor Productions, pleased the crowd with his musical talent. His instruments included keyboards, a breath controller and the Yamaha VI70M physical Modeling box which incorporates Sonidius-X™ technology (see Brainstorm, Summer 1997). With these instruments, Gary provided music which included synthesized drum, flute, guitar and saxophone sounds. Overheard was the comment, "I read about Sonidius-X™ in the OTL newsletter, but I had no idea how amazing it is!"

Also for entertainment, OTL showed a new video about the office entitled Connections. The video was produced by the Stanford Commercial and Professional Development (SCPD) and is being used to help introduce inventors, industry personnel and other interested parties to OTL.

However, the main entertainment was conversation. Inventors, lawyers, patent agents, associates and OTLers all converged to enjoy the beautiful weather and one another's company for a momentous occasion in OTL history.

Thank you for joining us in our celebration!
OTL's Homepage Gets a Facelift...
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technology in this section every month.

“Corporate User Info” describes many of the possible interactions between Stanford and industry. You can even see what OTL’s boilerplate license agreements look like.

For the University members of the audience, the next category on the menu will be of interest. This section includes information on the OTL Graduate Fellowship Fund, the OTL Research Incentive Fund, the patent process, Stanford policies associated with technology transfer, a marketing abstract example and much more.

The marketing abstract example is of particular note. OTL encourages inventors to be more involved in the marketing of their inventions. Besides discussing possible industry interests with the Associate handling the invention, OTL would like to elicit the aid of the inventors to write the abstract for the marketing letters. Inventors know their technology best and therefore can write more succinct and comprehensive description.

“Entrepreneurial Information” is a page of links to sites to help Stanford’s many budding entrepreneurs. OTL would like to help companies developing from the Stanford community and hope this site will be of assistance.

“Technology Transfer” is a page of links to other Universities, research centers, patent information sites, and technology transfer information sites.

Now linked to our site is the Sondius-XG™ homepage. This link will transport the user to Sondius-XG™ land, the future in sound synthesis.

Invention disclosures can now be submitted online, but to ensure the security of the information being transmitted, the submitter must have a Stanford SUNet ID. This electronic submission has been added to ease the disclosure process for our inventors and to facilitate the transfer of information to the OTL database.

Last on the list is “Search the Available Technologies Database.” Though this database has been up and running for a while, a new search engine has recently been added that will facilitate finding particular technologies that are available for licensing from Stanford.

OTL currently has descriptions of many of its available technologies located in the online database. However, please contact OTL if the search does not prove fruitful. Some technologies have not yet been released to the web. The database is updated daily.

Please contact Mary Watanabe at mary@otlmail.stanford.edu with any comments or insights on our reformatted and updated homepage.

OTL Fiscal Year 1996-97 (Preliminary Figures)
Total Income: $51.8 Million (M)
Cohen-Boyer DNA Patents:
Total Income: $38.5 M
New Licenses: 31
All Other Technologies:
Total Income: $13.3 M
New Licenses: 122
Companies in which Stanford took equity: 8
Distribution:
OTL Budget: $1.8 M
Other Institutions: $17.9 M
SU Departments: $8.5 M
SU Schools: $8.4 M
Inventors: $7.6 M
Research Incentive Fund: $3.7 M

than other methods.

“I believe that once the key reagent - the transposase or integrase enzyme - becomes widely available,” said Brown, “genetic footprinting will become the method of choice for rapidly dissecting the functional organization of cloned genes.”

For more information on genetic footprinting, please contact Jessica Smith at (650) 723-1586 or jessica@otlmail.stanford.edu.