

# First HFSP Awardees Meeting

## Turin, Italy

### June 17-20, 2001

By Geoffrey Montgomery

#### Introduction



The idea of gathering together awardees of the Human Frontier Science Program (HFSP) at the completion of their grants or fellowships, said General Secretary Torsten Wiesel in his welcoming remarks at the first annual awardees meeting in Turin, "seemed very much in the spirit of this program". For the basic aim of the program, said Dr. Wiesel, is to bring together vanguard groups of scientists from different nations, different disciplines, different cultures: "to create an international community of HFSP scientists who treasure excellence in intellectual pursuits." The June 17-20, 2001 meeting was held high on a hilltop at the Villa Gualino, which with its magnificent views of the red-tiled roofs of Turin in the valley below and the Italian Alps snow-capped in the distance, provided a beautiful setting for interactions between scientists from the HFSP Grants and Fellows programs. A series of oral presentations were held in a well-equipped conference room; there were poster displays by HFSP fellows; and informal "chalkboard sessions" were organized around various sub-disciplines, allowing small groups of scientists to present ongoing work and exchange questions and ideas freely. There was also a Baroque concert with period instruments at the Palazzo Barolo; a sumptuous, many-coursed banquet; lunches at the Villa's cafeteria; and many lively coffee-break conversations on the Villa's sunny verandah.

In one of these coffee-break conversations, HFSP President Masao Ito spoke of the uniqueness and importance of the HFSP program's effort to create, nourish and sustain international networks of scientists. Science is preeminently an international cultural enterprise; yet discussions within individual nations about the support and funding of scientific research, Dr. Ito said, were nearly always rather narrowly framed around questions of what best served the national interest. Moreover, he noted, while most existing life science funding agencies remained organized along traditional disciplinary boundaries, it is now recognized that future progress in the life sciences will depend more and more on combining approaches from different fields of science, an interdisciplinary perspective that is a hallmark of the projects HFSP supports.

#### *Guides for coping with biological complexity*

All the natural sciences are converging on biology, the pioneering molecular biologist Sydney Brenner said on the first day of the conference. With characteristic wit and style, Brenner delivered an opening plenary lecture, titled "Central Problems in Biology in the New Millennium". The central problem, Brenner said, "was to explain the oldest problem in biology: how like begets like." Organisms, he continued, "are unique in nature because they contain an internal description of themselves" - a description written in their DNA sequences. Yet the flood of raw information pouring out of the new science of genomics "has generated a crisis in the biological sciences." There is a great difference, Brenner said, "between information and knowledge. How can we turn this explosion of information into knowledge we can use?"

As will be highlighted in this brief report, Brenner's suggestions for coping with this crisis, and with the immense complexity of biological systems in general, were well reflected in many of the interdisciplinary studies pursued by HFSP grantees and discussed in the oral presentations and posters. One of Brenner's major points of emphasis was that organisms are *modularly* organized. That is, while the human genome contains coding sequences for some 30,000 proteins, the life of a human

cell, like the life of a human city, should not be viewed as a matrix of interactions between 30,000 distinct and separate entities. Cities are organized around modular structures: apartment houses, office buildings, transportation lines and vehicles for taking people from place to place. Similarly, Brenner said, "proteins don't act alone, they act in *assemblies*." For instance, there are 65 proteins in the complex that splices out introns from genes, and 100 protein components in the tunnel-like nuclear pore complex. Thus from this modular perspective the complexity of the cell can be reduced from 30,000 distinct entities to perhaps 3,000 modular, multi-protein complexes. Moreover, these multi-protein modules are not simply floating and interacting freely in a kind of intracellular ocean, but are instead generally localized to specific cellular compartments, a fact that reduces by another two orders of magnitude the matrix of molecular interactions with which biologists must grapple. Analysis of such multi-protein modules localized to various cell compartments—from the histone assemblies packaging DNA in chromosomes to the adhesion molecules linking neurons in the brain—were a major focus of presentations at the HFSP awardees meeting.

*Mapping  
organismic space  
onto genetic space*



What biologists are seeking to understand in the new post-genomic era, said Brenner, is "how organismic space maps onto genetic space." And in the very first talk of the Turin conference, Paul Brakefield (Leiden University, The Netherlands) described a collaborative research project, undertaken with the laboratories of Sean Carroll (University of Wisconsin, USA) and Vernon French (University of Edinburgh, U.K.), demonstrating how this most fundamental of questions was finally becoming approachable by biological science. Brakefield, an evolutionary biologist by training, spoke of his lifelong "fascination with biodiversity. What are the roots of biodiversity?" Until recently, it was impossible to picture in any concrete way the genetic processes by which the vast array of animal and plant

forms populating the earth evolved from a small set of common ancestors, or to determine how these plants and animals, through new genetic variations, continue to create novel forms that are acted on by natural selection. But this situation has changed dramatically over the last decade and a half, as a universally-conserved "toolkit" of developmental control genes and developmental signaling pathways have been unveiled in a few model organisms, such as the *Drosophila* fruitfly and the *C. elegans* nematode (a worm, it should be remarked, introduced into modern biology by Brenner). Work in the Carroll laboratory demonstrated that the key developmental toolkit gene *Distalless* and the hedgehog signaling pathway are involved in the generation of the striking eyespots present on the wings of many butterflies and moths. These eyespots, which vary greatly in color, size, shape and in their placement on wings, have been intensively studied by evolutionary biologists, and have been shown in different species in the wild to be used during sexual selection and as predator-avoidance and predator-deflection devices. Thus insect eyespots are an ideal, and aesthetically beguiling, model system for studying how variations in "genetic space" - that is, in sequences of DNA - may generate variations in "organismic space" - the visible forms of animal bodies.

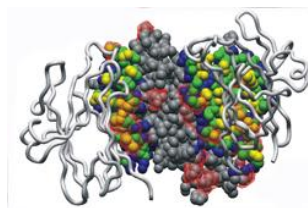
Brakefield and his colleagues have combined artificial selection experiments and analysis of mutants of an African butterfly, *Bicyclus anynana*, with gene expression, tissue transplant, and quantitative trait loci analysis. The HFSP grant, said Brakefield in a conversation after his seminar, has allowed frequent visits between members of the international and interdisciplinary team, and the deployment of technologies in his own laboratory, such as gene expression monitoring through confocal microscopy, that would not otherwise have been possible. Together the investigators have mapped some of the variations in eyespot forms to known developmental genes such as *Distalless*. And here Brakefield enlarged upon one of Brenner's key themes: that species in a phylum contain the same basic repertoire

of genes, and that the major differences between these species — and almost certainly the major differences between closely-related species such as chimpanzees and humans — arise not from the evolution of new protein-coding sequences, but instead from evolutionary changes in flanking regulatory sequences. The regulatory elements of developmental genes tend to be long and intricate; it is these regulatory elements that determine how adjoining coding sequences are deployed at different developmental stages in different tissues to shape animal bodies. "And I suspect," said Brakefield, "that these regulatory elements occur as *modules*." That is, a gene like *Distalless* seems to be controlled by a modular array of DNA regulatory sequences, and different modules independently subserve different functions - one set driving *Distalless*'s control of limb formation in the trunk of the body early in development, another set driving its control of eyespot formation in wings towards the very end of development. This independent, modular organization is highly "evolvable," so that changes in the eyespot regulatory elements may permit the eyespots of an insect to change dramatically without altering the shape of its limbs.

"If we can succeed [in mapping eyespot changes to specific DNA changes in these regulatory modules], said Brakefield, who will continue his collaboration with the Carroll and French labs beyond the course of the HFSP grant, "we really will have integrated evolutionary biology with developmental biology in a very powerful way. If we do it for traits such as eyespot development where we also know something about what is going on in functional terms in natural populations, we will really in my mind be telling the whole story: from the genetic variation, which produces evolutionarily relevant phenotypes, through to what actually happens to those phenotypes during natural selection in the wild. To me that's the great goal."

In his plenary talk, Brenner spoke of the additional scientific value accruing from earth's biodiversity: "such is the diversity of organisms," he said, that an investigator could be reasonably confident that somewhere on earth "there is an organism that will display in some exaggerated and convenient form an aspect [of a biological phenomenon you wish to study] that allows you to do important experiments." The use of different organisms was well illustrated in several other HFSP-supported investigations into developmental mechanisms. To name a few: Krzysztof Jagla (INSERM, France) presented a collaborative research project that, working in *Drosophila*, mice and zebrafish, identified a group of conserved transcription factors, the ladybird homeobox genes, as key regulators of muscle and heart development. Patrick Lemaire (Institute of Developmental Biology of Marseilles, France) presented collaborative studies of primary body axis formation utilizing both amphibians and ascidians—small, transparent and convenient invertebrate chordates which were once a favorite of classical embryologists, fell out of fashion, and which are now experiencing a renaissance due to the application of modern molecular methods. HFSP Fellow Dana Schroeder (The Salk Institute, USA) presented a poster describing provocative new insights, using mutants of the *Arabidopsis* weed, into the molecular mechanisms by which light regulates plant growth and development. And Elena Alvarez-Buylla (Instituto de Ecología, Mexico) made the most exotic use of natural biodiversity with her intriguing collaborative studies of the molecular genetics and evolution of *Lacandonia schismatica*, a native of the Lacandon forest in Mexico, and the world's only known flowering plant with inverted reproductive whorls.

*Wiring up nervous  
systems and  
probing brain  
functions*



The Awardees meeting had particularly strong representation in the area of developmental neurobiology. To cite some examples: Andrew Boyd (Queensland Institute of Medical Research, Australia) presented collaborative research on ephrin ligands and receptors, which are critical regulators of many aspects of cell migration and axonal guidance. To date, knock-out transgenic mouse models of ephrin function have proved largely disappointing, displaying little or no phenotypes, presumably due to genetic redundancy in the

large ephrin receptor and ligand family. However, Ephrin A4 knockouts generated by Boyd's laboratory produced mice with strong motor phenotypes: a "kangaroo-like gait, and global clumsiness and paucity of movement," providing important new material in which to investigate ephrin function in the nervous system. HFSP fellow Thomas Biederer (University of Texas, Southwestern Medical Center; USA) presented his discovery of a possible new synaptic adhesion molecule. And in similar vein, HFSP fellow Shernaz Bamji (University of California, San Francisco; USA) presented a fascinating poster on her recent studies of a key class of synaptic adhesion proteins, the cadherins.



As a graduate student at McGill University in Montreal, Bamji had become deeply interested in the problem of spinal cord regeneration, and met many patients who had suffered spinal injuries. At that time, Bamji was studying neurotrophins, key molecular factors involved in neuronal survival and death. For her postdoctoral studies, however, and in preparation for her career as an independent scientist, Bamji wanted to advance along the pathway of understanding how damaged neurons might be made functionally whole again. She knew that many labs were studying cell survival and death, and many others were looking at the problem of axonal regeneration. "I thought the next big thing that people would want to know," she said, "is after you learn more about how to keep these neurons alive, after you facilitate the regeneration of their axons, how are you going to hook them up appropriately with their synaptic partners? And I found that the cadherins are a very good candidate for doing this."

At the HFSP meeting, Bamji presented two important new sets of findings relating to cadherin function. Working in vivo with a special transgenic mouse model provided by a collaborator, she showed evidence that the cadherins not only function as molecular "glue" to bind pre-and post-synaptic membranes together: additionally, through the phosphorylation of an associated protein, B-catenin, they may be involved in synaptic plasticity during adult life. In the second set of studies, working with cultured hippocampal cells in vitro, Bamji made extremely intriguing observations suggesting that cadherins, working through B-catenin and another linked multi-protein module called the Lin-2 complex, may act during development to mark "hot spots" on neurites where future synapses will be laid down. "This work is quite interesting," she says, "because it indicates that not only can neurons use cadherin molecules to find the correct synaptic partner, but also that when they have found their partner, the cadherins might facilitate synaptic formation."

In Montreal as a graduate student, Bamji applied for postdoctoral fellowships from HFSP and from the Canadian Medical Research Council to support her planned cadherin studies in Louis Reichardt's UCSF laboratory. She was awarded both fellowships and accepted the one from HFSP. Why? "It was more prestigious," she said, and the additional money for research included in the HFSP fellowship gave her added confidence that she would be able to pursue leads in her research even if her host laboratory lacked key resources. Only a week into her stay in the Reichardt lab, HFSP funds permitted her to attend a conference at which she learned that the B-catenin transgenic mouse she had planned to make in San Francisco had already been made by a German laboratory; she entered into collaboration with this laboratory and used the mouse in her in vivo studies of cadherin function. At her poster at the awardees meeting, she said that a group of HFSP fellows had gone to dinner together in Turin the evening before, "and we were talking about the freedom that HFSP fellowships give you."

If the central problem of genomically-oriented biology is, as Brenner said, how organismic space maps onto genetic space, then it might be said that the central problem of the brain sciences is how outside-world space maps onto neural space: how cells of the brain provide "an internal description" of past and present

experiences. Stuart Firestein (Columbia University, USA) presented an innovative collaborative research project aimed at understanding how olfactory information is encoded by cells at various stages of the olfactory pathway. These interdisciplinary studies of olfactory processing employed sophisticated transgenic techniques and physiological recordings, as well as molecular pharmacological methods by which one of the major unsolved issues in olfaction - how olfactory receptors actually recognize the odoriferous molecules we "smell" - was addressed. Indeed, Firestein's presentation showed that studies of the "chemistry of organization" (see below) are beginning to make major contributions to our understanding of neural functions. Vittorio Gallese (University of Parma, Italy) presented a continuing collaborative series of studies of a remarkable set of cortical cells called "mirror neurons." Mirror neurons fire both when a macaque monkey observes a specific action - such as a reaching motion by a hand—and when that same action is performed by the monkey. This has suggested that a common neural code exists linking the observation of actions with internal reproductions of these actions; this provocative work has also been pursued using a variety of brain imaging methods in humans. Edmund Rolls (University of Oxford, UK) presented collaborative physiological and computational studies of how the hippocampus, the mammalian brain region critical for the formation of long-term memories, represents spatial information about objects. Unlike many areas of the visual cortex, in which there is a strongly topographic arrangement of cells responding to different points of visual space, there appears to be no systematic organization of space cells in the hippocampus, a network property, Rolls said, predicted by the computational model developed by his team.

*Finding a common language in the chemistry of organization*

The First Annual HFSP awardees meeting was designed to include both brain scientists and molecularly-oriented scientists, in order to better forge interdisciplinary interactions between the two fields. Yet, as Brenner noted in his plenary address, ambitious interdisciplinary projects in the past have floundered because "nobody spoke the same language." However, said Brenner, "it is important to realize that we do have a very important common language, and that is the language of chemistry." He briefly reviewed the application of chemistry to biological problems, which began in the middle of the 19th century, with the study of how biological systems handle matter and energy: the chemistry of biological material and energy formed the basis of classical biochemistry. Then came the mid-20th century revolution in molecular biology, which "gave us the chemistry of information—a remarkable statement, when you think about it, that you could study biological information by looking at chemical aspects" of biological entities like DNA and RNA. "And I predict," said Brenner, "that the next phase in which chemistry will be important will be the chemistry of organization, the chemistry of how the components of biological systems interact. We will want to know how lots of [biological molecules] recognize each other"

Indeed, probing the nature of molecular recognition was the subject of the meeting's second plenary lecture, by leading structural biologist, Wayne Hendrickson (Columbia University College of Physicians and Surgeons, USA), who gave a tour de force presentation that focused on the structure of two HIV virus proteins called gp120 and gp41. The HIV virus uses these proteins to recognize and bind to T-cells of the human immune system; and these proteins present perhaps the best target for AIDS vaccines. Nevertheless, Hendrickson's studies of these proteins elucidated a variety of cunning tactics the HIV virus has evolved to evade antibodies and other immune responses, tactics which have made the design of an effective AIDS vaccine one of the greatest challenges faced by biomedical science.

In his introductory remarks, Hendrickson noted that this "is the heyday for structural biology," remarking how "glorious it is to see what we can learn and understand thanks to seeing atomic detail, because as we've seen from many of the talks and



posters here, it's not just atomic detail that we get, it's the big biological picture that emerges from this detail." Projects aimed at elucidating the structural biology and "chemistry of organization" of modular cellular complexes included collaborative studies by Anita Corbett (Emory University School of Medicine, USA) of the nuclear pore complex, which regulates the traffic of macromolecules between the nucleus and the cytoplasm. These studies used traditional biochemical and crystal structure methods, as well as novel techniques such as atomic force microscopy and fluorescence resonance energy transfer. HFSP fellow Alexey Rak (Max-Planck Institute for Molecular Physiology; Dortmund, Germany) probed the molecular-recognition machinery involved in the transport of vesicles between the membranous organelles of eukaryotic cells, presenting an extremely impressive series of new crystal structures of proteins involved in this process. Irwin Davidson (IGBMC, France) presented collaborative structural and functional analysis of molecules of the protein-complex regulating RNA polymerase II transcription. And HFSP fellow Kerstin Bystricky (Institute for Molecular Biology and Biophysics, Zurich, Switzerland) presented the fruits of two years of labor, during which, for the first time, she constructed fully synthetic chromatin arrays in which to study higher order chromatin organization, and the "histone code" by which DNA is packaged inside the nuclei of living cells.

*Unveiling new  
insights and  
creating new tools  
at the frontiers of  
biology*

In the conference's concluding afternoon session, Louise Cramer (University College London, UK) gave a spectacular demonstration of how chemistry can provide not only a common language for biologists, but powerful new tools for probing critical life processes. Cramer's long-standing interest, developed as an HFSP fellow in the Harvard Medical School laboratory of Tim Mitchison, has been to understand how the different elements of the cytoskeleton mediate cell movements. Cell migration, for instance, is at the heart of many developmental processes, and of axonal guidance, immune cell function, wound healing, and the invasive movement of metastatic tumor cells. Cramer described the comprehensive and highly ambitious efforts by an HFSP-supported team of chemists and biologists, which included Mitchison, aimed at isolating and synthesizing small molecule inhibitors (and activators) that could be used to target and probe the functions of specific cytoskeletal proteins. In a beautifully presented talk, in which video-microscopy and animation were integrated into a lucid display of models and data, she highlighted the insights provided by three inhibitors---two of which were newly discovered during the course of the project---into the process of cell migration and mitosis. Particularly interesting, said Cramer, was the isolation and synthesis of an inhibitor, called blebbistatin, of myosin type II, which offered new perspectives on the different roles the classical "movement molecules" actin and myosin play in migrating cells. Video-microscopy of blebbistatin-treated cells showed how inhibition of myosin II resulted in the loss of the morphological polarity associated with directed cell movement, and the apparent inability of the cell body to push off from the substrate and of the tail of the cell to retract. Another inhibitor studied during the course of the project had shown that actin disassembly is tightly linked to the protrusion of the leading edge, or cell margin, of migrating cells. "And this is really exciting to us," said Cramer, "because we've often thought that cell migration is the complex coordination of different actin-force generating mechanisms, one at the front and one in the middle, and this is the first direct data really showing that myosin is needed for the middle and rear of the migrating cell, but actin assembly and disassembly is acting at the front of the cell."

In his welcoming remarks, Torsten Wiesel emphasized how recent changes in the HFSP program were designed to better promote "a sense of adventure and risk-taking" by HFSP grantees. While NIH proposals, for example, often require the investigator to have already performed a large bulk of the experimental project for which support is being sought, HFSP, said Dr. Wiesel, aims to provide "seed money to start projects you otherwise could not have, and in this way create new frontiers of science."

The pioneering joint project described by Louise Cramer seemed to marvelously

exemplify many of the qualities described by Dr. Wiesel. The HFSP is working, he said, "to create a track within the system" whereby creative young scientists can advance from productive postdoctoral studies to independent careers as frontier scientists running labs of their own. Cramer began her studies of the cytoskeleton as an HFSP fellow; and at the opening of her talk she noted that the pioneering new project she described "would not have been possible without a Human Frontier collaborative grant. Also I'm the youngest member of the team, and I started my independent career at the beginning of this grant. So Human Frontier is greatly responsible for my lab beginning to flourish now." Cramer concluded her presentation by noting that the HFSP grant had served its seed function extremely well, as "we will be continuing our collaboration beyond the end of the grant period," characterizing other intriguing inhibitors of cytoskeletal function that are now in the pipeline, working with "the hope that in the future we'll be seeing many more useful inhibitors that will be freely available to the biological community."