

Community Sequencing Program: Project Proposal
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Project Title: PHYSCOME: The Moss *Physcomitrella* Genome Project

Abstract: PHYSCOME: The Moss *Physcomitrella* Genome Project

The moss, *Physcomitrella patens*, is becoming widely recognized as an experimental organism of choice not only for basic molecular, cytological, and developmental questions in plant biology, but also as a key link in understanding plant evolutionary questions, especially those related to genome evolution. *Physcomitrella* is well-placed phylogenetically to provide important comparisons with the flowering plants. In terms of evolutionary distance, *Physcomitrella* is to the flowering plants what the *Drosophila* is to humans! Having the full *Physcomitrella* genome available would greatly inform bioinformatic comparisons and functional genomics in plants, just as the mouse, *Fugu*, and *Drosophila* genomes have informed animal biology.

Over 20 labs worldwide perform molecular analyses. Sexual crossing, somatic hybridization and the generation of transgenic plants is routine. cDNA, genomic and insertional libraries, and RNA interference lines are available to study the function of key genes and processes. The simple morphology allows informative imaging of tagged macromolecules and the direct observation *in vivo*, of intracellular processes involved in tip growth and positioning of the plane of cell division, key processes in plant morphogenesis. These methods are allowing the study in moss of critical higher plant processes, e.g. phytohormone signalling (i.e. auxin, cytokinin and especially abscisic acid), cell polarity, tropisms, and photomorphogenesis (i.e. phytochrome and cryptochrome). *Physcomitrella* is the only plant that undergoes homologous recombination with a frequency that allows easy targeting of genes for replacement and elimination. This has been crucial for the development of other model systems such as yeast, allowing gene function in higher organisms to be predicted. To extract completely the full potential of the unique experimental system of *Physcomitrella*, and to apply data from this organism to higher plant models such as *Arabidopsis* and rice, a comprehensive genome sequencing project is now required.

Physcomitrella is uniquely positioned to utilize a fully sequenced genome. A public database of over 80,000 ESTs, arranged in about 22,000 contigs is available, as is a BAC library and an Agilent gene chip containing 60-mers of each contig. These resources are gaining usage within the wider plant community for comparative genomic and gene expression studies. Through workshops giving training in moss techniques, and an annual meeting where experimental results are displayed and discussed, this system continues to gain interest, in both the U.S. and worldwide. We now have commitments from an international group composed of representative from the U.S., U.K., Germany, and Japan to request support for a comprehensive genome sequencing project.

This request to the U.S. JGI is to provide raw sequence data from the ~500Mb *Physcomitrella* genome that will be supplemented with funding support from our collaborators to aide in assembly, annotation, gap closure, etc. Our plan would be to center “genome finishing” at one site, e.g. the Genome Sequencing Center at Washington University. The international moss community is committed to establishing a map of the *Physcomitrella* genome, initially using molecular markers (AFLPs and RFLPs), and efforts are already being coordinated through our international team to assemble an integrated physical, genetic and molecular map. Thirteen different isolates of *Physcomitrella patens patens* and three of *Physcomitrella patens californica* are available to Dr. D. Cove who will set up crosses between suitable ecotypes and a standard laboratory strain to generate progeny for analysis. We will include in this comprehensive genome sequencing program a substantial genetic and physical mapping component. EST

sequences will be mapped to BAC clones and placed within BAC contigs, in order to provide a skeleton of molecular markers to assist in anchoring the assembly of shotgun-sequence-derived contigs.

Scope of Work:

A full understanding of the *Physcomitrella patens* genome will only be achieved through a comprehensive genome sequencing program - a project of such scope as to require the commitment of considerable resources on an international scale. We propose a five to eight-fold coverage of the entire genome of about 500 Mb. An important additional component of such a project is the determination of both a genetic and physical map of the *Physcomitrella* genome. We believe we as a community are now in a strong position to initiate such an analysis through an international effort which includes a team representing not only the U.S. (Drs. Quatrano and Mishler) but the U.K. (Drs. Cove and Cuming), Germany (Dr. Reski) and Japan (Dr. Hasebe) to apply molecular marker technology to generate a genetic linkage map of *Physcomitrella* that can subsequently be integrated with a physical map derived from current (and future) genomic and EST resources. This will be based on a combination of AFLP and RFLP linkage mapping, BAC-fingerprinting and end-sequencing, and the placement of molecular markers (ESTs, AFLP and RFLP sequences) on BAC contigs, using resources developed in the course of the different sequencing programs. We expect substantial additional sequence resources from our collaborators at the National Institute for Basic Biology (Okasaki, Japan) will also be available for this project. The very active research being undertaken both at this Institute and at the University of Freiburg (Germany), underlines the vigorous international interest in *Physcomitrella* genomics.

Technical Information and Available Resources:

The genome size of *Physcomitrella* is ~500 Mb on 27 chromosomes (Schween *et al.*, 2003). The G+C content of coding regions is 50% (compared with 44% for *Arabidopsis*). In the course of the EST sequencing program, we constructed a BAC library from *Physcomitrella* that we estimate to contain *ca.* 50% of the *Physcomitrella* genome (based on our analysis of insert sizes and the frequency of successful gene isolation thus far). Additionally, we have obtained substantial sequence information from four BAC clones, comprising some 400 Kb, that sheds some light as to the nature of the *Physcomitrella* genome:

- (i) The *Physcomitrella* genome appears to be similar to other large eukaryotic genomes, in that it contains genes interspersed among long stretches of non-coding DNA. More specifically, one BAC region of approximately 53kb resulted in 12 strong BLASTN hits on Physcobase ESTs, including one duplicated gene pair ("putative BURP domain protein").
- (ii) The non-coding regions tend to be relatively AT-rich, and include some highly repetitive motifs that make the assembly of long contigs challenging.
- (iii) Like most other plant genomes, the *Physcomitrella* genome contains a significant number of sequences that can be identified as of retrotransposon origin. – The representation of retrotransposon-related sequences in the EST database strongly indicates that a population of these sequences are active in *Physcomitrella*.

Scientific Importance:

There is a growing national and international community that represents a broad biological community that will utilize this resource. At the Sixth Annual Moss International Conference (*Moss 2003*) this past September in St. Louis, over 75 scientists attended representing over 20 countries. Although the areas represented at this gathering included primarily cell/molecular /genetic presentations, an immediately preceding meeting at the Missouri Botanical Garden on bryophyte molecular systematics resulted in considerable overlap in attendance. Attendees from both disciplines interacted on topics of mutual interest. This group unanimously agreed that sequencing the entire genome of *Physcomitrella patens* is of the highest priority; it would produce a quantum leap in the study of mosses and, because of the comparative method, the whole of green plants and by extension the eukaryotes. *Moss 2004* will be held from Sept. 12-15, 2004 in Freiburg, Germany (www.plant-biotech.net/moss2004).

Post-sequencing Plans:

The international moss community, motivated and organized at the Moss 2003 meeting, and facilitated since via the IBRIS email group, is committed to establishing a map of the *Physcomitrella* genome, initially using molecular markers. To this end, thirteen different isolates of *Physcomitrella patens patens* and three of *Physcomitrella patens californica* have been sent to Dr. R. Reski at the University of Freiberg, Germany, who is in the process of analyzing their karyotype and determining their genome sizes. Dr. D. Cove will then set up crosses between suitable ecotypes and a standard laboratory strain to generate progeny for analysis. Funding is currently being sought by the U.K. lab to carry out this analysis at Leeds University, England. If funding is forthcoming a genome framework should be available by mid 2005, into which BAC sequences can then be integrated.

Technical Challenges:

Because of the content of repetitive, low-complexity and multicopy sequences, we will include in this genomic sequencing program a substantial genetic and physical mapping component, in which EST sequences will be mapped to BAC clones and placed within BAC contigs, in order to provide a skeleton of molecular markers that should assist in anchoring the assembly of shotgun-sequence-derived contigs. Additionally, the development of a linkage map based on molecular markers (AFLPs and RFLPs), now being pursued by the U.S. and U.K. labs, will provide genetic markers that can be placed directly on the physical map. This should be greatly facilitated by the planned construction of a more comprehensive large-insert BAC library of *Physcomitrella*. If this request for sequencing the *Physcomitrella* genome is approved, we would immediately coordinate our efforts with the genome sequencing group at JGI in constructing this library.

Scheduling Requirements:

We do not have any specific scheduling requirements.

Project Description:

The genomics revolution has impacted all fields of biology; from protein/cell structure to the structure of populations. One of the greatest uses of the data and approaches associated with this advance is in the area of comparative genomics. A recent synthesis of phylogenetics and genomics – two fields once estranged – is beginning to form a new field that could be called "phylogenomics" (Eisen, 1998). Much can be learned about function of genes by examining them in one organism. However, a much richer array of tools is available using a comparative approach. Close sister-group comparisons between lineages differing in a critical phenotype (e.g., desiccation or freeze tolerance) can allow a quick narrowing of the search for genetic causes. Dissecting a complicated, evolutionarily advanced genotype/phenotype complex (e.g., development of the angiosperm flower), by tracing the components back to simpler ancestral systems, can lead to quicker understanding.

Using such a comparative approach has and will continue to impact heavily our understanding of the evolution of macromolecular, cellular, and population structure(s), as well as the evolution of whole genomes and basic cellular processes. It will also aid us in the major challenge in the next decade, i.e. to elucidate the unknown function(s) of a large number of genes in completely sequenced genomes, and to discover how these genes interact to perform specific biological processes. For example, Stuart, et. al. (2003) used microarray data from four completely sequenced genomes (yeast, nematode, insect and human) to show co-expression relationships that have been conserved across a wide spectrum of animal evolution. Not only did they elucidate the function of several unknown genes in basic core functions (e.g. cell cycle, secretion, cell proliferation), they identified gene networks that were "animal specific" as well as interactions between newly evolved and ancient gene sets.

Hence, comparative genomics allows one to use not only sequence similarities, but also phylogenetic relationships to confirm and/or to establish gene function and interactions.

Understanding of relationships of the green plants.

The renowned "Deep Green" collaboration (see below) has assembled over the last decade a large and well-resolved phylogeny for the green plants. Results to date suggest that the green plants appear to be composed of two major lineages and a residuum of unicellular micromonadophytes (Fig. 1; for an elaborate tree shown in hyperbolic space see: <http://ucjeps.berkeley.edu/TreeofLife/hyperbolic.php>). One of these major lineages contains the bulk of the classical green algae (Chlorophyceae, Pleurostrophyceae, and Ulvophyceae). The other contains the land plants plus some of the former green algae. Within the land plants, there are four major lineages, i.e., liverworts, hornworts, mosses, and the tracheophytes (vascular plants). Within the tracheophytes, the lycophytes (club mosses) are sister to all other tracheophytes. Ferns and their allies are sister to the seed plants. Within the seed plants, morphological and molecular data currently provide conflicting topologies for the five extant lineages (cycads, *Ginkgo*, conifers, Gnetales, and angiosperms). Further work is underway to resolve relationships among lineages of seed plants, and other uncertain places in the phylogeny, funded by the National Science Foundation Tree of Life program (see: <http://ucjeps.berkeley.edu/TreeofLife/>).

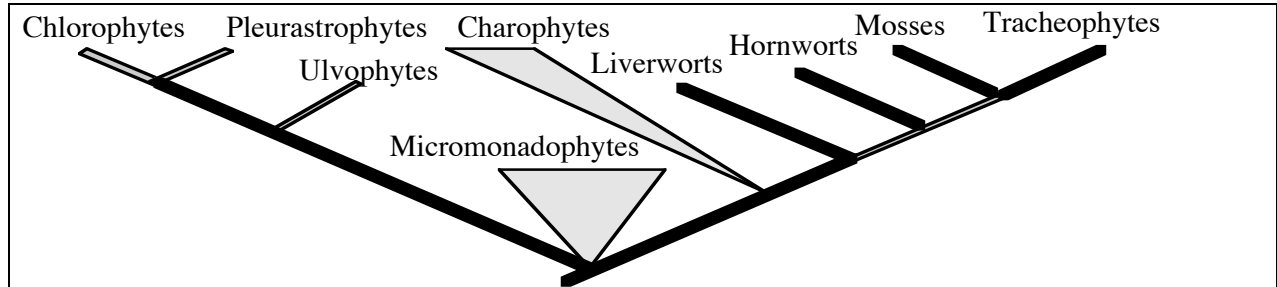


Fig. 1. A summation of the currently hypothesized cladistic relationships of green plants, along with an indication of places in the phylogeny where uncertainty is greatest (stippled). *Chlamydomonas* is a member of the farthest left clade, while flowering plants are nested well up in the farthest right clade. *Physcomitrella* is a moss.

The more robust parts of the current cladogram can serve as a framework for evolutionary interpretations. It appears reasonably well supported, for example, that multicellularity arose at least twice in the green plants. The diversification of life-history strategies is becoming clearer; from a primitively haplontic life cycle, alternation of generations and diploid-dominant life-cycles arose at least twice each. The habitat transition in the movement of plants to land was from fresh water, not from salt water. Within the land plants, several morphological transformations can be reasonably postulated at present, such as the origin of branched, multisporangiate plants from unbranched, unisporangiate ones, and the radiation of types of conducting cells. In addition to these ecological and morphological inferences, we could begin to understand genome evolution in the same manner, once some major gaps in the phylogenetic coverage are filled.

The phylogenetic distribution of current plant genomics, and the need for a completely sequenced moss genome.

Two angiosperm genomes have been sequenced, the dicot *Arabidopsis* (<http://arabidopsis.org/>) (The Arabidopsis Initiative, 2000) and the monocot rice (<http://rgp.dna.affrc.go.jp/>) (Goff, *et al.*; Yu, *et al.*, 2002). More recently, the genome of a single-celled alga, *Chlamydomonas*, that belongs to the other main lineage of the green plant evolutionary tree (a member of the Chlorophyceae, in the far left-hand branch of Fig 1), has been completed (<http://genome.jgi-psf.org/chlre1/chlre1.home.html>), and a few other angiosperms have genome projects underway (including the *Populus* project at JGI).

Unlike the case in animal genomics, where the mouse, *Fugu*, *Drosophila*, and *Caenorhabditis* genomes (spaced nicely along the phylogenetic tree sequentially more distant from humans) allow powerful evolutionary comparisons with the human genome, no evolutionary “intermediate” plant genomes have been completed to supplement those available in angiosperms. The separation between *Chlamydomonas* and the angiosperms is about 1 billion years and an enormous gap in complexity. To sequence a genome that is intermediate

phylogenetically, from a plant that is intermediate in complexity, would link together these widely separated lineages, thus greatly increasing the power of comparative genomics. With such information, not only could gene function and relationships be identified in plants, but also how such associations may be conserved between plants, fungi, and animals. One strategic phylogenetic link between the aquatic, single-celled plants (e.g. *Chlamydomonas*) and the flowering plants (e.g. *Arabidopsis* and rice) that is missing is a representative of the first multicellular, non-vascular land plants, i.e. the bryophytes. The moss *Physcomitrella patens* is the plant in this group that is most heavily studied and that possesses a large number of traits that makes it the preferred target for the next whole genome of a plant to be sequenced. It is an important system in its own right, and perfectly positioned phylogenetically (separated by approximately 500 million years from the flowering plants) to begin to fill in the gap between *Chlamydomonas* and the angiosperms.

Physcomitrella patens – an emerging model system.

Physcomitrella has been developed as a model system to study plant gene function (Cove, 2000; Reski, 1998;1999). The moss can be cultured axenically on a very simple defined medium containing only inorganic salts. Growth is rapid both on agar or in liquid media. Because of *Physcomitrella*'s capacity for tissue regeneration, tissue for DNA, RNA, protein analysis or for protoplast isolation, can be obtained simply by inoculating tissue that has been blended on to agar medium overlaid with cellophane, and incubating for a week. This generates about 500 mg fresh weight of tissue, and can yield about 5×10^6 protoplasts, with a regeneration rate in excess of 90%. The life cycle can be completed in sterile culture in about two months, and since the dominant phase of the life cycle, the gametophyte, is haploid, mutant phenotypes can be observed directly in progeny. This removes the need to carry out F2 or test crosses, halving the number of generations required for genetic analysis, and eliminating the need to produce inbred lines. Mutant isolation and characterization is also facilitated by the predominantly haploid nature of its life cycle, as mutant phenotypes can be observed directly following mutagenesis. Diploid gametophytes can be obtained by regeneration of the diploid tissue of the sporophyte generation, but are also obtained in the laboratory by the selection of somatic hybrids, following protoplast fusion. The diploid lines generated allow dominance and complementation to be determined.

Physcomitrella utilizes the same hormones as flowering plants, and the effects of auxins, cytokinins and ABA (Knight, *et.al.*, 1995) have been studied, utilizing mutants with altered response to these hormones. Protonema, the filamentous developmental stage, provides unrivalled opportunity for the study of cell polarity. Filaments extend by growth only at the tip of the apical filament cell. Growth direction is sensitive to both light and gravity inputs and mutants abnormal in their directional responses to these stimuli have been characterized physiologically. In addition, mosses are a rich source of secondary metabolites which has led to considerable commercial interest.

Physcomitrella may be transformed, either using PEG-mediated direct DNA uptake by protoplasts (Schaefer *et al.*, 1991) or microprojectile bombardment of intact tissue (Sawahel *et al.*, 1992). Both techniques give high rates of transformation. The finding that recombination

occurs at a very high frequency between sequences in transforming DNA that are homologous to genomic sequences (see Schaefer, 2002), has reinforced the utility of *Physcomitrella* as a model organism. The frequency of targeting genes for disruption and/or replacement is as efficient as yeast and orders of magnitude more efficient than any other plant system. The recently, RNA interference (RNAi), both transient and stable has been shown to occur in *Physcomitrella* (Bezanilla, *et al.*, 2003), allowing the investigation of functional redundancy (exhibited with multigene families), as well as lethality (especially in the predominantly haploid life cycle). Together the homologous recombination and RNAi techniques place moss in a unique experimental position to understand the functions of plant genes and their interactions.

The moss genome is estimated to be about four times the size of *Arabidopsis* and slightly larger than rice, not large by the capacity, cost and speed of DNA sequencing today. Resources that have been developed for molecular studies, include a very extensive EST collection, comprising 80,000 ESTs in the public database (moss.nibb.ac.jp) and over 100,00 in a private database (Rensing, *et al.*, 2002). The transcriptome has been characterized (Nishiyama, T., *et al.*, 2003) and not unexpectedly, this analysis has revealed new gene sequences not found in other plants or animals, but it has also identified many genes with high sequence similarity to genes from cyanobacteria, algae and flowering plants, with about two-thirds of the moss sequences having a high degree of similarity with *Arabidopsis*. In addition to these resources, cDNA and genomic libraries are available, as are plasmids suitable for transient and stable transformation by bombardment and DNA uptake into protoplasts (www.moss.leeds.ac.uk). A BAC library is also available and will be used to generate a physical/genetic map/molecular map of the moss genome.

Relationship of this proposal to existing groups and collaborations.

"Deep Green" -- The Green Plant Phylogeny Research Coordination Group (GPPRCG). Back in the early 1990's, considerable data were becoming available bearing on green plant phylogeny, and the community was clearly poised for rapid progress in this area as a result of recent technological, theoretical, and computational improvements. However, several obstacles remained. No mechanism existed for attacking this major effort in a cooperative, coordinated manner. Data sets derived from different molecules and different morphological character systems rarely included the same basic taxa, thus they could not be compared. Current analytical software, and the concepts behind it, needed improvements to handle analyses of this size and complexity, as did data storage and retrieval software. The GPPRCG was formed in 1994 in order to remedy these shortcomings by facilitating interactions between distinct research groups, and has been spectacularly successful. A full account of the meetings and progress of the GPPRCG can be found at: <http://ucjeps.berkeley.edu/bryolab/greenplantpage.html>.

The GPPRCG has continued to function after its initial grant expired, and now consists of a set of seven independent but coordinated research grants (listed with links from <http://ucjeps.berkeley.edu/TreeofLife/related.php>). The present proposal would interface with all these projects, facilitated by Mishler's role as chair of the GPPRCG executive committee. The GPPRCG has always placed a heavy emphasis on student involvement and training. The present

proposal will continue that tradition, expanding training activities from workshops and symposia into the laboratory, facilitated by workshops sponsored jointly with the "Deep Gene" RCN.

Related Research Coordination Networks. Two related Deep Green NSF RCN proposals were funded -- one ("Deep Gene" Mishler, PI) to coordinate genomics and phylogenetics, the other ("Deep Time" D. Soltis, PI) to coordinate paleontology and phylogenetics. The currently proposed Physcome Project will interface with both of these RCNs (particularly the former, which is dedicated to furthering phylogenomics in plants). Joint meetings of these networks will maintain communication and lead to joint sponsorships and colloquia (e.g., a joint workshop on phylogenetics for molecular biologists and paleobotanists). In addition, we have close ties with the Gramene NSF RCN and the Plant Ontology Consortium which will prove very useful for incorporating our results into general discussions of bioinformatics and the structuring of databases.

NSF Plant Genome projects. We will also interface closely with projects currently funded by the NSF Plant Genome Program, in particular The Floral Genome Project (FGP). The FGP focuses on the origin, conservation, and diversification of the genetic architecture of the flower. The FGP involves extensive EST and finished sequencing of genes expressed in early flower development, expression analysis, and phylogenetic study of these florally expressed genes. All informatic tools derived by the FGP will be available to this project, and comparisons of our moss data with data from the reproductive tissues in the FGP will be mutually beneficial. Another project currently funded by the NSF Plant Genome Program is Regulation of Inflorescence Architecture in Maize. This project is developing comparative genomic data for reproductive tissues from members of the grass family, again, providing valuable comparisons on a mutually informative basis.

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1978. M.S. in Biology, California State Polytechnic University, Pomona, CA
1984. Ph.D. in Biology, Harvard University, Cambridge, MA

APPOINTMENTS:

- Duke University: Assistant Professor of Botany, 1984-90; Associate Professor, 1990-93; Director of Undergraduate Studies in Biology, 1992-93
University of California at Berkeley: Associate Professor, Dept. of Integrative Biology, 1993-1996; Professor, 1996-present; Director, University and Jepson Herbaria, 1993-present; Associate Director, Center for the Study of California Environments and Biological Diversity, 2001-

ACTIVITIES IN PROFESSIONAL ORGANIZATIONS:

- American Bryological and Lichenological Society (Member-at-large, Exe. Com., 1991-93; President-Elect, 1995-1997; President, 1997-1999). American Society of Naturalists (Editorial Board, American Naturalist, 1990-94). American Society of Plant Taxonomists (Editorial Com., Systematic Botany, 1989-92). California Botanical Society (Council Member, 1993-95). International Association for Plant Taxonomy (Editorial Board, Taxon, 2000-present). Society of Systematic Biologists (Nomination Com., 1990; Associate Editor, Systematic Biology, 1992-95).

CURRENT GRANT SUPPORT:

- NSF Research Coordination Network Grant, "Beyond 'Deep Green': Toward an Integration of Plant Phylogenetics and Plant Genomics," University of California, Berkeley, 2001-2006, \$496,434.
NSF Research Grant, "ATOL: Collaborative Research: Deep Green Plant Phylogenetics: Novel Analytical Methods for Scaling from Genomics to Morphology," 2002-2007, \$683,000.
NSF Research Grant, "ITR Collaborative Research: Building the Tree of Life -- A National Resource for Phyloinformatics and Computational Phylogenetics," 2003-2008, \$1,229,000.

FIVE RELEVANT PUBLICATIONS (of 99 lifetime):

- 2000** A.E. Newton, C.J. Cox, J.G. Duckett, J. Wheeler, B. Goffinet, T.A.J. Hedderson, and B.D. MISHLER. Evolution of the major moss lineages: phylogenetic analyses based on multiple gene sequences and morphology. The Bryologist 103: 187-211.
2000 M.J. Oliver, Z. Tuba, and B.D. MISHLER. The evolution of vegetative desiccation tolerance in land plants. Plant Ecology 151: 85-100.
2000 B.D. MISHLER. Deep phylogenetic relationships among "plants" and their implications for classification. Taxon 49: 661-683.
2000 C. La Farge, B.D. MISHLER, J. Wheeler, D. Wall, K. Johannes, S. Schaffer, and J. Shaw. Phylogenetic relationships within the haplolepidaceous mosses. The Bryologist 103: 257-276.
2000 L. R. Stark, B.D. MISHLER, and D.N. McLetchie. The cost of realized sexual reproduction: assessing patterns of reproductive allocation and sporophyte abortion in a desert moss. American Journal of Botany 87: 1599-1608.

FIVE ADDITIONAL SIGNIFICANT PUBLICATIONS:

- 2002** B.D. MISHLER. Phylogeny. In B. K. Hall and W. M. Olson (eds.) Keywords and Concepts in Evolutionary Developmental Biology, pp. 298-308. Harvard University Press.
- 2001** L. R. Stark, D.N. McLetchie, and B.D. MISHLER. Sex expression and sex dimorphism in sporophytic populations of the desert moss *Syntrichia caninervis*. Plant Ecology 157: 181-194.
- 2000** J. Shaw, L.E. Anderson, and B.D. MISHLER. Paedomorphic sporophyte development in *Bruchia flexuosa* (Bruchiaceae). The Bryologist 103: 147-155.
- 1999** E. De Luna, A.E. Newton, A. Withey, D. Gonzalez, and B.D. MISHLER. The transition to pleurocarpy: a phylogenetic analysis of the main Diplolepideous lineages based on *rbcL* sequences and morphology. The Bryologist 102: 634-650.
- 1997** L.A. Lewis, B.D. MISHLER, and R. Vilgalys. Phylogenetic relationships of the liverworts (Hepaticae), a basal embryophyte lineage, inferred from nucleotide sequence data of the chloroplast gene *rbcL*. Molecular Phylogenetics and Evolution 7: 377-393.

SYNERGISTIC ACTIVITIES:

President, American Bryological and Lichenological Society, 1997-99; instituted new programs for supporting student travel to meetings, started the first society website.

Chair, Executive Committee, Green Plant Phylogeny Research Coordination Group (*Deep Green*) 1994-

Member, Steering Committee, Systematics Agenda 2000.

Program Committee, XVI International Botanical Congress, 1999.

Workshops for the general public on bryophytes, molecular systematics, and Deep Green, as part of the Jepson Herbarium Weekend Workshop series, 1995-

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Norton G. Miller (New York State Museum), P. F. Stevens (Missouri Botanical Garden), and C. E. Wood, Jr. (Harvard University), Ph.D. advisors. [no postdoctoral advisor]

Thesis and postdoctoral sponsor:

Efraim De Luna (Instituto de Ecologia, Xalapa), Steven Jessup (Southern Oregon University), Kirsten Fisher (current, UC Berkeley), Louise Lewis (University of Connecticut), Zack Murrell (Appalachian State University), Angela Newton (British Museum), Terry O'Brien (Rowan University), Kathleen Pryer (Duke U), Steven Rice (Union College), Allan Risk (Morehead State University), Patricia Sánchez-Baracaldo (Bristol University), Owen Schwartz (Australian National University), Dennis Wall (Stanford U), John Wheeler (U. Wisconsin, River Falls), Alison Withey (San Diego Supercomputer Facility), Kwok Leung (Joseph) Yip (Hong Kong Herbarium)

a. Education

Colgate University, Hamilton, NY	Botany	A.B.	1962
Ohio University, Athens, OH	Botany	M.S.	1964
Yale University, New Haven, CT	Biology	Ph.D.	1968

b. Appointments

1998-Pres.	<i>Spencer T. Olin Professor/Chairman of Biology</i> , Washington Univ., St. Louis, MO
2003-2007	Co-Director, Division of Biology and Biomedical Sciences, Washington University
1992-1997	<i>Chairman</i> , Department of Biology, University of North Carolina, Chapel Hill, NC
1989-1998	<i>John N. Couch Professor of Biology</i> , Univ. of North Carolina, Chapel Hill, NC
1985-1989	<i>Research Manager-Molecular Biology</i> , Central Research Department, DuPont Co.
1984-1986	<i>Director</i> , Center for Gene Research and Biotechnology, Oregon State University
1968-1986	<i>Assistant, Associate, Professor of Botany</i> , Oregon State University, Corvallis, OR

c. Activities or Accomplishments (selected)

1979	Elizabeth P. Richie Distinguished Professor Award, Oregon State University
1989	Distinguished Alumni Award, Ohio University
1998	Fellow, Robinson College, Cambridge University (UK)
2001	Fellow, Samuel Noble Foundation
2002	Fellow, St. Louis Academy of Science
2002	Fellow, American Association for the Advancement of Science
1989-2003	<i>Coeditor (1989-1995) and Editor-in-Chief (1998-2003), The Plant Cell</i>
1991-1998	Board of Reviewing Editors, <i>Science</i>
1992-1993	<i>President</i> , American Society of Plant Physiologists
1997-2001	<i>Member</i> , Advisory Committee for Biological Sciences, National Science Foundation,
1998-Present	Board of Directors, Boyce Thompson Inst. for Plant Research, Ithaca, NY
1999-2004	Scientific Advisory Board, Biolex, Inc., Pittsboro, NC
2002-Present	Board of Directors, Chromatin, Inc., Chicago, IL

d. Current Research Support

Title: "Genetic Approaches to Study Polarity in Plants," NSF IBN-0112461, 08/01/01-12/31/04

Total Costs: \$373,863

Title: WU/Monsanto, "Regulation of Complex Signaling," 1/02 through 12/05 – Total Costs: \$198,562

Title: NIH "A Partnership Linking Formal and Informal Education," 5R25RR15603, 7/01/02 through 6/30/05 – Total Costs: \$894,557

Title: NSF, "FIBR Planning: A Systems Approach to Study Redox Regulation of Photosynthetic Organisms," EF-0307212(H. Pakrasi, P.I., Bijoy Ghosh, R. Quatrano, Co-PI's), 2/01/03 through 1/31/04, Total Costs: \$50,000

e. Research Publication (selected; 135 total)

1. Quatrano, R. S. (1968). "Rhizoid formation in *Fucus* zygotes: Dependence on protein and RNA synthesis." *Science* 162: 468-470.
2. Hogsett, W. E. and R. S. Quatrano (1978). "Sulfation of fucoïdan in *Fucus* embryos. III. Required for localization in the rhizoid wall." *Journal of Cell Biology* 78: 866-873.
3. Kropf, D. L., B. Kloareg and R. S. Quatrano (1988). "Cell wall is required for fixation of the embryonic axes in *Fucus* zygotes." *Science* 239(187-190).
4. Marcotte, W. R. J., C. C. Bayley and R. S. Quatrano (1988). "Regulation of a wheat promoter by abscisic acid in rice protoplasts." *Nature* 335: 454-457.
5. Feldmann, K. A., M. D. Marks, M. L. Christianson and R. S. Quatrano (1989). "A dwarf mutant of *Arabidopsis* generated by T-DNA insertion mutagenesis." *Science* 243: 1351-1354.

6. Guiltinan, M. J., W. R. Marcotte, Jr. and R. S. Quatrano (1990). "A plant leucine zipper protein that recognizes an abscisic acid element." *Science* 250: 267-271
7. Knight, C. D., A. Sehgal, K. Atwal, J. C. Wallace, D. J. Cove, D. Coates, R. S. Quatrano, S. Bahadur, P. G. Stockley and A. C. Cuming (1995). "Molecular responses to abscisic acid and stress are conserved between moss and cereals." *The Plant Cell* 7: 499-506.
8. Vasil, V., W. Marcotte, L. Rosenkrans, S. M. Cocciolone, I. K. Vasil, R. S. Quatrano and D. R. McCarty (1995). "Overlap of VP1 and ABA Response Elements in the Em Promoter: G-box elements are sufficient but not necessary for VP1 transactivation." *The Plant Cell* 7: 1511-1518.
9. Bouget, F.-Y., S. Gertulla, S. L. Shaw and R. S. Quatrano (1996). "Localization of actin mRNA during the establishment of cell polarity and early cell divisions in *Fucus* embryos." *The Plant Cell* 8: 189-201.
10. Cove, D. J., R. S. Quatrano and E. Hartmann (1996). "The alignment of the axis of asymmetry in regenerating protoplasts of the moss, *Ceratodon purpureus*, is determined independently of axis polarity." *Development* 122: 371-379.
11. Shaw, S. and R. S. Quatrano (1996). "The role of targeted secretion in the establishment of cell polarity and the orientation of the division plane in *Fucus* zygotes." *Development* 122: 2623-2630.
12. Razik, M. A. and R. S. Quatrano (1997). "Effect of the nuclear factors EmBP1 and Viviparous1 on the transcription of the Em gene in HeLa nuclear extracts." *The Plant Cell* 9: 1791-1803.
13. Schultz, T., J. Medina, A. Hill and R. S. Quatrano (1998). "14-3-3 proteins are part of an ABA/VP1 response complex in the Em promoter and interact with VP1 and EmBP1." *The Plant Cell* 10: 837-848.
14. Quatrano, R. S. (1978). "Development of cell polarity." *Annual Review Plant Physiology* 29: 487-510.
15. Rock, C., and R. S. Quatrano. (1995). The role of hormones during seed development. In P. Davies, ed. *Plant Hormones: Physiology, Biochemistry and Molecular Biology*. Kluwer Academic Publishers, Dordrecht, The Netherlands. pp. 671-697
16. Fowler, J. E. and R. S. Quatrano (1997). "Plant cell morphogenesis: plasma membrane interactions with the cytoskeleton and cell wall." *Ann Rev Cell & Develop Biol* 13: 697-743.
17. Quatrano, R. S. (1997). Cortical asymmetries direct the establishment of cell polarity and the plane of cell division in *Fucus* embryos. Cold Spring Harbor Symposium on Quantitative Biology, Cold Spring Harbor, NY.
18. Quatrano, R. S. (ed.) (2001). *Plant Genomics: Emerging Tools*. (Rockville, Maryland; American Society of Plant Biologists), 319 pp.

Most Recent

1. Ivanchenko, M., Z. Vejlupkova, R. Quatrano and J. E. Fowler (2000). "Maize ROP7 GTPase contains a unique, CaaX box-independent plasma membrane targeting signal." *The Plant Journal* 24(1): 79-90.
2. Celia D. Knight, David J. Cove, Andrew C. Cuming, Ralph S. Quatrano (2002). "Moss Gene Technology" *Molecular Plant Biology*. P. M. Gilmartin and C. Bowler, Oxford University Press. 2: 285-301.
3. Bezanilla, M., Horton, A.C., Sevens, H.C. and Quatrano, R.S. (2003). "Phylogenetic analysis of new plant myosin sequences" *J. Mol. Evolution* 57: 229-239
4. Belanger KD, Wyman AJ, Sudol MN, Singla-Pareek SL, Quatrano, R. S. (2003). "A signal peptide secretion screen in *Fucus distichus* embryos reveals expression of glucanase, EGF domain-containing, and LRR receptor kinase-like polypeptides during asymmetric cell growth." *Planta* 217:931-950
5. Bezanilla, M., Aihong Pan, Ralph S. Quatrano (2003). "RNAi in the moss *Physcomitrella patens*." *Plant Physiology* 133: 1-5
6. Christensen, T. M., Zuzana Vejlupkova, Yogesh K. Sharma, Kirstin M. Arthur, Joseph W. Spatafora, Carol Albright, Robert B. Meeley, J. P. Duvick, Ralph S. Quatrano, John E. Fowler (2003) "Conserved Subgroups and Developmental Regulation in the Monocot *rop* Gene Family" *Plant Physiology*. *In Press*

f. Mentor

M.S./Ph.D. Students = 20

Postdoctoral Fellows = 33

Curriculum vitae - David John Cove

- 1960 B.A., University of Cambridge (Natural Sciences Tripos, Genetics, Class I)
1963 Ph.D., University of Cambridge. (Studies of the biochemistry and genetics of inorganic nitrogen metabolism in the fungus, *Aspergillus nidulans*)
1963 - 1964 Medical Research Council Personal Research Fellowship.
1964 - 1967 University Demonstrator, Department of Genetics, University of Cambridge.
1964 - 1977 Staff Fellowship, Trinity Hall, Cambridge.
1967 - 1977 University Lecturer, Department of Genetics, University of Cambridge.
1976 - 1977 Visiting Professor, University of Mainz
1976 - 1977 CIBA-Geigy Senior Research Fellowship, Leverhulme Research Fellowship
1978 - 2002 Professor of Genetics, University of Leeds.
1978 - 1993 Head of Department of Genetics, University of Leeds.
1992 - 1993 Senior Research Fellow, University of North Carolina, Chapel Hill, Leverhulme Research Fellow
1993 Visiting Research Professor, Free University, Berlin.
1993 - 1997 Chairman, School of Biology, University of Leeds.
2002 - Leverhulme Emeritus Research Fellowship
2002 - Clark Way Harrison Distinguished Visiting Professor, Washington University in St. Louis
2002 - Visiting Professor, University of Leeds

Recent Publications

- Cove, D.J., Quatrano, R.S. & Hartmann, E. 1996. The alignment of the axis of asymmetry in regenerating protoplasts of the moss, *Ceratodon purpureus*, is determined independently of axis polarity. *Development* 122, 371 - 397.
- Kammerer, W & Cove, D.J. 1996. Genetic analysis of the result of re-transformation of transgenic lines of the moss, *Physcomitrella patens*. *Molecular and General Genetics* 250, 380-382.
- Lamparter, T, Esch, H., Cove, D.J., Hughes, J. & Hartmann, E. 1996. Aphototropic mutants of the moss *Ceratodon purpureus* with spectrally normal and with spectrally dysfunctional phytochrome. *Plant, Cell and Environment* 19, 560-568.
- Russell, A.J., Knight, M.R., Cove, D.J., Knight, C.D., Trewavas, A.J. and Wang, T.L., 1996. The moss, *Physcomitrella patens*, transformed with apoaequorin cDNA responds to cold shock, mechanical perturbation and pH with transient increases in cytoplasmic calcium. *Transgenic Research* 5, 167-170.
- Lamparter, T, Esch, H., Cove, D.J., & Hartmann, E. 1997. Phytochrome control of phototropism and chlorophyll accumulation in the apical cells of protonemal filaments of wild type and an aphototropic mutant of the moss *Ceratodon purpureus*. *Plant Cell Physiology* 38, 51-58.
- Sheffield, E., Douglas, G.E. & Cove, D.J. 1997. Growth and development of fern gametophores in an air;ift fermenter. *Plant Cell Reports* 16, 561-564.
- Wagner, T.A., Cove, D.J. & Sack, F.D. 1997. A positively gravitropic mutant mirrors the wild-type protonemal response in the moss *Ceratodon*. *Planta* 202, 149 - 154.
- Hoffman, A.H., Codon, A.C., Ivascu, C., Russo, V.E.A., Knight, C., Cove, D., Schaefer, D.G., Chakparonian, M. & Zryd, J.-P. 1999. A specific member of the Cab multigene family is

- efficiently targeted and disrupted in the moss *Physcomitrella patens*. *Molecular and General Genetics* 261, 92 - 99
- Machuka, J., Bashiardes, S., Ruben, E., Spooner, K., Cuming, A., Knight, C. & Cove, D. 1999. Sequence analysis of expressed sequence tags from an ABA-treated cDNA library identifies stress response genes in the moss *Physcomitrella patens*. *Plant and Cell Physiology* 40, 378 - 387.
- Cove, D.J. 2000. The generation and modification of cell polarity. *Journal of Experimental Botany* 51, 831 – 838
- Cove, D.J. & Quatrano, R.S. 2004. The use of mosses for the study of cell polarity. In: “New Frontiers in Bryology” eds: A.J.Wood, M.J.Oliver & D.J.Cove. Kluwer (in press).
- Reski, R. & Cove, D.J. 2004. Quick guide: *Physcomitrella patens*. *Current Biology* (in press).

Name:	D.o.B	Gender
Dr. Andrew C. Cuming	3 Jan 1952	M

Education

BA (Oxon) Botany 1973

PhD (Cantab) Plant Biochemistry 1976

Professional career

Postdoctoral Fellow: Dept. of Biochemistry, University of Toronto 1977-8

Postdoctoral Fellow: Dept. of Biological Sciences, University of Warwick 1978-4

SERC Advanced Fellow: Dept. of Biological Sciences, University of Warwick 1981-84

Lecturer in Genetics, Leeds University 1984-1994

Senior Lecturer in Genetics 1994-2004

Major research grants awarded

1981: SERC (£28,126) *The expression of genes encoding ribosomal proteins during wheat embryo development*

1984: SERC (£30,640) *The structure of genes encoding wheat ribosomal proteins*

1986: SERC (£44,460) *Organisation and expression of an ABA-regulated gene in *Triticum aestivum* L.*

1988: SERC (£50,880) *Gene regulation in cereal embryo developmental mutants*

1989: SERC (£57,630) *Selective proteolysis in cereal embryo development*

1989: AFRC (83,047) *Characterisation of protein factors regulating transcription of a family of ABA-modulated genes in wheat embryos*

1991: EC (£88,270) *Protein engineering of the chloroplast light-harvesting complex of photosystem II*

1992: AFRC (£65,000) *Genetic engineering of light-harvesting complexes in *Arabidopsis thaliana**

1993: SERC (£142,662) *Molecular characterisation of a substrate-specific protease in wheat embryos*

1999: BBSRC (£540,064) *Expressed sequence tags for homologous recombination in the moss *Physcomitrella patens**

Recent and related publications (5)

Knight, CD *et al* (1995) Molecular responses to abscisic acid and stress are conserved between moss and cereals. *Plant Cell* **7**: 499-506

Machuka *et al* (1999) Sequence analysis of expressed sequence tags from an ABA-treated cDNA library identifies stress response genes in the moss *Physcomitrella patens*. *Plant & Cell Physiol* **40**: 378-387

Quatrano RS *et al* (2000-2003): Leeds/Wash U Moss EST Project. (21215 EST sequences deposited in GenBank in the course of the BBSRC-sponsored "PEP" programme). <http://www.ncbi.nlm.nih.gov>

Caliskan M *et al* (2003) Isolation and localization of new germination-related sequences from wheat embryos. *J. Biochem. Mol. Biol.* **36**: 580-585

Caliskan M *et al* (2004) The formation of wheat (*Triticum aestivum* L.) embryogenic callus involves peroxide-generating germin-like oxalate oxidase. *Planta*: In Press

CURRICULUM VITAE

Mitsuyasu Hasebe

Professor
National Institute for Basic Biology
38 Nishigonaka, Myodaiji-cho, Okazaki 444-8585, JAPAN
Tel: +81-564-55-7546
FAX: +81-564-55-7546
E-mail: mhasebe@nibb.ac.jp

NATIONALITY: Japanese

DATE OF BIRTH: March 6, 1963

EDUCATION: 1987, B.Sc. Department of Botany, University of Tokyo, Japan

1992, Ph.D. Department of Botany, University of Tokyo, Japan

Thesis title: Molecular Phylogeny of vascular plants

EMPLOYMENT:

1991-1996 Assistant Professor, Botanical Gardens, Faculty of Science, University of Tokyo, Tokyo, Japan

1993-1995 Visiting Research Scholar, Department of Botany and Plant Pathology, Purdue University, Indiana, U.S.A.

1996-2000 Associate Professor, National Institute for Basic Biology, Okazaki, Japan

2000-present Professor, National Institute for Basic Biology, Okazaki, Japan

RESEARCH INTERESTS

Evolution of development in green plants

Evolution of generations in plants

Phylogeny of green plants

EDITORSHIPS

1997-2000 Editorial Board, Journal of Plant Research

1998-present Editorial Board, International Journal of Plant Science

1998-2002 Section Editor of Genetics and Systematics, Plant Biology

AWARDS

1997 Young Scientist Prize (The Botanical Society of Japan)

2001 Young Scientist Prize (The Japanese Society of Evolution)

RECENT RESEARCH PUBLICATIONS:

40) Kofuji, R., Sumikawa, N., Yamasaki, M., Kondo, K., Ueda, K., Ito, M. and Hasebe, M. 2003. Evolution and divergence of MADS-box gene family based on genome wide expression analyses. Mol. Biol. Evol. 20: 1963-1977.

- 39) Sakakibara, K., Nishiyama, T., Sumikawa, N., Kofuji, R., Murata, T. and Hasebe, M. 2003. Involvement of auxin and a homeodomain-leucine zipper I gene in rhizoid development of the moss *Physcomitrella patens*. *Development* 130: 4835-4846.
- 38) Nishiyama, T., Fujita, T., Shin-I, T., Seki, M., Nishide, H., Uchiyama, I., Kamiya, A., Carninci, P., Hayashizaki, Y., Shinozaki, K., Kohara, Y., and Hasebe, M. 2003. Comparative genomics of *Physcomitrella patens* gemetophytic transcriptome and *Arabidopsis thaliana*: Implication for land plant evolution. *Proc. Natl. Acad. Sci. USA* 100: 8007-8012.
- 37) Wolf, P.G., Rowe, C.A., Sinclair, R.B., and Hasebe, M. 2003. Complete nucleotide sequence of the chloroplast genome from a leptosporangiate fern, *Adiantum capillus-veneris* L. *DNA Res.* 10: 59-65.
- 36) Tanabe, Y., Uchida, M. Hasebe, M., and Ito, M. 2003. Characterization of the *Selaginella remotifolia* MADS-box gene. *J. Plant Res.* 116: 71-75.
- 35) Itoh, Y., Hasebe, M., Davies, E., Takeda, J., and Ozeki, Y. 2003. Survival of *Tdc* transposable elements of the *En/Spm* superfamily in the carrot genome. *Mol. Gen. Genomics* 269: 49-59.
- 34) Rivadavia, F., Kondo, K., Kato, M., and Hasebe, M. 2003. Phylogeny of the sundews, *Drosera* (Droseraceae) based on chloroplast *rbcL* and nuclear 18S ribosomal DNA sequences. *Amer. J. Bot.* 90: 123-130.
- 33) Iwakawa, H., Ueno, Y., Semiarti, E., Onouchi, H., Kojima, S., Tsukaya, H., Hasebe, M., Soma, T., Ikezaki, M., Machida, C., and Machida, Y. 2002. The *ASYMMETRIC LEAVES2* gene of *Arabidopsis thaliana*, required for formation of a symmetric flat leaf lamina, encodes a member of a novel family of proteins characterized by cysteine repeats and a leucine zipper. *Plant Cell Phys.* 43: 467-478.
- 32) Imaizumi, T., Kadota, A., Hasebe, M. and Wada, M. 2002. Cryptochrome light signals control development to suppress auxin sensitivity in the moss *Physcomitrella patens*. *Plant Cell* 14_373-386.
- 31) Henschel, K., Kofuji, R., Hasebe, M., Saedler, H., Munster, T. and Theissen, G. 2002. Two ancient classes of MIKC-type MADS-box genes are present in the moss *Physcomitrella patens*. *Mol. Biol. Evol.* 19: 801-814.
- 30) Shindo, S., Sakakibara, K., Sano, R., Ueda, K. and Hasebe, M. 2001. Characterization of a *FLORICAULA/LEAFY* homologue of *Gnetum parvifolium*, and its implications for the evolution of reproductive organs in seed plants. *Int. J. Plant Sci.* 162: 1199-1209.
- 29) Himi, S., Sano, R., Nishiyama, T., Tanahashi, T., Kato, M., Ueda, K., and Hasebe, M. 2001. Evolution of MADS-box gene induced by *FLO/LFY* genes. *J. Mol. Evol.* 53: 387-393.

- 28) Hiwatashi, Y., Nishiyama, T., Fujita, T. and Hasebe, M. 2001. Establishment of gene-trap and enhancer-trap systems in the moss *Physcomitrella patens*. *Plant J.* 28: 105-116
- 27) Sakakibara, K., Nishiyama, T., Kato, M., and Hasebe, M. 2001. Isolation of Homeodomain-Leucine Zipper Genes from the Moss *Physcomitrella patens* and the Evolution of Homeodomain-Leucine Zipper Genes in Land Plants. *Mol. Biol. Evol.* 18: 491-502.
- 26) Yoshimoto, Y., Higeta, D., Ito, Y., Yoshida, H., Hasebe, M. and Ozeki, Y. 2000. Isolation and characterization of a cDNA for Phenylalanine ammonia-lyase (PAL) from *Dianthus caryophyllus* (carnation). *Plant Biotechnology* 17: 325-329.
- 25) Sano, R., Ito, M., Kurita, S. and Hasebe, M. 2000. *Deparia formosana* (Rosenst.) as the new name for *Diplazium formosanum*. *Acta Phytotaxa Geobot.* 51: 17-20.
- 24) Sano, R., Takamiya, M., Kurita, S., Ito, M. and Hasebe, M. 2000. *Diplazium subsinuatatum* and *Di. tomitaroanum* should be moved to *Deparia* according to molecular, morphological, and cytological characters. *J. Plant Res.* 113: 157-163.
- 23) Nishiyama, T., Hiwatashi, Y., Sakakibara, K., Kato, M. and Hasebe, M. 2000. Tagged mutagenesis and gene-trap in the moss, *Physcomitrella patens* by shuttle mutagenesis. *DNA Res.* 7: 1-9.
- 22) Sano, R., Takamiya, M., Ito, M., Kurita, S. and Hasebe, M. 2000. Phylogeny of lady fern group, tribe Phymatidae (Dryopteridaceae) based on chloroplast *rbcL* gene sequences. *Molec. Phyl. Evol.* 15: 403-413.
- 21) Yokoyama, J., Suzuki, M., Iwatsuki, K. and Hasebe, M. 2000. Molecular phylogeny of *Coriaria*, with special emphasis on the disjunct distribution. *Molec. Phyl. Evol.* 14:11-19.
- 20) Shindo, S., Ito, M., Ueda, K., Kato, M. and Hasebe, M. 1999. Characterization of MADS genes in the gymnosperm *Gnetum parvifolium* and its implication on the evolution of reproductive organs in seed plants. *Evolution and Development* 1 : 180-190.
- 19) Aso, K., Kato, M., Banks, J.A. and Hasebe, M. 1999. Characterization of homeodomain-leucine zipper genes in the fern, *Ceratopteris richardii* and the evolution of the homeodomain-leucine zipper gene family in vascular plants. *Molec. Biol. Evol.* 16: 544-552
- 18) Hasebe, M., Wen, C.-K., Kato, M. and Banks, J.A. 1998. Characterization of MADS homeotic genes in the fern *Ceratopteris richardii*. *Proc. Natl. Acad. Sci. USA* 95: 6222-6227.
- 17) Hasebe, M., Ando, T. and Iwatsuki, K. 1998. Intrageneric relationships of maple trees based on the chloroplast DNA restriction fragment length polymorphisms. *J. Plant Res.* 111: 441-451.
- 16) Hasebe, M. and Banks, J.A. 1997. Evolution of MADS gene family in plants. In K. Iwatsuki and P.H. Raven eds., *Evolution and Diversification in Land Plants*, Springer-Verlag, Tokyo, pp179-197.

Prof. Dr. Ralf Reski's Curriculum Vitae

Director Plant Biotechnology, University of Freiburg, Germany.

Born in Gelsenkirchen, Germany, on 18th November 1958, married, two children.

- 2004** Guest editor Plant Biology.
- 2003 - 2006** Permanent member of the Habilitation-Board, Faculty of Biology, Freiburg University.
- Since 2003** Advisory Board of BioPro GmbH, a biotechnology enterprise of the federal state of Baden-Württemberg.
- Since 2002** Editorial Board of Plant Cell Reports.
- Since 2002** Chairman of the advisory board at greenovation Biotech GmbH.
- 2002 - 2004** Managing Director/Co-Director of the Institute Biology II, Freiburg University.
- 2001 - 2002** Life Science co-ordinator of Freiburg University.
- Since 2001** Director Plant Biotechnology of the Center for Applied Biosciences (ZAB) at Freiburg University.
- Since 2000** Advisory Board of InnoPlanta e.V..
- 1999 - 2002** Scientific co-ordinator of the BioRegio Freiburg.
- 09.09.1999** Co-founder of greenovation Plant Biotechnology GmbH.
- Since 1999** Co-ordinator of the German National Science Foundation (DFG) Program "Molecular Analysis of Phytohormone Action".
- Since 1999** Full Professor (C4), Chair Plant Biotechnology at Freiburg University.
- 1997-1999** Ranked 1st for an Associate Professorship in Norway (Oslo), ranked 3rd for a full professorship in Berlin (HU), ranked 2nd for a full professorship in Bremen, and ranked 1st for full professor-ships in Dresden (TU), Rostock and Freiburg.
- 1996-1999** Heisenberg-Fellow of the German National Science Foundation (DFG) at Freiburg University.
- 1994** Habilitation awarded in "General Botany".
- 1990-1996** Assistant Professor (C1) at Hamburg University, Department of Cell Biology.
- 1990** Ph.D. Degree (Dr. rer. nat.) at Hamburg University, Department of Genetics.

Recent Publications

- 56) Rensing, S.A., S. Rombauts, A. Hohe, D. Lang, E. Duwenig, P. Rouze, Y. Van de Peer, **R. Reski** (2002): The transcriptome of the moss *Physcomitrella patens*:

- comparative analysis reveals a rich source of new genes. http://www.plant-biotech.net/Rensing_et_al_transcriptome2002.pdf.
- 57) **Reski, R.** (2002): Rings and networks: the amazing complexity of FtsZ in chloroplasts. *Trends Plant Sci.* 7, 103-105.
 - 58) Richter U., J. Kiessling, B. Hedtke, E. Decker, **R. Reski**, T. Börner, A. Weihe (2002): Two *RpoT* genes of *Physcomitrella patens* encode phage-type RNA polymerases with dual targeting to mitochondria and plastids. *Gene* 290, 95-105.
 - 59) Schipper, O., D. Schaefer, **R. Reski**, A. Fleming (2002): Expansins in the bryophyte *Physcomitrella patens*. *Plant Mol. Biol.* 50, 789-802.
 - 60) Schween G., S. Fleig, **R. Reski** (2002): High-throughput-PCR screen of 15,000 transgenic *Physcomitrella* plants. *Plant Mol. Biol. Rep.* 20, 43-47.
 - 61) Zank, T.K., U. Zähringer, C. Beckmann, G. Pohnert, W. Boland, H. Holtorf, **R. Reski**, J. Lerchl, E. Heinz (2002): Cloning and functional characterisation of an enzyme involved in the elongation of Δ^6 -polyunsaturated fatty acids from the moss *Physcomitrella patens*. *Plant J.* 31, 255-268.
 - 62) Rensing, S.A., S. Rombauts, Y. Van de Peer, **R. Reski** (2002): Moss transcriptome and beyond. *Trends Plant Sci.* 7, 535-538.
 - 63) Schlink, K, **R. Reski** (2002): Preparing high-quality DNA from moss (*Physcomitrella patens*). *Plant Mol. Biol. Rep.* 20, 423a-423f.
 - 64) Sperling, P., T. Egener, J. Lucht, **R. Reski**, P. Cirpus, E. Heinz (2002): Identification of a Δ^5 -fatty acid desaturase from *Physcomitrella patens*. In: *Advanced Researches on Plant Lipids*. N. Murata, M. Yamada, I. Nishida, J. Sekiya, H. Wada (eds.), pp. 113-116. Kluwer Academic Publishers, Dordrecht.
 - 65) **Reski, R.** (2003): *Physcomitrella patens* as a novel tool for plant functional genomics. In: *Plant biotechnology 2002 and beyond*. I.K. Vasil (ed.) Kluwer Acad. Publ., 205-209.
 - 66) Schween, G., A. Hohe, A. Koprivova, **R. Reski** (2003): Effects of nutrients, cell density and culture techniques on protoplast regeneration and early protonema development in a moss, *Physcomitrella patens*. *J. Plant Physiol.* 160, 209-212.
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Wed 19th Feb. 2004

Dear Ralph,

I strongly support your proposal to DoE to seek funding for the initiation of genomic sequencing in *Physcomitrella patens*. This is a logical continuation of our BBSRC-supported EST sequencing programme the success of which was due in large part to the close research collaboration between Washington University and Leeds University.

A full understanding of the *Physcomitrella* genome will only be achieved through a comprehensive genome sequencing programme - a project of such scope as to require the commitment of considerable resources on an international scale. An important component of such a project is the determination of both a genetic and physical map of the *Physcomitrella* genome. We at Leeds University are now in a strong position to initiate such an analysis, and are seeking funding in the UK to apply molecular marker technology to generate a genetic linkage map of *P. patens* that can subsequently be integrated with a physical map derived from current (and future) genomic and EST resources. This will be based on a combination of AFLP and RFLP linkage mapping, BAC-fingerprinting and end-sequencing, and the placement of molecular markers (ESTs, AFLP and RFLP sequences) on BAC contigs, using resources developed in the course of the EST sequencing programme. The substantial additional sequence resources developed in Japan at the National Institute for Basic Biology will also be available for this, and the very active research being undertaken both at this Institute and at the University of Freiburg, underlines the vigorous international interest in *Physcomitrella* genomics.

In the course of the EST sequencing programme, we constructed a BAC library from *Physcomitrella* that we estimate to contain *ca.* 50% of the *Physcomitrella* genome (based on our analysis of insert sizes and the frequency of successful gene isolation thus far). Additionally, we have obtained substantial sequence information from four BAC clones, comprising some 400 kb, that sheds some light as to the nature of the *Physcomitrella* genome.

- (i) The *Physcomitrella* genome appears to be similar to other large eukaryotic genomes, in that it contains genes interspersed among long stretches of non-coding DNA.
- (ii) The non-coding regions tend to be relatively AT-rich, and include some highly repetitive motifs that make the assembly of long contigs challenging.
- (iii) Like most other plant genomes, the *Physcomitrella* genome contains a significant number of sequences that can be identified as of retrotransposon origin. – The representation of retrotransposon-related sequences in the EST database strongly indicates that a population of these sequences are active in *Physcomitrella*.

Because of the content of repetitive, low-complexity and multicopy sequences, I would recommend that any genomic sequencing programme include a substantial genetic and physical mapping component, in which EST sequences can be mapped to BAC clones and placed within BAC contigs, to provide a skeleton of molecular markers that should assist in anchoring the assembly of shotgun-sequence-derived contigs. Additionally, the development of a linkage map based on molecular markers (AFLPs and RFLPs) will provide genetic markers that can be placed directly on the physical map. This should be greatly facilitated by the planned construction of a more comprehensive large-insert BAC library.

Best wishes for success in your application. We shall be happy to provide whatever support we can.

Sincerely,

Dr. Andrew C. Cuming

2/17/04(email)

Dear Ralph and David,

It is very nice to hear that *Physcomitrella* genome project will be submitted to Department of Energy, USA. As we discussed in the last international moss meeting held in St. Louis you hosted, I am willing to work to promote the *Physcomitrella* genome project in Japan. Genome projects of MEXT in Japan will be renewed in 2005, and *Physcomitrella* genome projects including (1) more full length cDNA libraries covering whole developmental stages, (2) EST sequences, and (3) whole genome sequencing are included in the proposal prepared by the government council. Japan government encourages an international collaboration for whole genome sequencing, and the good situations in US, UK, and Germany are very helpful to promote the work in Japan. I really hope that the international collaboration on *Physcomitrella* genome sequencing will be successful.

Best wishes,

Mitsuyasu

Mitsuyasu Hasebe, PhD

Professor

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2/19/04(email)

Dear Prof. Quatrano,

Thank you very much for your efforts to obtain funding for sequencing the complete genome of the model moss *Physcomitrella patens*.

So far, we have sequenced more than 115,000 expressed sequence tags from this moss and built a comprehensive database from them.

All our analyses indicate that this moss harbours several hundreds of genes not being described from seed plants before.

As the last common ancestor of mosses and seed plants lived about 450 MYA, the full genome sequence of this moss will have a tremendous impact in basic as well as applied science.

I will be happy to participate in an international steering board with you, Prof. Cove (UK) and Prof. Hasebe (Japan) to co-ordinate such a sequencing effort.

In parallel to your efforts we are applying for German national funds for this sequencing project, so that financial burdens might be shared with different countries. We are also proud to be the host of the international moss community from Sept. 12-15, 2004 in Freiburg, Germany. Please check www.plant-biotech.net/moss2004 for details.

Best regards,

Ralf Reski

Professor, Head and Director Plant Biotechnology

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February 19, 2004

Dr. Brent Mishler
Director of the University and Jepson Herbaria
Department of Integrative Biology
1001 Valley Life Sciences Building, #2465
Berkeley, CA 95720-245

Dear Brent,

I am writing to express our support for your proposal to JGI to sequence the genome of the moss *Physcomitrella patens*. The plant community would be well served by having this genome and the genome of *Selaginella moellendorffii* sequenced and annotated. We will do whatever we can to collaborate and integrate the data generated from both projects. From my many conversations with plant biologists, these two species are obvious choices for genome sequencing. They each represent different yet important nodes of the plant evolutionary tree and are phylogenetically well spaced. Having their sequences in hand will have a significant impact on our understanding on the organization and evolution of plant genomes and will contribute greatly to gene discovery and gene annotation of all plant genomes, including crops.

I am very excited by the possibility of us working with JGI to develop informatics and WWW tools to make all of our results meaningful to the public. We also look forward to collaborating with you on any educational or outreach opportunity provided by "Deep Green".

Sincerely,

Jo Ann (Jody) Banks

