



## *Paracoccidioides brasiliensis* - Special Session

### INTRODUCTION

Paracoccidioidomycosis (PCM) is the most important systemic mycosis in Brazil, and it is widespread throughout Latin America. An estimated 10 million individuals are infected, with about 2% developing the disease. This mycosis has also been reported among patients with AIDS in Latin America. PCM presents several clinical forms, ranging from a localized and benign disease to a progressive and potentially lethal systemic infection. All patients from whom the fungus is isolated should be treated, and pulmonary fibrosis is still a common sequel, despite the availability of new antifungal drugs for therapy.

The etiological agent of PCM is the pathogenic ascomycete fungus *Paracoccidioides brasiliensis*, which has a close relationship to other pathogenic fungi, such as *Blastomyces dermatitidis* and *Histoplasma capsulatum*. *Paracoccidioides brasiliensis* exists as a mycelium in the soil and as a yeast in the host. The unicellular hyphae and fungal propagules, or conidia, are uninucleate, while the yeasts are multinucleate. Information on genome size, chromosome organization, mechanisms that contribute to dimorphism, pathogenicity, and virulence are scarce. In addition, a strong variability among the fungus isolates has been reported, with genotypic differences showing some correlation with virulence, geographic distribution and susceptibility or resistance to drugs.

Our group has been working for the last eight years in order to identify the differentially expressed genes in the dimorphism of *P. brasiliensis* and also to look for genes encoding proteins that are immunogenic in humans during infection. Several genes that are up-regulated in mycelial and yeast cells have been previously identified by proteome analysis, and by immunoscreening of cDNA libraries from mycelium and yeast cells, using patient's sera.

In order to accelerate and improve the discovery of differentially expressed genes in both mycelial and yeast cells, the "Functional and Differential Genome Project of *Paracoccidioides brasiliensis*" was carried out with the collaboration of 11 public and private institutions located in the Midwest region of Brazil; these institutions constitute the PbGenome Network. The main goals of the project were the identification of genes: (i) differentially expressed in mycelium and yeast cells; (ii) related to the general and differential metabolism of this pathogen in mycelium and yeast cells; (iii) related to signal transduction pathways involved in dimorphic transition, proliferation, cell wall construction, osmoregulation, and high temperature growth; (iv) related to host-pathogen interaction, and (v) related to drug targets and essential ortholog-

to-*C. albicans* genes. Ultimately, the functional analysis of these genes may lead to the identification of drug targets for disease control.

The strategy employed was to sequence ESTs (expression sequence tags) derived from non-normalized cDNA libraries from both mycelium and yeast cells from *P. brasiliensis* (Pb01 isolate), cultivated at 23° and 36°C, respectively. The project resulted in the sequencing of 25,511 clones, which yielded 6,022 groups that represent *P. brasiliensis*-expressed genes. This transcriptome analysis covered about 80% of the gene content, since it is estimated that *P. brasiliensis* has ~8,000 genes. The 6,022 clusters were submitted to categorization analysis: 2,129 were successfully annotated, 1,604 were non-conclusively annotated and 2,289 were not identified. We classified the PbAESTs (*P. brasiliensis* assembled ESTs) into 18 functional COG categories, as follows: cellular metabolism (29%); transcription (12%); protein synthesis (10%); energy production (9%); control of cellular organization (4%), and other categories.

Differentially expressed genes in mycelium and yeast cells were detected by statistical comparison of the number of sequences in corresponding PbAESTs. We have identified up-regulated genes in mycelium and yeast cells, most of them corroborated by cDNA microarray and confirmed by Northern blot analysis. The transcriptome of *P. brasiliensis* generated an overview of the metabolic pathways that occur in mycelium and yeast cells and are detailed in this issue. Furthermore, we were able to point out the main metabolic features of mycelium and yeast cells, as detected in the differential transcriptome by EST analysis. The overall analysis indicates that ATP production through alcohol fermentation and the respiratory chain occurs in a biased pattern, the former being preferential in the yeast form and the latter, in mycelium. Also, the transcriptome enabled us to describe: (i) the main signal transduction pathways that are probably operating in this pathogen, (ii) possible genes involved in host-pathogen interaction, and (iii) essential genes and others related to drug targets. The functional genome project described in this issue has provided us for the first time with a broad view of the pathogen's metabolism and has also given new insights into biological features that had been elusive to investigation till now.

This collection of 15 papers presented in this issue is a consequence of the *P. brasiliensis* transcriptome publications (*Yeast* 20: 263-271, 2003; *FEMS Immunol. Med. Microbiol.*, in press, and *J. Biol. Chem.*, in press). These 15 papers provide a more comprehensive view of some specific features considered relevant to the understanding of basic and applied knowledge on cell differentiation, drug targets and virulence of *P. brasiliensis*.

I am thankful to all the researchers who worked in the PbGenome Network project and to the MCT/CNPq (Ministry of Science and Technology and National Council for Research and Technological Development) for their financial support. I would also like to add that this initiative included training of 204 teachers and students from public and private universities, researchers from Embrapa Gado de Corte and Embrapa Pantanal located in the Brazilian Midwest States of Mato Grosso do Sul (MS) and Mato Grosso (MT). Six courses were given in both states: Molecular Biology, Biochemistry/Biophysics, Genetic Engineering, DNA Sequencing/Bioinformatics, Molecular Markers, Microbiology/Enzymology. In addition, about 20 researchers from nine Institutions from MT and MS were trained in basic techniques for EST sequencing and gene annotation at the University of Brasília and the Federal University of Goiás.

Maria Sueli Soares Felipe  
Full Professor  
University of Brasília

Introduction

The PbGenome Project page is available at [www.biomol.unb.br/Pb/](http://www.biomol.unb.br/Pb/)

Guest Editor and Project Coordinator

Maria Sueli Soares Felipe

Laboratório de Biologia Molecular

Departamento de Biologia Celular

Instituto de Ciências Biológicas

Universidade de Brasília - Asa Norte

70910-900 Brasília, DF

Brazil

E-mail: [msueli@unb.br](mailto:msueli@unb.br)

Associate Editor for the Special Session on *Paracoccidioides brasiliensis*

Vasco Azevedo

Departamento de Biologia Geral,

Instituto de Ciências Biológicas,

Universidade Federal de Minas Gerais

Av. Antonio Carlos, 6627, Pampulha, Caixa Postal 486

31270-901 Belo Horizonte, MG

Brazil

E-mail: [vasco@mono.icb.ufmg.br](mailto:vasco@mono.icb.ufmg.br)

Principal Lab Home Pages:

<http://www.icb.ufmg.br/~vasco/genmic/>

<http://www.icb.ufmg.br/~vasco/coryne>

<http://www.icb.ufmg.br/~prodap/2001/baclact/>