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Retrospective

RETROSPECTIVE

The birth of enthusiasm and passion for science

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One sentence summary: Retrospective on the life and work of Claudina Rodrigues-Pousada.
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ABSTRACT

In this paper I describe the main aspects of my career and focus on the retrospective on my life and my work.

Keywords: retrospective; *D. gigas*; Tetrahymena; Saccharomyces cerevisiae

My story, as related to me by family members, begins as a young girl who, when given a children's book on the life of Marie Curie, became instantly fascinated with the exciting experiments and life of that famous scientist, her setbacks and discoveries. It was at that moment that I decided that I myself wanted to become a woman of science.

WAR ENDS—WITHOUT CELEBRATION

I was born in, 1941, in Tadim, a small village of Celtic origin in the North of Portugal. It was a crucial and harsh year for Europe, gripped in the midst of the World War II. The government of my country, led by Salazar, was a keen Hitler supporter, and when the war ended, Salazar forbade celebrative demonstrations and enforced widespread political imprisonment.

My parents led modest lives, my mother a Primary School Teacher and my father working in the area of commercial automobiles. Between six and ten years old, I attended the elementary school, where my mother taught. My parents liked to draw their children's attention, my two brothers, sister and I, towards the observation of natural events. I remember seeing fireflies emit a calm, soft luminescence under artificial light which, years later as an undergraduate student, I learned was due to a chemical reaction in which luciferin is converted to oxyluciferin by the enzyme luciferase. Later, during my graduate studies, I employed luciferase reporter assays as a tool to study gene transcription.

THE INFLUENCE OF A YOUNG ENTHUSIASTIC TEACHER

Having finished the seven years of high school, I was accepted as an undergraduate in mathematics at the Faculty of Science of the University of Oporto. I truly excelled in math, physics and chemistry, but became disappointed with my Math Professor and transferred to the Faculty of Pharmacy due to my love for inorganic, organic and biochemistry. Biochemistry was taught by Professor Carvalho Guerra, newly arrived from the Washington University School of Medicine in St Louis, Missouri, where he had undertaken his Ph.D. studies (Fig. 1). He was indeed an excellent Professor, allowing us to ask questions during his lectures if we failed to understand his points. In 1968, this method of teaching was not practiced by other Professors. He also liked to invite professors from other faculties to present subjects about which he was less knowledgeable. As a result, molecular biology was taught by Professor Luis Archer, who had also undertaken his Ph.D. studies in the United States. I was really enthusiastic about everything I was learning and received top scores in all disciplines. In the fifth year of my undergraduate studies, the faculty organized a visit to the Gulbenkian Research Center located in Oeiras, Lisbon, which in the 1960s was the country's top research institute. Thoroughly impressed with the laboratories and the research projects, I left the Research Center with a firm intention to engage in a scientific career within the field of biochemistry.

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Figure 1. This photograph was to celebrate the fifth year of the graduation. We were using a high hat and walking stick. Professor Carvalho Guerra first on the left, Author and 3-year-old son, Renato on the right.

Figure 2. (left panel) Author in front of the Institute of Biologie Physico Chimique, 1973. (right panel) Author with sons (left to right) Luis, Pedro and Renato.

AN UNINTERESTING BUT USEFUL TECHNIC

After graduating in 1968, I expressed my ideas about science and my deeply felt desire to engage in a career in scientific research to Professor Guerra. He strongly advised me to visit the laboratories of Microbiology and Pharmacology in the Gulbenkian Research Center and speak with the respective directors. As I had to move to Lisbon, where my husband was working as a civil engineer, I followed his advice. Arriving at the Gulbenkian Research Center without any previously arranged interview, I was first interviewed by Dr Van Uden, the Director of the Microbiology laboratory, and who was a native of the Netherlands (Fig. 2, left panel). I had a long interview, but he did not seem to be interested in engaging my collaboration. Unwilling to give up, I visited the Pharmacology Laboratory headed by Dr Peres Gomes. In contrast with Dr Van Uden, he was very gentle and interested in knowing what I wanted to do. I told him that I was interested in working in biochemistry and, given that his laboratory was in this area, I would be very happy to join his group. We talked about many things related to pharmacology and biochemistry, and after the hour interview, he told me that he would write me within a month with his answer. After a month of anxious waiting, I received his letter scheduling another interview for 15 December 1968. I was introduced to the biochemistry group led by Dr Maria Celeste Lechner, with whom I spoke and joined her group as a junior fellow three weeks later. I remained with her until September, 1973. My first research project was neither stimulating nor interesting, but at least it was an opportunity to learn about cellular fractionation, a useful technic for my work describing the processes of drug detoxification. Cytochrome P450s (CYPs) have long been interesting because of their critical function in detoxification and/or activation of xenobiotics e.g. drugs, pesticides, plant toxins, chemical carcinogens and mutagens as well as their role in the metabolism of endogenous compounds such as hormones, fatty acids and steroids.

To study these processes, rats were injected with phenobarbital for different periods of time after which they were killed. Their harvested livers were homogenized and cytochrome P450 and the aniline hydroxylases analyzed in their microsomal fractions. The fact that we observed an increase in the levels of cytochrome P450 and hydroxylase activities led us to postulate that their respective mRNAs were probably increased due to a decrease in RNase activity. Although this decrease was in fact observed, more recent studies indicated this interpretation to be incorrect. Indeed, multiple P450 genes are co-overexpressed/upregulated following stimulation.

PARIS: A FASCINATING WORLD OF AGONY AND ECSTASY

In early 1973, I decided to pursue my Ph.D. in Paris. Grants I received from the French Government and Calouste Gulbenkian Foundation, allowed me to remain in Paris for several years despite its famously high cost of living. This was a tough decision for me, however, because I was already the mother of a nine-year-old son and four-year-old twins, who remained at home in Portugal, with their father and a maid (Fig. 2, right panel). During the seventies, Portuguese women scientists were not accepted and, as such, I had little choice but to be strong if I wanted to pursue a scientific career. Male scientists received sufficient financial support for their families to accompany them, a benefit not afforded to their female counterparts. Most gratefully, my sons never criticized my decision to study in Paris and grew into fine young men. Renato and Pedro are professors at the Universities of Acquila (Italy), and Coimbra (Portugal) and Luis is in London studying History, and teaching Portuguese, English and French.

In September 1973, I joined the Institute of Biologie Physico Chimique, where I was mentored by Professor Donal Hayes, a person I much appreciated (Fig. 3, upper panel). He was a wise, sensitive and dedicated supervisor, who expended great effort to help me identify grant opportunities to finance frequent visits home to see my sons. As a young woman and coming to Paris from, at the time, an underdeveloped country, I was very enthusiastic to immerse myself into such a developed and enlightened society. What a fantastic world was opened to me! In spite of having emotional problems generated by the absence of my sons, I do not regret my stay in Paris.

The Institute was located in the typical ‘quartier latin’, facing Pierre et Marie Curie street close to Gay-Lussac and Claude Bernard streets. The Institute had, and still has, many laboratories whose members, in contrast to those in Portugal, were highly interactive. As such, we attended many lectures by invited scientists as well as those presented at the nearby College de France, where we could also attend courses. Among these extraordinary opportunities, I was fortunate to attend lecturers presented by Nobel Prize winners François Jacob and Jacques Monod as well as by François Gros and Jean-Pierre Changeux. (Fig. 3, lower panel).

Figure 3. (upper panel) Author with her supervisor, Professor Donal Hayes, his wife, Francoise (also a scientist) and daughter, Hélène. (lower panel) Dr Andre Goffeau, the Author, and Dr Peter Phillipsen at lunch, during the 1995 Workshop of the Yeast Genome.

IT WAS ABOUT MORE THAN JUST TRAVEL

Of the several projects Donal offered me, my favorite was the maturation of ribosomal and messenger RNAs. Thus, began my passionate work in preparation for my Ph.D. degree in Biochemistry. Thanks to my mentor’s open mindedness, I was given the opportunity to learn several technics I needed to perform my research from members of the Molecular Biology Department led by François Gros. From the moment I met François, I developed a great appreciation for him and his friendship. (Fig. 4, upper panel) I published several papers in European Journal of Biochemistry whose publishing criteria were not dependent on an impact factor. Our mentors also wanted us to attend Congresses abroad, which were of enormous benefit. This was not an option offered to graduate students at the Institute in my home country, because its Directors subscribed to the narrow-minded opinion that students were really interested only in an opportunity to travel. By the end of 1979, my Ph.D. thesis, entitled ‘The ribosome biosynthesis in the protozoa ciliate *Tetrahymena*’ was ready.

Figure 4. (upper panel) Author in 1994 with a group of her students. Peter Bossier is in the blue shirt, Hettema is in black next to Pascale Dupuis in the white lab coat, and Helena Soares is seated and behind standing is Lisete Fernandes. (lower panel) Opening ceremony of the 27th FEBS Congress in 2001, Lisbon.

SORRY, YOU DON'T GET TO PICK

I was employed by the Gulbenkian Institute of Science as an Assistant researcher while completing my Ph.D. Therefore, upon receiving it and returning to Portugal in 1979, I was promoted to Principal Researcher and began organizing my own lab. When thinking about the areas of research my future lab might follow, I considered working with the yeast *Saccharomyces cerevisiae*. There was a laboratory working with this biological model in the Institute, and its genetics were already well established in many laboratories around the world but not in Portugal. To my astonishment and dismay, however, the head of the department did not allow me to do so. In those days, the directors of Portuguese laboratories wielded tremendous authority. Therefore, I decided to continue working with *Tetrahymena*. I chose to investigate microtubule biosynthesis because this ciliate harbors a wide variety of microtubules in its cortex, cilia and cytoskeleton. By cloning and sequencing the α-tubulin and two β-tubulin genes, we became the first laboratory in Portugal to clone and sequence DNA (Barahona *et al.* 1988). My idea was to understand the functional differences underlying the great variety of tubulins in this ciliate. The number of genes, we found, was much smaller than the previously identified protein isoforms. We demonstrated the difference to derive from post-translational modifications, e.g. acetylation, phosphorylation and glutamylation. Within this context, my lab also characterized the *Tetrahymena* cytosolic chaperonin CCT, a hetero-oligomeric complex formed by two rings connected back-to-back, each composed of eight distinct subunits (CCTα to CCTζ). The CCT complex mediates folding of a wide range of newly synthesized proteins, with actin, α, β and γ tubulin being the bulk of its substrates (Soares *et al.* 1994).

I was also interested in understanding how gene expression was reprogrammed to cope with adverse conditions. We found that *Tetrahymena* responds to heat by inducing molecular chaperones and repressing normal protein synthesis as had been shown to occur in other biological models. We also showed the absence of cilia-induced molecular chaperone mRNAs with the concomitant destabilization of mRNAs expressed under physiological conditions. As I was launching my laboratory, French Government support permitted us to invite French lecturers, e.g. Donal Hayes, François Gros, Andre Sentenac, Michel Jacquet and Janine Beisson, to visit and discuss our results. François Gros, a longstanding friend, was especially helpful by supplying consumables that were not easily available in my home country.

DECIPHERING THE YEAST GENOME: A TEAM EFFORT

The first phase of the European Yeast Genome Sequencing Project was restricted to a small number of laboratories, but subsequently opened to the larger yeast community by Andre Goffeau. In conversation with Rudi Planta, Andre mentioned that he wanted to engage a Portuguese laboratory in the project, but, to his knowledge, no one in Portugal was sequencing. Fortunately, Rudi replied that there was indeed such a laboratory. As such, I decided to apply and join the network. We participated in sequencing Chromosome XI, coordinated by Bernard Dujon, and Chromosome VII, coordinated by Hervé Thettelin, both published in Nature. (Fig. 4, lower panel) Supported by the EU under the BIOTECH II programme, we published several yeast sequencing Reports in a specially created section of the journal Yeast. The most important one was a report on the gene YKL188c, located on the left arm of Chromosome XI, which encoded a homolog of the human ALD Protein (ALD:adrenoleukodystrophy, an X-linked disease) (Bossier *et al.* 1994). Soon after, I received a phone call from Henk F. Tabak in the Netherlands asking whether we might be interested in collaborating with his group. We received other phone calls as well, but Andre advised us to work with the Dutch team. Following his opinion and I invited Ewald H.Hettema, one of Henk's Ph.D. students, into the lab, where he generated a YKL188c mutant and showed that the deletion had no effect on growth in media containing glucose, acetate, glycerol or the fatty acids laureate (C12:0) or myristate (C14:0) as carbon source. However, growth on solid media containing either oleate (C18:1) or palmitate (C16:0) was impaired. The gene was subsequently designated as *PAT1* (peroxisome ABC transporter). After returning to his lab, Ewald carried out many biochemical analyses and verified that Pat1p and Pat2p were involved in the uptake of activated very long chain fatty acids (VLCFAs: $>$ C22), which are degraded in peroxisomes via β-oxidation. We were very happy with this collaboration because Ewald was a hard worker, with excellent intellectual capabilities and he immediately fit socially with the lab, being appreciated by all of our students. Thanks to this collaboration, a paper was published in EMBO J in which we suggested that the human X-ALD ABC transporter protein is more similar to Pat1p and Pat2p than to any other yeast ABC transporter and might be involved in the uptake of activated VLCFAs into human peroxisomes (Hettema *et al.* 1996). This work was subsequently pursued by Tabak's lab because I wanted to continue studying stress responses in yeast.

Figure 5. (upper panel) Author with the organizing team (mostly my students) for the second International Congress of CSSI on Stresses in Biology and Medicine, 2005, Tomar, Portugal. (lower panel) Fin Pol Committee meeting at the 22nd International Conference of Yeast Genetics and Molecular Biology in Bratislava, Slovak Republic (2005). Left to right Drs Ian Dawes (Pope), Terrance Cooper, Author, Florian Bauer.

APPROVED AT LAST—DISCOVERY OF YAP2 AND PLAYING WITH THE EXTENDED FAMILY OF YAP TRANSCRIPTION FACTORS

When Portugal became part of the European Community, our Spanish colleagues António Jimenez and Juan Pedro Ballesta invited me to participate in a joint project. These researchers were already working with yeast, and I realized that it was time to start working with yeast in my laboratory as well. I also had the great support of Rudi Planta to pursue this objective, and due to this advice from foreign colleagues, my previous departmental rejection regarding yeast research was now reversed. I was set to go. (Fig. 5, upper panel) A Belgian post-doc, Peter Bossier, from Mick Tuite's laboratory, was a great asset in launching our laboratory into the genetics of *S. cerevisiae*. Peter and I discussed the area of our research and decided to start by analyzing the mechanisms of thermotolerance. We planned to determine whether yeast cells acquired thermotolerance independent of heat-shock protein synthesis. We found papers reporting that low concentrations of 1,10-phenantroline produced growth arrest of *S. cerevisiae* in G1 and entry into the G0 phase. We also learned that this was probably due to its high affinity for and chelation of both Zn^{2+} and Fe^{2+} ions, which forced the cells into stationary phase. We used 1,10-phenantroline plates to selected transformants of a genomic library that were able to grow under these conditions. One of these clones contained the previously described *YAP1* gene, while the other contained a new member of the yeast AP-1 family, which we designated *YAP2*, due to its high homology with the AP-1 DNA-binding domain (Bossier *et al.* 1993). These proteins contain a basic DNA-binding domain and leucine zipper motif and bind *in vitro* to the same cis-element. (Fig. 5, lower panel) Overexpression of *YAP1* and *YAP2* alleviates the growth arrest of yeast caused by low concentrations of 1,10-phenanthroline and display pleiotropic drug resistance. On the other hand, a *yap2* null mutant had increased thermotolerance, under conditions of starvation caused by 1,10-phenanthroline (Bossier *et al.* 1993).

ZIPS, YAPS AND ARSENIC AS BOTH A THERAPEUTIC AND POISON

I then sent one of my Ph.D. students to attend a course on transcriptional regulation in Spetzes, Greece, financially supported by FEBS and EMBO. There she met Kevin Struhl and had the initiative to talk to him, thereby piquing his interest in our work. This, in turn, led to one of several very productive collaborations. Given that we were already studying two bZIP proteins, Yap1 and Yap2, we decided to search the complete yeast genome with a degenerate motif based on the sequence of a large number of basic regions in bZIP proteins from various organisms. We found 14 bZIP proteins, including Gcn4, Yap1, Yap2 and Met28 as well as the ATF/CREB proteins, Acr1/Sko1 and Hac1. Based on amino acid differences in two critical positions in their basic domains, we divided eight of the bZip proteins into two families, one previously known and a novel family consisting of Yap1-8. Investigation of their DNA-binding specificities and inherent activation potentials revealed that Yap1 and Yap3 were most active,

Figure 6. Ph.D. jury for one of my first Ph.D. students, Ana Neves (fourth from left), Author (third from right) and Dean of the University (fourth from right). Professor Rudi Planta from the Netherlands (third from left) participated as the main examiner.

but whereas the function of Yap1 is now very well described, the same is not true of Yap3 (Rodrigues-Pousada, Menezes and Pimentel 2010).

Since, then, we have concentrated our efforts on investigating the regulatory mechanisms whereby Yap proteins mediate genetic reprogramming that occurs in response to various stresses. Yap1 is regulated at the level of its subcellular localization, and we showed that the same is true for Yap2 and Yap8 (Rodrigues-Pousada, Menezes and Pimentel 2010). Under normal physiological conditions, exportin Crm1 binds to the Yap1 nuclear export signal (NES), resulting in its export to the cytoplasm. (Fig. 6) However, when cells are exposed to oxidative stress, Yap1 remains in the nucleus, activating antioxidant target genes e.g. *GSH1 SOD1* and *TRX2*. In collaboration with Michel Toledano, we concluded that the decision of whether or not Yap1 exits from the nucleus is regulated by two cysteine-containing redox centers within its C-terminus (Rodrigues-Pousada, Menezes and Pimentel 2010). In oxidative conditions, these redox centers mask and inactivate the NES, either by forming an intramolecular bridge in the presence of H_2O_2 or binding directly to electrophiles such as diamide. We additionally found that Yap1 is not only a transcriptional activator, but also a repressor, downregulating the expression of *FET4*, (a low-affinity iron and divalent metal ion transporter) (Caetano *et al.* 2015).

Yap2 and Yap8 intracellular localization is regulated by mechanisms similar to those we demonstrated for Yap1. Yap2 relocates to the nucleus in response to Cd^{2+}, whereas arsenic compounds are the important ligands for Yap8, alias Acr1 (arsenic compounds resistance 1) (Rodrigues-Pousada, Menezes and Pimentel 2010). In contrast to Yap1, which functions more broadly, Yap8 has a very restricted DNA-binding specificity, orchestrating only the expression of *ACR2* and *ACR3* genes. Using the *S. pombe* Pap1-DNA complex crystal structure (a Yap8 ortholog), bioinformatic tools, together with *in vivo* and *in vitro* approaches, we identified several conserved residues responsible for the exquisite specificity of Yap8-DNA binding and confirmed their importance *in vivo* with appropriate substitution mutations in a 2013 Plos One paper.

We then turned our attention to the contiguous *ACR1*, *ACR2* and *ACR3* genes. The latter two encode arsenate-reductase and a plasma membrane arsenite efflux protein, respectively, while sharing the same promoter. Although arsenic is often thought of as a deadly poison, it is one of the oldest drugs in the History of mankind; first used to treat cutaneous ulcers, later periodic fever and malaria, and currently, it is used in the treatment of acute promyelocytic leukemia (PML). These paradoxical biological effects led us to investigate the toxic effects of arsenic and demonstrate that it induces oxidative stress and destabilizes iron homeostasis in yeast and mammalian cells (Batista-Nascimento *et al.* 2013).

To investigate the stress-induced Yap4/Cin5 and Yap6 transcription factors, I established wonderful collaborations with Kevin Struhl, Michel Toledano and Dennis Thiele to perform transcriptional arrays as this technic simply did not exist in Portugal at the time. Primarily regulated by Hog1 signaling, transcription of these genes is subject to Msn2 (but not Msn4) via two proximal stress response elements (AG4) in their promoters. (Fig. 7, upper panel) Yap4 is, however, also induced by oxidative stress in a Yap1- and Msn2-dependent manner (Rodrigues-Pousada, Menezes and Pimentel 2010). Michael Snyder and his coworkers elucidated the temporal binding order of key yeast transcription factors (Hot1, Msn1, Msn2, Msn4, Skn7, Smp1 and Yap6) to their respective target genes under hyperosmotic conditions and found that this binding correlated with distinct classes of Yap4- and Sko1-binding patterns and distinct gene targets. This correlated well with our mutant and expression analyses, associating Yap6 with various stress responses.

BIG BACTERIA, ACADEMIA AND INDUSTRY

I have focused only on the work we performed in *Tetrahymena* and *Saccharomyces cerevisiae*. However, because we were, at the time, the only lab in Portugal engaged in molecular biology, we were also invited by other laboratories to decipher the complete genome of the bacterium *Desulfovibrio gigas*. I established collaboration with the company STAB-VIDA and the bioinformatic team led by Jerónimo Cruz (Centro de Pesquisa René Rachou—FIOCRUZ, Belo Horizonte, Brazil), who helped us with genome assembly and automatic annotation. We also subsequently participated in the manual curation of the annotated sequence. The skills I garnered in the yeast sequencing project were of upmost

Figure 7. (upper panel) Left to right: Tiago Outeiro, one of my brightest students and now Professor at the University of Gottingen, the Author responsible for this FEBS activity and the chair of the Young Scientist Forum Francesco Ruas (Torino). (lower panel) The Author when she received the FEBS Diplome d'honneur in 2009, Prague, Check Republic. Left to right: Professor Jose Mariano Gago, Minister for Science and Technology, Author, and Professor João Sentieiro, President of the Foundation for Science and Technology.

importance to the organization of our future work. Also, our collaboration with a biotech company was considered a prime example of fruitful collaboration between academia and industry.

We dedicated a Microbiology Open paper to the memory of the scientists Jean LeGall, who discovered *D. gigas*, and António V. Xavier, who worked with this organism for many years and stimulated us to pursue our studies with it. The designation *gigas* arose from the bacterium's uncharacteristically large size relative to other *Desulfovibrio* species. We identified the genes encoding FtsZ, the tubulin homolog responsible for cell division, and MreB, related to actin and involved in cell elongation of rod-shaped bacteria. The *D. gigas* genome contains an inhibitor of FtsZ assembly, the minCDE system, which may suggest the involvement of the encoded polypeptides in the distinguishing size of this bacterium.

PARIS LESSONS MAKING LEADERS

Although I supervised 30 Ph.D. students and published over 100 papers, I have also taught undergraduate students. Indeed, when I returned from Paris in the 1980s, I realized that the students, who joined my lab, were inadequately versed in the main concepts of Molecular Biology. The University, at that time, was old-fashioned, outdated, and subjects such as protein synthesis, transcription, replication and their respective regulation were completely ignored.

I soon realized that to engage students in scientific lab discussions that generated a critical intellectual mass in this field I would have to teach them myself. Moreover, the Universities of Oporto and Lisbon were launching new graduate courses in biochemistry, which included genetics and molecular biology. Given my expertise, I was invited to teach the students attending these Universities, an opportunity which I accepted with great enthusiasm as a good contribution to their education. The courses, I attended in the College of France, were invaluable in the preparation of my courses. Every Monday, early in the morning, I would catch the train to Oporto, where I spent the whole day in the University teaching their fantastic students. In order to introduce a new concept, I enjoyed proposing an experiment and then allowing them, through their own reasoning, to arrive at its interpretation. We also reviewed scientific papers related to the topic I was teaching, where students were asked to present the main conclusions and critiques of them to their colleagues. I could see members of the class with leader characteristics in our very lively discussions. The best of them are now actually the great scientists of my country and engaged in exceptional careers.

Arriving home in Lisbon from Oporto late in the evening, my heart was jumping with happiness. The fact that I was a scientist helped me to deliver the best lectures. At the University of Lisbon, I was asked to give a 4-week course of lectures and labs. My best students worked with me in these practical lab classes. It helped me and taught them to become good lab instructors. I went as far as taking some equipment and consumables from my lab for these practical classes because a budget was not provided for any of my lectures. We organized a small project, giving students the opportunity to learn several technics, e.g. sequencing Southern, Northern and Western blots. One day, I was very gratified upon learning that one of my previous undergraduate students was using the technics she learned during my course in her research project. Although a full professor at the University of Oporto with a 30% total salary, I taught at the University of Lisbon free of charge. Being a teacher and a mentor was incredibly rewarding and nothing has given me more satisfaction than to follow the careers of my best students.

AWARDS AND REWARDS OF ORGANIZING SCIENTIFIC EVENTS

I was elected member of the European Molecular Biology Organization (EMBO) in 1994 and received the prize in Genetics awarded by the Institute of Medical Genetics of Oporto. In 2001 and 2002, I received, together with a few colleagues (70), the Excellence in Science prize of 10 000€, given by the Minister of Science and Technology Professor, Graça Carvalho. I was President of the Executive Committee of the Federation of the European Biochemical Societies (FEBS) from 2002–03, 2005–07 and from 2011-present have chaired the FEBS Working Group on the Careers of Young Scientists. This is a work I truly enjoy because of its many contacts with young Ph.D. and post-doctoral students. I also received the 2009 FEBS Diplôme d'honneur, during a ceremony honored with the presence of our Minister for Science and Technology, Professor José Mariano Gago and the President of the Portuguese Foundation for Science and Technology, Professor João Sentieiro (Fig. 7, lower panel). In 2010, I received the 'Seeds of Science' prize from the on-line journal 'Ciência Hoje' (Science Today') and, in 2011, was elected as a fellow of the American Association for the Advancement of Science for 'distinguished contributions to the fields of stress regulation of

gene expression and yeast genetics and genomics’. These were the same reasons that led to my being elected senior fellow of the Cell Stress Society International.

The organization of scientific meetings is another activity I enjoy and fondly remember as it provides opportunities for me and my students to interact with other senior scientists. It is never a simple task, but our enthusiasm has always overcome the unexpected problems that always arise. In 1995, I was President of the Organizing Committee hosting the XVII International Conference on Yeast Genetics and Molecular Biology (951 participants). I then organized a 1997 EMBO workshop on Gene Expression Regulation Under Stress Response. It prompted my invited American speakers to subsequently apply for a biannual Gordon Research Conference on Stress Response and Molecular Chaperones. In 2001, I was President of the organizing and scientific committees of the joint congress of the 27th FEBS and Pan-American Association for Biochemical and Molecular Biology (2090 participants). This event was a real challenge, because a few months before the meeting was to open, the Congress Center Director decided to remodel the building leaving me fearing that it would not be ready in time. Fortunately, it was with the Minister for Science and Technology attending the opening ceremony. In 2005, I organized the Second International Congress on Stress responses in Biology and Medicine held in Tomar, Portugal (350 participants). The reasons I enjoyed organizing scientific events is because it has been my deeply felt belief that in so doing, I provide opportunities for young scientists to meet their seniors. Recently, one of my young colleagues told me that all of the collaborations in which he is now participating were initiated in one of the Congresses I organized.

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Conflict of interest. None declared.

REFERENCES

Barahona I, Soares H, Cyrne L *et al.* Sequence of one alpha- and two beta-tubulin genes of *Tetrahymena pyriformis*. Structural and functional relationships with other eukaryotic tubulin genes. *J Mol Biol* 1988;**202**:365–82.

Batista-Nascimento L, Toledano MB, Thiele DJ *et al.* Yeast protective response to arsenate involves the repression of the high affinity iron uptake system. *Biochim Biophys Acta* 2013;**1833**:997–1005.

Bossier P, Fernandes L, Rocha D *et al.* Overexpression of YAP2, coding for a new yAP protein, and YAP1 in *Saccharomyces cerevisiae* alleviates growth inhibition caused by 1,10-phenanthroline. *J Biol Chem* 1993;**268**:23640–5.

Bossier P, Fernandes L, Vilela C *et al.* The yeast YKL741 gene situated on the left arm of chromosome XI codes for a homologue of the human ALD protein. *Yeast* 1994;**10**:681–6.

Caetano SM, Menezes R, Amaral C *et al.* Repression of the low affinity iron transporter gene FET4: a novel mechanism against cadmium toxicity orchestrated by Yap1 via ROX1. *J Biol Chem* 2015;**290**:18584–95.

Hettema EH, van Roermund CW, Distel B *et al.* The ABC transporter proteins Pat1 and Pat2 are required for import of long-chain fatty acids into peroxisomes of *Saccharomyces cerevisiae*. *EMBO J* 1996;**15**:3813–22.

Rodrigues-Pousada C, Menezes RA, Pimentel C. The Yap family and its role in stress response. *Yeast* 2010;**27**:245–58.

Soares H, Penque D, Mouta C *et al.* A *Tetrahymena* orthologue of the mouse chaperonin subunit CCT gamma and its co-expression with tubulin during cilia recovery. *J Biol Chem* 1994;**269**:29299–307.