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JOINT DOCTORAL
PROGRAMMES

Programa Inter-Universitário de Doutoramento
em Biologia de Plantas - BioPLant
Joint Doctoral Programme in Plant Biology

1^a Workshop BioPlant

10 de Julho 2009 / *July 10, 2009*

PROGRAMA / PROGRAMME

Livro de resumos / Book of abstracts



Universidade do Minho



universidade de aveiro

U. PORTO

Bem vindos à 1ª Workshop do Programa Inter-Universitário de Doutoramento em Biologia de Plantas – BioPlant, que reúne docentes e investigadores provenientes de três Universidades, com o propósito de preparar de forma integrada e sinérgica a formação avançada da primeira edição do BioPlant. Vamos estar aqui à descoberta da ciência que fazemos ou queremos fazer, da formação que ministramos ou queremos ministrar, para diagnosticar de que forma podemos interagir para criar um resultado que seja muito mais do que a soma das suas partes.

E que resultado queremos? Queremos um Programa Doutoral de referência, que ofereça formação de excelência. Queremos um Programa Doutoral evolutivo, que ofereça formação avançada de qualidade crescente, ao servir ele próprio como motor para a dinamização e internacionalização da ciência que fazemos. Queremos um Programa Doutoral que salte os muros da Universidade, e vá ao encontro de necessidades de empresas e explorações para as quais temos competências para dar resposta.

*Queremos muito? Talvez, mas está **nas nossas mãos** fazer com que aconteça!*

Welcome to the 1st Workshop of the Joint Doctoral Programme in Plant Biology – BioPlant, which gathers together professors and researchers coming from three Universities, with the purpose of preparing, in an integrated and synergic way, the advanced courses of the first edition of BioPlant. We'll be here to discover the science that we do or wish to do, the courses that we teach or wish to teach, in order to diagnose in which way we may interact to create a result that becomes more than a sum of its parts.

And what result do we want? We want a Doctoral Programme of reference, offering an education of excellence. We want an evolutionist Doctoral Programme, offering advanced education of growing quality, as it will itself empower the dynamics and internationalization of the science we do. We want a Doctoral Programme that jumps over the walls of University, and comes across needs of companies and producers to which we have competences to respond.

*Do we want too much? Maybe, but it's **in our hands** to make it happen!*

Mariana Sottomayor
Directora do BioPlant 2009/2010

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Apoios / Support

O BioPlant tem o privilégio de usufruir de um financiamento da **Fundação Calouste Gulbenkian** ao abrigo do seu Programa de Reforço da Capacidade Científica para Projectos Inter-Universitários de Doutoramento, obtido em concurso nacional em que apenas dois programas foram subsidiados. Este financiamento dotou o BioPlant com 180.000 EUR destinados a financiar exclusivamente despesas de funcionamento durante os próximos três anos.

Organização da Workshop / Organization of the workshop

Mariana Sottomayor – Departamento de Botânica, FCUP
Paula Melo - Departamento de Botânica, FCUP
Carmencita Lino - EPALMO

Comissão Directiva BioPlant / Executive Committee BioPlant

Mariana Sottomayor UP - *Directora do BioPlant 2009/2010*
Alberto Dias UM
Conceição Santos UA

Comissão Científica BioPlant / Scientific Committee BioPlant

Mariana Sottomayor UP - *Presidente da CC 2009/2010*
Alberto Dias UM
Conceição Santos UA
João Seródio UA
Manuel Ferreira UM
Paula Melo UP

Instituições Promotoras / Promoting Institutions

Departamento de Botânica da Faculdade de Ciências da Universidade do Porto (DB-FCUP) – *Instituição de acolhimento 2009/2010*
Departamento de Biologia da Universidade de Aveiro (DB-UA)
Departamento de Biologia da Universidade do Minho (DB-UM)

Programa / Programme

09:30 – **Abertura / Opening.**

Isabel Salgado Labouriau, Vice-Directora da FCUP.

09:45 – **Apresentação do Bioplant / Presentation of Bioplant.**

Mariana Sottomayor, FCUP/IBMC, Directora do Bioplant.

10:30 – Coffee Break e/and Posters

11:00 – **PlantUM – “Plantas na Universidade do Minho” / PlantUM - “Plants at the University of Minho”.**

Alberto Dias, DB-UM/CITAB.

11:10 – **Oferta formativa do Departamento de Biologia da Universidade do Minho | CITAB ao Bioplant / Education offer from the Department of Biology of the University of Minho | CITAB to Bioplant.**

Manuel Ferreira, DB-UM/CITAB.

11:30 - **Oferta Formativa do Departamento de Biologia da Universidade do Minho | BioFIG Ao Curso Doutoral em Biologia de Plantas – BioPlant / Education offer from the Department of Biology of the University of Minho | BioFIG to the Doctoral Programme in Plant Biology - Bioplant.**

Rui Tavares, DB-UM/BioFIG.

11:50 – **Envolvimento de proteínas arabinogalactânicas na reprodução sexuada de *Arabidopsis thaliana* / Role of arabinogalactan proteins in *Arabidopsis thaliana* sexual reproduction.**

Silvia Coimbra, FCUP/BioFIG.

Tráfego intracelular, desenvolvimento vegetal e stress / Intracellular trafficking, plant development and stress.

Susana Pereira, Fernanda Fidalgo, FCUP/BioFIG.

12:10 – **CIBIO-UP: Biodiversidade e ecologia da conservação / CIBIO-UP: Biodiversity and Conservation Ecology.**

João Honrado, FCUP/CIBIO.

12:20 – **Apresentação do Laboratório de Palinologia. Pólen e as suas aplicações / Presentation of the Palynology laboratory. Pollen and its applications.**

Ilda Noronha, FCUP/ ENVISED–CGUP.

12:30 - Almoço / Lunch

14:00 – Sessão de Posters / Posters Session

15:00 - **Biotecnologia vegetal e citômica em Aveiro: uma visão geral** / *Plant biotechnology and cytomics at Aveiro: an overview.*
Conceição Santos, DB-UA/CESAM.

15:10 – **Impacto das alterações climáticas nas plantas: estratégias e marcadores** / *Climate changes impact in plants: strategies and endpoints.*
Glória Pinto, DB-UA/CESAM.

15:20- **Estudos da flora e vegetação: biodiversidade, estudos de impacto ambiental e ecoturismo** / *Flora and vegetation studies: biodiversity, environmental impact studies and ecotourism.*
Paulo Silveira, DB-UA/CESAM

15:30 – **Biodiversidade dos Ecossistemas Aquáticos** / *Biodiversity of Aquatic Ecosystems.*
Isabel Sousa Pinto, FCUP/CIMAR.
Laboratório de Ecofisiologia Microbiana / *Laboratory of Microbial Ecophysiology.*
Olga Lage, FCUP/CIMAR.

15:40 - **Ecofisiologia da fotossíntese em algas** / *Ecophysiology of photosynthesis in algae.*
João Seródio, DB-UA/CESAM.

15:50 - **Microbiologia Celular e Aplicada: tópicos de investigação em interações plantas - microorganismos** / *Cellular and Applied Microbiology: research topics in plant-microbes connections.*
Fernando Tavares, Paula Tamagnini, FCUP/IBMC.

16:00 – Coffee Break e/and Posters

16:30 – **Microbiologia da rizosfera** / *Rhizosphere microbiology.*
Newton Gomes, DB-UA/CESAM.

16:45 - **Biologia de plantas no IBMC. Comunicação da ciência no DB-FCUP e IBMC** / *Biology of plants at IBMC. Communication of science at DB-FCUP and IBMC.*
Mariana Sottomayor, FCUP/IBMC.

17:00 – **Debate: BioPlant – objectivos e estratégias.** / *Debate: BioPlant - objectives and strategies.*

18:00 – Chill Out

Lista de Participantes / List of Participants

Nome	Contacto	Áreas / Linhas de Investigação
Ana Cristina Gomes da Cunha	accunha@bio.uminho.pt DB-UM IBQ – Integrative Biology and Quality; CITAB – Centro de Investigação e de Tecnologias Agro-Ambientais e Biológicas	In vitro plant culture Lipid analysis (In vitro) Plant stress physiology Biological activities of plant extracts and other natural products (mainly allelopathic effects)
Alberto Dias	acpdias@bio.uminho.pt DB-UM	Secondary metabolism, metabolites and Metabolomics; phenolics, PUFAs, lipids, Antioxidants, neuroprotection and anti – ageing plant metabolites. Biopesticides and plant biotic stress.
Celeste Dias	celeste.dias@ua.pt DB-UA CESAM	Ecologia/ Ecofisiologia de plantas; Stress Fisiológico; Biotecnologia de Plantas.
Cristina P. Vieira P10	cgvieira@ibmc.up.pt Molecular Evolution Group at IBMC	Plant Molecular Evolution and Population Genetics
Cristiana Vieira	cristianavieir@gmail.com CIBIO: Biodiversity and Conservation Ecology Group	Caracterização ecológica e biológica de comunidades vegetais Brioflora de Portugal Continental Biomonitorização de habitats e espécies raras Requalificação ecológica de comunidades fluviais.
Domingos Almeida	dalmeida@fc.up.pt CBOF – Interface A4 FCUP / Frulact SA	Fisiologia da maturação e da senescência Metabolismo de paredes celulares em frutos Cinética respiratória em frutos minimamente processados (aplicado) Ecofisiologia de roseiras (efeito da humidade relativa na regulação estomática e implicações para a qualidade pós-colheita de rosas)
Fernanda Fidalgo P12/13/14/ 15	ffidalgo@fc.up.pt DB-FCUP BioFIG Center for Biodiversity, Functional and Integrative Genomics	Bioquímica e Fisiologia Vegetal - Stresse oxidativo, sistemas antioxidantes e tolerância das plantas a stresses abióticos - Mecanismos de adaptação das plantas a contaminantes ambientais - Bio-indicadores de stresse em ecossistemas dunares litorais

<p>Fernando Tavares P18/19</p>	<p>ftavares@ibmc.up.pt DB-FCUP IBMC</p>	<p>Detecção, identificação e Genotipagem de Microrganismos fitopatogénicos. Caracterização da resposta ao stress oxidativo durante infecções bacterianas (simbióticas, mutualísticas ou patogénicas) em plantas. Regulação, fisiologia e evolução.</p>
<p>Glória Catarina Cintra da Costa Pinto</p>	<p>gpinto@ua.pt UA Laboratory of plant biotechnology and cytomics/ CESAM</p>	<p>Plant Ecophysiology, functional genomics, plant performance, genetic control, plant biotechnology</p>
<p>Harald Meinberg P11</p>	<p>meimberg@mail.icav.up.pt PI Plant Evolution Group, CIBIO</p>	<p>Adaptation and selection patterns during Biological invasions, Horizontal gene flow and Hybrid speciation, Polyploid speciation</p>
<p>Helena Carvalho P16</p>	<p>mhcarval@ibmc.up.pt Molecular Biology of Nitrogen Assimilation-IBMC</p>	<p>Assimilação do Azoto nas plantas Simbiose entre Leguminosas e Rizhobium, Fixação Biológica do Azoto. Regulação de enzimas chave no metabolismo do azoto.</p>
<p>Helena Isabel da Costa Ribeiro</p>	<p>helena.ribeiro@fc.up.pt ENVISED – CGUP</p>	<p>Palinologia Modelação</p>
<p>Hernâni Varanda Gerós</p>	<p>geros@bio.uminho.pt IBQ / CITAB</p>	<p>Plant Biochemistry / Nutrient metabolism and transport / Sress Biology</p>
<p>Ilda da Conceição Abreu de Noronha P17/20/21/22</p>	<p>ianoronh@fc.up.pt DB-UP ENVISED – CGUP</p>	<p>Anatomia e Estrutura das Plantas; Biologia floral; Palinologia;</p>
<p>Isabel Mina P1</p>	<p>icapmina@bio.uminho.pt DB-UM (CITAB) – Grupo de Biologia Integrativa e Qualidade (IBQ)</p>	<p>Ecologia e Actividade Biológica em “Zonas Húmidas Construídas”(FitoETARs)</p>
<p>Isabel Laboriau</p>	<p>islabour@fc.up.pt DMA-FCUP</p>	

<p>João Honrado P23/24/25</p>	<p>jhonrado@fc.up.pt DB-FCUP CIBIO: Biodiversity and Conservation Ecology Group</p>	<p>Ecologia Funcional Ecologia da Paisagem Ecologia da Vegetação e Fitosociologia</p>
<p>Joao Serodio P3</p>	<p>jserodio@ua.pt DB-UA CESAM – Centro de Estudos do Ambiente e do Mar</p>	<p>Microalgal ecophysiology and primary productivity. Diatom photophysiology. Photosynthesis, Photoprotection and Photoinhibition. Photosynthetic symbioses. Pulse Amplitude Modulation Fluorometry Spectral reflectance analysis</p>
<p>Jorge Teixeira</p>	<p>jteixeira@fc.up.pt DB-FCUP BioFIG Center for Biodiversity, Functional and Integrative Genomics</p>	<p>Bioquímica e Fisiologia Vegetal - Stresse oxidativo, sistemas antioxidantes e tolerância das plantas a stresses abióticos - Mecanismos de adaptação das plantas a contaminantes ambientais - Bio-indicadores de stresse em ecossistemas dunares litorais</p>
<p>Jose Pissarra P33/34/35/36</p>	<p>jpissarr@fc.up.pt DB-FCUP BioFIG Center for Biodiversity, Functional and Integrative Genomics</p>	<p>Bioquímica e Fisiologia Vegetal Trânsito de Proteínas e Desenvolvimento - Identificação de determinantes de endereçamento intracelular - Especialização das vias de trânsito durante o desenvolvimento</p>
<p>Manuel Ferreira</p>	<p>mferreira@bio.uminho.pt DB-UM</p>	
<p>Maria da Conceição Santos</p>	<p>csantos@ua.pt DB-UA Laboratory of plant biotechnology and cytomics, CESAM</p>	<p>Biotecnologia vegetal. Toxicologia ambiental. Citomica estrutural e funcional vegetal. Genómica.</p>
<p>Maria Eugenia Santos Nunes</p>	<p>enunes@fc.up.pt CIBIO /FCUP</p>	<p>(1) Taxonomia, reconstrução filogenética e evolução, (2) adaptação ao stress e (3) compostos bioactivos.</p>
<p>Maria Helena Silva P4/5/8</p>	<p>hsilva@ua.pt DB-UA CESAM</p>	<p>Biodiversidade e ecofisiologia das plantas vasculares das zonas litorais: estudo das plantas de sapal (produtividade, factores de stress, interacção planta-rizosfera)/ Biodiversity and ecophysiology of vascular plants from littoral zones: study of saltmarsh plants (productivity, stress factors, interaction plant-rizosphere).</p>

Maria Isabel Amorim	mpamorim@fc.up.pt DB-FCUP	Biologia Molecular e Celular. Biologia Funcional e Genética de Plantas. Aerobiologia e imunoalergologia de esporos e polens
Mariana Sottomayor P26/27/28	msottoma@ibmc.up.pt DB-FCUP	1 - Functional studies of class III peroxidases and their impact in the chemistry and properties of vacuoles and cell walls in Arabidopsis. 2 - Interactions of class III peroxidases with arabinogalactan proteins. 3 - Subcellular sorting of class III peroxidases in Catharanthus and Arabidopsis. 4- Class III peroxidases and indole alkaloid metabolism in Catharanthus. 5 - Mechanisms of transport and accumulation of medicinal indole alkaloids in Catharanthus. 6 - Phytochemical screening and bioactivities of Catharanthus.
Miguel Angelo Faria	mfaria@ff.up.pt Serviço de Bromatologia FFUP REQUIMTE	(1) Taxonomia, reconstrução filogenética e evolução, (2) adaptação ao stress e (3) compostos bioactivos.
Newton Carlos Marcial Gomes P6	gomesncm@ua.pt Ecosistemas Marinhos e Modelação (EMM), CESAM	- Investigação das comunidades microbianas do solo e interações planta-microrganismos. - Desenvolvimento de técnicas moleculares para detecção de genes funcionais e o estudo de comunidades microbianas em amostras ambientais. - Avaliação multidisciplinar de ecossistemas estuarinos expostos a contaminantes antropogénicos e influência das alterações globais. - Estudar os padrões estruturais e funcionais de comunidades microbianas em diferentes ecossistemas.
Olga Maria Lage P30/31/32	olga.lage@fc.up.pt DB-FCUP Laboratório de Ecofisiologia Microbiana – CIIMAR	Ecophysiological studies of Planctomycetes Transmission electron microscopy (TEM) Cell wall signaling and DNA protection against environmental damage in Planctomycetes (com Rui Oliveira, Departamento de Biologia, Universidade do Minho)
Patrícia Macedo	pduarte@ibmc.up.pt IBMC	Plant biology namely cellular biology and physiology. Study of the sorting of soluble proteins in the plant secretory pathway.
Paula Melo P29	pmmelo@ibmc.up.pt DB-FCUP IBMC	Simbiose Regulação pos-transcrição da enzima glutamina sintetase

<p>Paulo Cardoso da Silveira P5/7/8/9</p>	<p>psilveira@ua.pt DB-UA Biodiversidade Funcional – CESAM</p>	<p>Sistemática e ecologia de plantas vasculares (flora de Portugal, Península Ibérica, Angola, Moçambique e Timor Leste), conservação da natureza, avaliação e monitorização de impactos ambientais (flora e vegetação).</p>
<p>Rosa Maria Ferreira Pinho P8</p>	<p>rpinho@ua.pt DB-UA</p>	<p>Sistemática e ecologia de plantas vasculares, estudos de impacto ambiental, planos de ordenamento do território, monitorização ambiental (descriptor flora e vegetação). Acções de divulgação e educação ambiental (ex. Biologia no Verão – Ciência Viva, Criação de Trilhos em Parques urbanos, visitas guiadas, etc.). Estudos de Flora ornamental (ex. Parque de Serralves e Mata do Buçaco).</p>
<p>Rui Manuel Tavares</p>	<p>tavares@bio.uminho.pt DB-UA BioFIG-Pólo da Universidade do Minho</p>	<p>Fisiologia Molecular de Plantas Genómica Funcional</p>
<p>Rui Pedro Soares de Oliveira P2</p>	<p>ruipso@bio.uminho.pt Programmed Cell Death research group. Molecular and Environmental Research Center (CBMA)</p>	<p>DNA degradation in apoptosis and DNA repair in the yeast <i>Saccharomyces cerevisiae</i> Search for natural compounds with antigenotoxic activity and mechanism of action Cell wall signalling and remodeling in the pathogenic yeast <i>Candida albicans</i>.</p>
<p>Silvia Coimbra</p>	<p>scoimbra@fc.up.pt DB-FCUP BioFIG – grupo AGPs /</p>	<p>Reprodução sexuada em <i>Arabidopsis</i>, nomeadamente o estudo do envolvimento das proteínas arabinogalactânicas na gametogénese e na interacção pólen-pistilo</p>
<p>Susana Pereira P33/34/35/36</p>	<p>mspereir@fc.up.pt DB-FCUP BioFIG Center for Biodiversity, Functional and Integrative Genomics</p>	<p>Bioquímica e Fisiologia Vegetal Trânsito de Proteínas e Desenvolvimento - Identificação de determinantes de endereçamento intracelular - Especialização das vias de trânsito durante o desenvolvimento</p>

Resumos dos Posters / Poster Abstracts

UNIVERSIDADE DO MINHO

P1 TRATAMENTO DE ÁGUAS RESIDUAIS - SOLUÇÕES DE BAIXA TECNOLOGIA

ISABEL MARIA CRAVO AGUIAR PINTO MINA, DB-UM/UM, CITAB/IBQ

Considerando a crescente construção de FitoETARs em países da comunidade europeia, incluindo Portugal, é nosso objectivo aumentar a eficiência destes sistema de tratamento, testando inovações introduzidas em protótipo(s) de FitoETARs construídos para o efeito. A avaliação da eficiência dos sistemas será feita pela não só pela determinação de parâmetros tradicionalmente utilizados em engenharia sanitária mas também recorrendo à monitorização de comunidades biológicas destes sistemas.

Palavra(s) Chave:

FitoETARs, macrófitas, bioindicadores, qualidade sanitária

P2 Studies of DNA repair and toxicity mechanism after an oxidative challenge by hydrogen peroxide in *Saccharomyces cerevisiae*: the protective effect of *Ginkgo biloba* leaf extracts

Rui Pedro Soares de Oliveira, DB-UM/CBMA

A reliable and sensitive technique for detection of DNA damage is the Single Cell Gel Assay (comet assay). In this procedure, cells are exposed to an electric field and damaged DNA moves out of the nucleus, which can be visualized by ethidium bromide staining, displaying a comet appearance under fluorescence microscopy. We show that there is a correlation between peroxide concentration and comet tail length and that DNA damage increases as the yeast population enters the post-diauxic phase, which is in agreement with the accumulation of DNA damage with aging. We show also that a water extract from *Ginkgo biloba* leaves protect DNA against oxidative stress. DNA repair kinetics was assessed as well by the comet assay after oxidative challenge and a subsequent recovery period. For this, we are using different mutants affected in NER and BER DNA damage repair pathways to determine the repair pathway involved in the oxidative damage response. To investigate the peroxide toxicity mechanism and to assess the protection mechanism of antigenotoxic compounds, we are testing the non-metabolizable glucose-analog 2-deoxyglucose in cell viability and in DNA damage after peroxide treatment. Our results show an increased viability concomitant with decreased tail length, suggesting that peroxide requires an active energetic metabolism for its toxicity.

UNIVERSIDADE DE AVEIRO

P3 Ecophysiology of photosynthesis in algae: oxidative stress, photoprotection and photoinhibition

João Serôdio, DB-UA/CESAM

Main lines of research are centered on the study of processes of photoprotection against photooxidative stress in marine algae. These include the operation of the xanthophyll cycle, effects of antioxidant agents, D1 protein turnover, and behavioural photoprotection. Most work has been done on diatoms, an important group of microalgae responsible for 25% of the carbon fixation in the biosphere. Diatoms have exceptionally high photoprotective capacity allowing to colonize and dominate in extreme environments such as coastal and estuarine shallow waters (phytoplankton) and intertidal flats (microalgal biofilms, microphytobenthos).

Another line of research addresses the photobiology of photosynthetic symbioses, biologically unique associations between marine invertebrates and algae. These ‘solar-powered’ animals depend on the photosynthates produced by the algal symbiont, attracting scientific interest due to remarkable interplay between animal physiology and behaviour and the photosynthetic metabolism.

Emphasis has been put on the use and development of non-destructive techniques to study *in vivo* the photosynthetic processes: Pulse Amplitude Modulation (PAM) fluorometry, which enables to monitor in real time the functioning of the photosynthetic apparatus, and spectral reflectance analysis, which permits to remotely assess photosynthetic pigment content, and infer on photophysiological processes.

P4 The role of salt marsh vegetation in sedimentary processes in the Aveiro lagoon (Portugal)

Sara Bárrios⁽¹⁾, Inês Silva⁽¹⁾, João Miguel Dias⁽²⁾, Helena Silva⁽¹⁾,

⁽¹⁾Departamento de Biologia, Universidade de Aveiro, Portugal, hsilva@bio.ua.pt

⁽²⁾Departamento de Física, Universidade de Aveiro

Rates of sediment accretion in the Aveiro lagoon saltmarsh are critically important in view of rising relative sea level. Sedimentation rates may be influenced by a range of factors such as the elevation of salt marsh, the duration of tidal flooding, tidal range, storms, organic matter, compaction and vegetation cover. Variations in sedimentation patterns also occur seasonally and may be influenced by biotic processes. The sediment samples were taken from places without vegetation and with different vegetation cover (*Spartina maritima*, *Sarcocornia perennis subsp. perennis* and *Halimione portulacoides*) according to a transect that was set up perpendicularly to the main channel. Four sampling sites were selected (Verdemilho, Barra, Varela-ria and Torreira), on the main channels of the lagoon and short term sediment depositions were studied: the measurements were taken monthly using seven replicates of pre-weighed nylon calibrated net (mesh- 15 µm; diameter- 8 cm) placed on the sediment and fixed with plastic coated paper clips. Every month the disks were carefully recollected into Petri dishes and the dry weight of sediment was registered. The results were discussed according to the predicted depth mean current values obtained by the application of a mathematical model developed for the Aveiro lagoon. Deposition (g. m⁻². dia⁻¹) was very variable in each zone, but Verdemilho usually presented the highest values of deposition and Torreira presented signs of erosion. According to the results, the role of each species in the sedimentation processes was highly influenced by the particular edaphic conditions of each saltmarsh and also by the phenology of the plant.

Note: published in Hydrobiologia 621: 33-47 (2009)

P5 Uma nova abordagem na descoberta da flora do Baixo Vouga

Lopes L.; Ezequiel J.; Maia P.; Silva I.; Bárrios S.; Lourenço A.; Pinho R.; Silva H.; Silveira P.
Departamento de Biologia, Universidade de Aveiro, Campus Universitário de Santiago 3810-193 Aveiro, Portugal. (lisia@bio.ua.pt)

A maioria da informação usada para identificação de plantas está contida nas floras e outras obras taxonómicas que, por vezes, são demasiado complexas e de difícil consulta por parte de quem demonstra potencial interesse pela diversidade vegetal como, por exemplo, os docentes e discentes do ensino básico e secundário. Existem no mercado, pequenos guias de identificação, mas estes resultam normalmente de adaptações ou simples traduções de obras estrangeiras, não estando completamente adaptados à realidade encontrada no campo.

Inserida no Projecto Radical, que se enquadra no 2º Programa Aveiro – Cidade Digital, uma equipa do Departamento de Biologia da Universidade de Aveiro, desenvolveu um software, que pretende facilitar a identificação da flora vascular, especialmente adaptada à área de estudo, o Baixo Vouga Lagunar (Ria de Aveiro), onde é possível encontrar *habitats* naturais, comuns a muitos outros pontos de Portugal. Esta ferramenta pode ser acedida via internet ou em DVD-ROM e inclui chaves de identificação para as espécies presentes naquele tipo de ecossistema, contempla também um glossário ilustrado (com fotografias e esquemas), de forma a facilitar a sua interpretação por parte dos leigos no tema. A identificação pode ser confirmada facilmente pela ligação que é estabelecida entre cada espécie constante nas chaves e a respectiva caracterização efectuada no âmbito do Projecto Biorede (1º Programa Aveiro – Cidade Digital) e que tem vindo a ser expandida e melhorada dentro do contexto do actual projecto (Radical).

Os objectivos deste projecto são: facilitar a divulgação científica da biologia, incrementar o estudo das espécies, cada vez mais ausente dos conteúdos programáticos do ensino básico e secundário e permitir a interligação entre diversos graus de ensino e a comunidade em geral.

P6 MICROBIOLOGIA DA RIZOSFERA

Newton Carlos Marcial Gomes, DB-UA/CESAM

O termo rizosfera designa a poção de solo ou sedimento que contacta directa ou indirectamente com a raiz das plantas e onde os processos mediados por microrganismos se desenvolvem sob a influência das raízes. A interacção entre microrganismos e plantas ao nível da rizosfera tem impacte na nutrição e saúde das plantas mas tem também repercussões ao nível do ecossistema uma vez que afecta processos microbianos de captura e transformação da matéria orgânica, nutrientes inorgânicos, metais e poluentes.

A dinâmica microbiana na rizosfera é afectada por diversos factores bióticos e abióticos dos quais se destacam a espécie da planta em causa e as propriedades físicas do solo ou sedimento.

A quantidade e qualidade dos exsudados das raízes bem como a morfologia radicular impõem um carácter de elevada especificidade na estrutura e actividade das comunidades microbianas da rizosfera que só pode ser descrita através da integração de abordagens microscópicas, fisiológicas e moleculares.

A microbiologia da rizosfera tem implicações directas em áreas relacionadas com agricultura, conservação dos recursos naturais, biorremediação e biotecnologia. O objectivo geral deste módulo é a compreensão das interacções microrganismo-planta ao nível da rizosfera, bem como das implicações fisiológicas e ecológicas que lhe estão associadas e das abordagens metodológicas ao estudo da microbiologia das rizosferas.

P7 ESTUDO PALINOLÓGICO DO GÉNERO *TYLOSEMA* (SCHWEINF.) TORRE & HILLC.

COUTINHO, AP¹; CASTRO, S²; SILVEIRA, P²; FIGUEIREDO, E³

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O estudo da morfologia polínica, através de microscopia óptica, de varrimento e/ou de transmissão, constitui uma importante ferramenta em estudos taxonómicos. Isto deve-se, essencialmente a duas características dos grãos de pólen: 1) a sua extrema resistência a degradações de natureza física, química e biológica; 2) e a sua elevada especificidade.

O objectivo deste estudo consistiu na análise de grãos de pólen pertencentes a quatro espécies do género *Tylosema* (Schweinf.) Torre & Hillc. e a uma população de indivíduos pertencentes a este género mas cujas características morfológicas colocam problemas taxonómicos. Assim, procedeu-se ao estudo de grãos de pólen acetolisados através de microscopia óptica e microscopia electrónica de varrimento, tendo os dados numéricos obtidos sido tratados por testes estatísticos adequados.

Os resultados obtidos permitiram efectuar a primeira descrição palinológica para o género *Tylosema* e possibilitaram a distinção dos diferentes grupos taxonómicos estudados. Apoiam ainda a delimitação de um novo *taxon* já individualizado pelas suas características morfológicas.

P8 Estudos da flora e vegetação: biodiversidade, avaliação de impactes e ecoturismo

Paulo Silveira, Helena Silva & Rosa Pinho, DB-UA/CESAM

Os estudos na área da biodiversidade estão longe de estar ultrapassados, novas questões/aplicações no âmbito da biodiversidade funcional, avaliação de impactos/monitorização ambiental e no âmbito do ecoturismo, têm despoletado um renovado interesse e necessidade de especialistas nestas áreas.

Pretende-se, com esta apresentação, demonstrar as valências da equipa nestas áreas, bem como a proposta de curso a implementar.

The biodiversity studies are far from being outdated, new questions/applications on the scope of functional biodiversity, environmental impact studies and ecotourism, have renewed the interest on this areas and the need for well prepared specialists. We pretend to resume the team's research on these areas and to introduce the proposed course.

P9 A ENERGIA EÓLICA E A CONSERVAÇÃO DA NATUREZA: UM CASO DE ESTUDO NA SERRA DO AÇOR

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A serra do Açor faz parte da chamada Cordilheira Central assim como a Serra da Estrela e a Serra da Lousã. Geologicamente, é constituída essencialmente por uma grande variedade de xistos, possuindo também algumas cristas quartzíticas. Insere-se biogeograficamente na Região Mediterrânica, embora possua grande influência Atlântica, com as encostas Norte e os cumes mais elevados no Sector Estrelense, as encostas e cumeadas mais ocidentais e as vertentes viradas a Sudoeste, no Subsector Beirense Litoral e com influencia do Superdistrito Zezerense, sobretudo nas encostas mais orientais, dispostas a Sudeste.

Nesta região são característicos os urzais de queiroga e tojo-molar (*Ulici minoris-Ericetum umbellatae*) e as comunidades rupícolas e fissurícolas, ricas em plantas com distribuição restrita e elevado estatuto conservacionista, como é o caso dos endemismos lusitanos, *Teucrium salviastrum* e *Murbeckiella sousae*. Podem ainda ocorrer pequenos bosquetes da aliança *Quercion robori-pyrenaica*, embora, geralmente, muito degradados e de pequena dimensão.

Os futuros Parques Eólicos de Toita (Concelhos de Pampilhosa da Serra e Góis), Arouca-Silva (Concelho de Pampilhosa da Serra) e Vale Grande-Burrela (Concelho de Arganil), situados na Serra do Açor, representam actualmente uma excelente alternativa aos recursos energéticos não renováveis. No entanto, apesar de ser uma energia limpa, ecológica e renovável, não é isenta de impactes. Assim sendo, a localização, instalação e manutenção dos parques eólicos, deve permitir a salvaguarda dos valores ecológicos locais. Com este trabalho pretendeu-se avaliar a importância destes valores ecológicos, no que concerne à flora e vegetação locais e definir áreas com diferente relevância florística, de forma a permitir o planeamento dos referidos parques eólicos minimizando os impactes resultantes da respectiva instalação.

A caracterização e relevância das comunidades vegetais teve como objectivo final a elaboração de uma Carta de Significâncias para a área de estudo, baseando-se nas seguintes etapas metodológicas:

- 1) Identificar a ocorrência de *habitats* e comunidades naturais constantes da Directiva 92/43/CEE - Directiva *Habitats*;
- 2) Identificar a ocorrência de espécies RELAPE (Raras, Endémicas, Localizadas, Ameaçadas ou em Perigo de Extinção) e/ou constantes da Directiva 92/43/CEE - Directiva *Habitats*;
- 3) Delimitar (por georeferenciação) as áreas com interesse conservacionista para a flora e vegetação.
- 4) Inferir acerca do valor florístico e importância de cada formação vegetal (utilizando uma metodologia semelhante à proposta para os planos de ordenamento das áreas protegidas-ICNB);

As Cartas de Significância ecológica produzidas permitiram planear a disposição de acessos e locais de implantação das torres aerogeradoras, de forma a preservar as zonas com formações vegetais/habitats ou comunidades de espécies protegidas de maior valor conservacionista, promovendo a conservação dos valores naturais da região.

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P10 Recombination at *Prunus* S-locus region *SLFLI* gene

Cristina Vieira, IBMC

In *Prunus*, the self-incompatibility (*S*-) locus region is smaller than 70 Kb. Two genes, the *S-RNase*, that encodes the functional female recognition component, and the *SFB* gene, that encodes the pollen recognition component, must co-evolve as a genetic unit in order to maintain functional incompatibility. Therefore, recombination must be severely repressed at the *S*-locus. Levels of recombination at genes in the vicinity of the *S*-locus have not been yet rigorously tested, thus it is unknown whether recombination is also severely repressed at these loci. In this work we looked at variability levels and patterns at *P. spinosa* *SLFLI* gene, that is physically close to the *S-RNase* gene. Our results suggest that the recombination level sharply increases near the *SLFLI* coding region. These findings are discussed in the context of theoretical models predicting an effect of linked weakly deleterious mutations on *S*-locus specificities relatedness. Moreover, we show that *SLFLI* belongs to a gene family of at least five functional genes and that *SLFLI* pseudogenes are frequently found in the *S*-locus region.

P11 The effect of multiple origins on ecological success in allotetraploid wild wheats of the genus *Aegilops* (Poaceae)

Harald Meimberg, CIBIO

Polyploid species can exhibit higher ecological tolerance than their progenitor species. This is often primarily attributed to the existence of heterosis resulting from entire genome duplication. Multiple origins of polyploid species may further promote their ecological success by providing genetic variability in ecological traits underlying local adaptation and range expansion. Here we show in a group of allopolyploid species in the genus *Aegilops* that range size and abundance are correlated with the number of inferred origins. We found that allopolyploid *Aegilops* spp. contain multiple chloroplast haplotypes identical to haplotypes of the diploid progenitor species, indicating multiple origins as the major source of variation. The number of inferred origins was correlated to the total area occupied by the allopolyploid and the tendency for the species to be common. These results strongly support the hypothesis that the introduction of genetic variability by multiple origins can increase the ecological amplitude and evolutionary success of allopolyploid species.

P12 PHYSIOLOGICAL STRESS INDICATORS IN SAND DUNE PLANTS AND ITS RELATIONSHIP TO COASTAL DYNAMICS

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Sandy coastal areas are highly dynamical systems, with strong sedimentary movements determined by the proximity to the ocean. In the Portuguese coastline, as in many other areas all over the world, coastal dynamics is nowadays mostly regressive, with sea level rise and fast inwards migration of the shoreline inducing dramatic changes in ecological conditions. Classical indicators of coastal dynamics include from geological features, mostly related to geomorphology and the sedimentary budget, to vegetation attributes like species richness and floristical structuring of discrete communities. However, in most cases geomorphology and vegetation tend to exhibit late and cumulative responses to changes in sea level and drift direction.

Plants growing under extreme ecological conditions can exhibit molecular and cellular indicators of physiological stress. When plants are subjected to environmental stress, such as salinity and drought, that disturbs the water balance and ion homeostasis of the cell, they have different strategies to avoid the adverse effects of stress injury. A common adaptive response to this stress condition is the synthesis and accumulation of compatible solutes such as proline and/or specific proteins that help cells to maintain their water balance and also to protect macromolecules in stressed cells. It was also stated that several kinds of stress, induce lipid peroxidation, which leads to disruption of membranes and deleterious effects on plant cells. In order to identify early indicators of regressive coastal dynamics, the levels of proline and malondialdehyde (an indicator of lipid peroxidation) were assessed in sea-couch (*Elytrigia boreo-atlantica*), a pioneer grass species that colonizes the higher beach and the embryonic dune and is therefore closely affected by changes in coastal dynamics.

Plants harvested from different locations which were known (from geomorphology and vegetation) to be facing strong regressive dynamics tended to accumulate higher levels of proline than plants from sites to which more stable conditions could be assigned, while lipid peroxidation exhibited an opposite pattern. These results suggest that the accumulation of proline has an adaptive significance, acting as an antioxidant that protects cells from free radical damage and reduces the lipid peroxidation-linked membrane deterioration under stress. Preliminary results allowed the identification of significant correlations between stress indicators and quantitative attributes of the vegetation-geoform system, which seems to indicate that plants facing different dynamical conditions exhibit distinct levels of both proline accumulation and lipid membrane peroxidation. Further studies are expected to provide additional insights on the relationship between stress indicators and classical ecological indicators and on the possibility of identifying early signs of regressive coastal dynamics.

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X Congresso Hispano – Luso
de Fisiologia Vegetal
September 18th – 21st, Alcalá de Henares, Madrid

P13 Tonoplast proton pumps and Na⁺/H⁺ exchange activity in potato cell lines and implications for salt tolerance

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Salinity is an important environmental stress reducing plant growth and productivity. Ion toxicity, osmotic stress and ion imbalance are the major constraints caused by salinity. Tonoplast enriched-vesicles isolated by a discontinuous sucrose gradient from control and 150 mM NaCl-tolerant calli lines were used as a model system to study the activity of V-H⁺ATPase and V-H⁺PPase and the involvement of Na⁺ compartmentation into the vacuole as a mechanism of salt tolerance in *Solanum tuberosum*. Both ATP- and PPi-dependent H⁺-transport, measured as the initial rates of ACMA fluorescence quenching, were higher in tonoplast vesicles from salt-tolerant line than in vesicles from control cells. Na⁺-induced dissipation of a pre-established PPi-dependent pH gradient was used as an experimental evidence for the involvement of a tonoplast Na⁺/H⁺ exchange system. The initial rates of Na⁺-dependent fluorescent recovery followed Michaelis-Menten kinetics and the V_{max} of proton dissipation was 2-fold higher in vesicles from salt-tolerant calli compared with the control cells. Both Na⁺ and Li⁺, but not K⁺, dissipated the ΔpH. The correlation between the increase of both the H⁺ pumping through V-H⁺ATPase and V-H⁺PPase and the activity of Na⁺/H⁺ exchange system in NaCl-tolerant cell line suggests that the accumulation of Na⁺ into vacuole represents an adaptative response to high salinity in *S. tuberosum*.



**XVI Congress of the
Federation of European
Societies of Plant Biology**

August 19th – 22st, 2008

P14 Genotypic Assessment by RAPD Markers and Ultrastructural Characteristics of a Potato Cell Line Tolerant to NaCl

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Soil salinity is an important environmental constraint to crop distribution and productivity. Potato (*Solanum tuberosum* L.), one of the most important food crops, is moderately salt sensitive and its improvement by traditional breeding programs has been slow. Tissue cultures may be relevant to plant improvement through selection of salt-tolerant cell lines and subsequent regeneration of plants. Besides those cell lines are a useful tool to study the mechanisms of salinity tolerance. In this work, we used the random amplified polymorphic DNA (RAPD) markers to investigate the occurrence of genetic polymorphism in a potato calli line grown with 150 mM NaCl. After DNA extraction from control and salt-tolerant lines, PCR was performed with 40 arbitrary primers, and eight revealed polymorphism in amplification products. Sixteen well resolved and reproducible bands were chosen as RAPD markers which showed that the salt-tolerant line differed genotypically from the control. Although this callus tissue displays a macroscopic aspect similar to the control, the ultrastructural characteristics were compared with the aim to unravel those one that may have adaptive value. Ultrastructural observations revealed that the integrity of cells grown with salt was not affected, however, they showed plastids less differentiated with a lower number of grana than in control cells. Round-shaped plastids with a less compact stroma displaying a higher number of large starch grains were also common features observed in NaCl-tolerant cells. In conclusion, RAPD analysis revealed that NaCl-adapted line is a somaclonal variant and the ultrastructural study showed changes essentially at the plastids. This variant line may be useful material for potato breeding programmes.

Key words: Salinity, potato, polymorphism, ultrastructure



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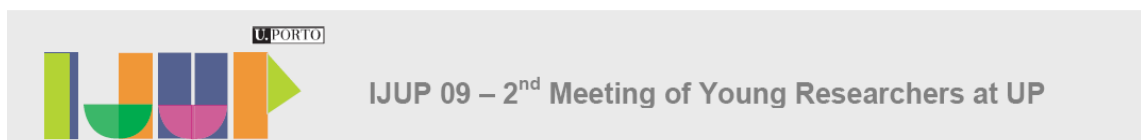
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P15 OPTIMISATION OF *IN VITRO* CULTURE CONDITIONS FOR OBTAINING CALLUS TISSUE AND DIRECT ORGANOGENESIS FROM *SOLANUM NIGRUM* L.

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Over 400 hyperaccumulator species have been reported to date, but generally these species bring up the problem of low biomass yield and growth rate. *Solanum nigrum* L. (black nightshade) is a pioneer species growing in polluted sites worldwide that has a fast growth rate and larger shoot biomass and has been recently found to be suitable to remediate As-, Cd- and Zn-polluted soils. Recently it was demonstrated that this plant species accumulates the organochlorine pesticide hexachlorocyclohexane (HCH) and polychlorinated biphenyls (PCBs). These studies point to a potential and multifaceted capability for *S. nigrum* to remediate both organic and inorganic pollutants from the environment, but further research is needed so that this species may be used as a phytoremediation tool itself, or as a source of candidate genes for producing better and improved transgenic plants suitable for cleaning the environment. Unconventional crop improvement methods, such as tissue culture techniques, can be used as a tool for obtaining different pollutant hyperaccumulator *S. nigrum* types. Furthermore, the complicated structure of the whole plant makes it difficult to separate systemic from cellular pollutant tolerance mechanisms and therefore tissue culture techniques may contribute to discriminate between cellular and whole plant response mechanisms to stresses. In order to be able to perform studies at the cellular level and/or to genetically manipulate this plant species, optimized protocols are necessary to be developed so that callus tissue can be obtained and *in vitro* regeneration of transformed plants can be performed. The main objectives of this work were, therefore, to establish *S. nigrum* callus tissue cultures and to develop an *in vitro* plant regeneration procedure. To achieve these objectives, two Murashige-Skoog derived culture mediums were used, M1 and M2, which differed only in their hormonal composition: M1 contained 1 mg/L NAA and 2 mg/L BA, while M2 consisted on 2 mg/L 2,4-D and 0.5 mg/L BA. The media were inoculated with cotyledons derived from sterile plantlets or leaf explants, previously sterilized with 70% ethanol for 5' followed by 11' in sodium hypochloride 20% plus Tween 20. Explants inoculated in M1 medium suffered direct organogenesis, originating full plantlets, while M2 medium originated callus tissue, independently of the type of explants used. Whatever the type of medium was used, cotyledons always responded more rapidly than leaf explants. The obtained callus tissue is characterized by a slow growth, which is possibly due to an increased oxidative metabolism, as evidenced by the darkening of the culture medium and the browning of the callus tissue. Further alterations to the medium composition will be attempted, such as the inclusion of ascorbate or citrate as antioxidant agents. The organogenesis protocol was established and can be used for future plant regeneration procedures, such as recovery of genetically modified tissues. When the callus tissue growth conditions are settled it can be used for future studies directed at cellular responses to pollutant exposure.



P16 Over expression of glutamine synthetase in root nodules of *Medicago truncatula* leads to enhanced nitrogen utilization efficiency

Ana Rita Seabra, Julie Cullimore and Helena Carvalho (IBMC)

Glutamine synthetase (GS) is a vital enzyme for the assimilation of ammonia into amino acids in higher plants and the enhancement of its activity has the potential to increase nitrogen utilization efficiency. In legume root nodules, GS is responsible for the assimilation of the ammonium released by symbiotic nitrogen fixation. To investigate how nodule GS activity affects plant performance, we have previously overexpressed GS1a cDNA specifically in root nodules of *Medicago truncatula* under the direction of a native leghemoglobin promoter.

In this study we have used these plants to examine the effects of increased nodule GS activity on phenotypic development, biomass production, rhizobial nitrogen fixation and nitrogen utilization efficiency and have used the tools of transcriptomics and metabolomics to identify the major transcript and metabolite changes associated with the altered nodule metabolism. Overall, the results indicate that *M. truncatula* overexpressing GS display enhanced growth phenotype as quantified by increases in biomass and seed production. Nodule GS activity was positively correlated with symbiotic nitrogen fixation activity and plant nitrogen utilization efficiency. These studies provide further support to the notion that it may be possible to increase nitrogen use efficiency by the manipulation of specific GS isoenzymes in transgenic crop plants.

This work is supported by projects POCTI/AGG/39079/2001 and FOOD-CT-2004-506223.

P17 Incidence of allergenic airborne tree pollen in the region of Porto (Portugal).

Abreu, I., Ribeiro, N., Ribeiro, H., Oliveira, M., & Cruz, A. 2008. XXVII EAACI Congress of the European Academy of Allergology and Clinical Immunology. Barcelona, Spain.
FCUP/ENVISED-CGUP

The aim of this work was to analyze the aerobiological behaviour of *Platanus* spp., *Acer* spp., *Salix* spp., *Betula* spp., *Populus* spp. and *Alnus* spp. pollen in the city of Porto (Portugal), to characterize its allergen protein profile and to identify the different reactivity levels in immunosensitive patients. Airborne pollen sampling was performed using a Hirst-type volumetric spore trap, from 2003 to 2007. The allergenic properties of these tree pollens were investigated using biochemical analyses (SDS-PAGE) and immunological techniques with serum IgE from immunosensitive patients.

The pollen from the selected species was present in the atmosphere from mid winter to early spring. All pollen extracts presented several bands with different molecular weights.

In immunoblotting analyzes, using allergenic patients sera, pollen extracts from less studied species (*Acer*, *Salix* and *Populus*) revealed IgE binding with three main groups of proteins comparable to allergens described in other species.

This information can be used in the planning of outdoor activities for allergic patients, in order to prevent allergen exposure, and in architectural landscape planning, to avoid the use of allergenic species.

P18 Cross-talk between oxidative stress and iron availability – the regulation of catalatic activity in *Frankia* spp.

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Background

Oxidative stress and iron availability have been implicated in numerous prokaryotic infective processes, including the symbiotic microbe-plant interactions. In Actinobacteria, the presence of a highly-conserved genomic pair composed of a ferric uptake regulator (*fur*) upstream of an hydroperoxidase-encoding gene further supports the intimate relation between iron and ROS¹. Moreover, in some actinobacterial species, Fur was identified as a redox-sensitive factor, able to repress the hydroperoxidase expression in the presence of Fe²⁺.

Objectives

This work addresses the multifaceted relationship between iron and ROS in *Frankia* strains ACN14a and CcI3, which differ in their oxidative-related genomic machinery and host ranges.

Methods

Frankia alni ACN14a and CcI3 were cultured in the presence of different iron concentrations and challenged with paraquat and hydrogen peroxide, two ROS sources. Growth and catalatic activity were monitored through time and catalases were visualized by non-denaturing PAGE. Electrophoretic shift mobility assays are underway to unveil whether Fur is binding to a putative promoter sequence located upstream of *fur-katA* in ACN14a.

Results & Conclusions

Our results suggest that iron has an important role in the regulation of the frankiae catalatic activity. In fact, in *Frankia* ACN14a the presence of different concentrations of iron *per se* does not seem to influence the basal levels of catalatic activity, opposite to what happens in the CcI3 strain. However, upon challenging the cells grown in different iron concentrations with an exogenous source of oxidative stress, CcI3 and ACN14a response patterns were strongly contrasted. These differences in the catalatic activity reflect the disparate genomic organization of these strains: while ACN14a has the *fur-katA* gene pair conserved, CcI3 has a divergently-transcribed *oxyR* upstream of a *katG*, a Gram-negative like pattern.

References:

¹ Santos, CL et al., *BMC Evolutionary Biology* **8** (1), 185 (2008)

P19 Selection and validation of *Xanthomonas fragariae* and *Xanthomonas axonopodis* pv. *phaseoli* molecular markers for detection and genotyping

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Background:

Members of the genus *Xanthomonas* are responsible for significant agricultural losses and strict quarantine measures are required to manage the infected areas and prevent dissemination. DNA-based methods are promising alternatives to the certified culture-based detection methods for rapid and reliable phytopathogen detection and typing.

Objectives:

- 1- Select and validate *Xanthomonas*-specific molecular markers, with emphasis for the unsequenced *X. fragariae* (*Xf*) and *X. axonopodis* pv. *phaseoli* (*Xaph*).
- 2- Assess the genotyping potential of the detection-validated markers.

Methods:

Xanthomonas-specific molecular markers were selected as described by Vieira *et al.* (2007).

Validation was carried out by PCR and dot blot hybridization techniques. PCR amplicons were sequenced to infer their typing resolution by multiple alignment and bootstrapping analysis.

Results:

Twenty *Xanthomonas*-specific molecular markers were selected *in silico* and PCR validation showed that nine markers were positive for *Xf* and *Xaph*. These markers were labelled and used as probes in dot blots spotted with DNA from a wide range of phytopathogenic bacteria. Obtained results further confirmed the markers specificity for xanthomonads.

Sequence analysis of the markers amplified from different *Xanthomonas* strains, including different strains of *Xf* and *Xaph* allowed clustering together members of the same species.

Conclusions:

Our approach allowed the selection of nine novel molecular markers for two unsequenced *Xanthomonas* species. The *Xf* and *Xaph* selected molecular markers were shown to be efficient for detection and genotyping.

References:

Vieira J, Mendes M V, Albuquerque P, Moradas-Ferreira P, Tavares F, A novel approach for the identification of bacterial taxa-specific molecular markers (2007), Lett. Appl. Microbiol. 44(5): 506-512.

P20 RELATIONSHIP BETWEEN BIOLOGICAL AND NON-BIOLOGICAL ATMOSPHERIC POLLUTANTS

Abreu, I., H. Ribeiro, M. Oliveira, S I V Sousa, F G Martins, M C Pereira & M C M Alvim-Ferraz 2006. 8th Internacional Congress on Aerobiology. Neuchatel, Suíça. FCUP/ENVISED-CGUP

The aim of this study was to investigate the relationship between allergenic bioaerosols (fungal spore and pollen) and non-biological pollutants (O₃ and PM₁₀) in the Porto city during 2004.

Daily airborne pollen and fungal spores were continuously sampled, using a 7-day Burkard volumetric trap.

Hourly mean concentrations of O₃ and PM₁₀ were monitored using UV-absorption photometry and beta radiation attenuation method, respectively. In this study, linear correlations between the allergenic bioaerosols and those non-biological pollutants were performed.

O₃ presented a positive correlation with pollen grains (0.192) and a negative correlation with fungal spores (-0.204) indicating that for higher O₃ concentrations the fungi decreased. This can be explained by the strong oxidant activity of O₃, probably interacting with the discharge mechanism of spores.

Concerning PM₁₀, a positive correlation with pollens (0.225) was observed.

P21 Análisis preliminar del contenido alérgico del polen de variedades de olivo de Portugal.

S. Morales, N. Ribeiro, L. Calado, M.I. Rodríguez-García, J.D. Alché & I. Abreu. XVI Simposio Internacional de Palinología de la A.P.L.E. Mallorca (Spain). September 2008. FCUP/ENVISED-CGUP

El presente estudio constituye un análisis preliminar del contenido del polen de diversos cultivares ampliamente distribuidos en Portugal en los siguientes alérgenos:

Ole e 1: destaca como el alérgeno mayoritario en el polen de olivo y juega un papel en la hidratación y germinación del polen de olivo.

Ole e 2: es una profilina con múltiples ligandos. Está implicada en funciones como el control de la organización del citoesqueleto de actina y la transducción de señales.

Ole e 5: es una proteína con actividad Cu/Zn superóxido dismutasa.

Ole e 9: es una proteína con actividad 1,3 B-glucanasa.

El análisis de isoformas de Ole e 1, Ole e 2, Ole e 5 y Ole e 9, así como de las proteínas reconocidas por dos sueros de pacientes alérgicos a polen de olivo (un paciente de origen portugués, y otro de origen español), se realizó mediante SDS-PAGE e inmunoblotting de 12 extractos proteicos crudos de polen de olivo de cultivares de origen portugués, o ampliamente distribuidas en este país (Maçanilha Tavira (1), Negrinha (2), Verdeal transmontana (3), Galega (4), Verdial de Serpa (5), Cobrançosa (6), Maçanilha almendral (7), Redondil (8), Ascolana (9), Carrasqueña (10), Conserva de Elvas (11) y Blanqueta (12). En los inmunoblots se ensayaron anticuerpos primarios y anticuerpos secundarios, éstos últimos conjugados con fluorocromos, con fosfatasa alcalina, o con peroxidasa.

La distribución de isoformas de alérgenos en cultivares portugueses muestra una amplia heterogeneidad, tanto en su número como en su nivel de expresión.

La heterogeneidad es especialmente manifiesta en el caso de los alérgenos Ole e 1 y Ole e 9 (los alérgenos con mayor relevancia clínica en el caso de los estudios realizados en España).

A pesar del número limitado de sueros de pacientes utilizados, se observan claras diferencias en la reactividad de éstos a los extractos de pólenes de distintos cultivares.

Las diferencias observadas pueden ser reflejo de características fisiológicas diferenciales del polen de los distintos cultivares. Por otra parte, pueden influir notablemente en la composición y la potencia alérgica de los extractos proteicos utilizados en la diagnosis y terapia de la alergia al polen de olivo.

P22 Characterization of the main fungal types present in the atmosphere of Porto (Portugal).

Oliveira M., Ferreira E., Amorim I., Abreu I. (2008). XVI Simposio de la Asociacion de Palinologos de Lengua Española. Palma de Maiorca, Espanha.
FCUP/ENVISED-CGUP

The aim of this work was to characterize the main fungal spore types present in the atmosphere of Porto. The aeromycological study was performed monthly, during 2007, using an Andersen one-stage sampler. After 5 days, colony forming units (CFU) were identified and counted.

Samples were collected for DNA extraction, according to manufactures instructions. 18S rRNA gene was amplified from each CFU using universal specific fungal primers pairs: FF1(5'-GTTAAAAGCTCGTAGTTGAAC-3') and FR1(5'-CTCTCAATCTGT CAATCCTTATT-3'); NS1 (5'-GTAGTCATATGCTTGTCTC-3') and NS4 (5'-CTCCGTCAATTCCTTTAAG-3')/or NS6 (5-'GCATCACA GACCTGTTATTGCCTC-3'). The purified PCR products were used in sequence reactions with the same set of primers. The obtained sequences were compared with fungal sequences in GenBank.

The Andersen sampling method can act as a complementary tool to allow the distinction between *Aspergillus* and *Penicillium* spores that are underrepresented in Hirst-type sampling. Also, it is possible to culture the viable spores enabling the use of several molecular techniques to identify the fungal species present in the atmosphere.

Using three different sets of primers, the sequences of the PCR products were identified as *C. cladosporioides*, *Aspergillus* spp., *A. ochraceus*, *Penicillium* spp. and *A. alternata*.

P23 Relationship between threatened species richness, habitat diversity and landscape structure: a case-study from a National Park in Northern Portugal

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The lack of information on biodiversity patterns and on the ecological processes underlying those patterns is often an obstacle in management plans for natural and semi-natural landscapes. This is particularly critical in protected areas, which are repeatedly used as models for the development of methodologies regarding the protection and management of biological diversity, since the required ecological data is most often available for these areas and the moderate human impact allows for different levels of disturbance to co-exist.

This work addresses the influence of landscape structure and distribution patterns of natural habitats on the local richness of threatened bryophyte and vascular flora. Both GIS-based spatial analyses and statistical techniques are used, since they have been proved to be powerful tools on the establishment of relationships between the variation of ecological factors and biodiversity patterns. A synthetic model relating species, habitat and landscape patterns is proposed using data from Peneda-Gerês National Park (Northern Portugal). The significance of this model for management purposes is discussed.

Keywords: Threatened species; Diversity; Landscape

P24 Early Indicators of Coastal Dynamics in Climate Change Vulnerability Assessment

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Global warming and other climate change related processes are expected to impact significantly on coastal dynamics, which may induce important ecological and economical consequences. Therefore early indicators of dynamic shifts are needed for vulnerability assessments, for monitoring protocols and for restoration processes.

In recent years, we have developed intensive studies on the relationship between coastal dynamics, dune morphology, and vegetation assembly. From general patterns of ecosystem structure to specific features of morphology and vegetation, these studies have provided strong evidence on the potential of this relationship for evaluation and monitoring purposes. Recently, we have been assessing the fine-scale assembly of dune vegetation under different coastal dynamics as recognised from previous studies and from quantitative features of local geomorphology. These detailed assessments have allowed the identification of a set of promising indicators that can be evaluated through cost-effective methodologies and are may thus prove to be suitable to support monitoring protocols. These indicators range from foredune morphology and spatial patterns of plant species to numerical indicators of diversity for entire vegetation profiles.

Even though the effectiveness of these indicators still has to be further validated, we believe they can provide the baseline for consistent assessments of local dynamics, for the development of predictive models to forecast climate change impacts, for detailed assessments of vulnerability and resilience of dune ecosystems to shifts in dynamics (e.g. driven by climate changes), and for restoration protocols directed towards the development of more resistant dune systems.

P25 Modeling Spatial Patterns and Dynamics of Alien Plant Invaders in Agro-Forestry Landscapes

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Biological invasions constitute the second threat to biodiversity around the world after habitat destruction. With plant species of Europe invading other continents and vice-versa, a process of “globalization of vegetation” is going on. Therefore, anticipating future invasions is a major task in conservation biology, and species distribution models (SDM) are useful tools in this regard. Here, we use a hierarchic modelling framework to: (i) understand the ecological constraints of alien plants and how they respond to environmental changes in agro-forestry landscapes, and (ii) to predict the current and future distribution for four alien tree and shrub species in the extreme Northwest area of Portugal. Ultimately, we intend to assess possible synergetic effects of climate and land use change on alien plant occurrence and dynamics. We use *Acacia longifolia*, *Acacia dealbata*, *Acacia melanoxylon* and *Hakea sericea* as model species. These species invade several natural habitats, but are more common in forest stands and agro-forestry landscapes characterized by intermediate levels of agricultural intensity. This study provides the theoretical framework for modelling invasion patterns in Mediterranean system and for supporting eradication strategies, by predicting the future occurrence and spread of these problematic alien species, under present and future climate and land-use change scenarios.

This study was financially supported by FCT (Portuguese Science Foundation), through PhD grant SFRH/BD/40668/2007 to J. Vicente.

P26 Fluorescence Activation Cell Sorting of *Catharanthus roseus* leaf protoplasts for isolation of cells accumulating medicinal indole alkaloids

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Catharanthus roseus accumulates in low levels the terpenoid indole alkaloids (TIAs) vinblastine and vincristine, used in cancer therapy, ajmalicine, used as an antihypertensive, and serpentine, used as sedative. Although much is known about the biosynthesis and regulation of TIAs, a number of biosynthetic steps remains unknown, and TIAs transport and accumulation inside the cells is largely uncharacterized, even if this information is crucial for any manipulation strategy to increase TIA levels.

We aim to characterize the transport and accumulation of TIAs in *C. roseus* cells and vacuoles through several strategies, including the differential characterization of alkaloid accumulating cells (idioblasts) in comparison with common mesophyll cells. This strategy involves the isolation of protoplasts from leaves and the separation of idioblast protoplasts from those of common mesophyll cells by Fluorescence Activation Cell Sorting (FACS), followed by differential proteomic, transcriptomic and metabolomic analysis.

Here, we show the results of protoplast isolation from *C. roseus* leaves and the optimization of FACS conditions leading to the isolation of two populations of mesophyll cells presenting different alkaloid accumulation (UV fluorescence). This achievement represents a crucial step for the success of our strategy.

P27 Sorting of class III peroxidases to the plant vacuole

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Vacuoles occupy most of the volume of plant cells and play key roles in plant physiology: they maintain the turgor pressure needed for support of the plant body and cell growth, perform the lytic functions of animal cell lysosomes, sequester and inactivate toxic compounds, participate in programmed cell death, accumulate defense proteins and thousands of secondary metabolites, participate in pH regulation and ion homeostasis, and store proteins to be used later as a source of amino acids or energy. Most of these functions depend on the correct vacuolar sorting of a variety of proteins which ultimately determine the characteristics and properties of the vacuole. Moreover, knowledge on the determination of vacuolar sorting is essential for the improvement of the protein content of seeds used as food, and for the development of highly productive plants and cell cultures as green factories for the production of medicinal phytochemicals, vaccines, antibodies, etc.

In our laboratory, we have worked for a long time with vacuolar class III peroxidases (Prxs) and their involvement in the biosynthesis of medicinal indole alkaloids in the plant *Catharanthus roseus*. Prxs form a large multigene family typical of plants that catalyzes the oxidation of small molecules or chemical groups at the expense of H₂O₂, and they may be localized in the cell wall or the vacuole. The determination of the vacuolar sorting of Prxs remains uncharacterized.

Here, we show that the C-terminal amino acid sequence of the main leaf vacuolar Prx from *C. roseus*, CrPrx1, is necessary and sufficient for the vacuolar sorting of GFP-CrPrx1 fusions, indicating that it constitutes a vacuolar sorting signal. The presence of a C-terminal vacuolar sorting signal directing the protein to a lytic vacuole, as is the case for CrPrx1, is unusual in plant cells, and indicates that the characterization of the Prx sorting pathway should add important knowledge concerning the general model of vacuolar sorting in plant cells.

P28 The Catharanthus alkaloids and class III peroxidases – the vacuolar link

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Catharanthus roseus (L.) G. Don is a medicinal plant producing several terpenoid indole alkaloids with important pharmacological activity, including the anticancer vinblastine and vincristine, the antihypertensive ajmalicine and the sedative serpentine. We have previously characterized the major class III peroxidase of *C. roseus* leaves, CrPrx1, and we have obtained evidence indicating that CrPrx1 is responsible for a key biosynthetic step in the biosynthesis of vinblastine and vincristine. Now, we have characterized a second class III peroxidase of *C. roseus*, CrPrx2, which is highly homologous to CrPrx1, the two genes presenting a mutually exclusive expression pattern regarding leaves and roots. CrPrx2 was partially purified and is able to oxidate ajmalicine, possibly contributing to the biosynthesis of serpentine.

We propose that the two peroxidases we have characterized in *C. roseus* are involved in the biosynthesis and metabolism of the important pharmacological terpenoid indole alkaloids produced by this plant. Of foremost importance in our reasoning, is the fact that the two enzymes have been localized in the vacuole, the exact compartment where the alkaloids are accumulated. The question remains, however, how enzymes with such a broad range of substrate specificity, localized in an organelle containing so many potential substrates as the vacuole, may be responsible for a defined specific reaction. Is it just mess, chance, or does it represent useful metabolic plasticity? Even more, may the peroxidase substrate be determined by metabolic channeling?

P29 Glutamine synthetase from *Medicago truncatula* is regulated by tyrosine nitration

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Glutamine Synthetase (GS), is a crucial enzyme in nitrogen metabolism as it catalyses the first step at which nitrogen is brought into cellular metabolism and thus the enzyme must be precisely regulated. Many studies have been devoted to understand how GS is regulated in plants, which have shown that the enzyme is subjected to tight controls operating at many different levels. Although the regulation of GS at the transcriptional level has been well studied, there is clearly a lack of information concerning its posttranslational regulation. Nitric oxide (NO) and its related species can induce important posttranslational protein modifications through S-Nitrosylation and nitration. In this study we have evaluated the effect of NO on GS activity. *In vitro* incubation of the enzyme with reactive nitrogen species producers (peroxynitrite or tetranitromethane) induced a dose-dependent loss of GS activity that could be related to an increase in nitrotyrosine immunoreactivity. Incubation of the enzyme with epicatechin, a selective nitration inhibitor, prevented both GS inactivation induced by reactive nitrogen species and tyrosine immunoreactivity. These results strongly suggest that the inhibition of GS activity is due to tyrosine nitration. Mass spectrometry analysis of *in vitro* nitrated purified GS1a, identified TYR 341 as a nitrated residue. The data point to a potential post-translational regulation of GS from *M. truncatula* by tyrosine nitration. Further experiments are underway to investigate whether GS nitration is reversible and the physiological significance of this posttranslational modification for plant nitrogen metabolism.

P 30 STUDY OF *PIRELLULA* SP. OJF20 LIFE CYCLE

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Planctomycetes, a fascinating main lineage of *Bacteria*, possess unique properties like lack of peptidoglycan, budding reproduction and membrane-bounded compartmentalization. Their ecological relevance has been well evidenced although their life and cell cycles have been poorly characterized. We aim to give some insights on these aspects by combining transmission electron microscopy (TEM), fluorescence microscopy (FM) and flow cytometry (FCM). *Pirellula* sp. OJF20 (EF589346), a member of a new independent cluster close to the *Rhodopirellula* genus (94% similarity in the 16S rDNA), was isolated from *Corallina* sp. surface sampled in a rocky beach in Porto. For TEM, cells were fixed in 2.5% glutaraldehyde in marine buffer, 1% osmium tetroxide and 1% uranyl acetate and included in epoxy resin. For FM and FCM, cells were labelled with FM 1-43 and propidium iodide, Dapi or Sybr green. As seen by light and electron microscopy, *Pirellula* sp. OJF20 has a life cycle similar to that of *Rhodopirellula baltica* reproducing by budding. Ultrastructural evidence seems to indicate that it can develop a spore-forming phase. Two completely mature, morphologically normal, new young cells appeared inside a much bigger mother cell. This seemed empty of cytoplasmic content but surrounded by a conventional cell envelope evidencing the presence of apical fimbria. *In silico* analysis of *R. baltica* genome indicates the presence of proteins related to sporulation. Further evidence is needed to support the co-existence of budding and spore formation as propagation means. The Spore-forming capacity would provide ecological advantages to an organism inhabiting a very polluted and stressful environment.

P31 *Planctomycetes* as potential oil producers

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Planctomycetes are still a very much unknown phylum of *Bacteria*, due to their quite recent awareness. Their biotechnological potential in many fields has not yet been explored even though their characteristics as a group are very particular and unique. During isolation experiments from the sediment of a freshwater aquarium we have obtained two colonies of planctomycetes (strains OJF2 and OJF8) that are in the process of description as a new genus. *Aquisphaera giovannonii* gen. nov., sp. nov. will be proposed. Routinely, they are cultured in PYGV medium (modified after Staley, 1968) having as carbon and nitrogen sources 0,025% of yeast extract, bacto peptone and glucose. As planctomycetes are known as non-tolerant high organic levels, growth of strains has been assayed in a medium with 4-fold the 3 compounds concentration. Both strains did not show any growth under these conditions. Thin sections of transmission electron microscopy and cell staining with Nile red and subsequent observation under fluorescence microscopy of strain OJF2 revealed the presence of prominent lipid droplets. Lipid profile is under characterisation. Comparative analysis of cells from both media will be analysed by flow cytometry. This has been the first evidence of planctomycetes as potential oil producers.

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P 32 Discovering Planctomycetes Biodiversity

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The Planctomycetes are a fascinating group of budding Bacteria characterised by membrane-bounded compartments within the cell and peptidoglycan-less cell walls [1]. They play an important role in the ecosystems due to their physiological diversity and ubiquity in many habitats as revealed, mainly, by molecular microbial ecological techniques like culture-independent 16S rRNA-based methods [2,3]. Despite the reported widespread distribution of Planctomycetes, precise knowledge of their ecological role in the environment is still very much unknown, mostly because of the relatively few species present in pure culture. Our main goals have been the isolation in pure culture of more strains, the characterisation of the existing and the new ones, their identification by the sequencing of the 16S rRNA gene and their infraspecies study by other genetic fingerprinting methods like the BOX-PCR.

Using the methodology previously described [4], more isolation experiments have been done and Planctomycetes living in association with the macroalgae *Porphyra dioica* collected in Mindelo and *Enteromorpha intestinalis* and *Laminaria* sp. collected in Foz, Porto have been obtained. Several aspects of the characterisation of some strains from the OJF *Planctomycetes* culture collection have been achieved. These include the study of growth rate, growth in different media, salinity and pH tolerance, Gram staining, nutritional requirements like carbon source, API and BIOLOG tests for strains *Isosphaera* sp. OJF2, *Isosphaera* sp. OJF8, *Rhodopirellula baltica* strain OJF23, *Pirellula* sp. OJF3, OJF7, OJF20 (Cor3), OJF22 (GrW3), OJF24 (FC25), OJF25 (UC16), OJF26 (UC17), OJF27 (CcC6), Cc2, CcC1.2, CcC8, Cor4, Ent1, FC17, FC18, MsF5, Pd1 and SM2. Ultrastructural studies (TEM) have been more focused on strains *Isosphaera* sp. OJF2, *Isosphaera* sp. OJF8, *Rhodopirellula baltica* strain OJF23, *Pirellula* sp. OJF7 and OJF20. The 16S rRNA gene oligonucleotide sequence of all the isolates has been obtained after DNA extraction and amplification of the gene by Polymerase Chain Reaction (PCR). These isolates are distributed mainly in 9 clusters, being 7 of them new independent ones from the already existing genera. The studies of two families of repetitive sequences were assayed as a method to generate genetic fingerprinting of *Planctomycetes*: the 124-127 bp enterobacterial repetitive intergenic consensus (ERIC) sequence and the 154 bp BOX element. This study has been done with about 50 strains. This cluster analysis allowed discriminating between strains, specially the many ones that we isolate and that are closely related to *R. baltica*.

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P33 Mapping cardosin A pathways: a powerful tool to question vacuolar

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Cardosin A is a vacuolar Aspartic Proteinase that undergoes several processing steps to acquire proteolytic activity. The precursor form has an ER signal peptide and probably enters the secretory pathway. The route taken by cardosin A to the vacuoles is essentially unknown. Does it go through the Golgi for further processing or does it go directly from ER to vacuoles is still an opened question. Cardosin A has two "plant complex type" glycosylation sites, suggesting a passage through the Golgi but our own studies suggest that a partial glycosylation may be indeed related to the direct transport from ER to the vacuole. The main objective of this work was to draw cardosin A vacuolar pathway. Two experimental systems were tested: *Arabidopsis thaliana* line stably expressing cardosin A and BY-2 cells expressing a cardosin-kaede fusion. Immunolocalisation of cardosin A and several endomembrane markers in *A. thaliana* germinating seeds suggests an ER – Golgi - PVC pathway. In BY-2 cells, cardosin-labelled structures observed corroborates the existence of an ER-Golgi pathway for cardosin A.

P34 DISSECTING CARDOSIN B TRAFFICKING PATHWAY

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Determining the sorting signals for protein targeting to the different intra- or extracellular locations has been widely investigated either for animal or plant proteins. Cardosin A and cardosin B are aspartic proteinases native to cardoon (*Cynara cardunculus*) that share high similarity (73%) at the protein level. However they show different subcellular localisations – cardosin A is vacuolar while cardosin B is a secreted protein. Studies have been undertaken in order to dissect their trafficking pathways and understand the signals responsible for their different sorting. In heterologous expression systems – either inducible expression in *Arabidopsis* or transient expression in tobacco – cardosin B has been found to localise in the vacuole and, under some circumstances, cardosin B is secreted. Fusions with fluorescent reporter protein mCherry allowed cardosin B visualisation in the vacuoles of tobacco epidermal cells. Cardosin B trafficking pathway was dissected by co-expressing cardosin B in tobacco with three different *Arabidopsis* Rab dominant negative mutants – RAB-D2a[NI], RAB-F2b[SN] and RAB-H1b[SN] –that act in particular intracellular transport steps. The results suggest that cardosin B goes through the Golgi to reach the vacuole and is secreted if the traffic to the vacuole is impaired. More studies will add new insights about cardosin B trafficking pathway in tobacco leaf epidermal cells.

P35 What Drives a Protein to the Plant Vacuole?

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The physical and biochemical compartmentalization of eukaryotic cells is achieved, in part, by the sorting of proteins to the correct subcellular location. Evidences indicate that small polypeptide domains are responsible for targeting proteins. Several pathways have been described either for animal, yeast and plant cells. Among these diverse pathways, sorting to the plant vacuoles is still a matter of debate: it seems to depend on a series of connected signals and receptors. Cardosin A, a vacuolar plant aspartic proteinase, is considered to be a good reporter to study trafficking pathways to the vacuole, giving the fact that cardosin B, despite sharing high similarity to cardosin A, is not targeted to the vacuole but secreted. It is known that there are a variety of signals directing proteins to the vacuole, depending on the type of vacuole, the type of organ and on the developmental stage. In the particular case of aspartic proteinases, is commonly accepted that the targeting signal is the PSI (Plant Specific Region) region, and many studies have been carried out as an attempt to verify this theory. In this work we made several cardosin A constructs with the removal of some key regions of the protein, cited as putative targeting signals.

P 36 Differentiation of cardosins trafficking pathways according to cell needs

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The seed has, in the context of other plant organs, some specific characteristics that make it a unique and compound structure, leading, among other features, to an adjustment of the intracellular trafficking pathways. Cardosins are plant aspartic proteinases (APs) isolated from flowers of *Cynara cardunculus* L. (cardoon) and they are considered to be good models to study intracellular trafficking given the fact that two similar proteins end up in different cell compartments in flower tissues: cardosin A is vacuolar and cardosin B is secreted. We focused our study on cardoon post-embryonic seed development with the aim to establish cardosins expression and sorting pathways in embryo cells and compare the obtained pattern with the one described previously in flowers. In storage organs cells, proteins that are typically to accumulated in protein bodies may not follow their ordinary pathway, but take a shortcut and go straight from endoplasmatic reticulum (ER) to their final destination. Given this, cardosin A and B expression was analysed during post-embryonic development in specific time-points of seed germination (dry, embedded, radicle emergence, hairy root and green cotyledons), through immunolocalisation assays with specific antibodies. We detected both cardosins inside protein bodies decreasing in amount along development. By definition, proteins destined to the protein bodies derive directly from de ER in dense vesicles, bypassing the Golgi apparatus (GA). In flowers, cardosin A is mainly accumulated in protein storage vacuoles and cardosin B is secreted to the extracellular matrix so, both proteins enter the secretory pathway and go through GA. Taking together these results, and comparing the route taken by cardosins in these organs, we consider that we are in face of a specialization of the trafficking pathways. Flowers and seeds are specialized and dynamic structures and both the physiological processes and the protein trafficking associated are under a high regulation control according to cell needs.