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Poster Presentations

Agnieszka Wilkowska, J Biotechnol Biomater 2017, 7:4 (Suppl)
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Enzymatic production of pectin-derived oligosaccharides from sugar beet pulp as a component of animal feed

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Statement of the Problem: Pectin-derived oligosaccharides (POS) are promising candidates for new-generation prebiotics. Sugar beet pulp (SBP) has a very high content of pectin, a complex polysaccharide mainly made up of three structural polymers: homogalacturonan (HG) and rhamnogalacturonans I and II. The combination of shorter HG chain length, high degree of acetylation and the large amount of side chains contributes to the poor gelling properties of sugar beet pectin. However, according to the SBP composition, this feedstock could be a suitable starting material for obtaining a variety of non-digestible oligosaccharides (NDO) with biological activity, such as oligogalacturonides (OGaU), arabino-oligosaccharides (AOS), and galacto-oligosaccharides (GaOS). The purpose of this study is to evaluate the potential of sugar beet pulp as a raw material for pectin-derived oligosaccharides production, potentially suitable as a prebiotic component of animal feed.

Methodology & Theoretical Orientation: The commercial enzymes applied included polygalacturonase, pectin lyase, pectinmethylesterase, arabanase and cellulase. Oligosaccharides with different degrees of polymerization (DP) were assessed using high performance anion exchange chromatography.

Findings: The pattern of obtained oligosaccharides was affected by the time of enzymatic hydrolysis – higher oligomers were hydrolysed to molecules of lower molecular weight. Enzymatic hydrolysis using arabanase yielded the highest concentration of oligosaccharides (DP 1-10) and was the most promising method for their production.

Conclusion & Significance: The application of sugar beet pulp for prebiotics production may be new and economically viable method of their utilization

Biography

Agnieszka Wilkowska is currently working as a Researcher in the Institute of Fermentation Technology and Microbiology, TUL. She has a strong background in enzymology as well as in biologically active compounds acquiring and preservation. Her expertise in probiotics and prebiotics resulted in several patents. Currently, her main field of interest is the production of pectin-derived prebiotics enriched in viable probiotic microbial biomass developed by simultaneous saccharification and fermentation (SSF) from a low-cost by-product

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Alessandra F Serain et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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UHPLC-MS/MS analysis of extracts of *Sinningia magnifica* (Gesneriaceae) from seedlings cultivable *in-vitro*, callus and *in-nature*

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The *Sinningia magnifica* belong to Gesneriaceae family which comprises of around 150 genera and 3500 species, distributed in tropics and subtropics around the world. In Brazil, phytochemical studies with *Sinningia* reported the identification of phenolic glycosides, anthocyanins and anthraquinones with biological properties. The *in-vitro* micropropagation is a way of maintaining healthy and uncontaminated explants for tissue culture application. The aim is to use the explants of S. magnifica to target callus and cell culture in suspension, which has been an excellent alternative for the exploration of secondary metabolites. In this work, initially a comparison was made between in-natura plant, *in-vitro* cultured seedlings and callus, to detect differences and similarities by UHPLC-MS and parity with some compounds of our research group by UHPLC-MS/MS. Thus, fruits of *S. magnifica* were collected and opened to release the seeds in Murashige e Skoog, 1962 (MS) medium, and cultivated in a photoperiod of 16/8 light/dark, 26±1ºC, with the leaves and the formation of calogenesis was induced with the use of growth hormones. Then, tubers and leaves of *S. magnifica in-nature* and *in-vitro*, and callus, which was dried at 40°C was collected, and subjected to the maceration with ethanol and determined the chemical composition by UHPLC-MS, and which was used in isolated compounds for comparison by UHPLC-MS/MS. The comparison showed common compounds and differences between the *in-natura* plant and the plant that grew in a controlled environment, as well as the callus, reinforced by the UHPLC-MS/MS analysis. Despite the positive results, further investigations are necessary to confirm the potential of this technique for the production of bioactive compounds and the real potential of the compounds for biological application.

Biography

Alessandra F Serain is currently a Pharmacist from the State University of Campinas and a PhD candidate with FAPESP scholarship, in the Biosciences and Technology Program of Bioactive Products of the DBV / IB - UNICAMP with the research project that seeks to perform a phytochemical study of two species of *Sinningia* for application in Photodynamic Therapy (PDT) in culture of human cells, and biotechnological study of plant cell culture. She has her experience of Scientific Initiation at BTPB-UNICAMP, 18 months with FAPESP scholarship, in the field of phytochemistry, PDT, essential oils and microbiological activity

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Paper-based diagnostic devices for evaluating the sperm quality

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Male infertility is the leading cause to approximately half of all infertility issues recorded worldwide. Many countries – especially developing ones – host various issues that have caused a rise in male infertility in recent years. With the rise of this issue, there are a limited number of treatments available for the poverty-stricken population. Mammalian sperm motility has traditionally been analyzed to determine fertility using computer-assisted semen analysis systems. To develop inexpensive and robust male fertility diagnostics, we created a paper-based tetrazolium-based colorimetric assay (MTT assay) and used it to estimate the sperm motility. After applying semen to the hydrophilic center circle of our patterned paper, a MTT assay data can be used to help estimate the percentage of motile sperm (sperm motility) in semen. When the porcine sperm (i.e. the model system that we used in this study) motility was influenced by two chemicals for both mitochondrial activity and glycolysis inside single sperms, we simultaneously recorded sperm motility and enzymatic reactivity through using a portable motility analysis system (iSperm) and a paper-based MTT assay, which was based on the calculation of the area mean value signal intensity. Using this paper-based device, we can evaluate fertility levels without consulting doctors. The duration and cost of one entire test are about 30 minutes and 0.1 USD, respectively. We believe that this paper-based assay system would be useful for fertility checks based on WHO references, without need of a microscope, at home. We wish to emphasize that our research could significantly advance a wide range of diagnostic developments.

Biography

Alex Ching is currently working on his internship under the expertise of Dr. Cheng through exploring and investigating in chemical-bioengineering at National Tsing Hua University, Taiwan. Through his developing passion to improve society through science, his simple and non-expensive model based on previous works and inspiration from other innovative scientists' fresh outlooks on this topic has opened a new perspective to improve healthcare products. After years of studying with a sizable measure of interning, he hopes researchers and individuals in developing countries with prospect of curtailing this issue may choose to use this new device to save funding.

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Aranha Netto L et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Plant cell and tissue cultures as experimental models for studies of coumarin and chlorogenic acid biosynthesis in *Mikania laevigata* (Asteraceae) and UHPLC-MS analysis

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Statement of the Problem and Aim: *Mikania laevigata*, guaco, is commonly used as treatment for respiratory system diseases due to its bronchodilator effect, bioactivity assigned to coumarin. Plant cell cultures are very promising tolls to produce secondary metabolites in vitro. They allow a rapid cell proliferation, they are under strict controlled conditions and are easy to manipulate. This research aimed to investigate the use of tissue and cell cultures of *M. laevigata* as experimental models applied to the study of coumarin and chlorogenic acids biosynthesis in this plant.

Methodology & Theoretical Orientation: Explants obtained from young leaves were sterilized and maintained on MS medium supplemented with 30 g.L-1 of sucrose and sub-cultured every 30 days. After several tests with different proportions of hormones to determine their optimal concentrations, medium containing 4.52 µM of 2,4-dichlorophenoxyacetic acid and 26 µM 6-benzilaminopurina was established as the best condition for callus induction for this species. Following the successful establishment of the callus culture, cell suspensions were established by inoculating cells from friable calli into liquid MS medium. Analysis for identification and quantification of the phenylpropanoid biosynthetic pathway products were done by UHPLC-MS, using methodology described by Melo.

Findings: The analysis of the chemical profile of *M. laevigata* calli, after 15, 30 and 60 days of subculture, detected mainly coumarin, caffeoylquinic acid and dicaffeoylquinic acid. The highest concentration of coumarin was observed after 30 days, 0.12 ± 0.01 mg.g-1 dry mass. The highest quantification of caffeoylquinic acid was observed after 15 days, which contained 5.53 ± 1.29 mg.g-1 of dry mass, while dicaffeoylquinic acid had a higher concentration after 60 days, with 36.39 ± 4.58 mg.g-1 dry mass.

Conclusion: Tissue and cell cultures of *M. laevigata* are promising biological models for application in studies of biosynthesis of its bioactive secondary metabolites. However, to confirm their full potential, the continuity of the investigations is necessary.

Biography

Aranha Netto L is currently a Master's student in the Plant Biology Program at the Institute of Biology - Unicamp. Her research investigates the use of plant cell and tissue cultures as biological models applied to the study of coumarin and chlorogenic acid biosynthesis in *Mikania glomerata* and *Mikania laevigate* (Asteraceae). She holds a Bachelor's degree in Biological Sciences from the State University of Campinas (2015), including a year studying at the University of East Anglia (UEA), England as part of the exchange program science without borders. Also in England, she did a research placement at Inspiralis Ltd.

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Aurelia Mandes et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Characterization of the advanced Ti-Ag coatings deposited by Thermionic Vacuum Arc method for biomedical applications

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The aim of this paper is to report on the results of the morphology, structure and wettability of the Ti-Ag films deposited by using the Thermionic Vacuum Arc (TVA) method. The morphology and the structure of the thin films surface were examined using a TEM (Phillips CM 120 ST, acceleration voltage of 120 kV, resolution point of 1.4 Å and a magnification of 1.2 M) and Scanning Electron Microscopy (SEM, Zeiss EVO 50 SEM). Wettability of the surface was calculated by the contact angle method. The measurement was performed by establishing the tangent angle of a sessile liquid drop on a solid surface, defined by the mechanical equilibrium of the drop under the action of three interfacial tension solid-vapour, solid-liquid and liquid – vapour, by meaning of the performed analysis software See System. All of these results were developed taking into account the Ti-Ag are difficult to be processed. However, Ti-Ag coatings are used in various medical applications due to bio- and chemically inert, being presented as prospective biomaterials in artificial hips, pins for setting bones and for other biological implants. Thermionic Vacuum Arc can be ignited in vacuum between a heated cathode surrounded by an electron focusing Wehnelt cylinder and an anode containing the material to be deposited (Ti-Ag). The TVA method is an original discharge type in pure vapour plasma, which can become one of the most suitable technology to significantly improve the quality of the surfaces covered with different materials. Two types of substrates were used in this work: silicon wafer and glass. Before the deposition, the substrates were chemically cleaned in ultrasonic bath with a highly effective special cleaner. The uniformity, low roughness and smoothness suggest that Ti-Ag thin films deposited by TVA technology could be considered as valuable advanced coatings titanium-based materials for biomedical applications

Biography

Aurelia Mandes has completed her PhD from PhD School at Physics Faculty from Bucharest University Romania. She is a Post-Doctoral Researcher at Ovidius University, Faculty of Applied Sciences and Engineering, Constanța, Romania. She has published more than 24 papers in reputed international journals (*J Appl Phys, Contrib Plasma Phys, - Eur. Phys. J. D*) and three Chapters in books at Wiley – VCH Publisher 2010, NOVA Publisher 2012 and IN TECH Publisher 2016. She has expertise in deposition and characterization of nanostructured thin films obtained by the Thermionic Vacuum Arc (TVA) technology

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Cabanes Cos J et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Semi-synthesis of lysine-betaxanthin and its fluorescent properties

Cabanes Cos J, Gandía-Herrero F, Jiménez-Atiénzar M, García-Carmona F and **Escribano-Cebrián J**
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Betalains are nitrogen-containing natural pigments that provide bright coloration to fruits, flowers, and roots of plants of the Caryophyllales order and present autofluorescence after excitation with blue light. They are divided into two groups: violet betacyanins, with absorbance spectra centered at wavelengths around λm = 536 nm, and yellow betaxanthins, with absorbance spectra centered at wavelengths around λm = 480 nm. Both groups share betalamic acid as their structural and chromophoric unit, which is condensed with *cyclo*-DOPA in the betacyanins and with amines and amino acids in the betaxanthins In this work, the semi-synthesis of lysine-betaxanthin from betanin, purified from red beet juice concentrate has been carried out. Basic hydrolysis of betanin released betalamic acid, whose aldehyde group was condensed with the amine group of lysine. Immediately after synthesis, lysine-betaxanthin was partially purified by solid phase extraction with a C-18 column. The pigment was characterized by absorbance spectroscopy and HPLC-DAD analysis. Electrospray ionization mass spectrometry analysis (HPLC-ESI-MS-MS) was applied to elucidate the pigment nature. Since lysine has two amine groups, one α and one ε, the *in-vitro* reaction of the amino acid with betalamic resulted in the formation of two adducts. In this work, the native fluorescence of lysine-betaxanthin has also been characterized, by using an aqueous solution of the pigment for registration of the fluorescence spectrum.

Biography

Cabanes Cos J, trained as a Biochemist at the Department of Biochemistry and Molecular Biology A of the University of Murcia (Spain). She got her PhD in 1986 and since then she has been working in Plant Biochemistry and Biotechnology. She has been teaching for more than 30 years Biology and Biotechnology both in University of Murcia. She also teaches Master's of Molecular Biology and Biotechnology of the University of Murcia. Currently, her research project combines different approaches and multiple techniques to study the functional capacity of a family of bioactive plant compounds -the betalains

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Catalina Andrea Lugo De Ossa et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Study heterotrophic growth of Chlorella spp. under different carbon-to-nitrogen and carbon-to-phosphorous ratios

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Microalgae have caused interest in recent years because of their particular way of accumulating lipids. These microorganisms can be cultivated in an autotrophic, mixotrophic, and/or heterotrophic way. Heterotrophic cultures decrease growth time, increase biomass concentration, and total lipid yields. Appropriate composition of the culture medium will favor the growth of the cells, for this reason commercial culture media have been modified to establish the effect of increasing or decreasing the concentration of some nutrients when producing biomass and lipids. However, to achieve good lipid yields, it is necessary to ensure a high concentration of biomass at an initial stage of cultivation. Lipids, being primary metabolites, can be induced by subjecting the microalgae to stress conditions depending on both the species and the abiotic factors. This work evaluated the heterotrophic growth of the native microalga *Chlorella* sp using glucose as carbon source and varying relations carbon/nitrogen and carbon/phosphorus to favor the production of biomass. In addition, the change of fatty acid composition changes with biomass production. Maximum biomass obtained was 9.25g/L and 8.67g/L for C/N of 25:1 and C/P of 200:1 during 7 days of cultivation, their productivities were 0.93g/L*d and 0.99g/L*d. Total fatty acid production was favored with C/N 50:1 and C/P 400:1 reaching 25.7% and 22% of total fatty acids in dry biomass, also higher fatty acid productivities in biomass of 41.16mg/L*d and 24.32mg/L*d with C/N 10:1 and C/P 200:1. Low C/N and C/P ratios stimulated biomass production, biomass lipid productivity, and decreased total fatty acid production. High C/N and C/P ratios improved the production of total fatty acids. In this way, the maximum production of biomass must be reached for further achieving the stage of nutritional exhaustion due to the deficiency of N and P in the culture medium. This causes the elongation of polyunsaturated fatty acid chains.

Biography

Catalina Andrea Lugo De Ossa is an Industrial and Environmental Microbiologist from Universidad de Antioquia, Medellin, Colombia, She belongs to Bioprocess Research Group in the microalgae investigation line. She participated in the design of culture medium for the induction and production of intracellular fatty acids, proteins, secondary metabolites and water bioremediation of heavy metals using wild microalgae isolated from water sources in Colombia.

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Production of bioactive pigments in cell lines of quinoa (*Chenopodium quinoa*)

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Betalains are water-soluble, nitrogen containing pigments present in most plants of the order Caryophyllales. Betalains are classified into two structural groups: betacyanins (violet) and betaxanthins (yellow). Both groups share betalamic acid as the structural and chromophoric unit. It is condensed with amines and amino acids in betaxanthins and with *cyclo*-DOPA in betacyanins. Betalains are bioactive molecules with high antioxidant and free radical scavenging activities and a strong chemopreventive potential. The present research was focused on the establishment of callus cell lines derived from the plant quinoa (*Chenopodium quinoa*). Quinoa belongs to the family Amaranthaceae and thus to the betalain producing order Caryophyllales. Cell lines were developed on semisolid medium and the betalamic pigments synthetized by different lines have been identified by HPLC-DAD-MS/MS, as well as the pigment content present in the seedlings used as explants. Violet callus cell lines of *Chenopodium quinoa* were developed from hypocotyls of differently colored Peruvian quinoa grains varieties (Figure C), recently described as source of betalains. In callus, the major pigment identified was betanin, while in seedlings one of the major pigments detected was the non-glycosylated and hydroxylated precursor of the violet compounds, betanidin. For the first time betalain producing callus lines derived from quinoa were established. This offers an opportunity to develop cell suspension cultures to be used as bio-factories in the production of bioactive pigments betalains. The controlled production of theses pigments to be used as functional natural colorants may be of interest for the food, pharmaceutical and cosmetic industries.

Biography

Josefa Escribano-Cebrián was trained as a Biochemist at the Department of Biochemistry and Molecular Biology of the University of Murcia (Spain). She got her PhD in 1986 and since then she has been working in Plant Biochemistry. She has publications in national and international journals. Currently, her research project combines different approaches and multiple techniques to study the functional capacity of a family of bioactive plant compounds-the betalains

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Kalada Itelima et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Causes of dampness in residential building walls in Jos Metropolis, Plateau State, Nigeria: Emphasis on microbial concentrations, biodegradation and health hazards

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Statement of Problem: Damp houses can be unhealthy and costly to remedy. Knowing more about the problems can reduce the worry and expense. Most dampness problems can be cured or minimized by simple remedial work but a few will need substantial outlays. In many instances, dampness in your home may not be present all year round and, depending on the source, may only become evident after a change in weather conditions, usually during the colder or wetter months or after periods of heavy or extreme rainfall. Jos Plateau is located on the North Central region of Nigeria. It is relatively undulating plain bounded by mountainous ridges. The prevailing climatic situation of the study area hinges upon alternate seasons of hot and cold weather conditions interspersed with an average period of about 6 months of rainfall, with July and August being the peak period. Dampness is the wetting of structural elements through moisture rise by capillary action. Dampness is one of the most serious structural defects in walls of buildings. Dampness in walls spoils paints and interior decorations, encourages the growth of bacteria and moulds, hampers aesthetics, and poses a threat to health of occupants through providing breeding conditions for mosquitoes. Dampness undermines structural integrity of wall elements, reduces thermal insulation property of building of building materials as well as affects the comfort not the occupants. Dampness causes damages to both structural and building materials and reconstruction efforts can be enormous. This study aimed at investigating the causes of dampness in the walls of some of residential buildings within Jos metropolis, Plateau State, Nigeria.

Methodology & Theoretical Orientation: The causes were diagnosed through tests, physical inspections and oral interviews. Oral interviews were held with the occupants of 500 randomly selected buildings in the study area. Fifty construction sites were also randomly selected for detailed study of the constructional practices and material adopted in building constructions within the study area. The fungi associated with the damped houses were also considered.

Findings: Results identified that about 80% of the houses investigated had damping defects at various degrees. Concentrations of fungi bacteria associated with the damp walls: *Aspergillus spp*. (55%), *Penicillium spp*.(24%), *Trichoderma sp* (12%), and *Fusarium sp*. (9%), while viable concentrations of Bacteria were <100 cfu/m^3 from damp houses. The microbial concentrations of none damp houses varies between 60%-70%.

Conclusion & Significance: The results also showed that rising damp through defective damp-proof membranes and efflorescence on walling unit serve as the major causes of dampness in residential building walls. Appropriate recommendations were made as preventive and remedial measures to the problem. For example, research shows that people living in well-insulated and adequately ventilated accommodation are less likely to visit their doctor or be admitted to hospital due to respiratory conditions than those living in damp homes.

Biography

Kalada Itelima qualified as an Architect from Rivers State University of Science and Technology Rivers State, Nigeria. He is presently the Director of Kalite Associate Nigeria. He has participated in conferences where he was actively involved in home and abroad. He is also involved in designing, building construction and consultancy services. He is giving community services in Jos Plateau, Nigeria, where he is residing and to other parts of the country

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Kaloyan K Petrov et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Inulin conversion towards fructose or lactic acid by Mn2+ mediated metabolic flux in *Lactobacillus paracasei*

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Presuming the important role of cations in carbohydrate metabolism, sugars transport, and potentially, in the regulation of genes transcription levels, it may be expected that they would exert a significant influence on inulin conversion by *L. paracasei*. The aim of this study was to obtain maximal amounts of two valuable bio-chemicals: lactic acid and fructose, by engagement of bivalent metal ions as a new instrumentality to govern the process of inulin conversion. The kinetics of batch processes of direct inulin conversion to LA showed that Mn2+addition accelerated the inulin hydrolysis and sugars consumption by allosteric activation of inulinase and enhancement of the glycolytic flux. The highest LA concentration was reached by 15 mM Mn2+ addition - 151 g/L, corresponding to 40% increase, with yield 0.83 g/g substrate. This LA amount is the highest ever obtained from inulin and discloses the role of Mn2+ as a powerful tool for LA production intensification. On the other hand, the exclusion of bivalent metal ions led to elevated expression of fosE gene, encoding fructan-ß-fructosidase - the key enzyme for inulin hydrolysis. Thus, if the process of inulin conversion is conducted by fed-batch fermentation mode (providing substrate excess) and in medium devoid of salts and microelements, maximal fructose concentrations could be achieved. During such process total amountof 675 g inulin was hydrolyzed, giving rise to formation of359 g/L fructose, along with 55.2 g/L LA, 34.8 g/L glucose,17.9 g/L sucrose, and about 25 g/L oligo sugars. In conclusion, the present study is the first that reveals the important role of bivalent cations on the overall process of inulin fermentation by *Lactobacilli.* The obtained fructose concentration is the highest ever received during microbial process (36% solution), allowing the developmentof a novel approach for fructose production via microbial fermentation of inulin.

Biography

Kaloyan K Petrov is the Head of the Department of Chemical and Biochemical Reactors in the Institute of Chemical Engineering, Bulgarian Academy of Sciences. His work is devoted to optimization of downstream processes by the techniques of bioprocess and metabolic engineering. His team develops biotechnologies for microbial production of platform chemicals and fuels by conversion of waste or renewable energy resources, including the cutting-edge biotechnologies for 2,3-Butanediol production from glycerol and starch by the use of natural and recombinant strains, and novel bio-processes for lactic acid synthesis by utilization of starch and inulin. Other topics of his work are the fermentative production of enzymes and valuable chemicals in respect of their industrial application, strain's improvement by gene engineering, microbiological and molecular biological tools, development and analysis of probiotics and prebiotics

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Effect of elicitors on the antioxidant activity of different plant cell cultures

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Statement of the Problem: Grapevine is a good source of antioxidant compounds like phenolic compounds which are important in human nutrition. Likewise, broccoli contains a wide range of nutrients, dietary fibre, and phytochemicals with health-related properties. On the other hand, safflower is a medicinal plant, which has red and yellow pigments that can be used as food colorants or in the cosmetic industry. Safflower also produces bioactive compounds such as tocopherols and carthamin. Due to the high value of these compounds, new strategies have been used in order to increase their production since their extraction from raw plant materials is often tedious, expensive and the extracts are often heterogeneous. In this way, we have developed a method of production of antioxidant compounds based on the elicitation of plant cell cultures with β-cyclodextrins (CD) separately or in combination with other elicitors such as methyl jasmonate, coronatine, NaCl, β-glucan and hexenol.

Methodology: We have evaluated the effect of CD, methyl jasmonate, coronatine, NaCl, glucan and hexenol on the antioxidant activity in extracellular medium of grapevine, broccoli and safflower cell cultures for 144 h of incubation.

Findings: In this work, we have observed that the highest levels of antioxidant activity were found in safflower cell cultures elicited with β-glucan, β-glucan+CD or hexenol+CD. In addition, we also detected high levels of antioxidant activity in grapevine cell cultures treated with CD and methyl jasmonate while no significant differences were observed in any of the treatments performed in broccoli cell cultures compared to control cells.

Conclusion: Grapevine and safflower cell cultures elicited were able to produce high levels of antioxidant compounds, and therefore, these elicited plant cell cultures can provide an alternative system, which is at the same time, a more sustainable, economical and ecological system for their production.

Biography

Lorena Almagro has completed her PhD from Murcia University and her Post-doctoral studies at the Institute of Molecular and Cell Biology in Porto (Portugal). In 2014, she had a Post-doctoral position in the University of Murcia. She has received her first award for Applied Research in a private Company by CEEIM (2009) and European Doctorate Awards (2013). She has published 25 papers in reputed journals and her work has been focused on the production and identification of bioactive compounds derived from different plant cell cultures under elicitation.

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Use of broccoli byproducts to obtain bioactive compounds

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Broccoli (*Brassica oleracea L. var. italica*) is an important crop since along with other Brassica vegetables its world commercial production is estimated over more than 90 million tons per year. Broccoli sprouts have attracted particular attention due to their high nutritional and functional values since they are an excellent source of a variety of vitamins (A, C, E, K, among others), essential nutrients and minerals, dietary fibre and many bioactive compounds among which stand out glucosinolates (i.e., glucoiberin, glucoraphanin, glucoalyssin, glucobrassicin, neoglucobrassicin) and carotenoids (β-carotene, lutein and zeaxanthin among them). In fact, the data available reveal broccoli to be a healthy food due to the beneficial biological effects of these bioactive compounds. In fact, glucosinolates are the most important health-promoting compounds commonly found in broccoli due to their antioxidant, antimicrobial, cardiovascular, antidiabetic, and antitumoral activities. On the other hand, broccoli byproducts, produced after harvest, can represent an important environmental problem. They have been used traditionally as an animal feedstuff, for fibre extraction and as a source of glucosinolate standards. However, the increase in broccoli cultivation in the last few years has made it difficult to find uses for the total amount of byproducts generated. For these reasons, broccoli byproducts have been proposed as a source of bioactive compounds. In this work, we have identified and quantified both hydrophilic and lipophilic bioactive compounds found in broccoli byproducts since they could be used as ingredients in the development of novel functional foods, thus adding value to them and reducing agricultural wastes.

Biography

María Angeles Pedreño holds a degree in Chemistry and a PhD in Sciences, Section Chemistry. She did a Post-doctoral stay in the Plant Biotechnology Department of the Agricultural School of Toulouse (ENSAT). In 1993, she got a permanent position as a Lecturer in Plant Physiology in the Department of Plant Biology, University of Murcia. She is a full Professor of Plant Physiology at the same University since 2006. She has published more than 130 papers in reputed journals and her research lines have been developed in the field of Plant Physiology and Biotechnology

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Penka M Petrova et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Amylolytic enzymes engaged in starch utilization by lactic acid bacteria: A transcriptome analysis

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Amylolytic lactic acid bacteria (ALAB) are diverse group of microorganisms that are capable to degrade starch and to convert it directly into sugars with lower molecular weight, lactic acid, and volatiles. They have numerous applications in food industry due to specific ability to improve the organoleptic properties and to increase the nutritional value of starch foodstuff, as well as in perspective biotechnologies for lactic acid production using renewable and abundant starch biomass as a feedstock. Here we report a comprehensive study of the amylolytic properties of 28 novel ALAB strains inhabiting the special niche of fermented cereals. The study describes the isolation of the first amylolytic representatives of *Lactobacillus sakei, Enterococcus faecium* and *E. durans*. By transcriptome analysis, it was revealed that seven different genes are engaged in starch degradation by ALAB, as their expression levels are species and strain specific. The most highly expressed in all strains were the genes encoding amylases and glycosyltranferases, in difference to the genes for α-glucosidases. One exception among the genus *Lactobacillus was L. sakei*, which although possessing extracellular amylase activity, owned very limited number of genes for starch hydrolysis (*glgB, agl*, and *treC*). Interestingly, only one of these genes was expressed – *treC*. It encodes an enzyme with a dual oligo-1,6-glucosidase and trehalose-6-phosphate hydrolase activity in *L. paracasei*, and *L. rhamnosus*, and therefore, it may be potentially responsible for the amylolytic activity of *L. sakei* too. Another enzyme, possibly interacting with starch in *L. sakei* is glucan 1,6-a-glucosidase (dextran glucosidase), encoded by dexB. However, since the last two genes encode intracellular enzymes, but amylase activity of the strain was extracellular, the most likely acting enzyme may be unrecognized and unexploited, similarly to other unknown, but putative glycoside hydrolases in lactobacilli.

Biography

Penka M Petrova is the Head of Gene Expression Laboratory at The Institute of Microbiology, Bulgarian Academy of Sciences. Her main interests are in the area of Microbiology and Molecular Biology of lactic acid bacteria (LAB) with special attention to probiotic starter cultures development including isolation and genetic characterization of LAB, searching for new enzymatic activities, prebiotics utilization and synthesis, genes cloning and expression. She is the author of more than 50 scientific publications and book chapters, cited more than 430 times. She is a leader of a number of research projects, funded by the National Scientific Fund, Republic of Bulgaria, Chr. Hansen A/S, and State Key Laboratory of Dairy Biotechnology of Bright Dairy & Foods Co. Ltd

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Obesity electrochemical nanobiosenor on nanowell array electrode

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In general, obesity is associated with significant disturbances in appetite and metabolic control system. Both neuropeptide hormones (leptin) and monoamine neurotransmitters have been recognized as obesity markers due to their essential roles in the regulation of food intake and energy expenditure. Here, we present the development of a leptin nanosensor as a multi-assay for obesity markers found in blood to create a personalized medical system. We have reported that nanowell array (NWA) can enhance electrochemical detection of molecular binding events by controlling the binding sites of the captured molecules. Using NWA biosensor based amperometric analysis; we have detected biological macromolecules such as DNA, protein or aptamers at low concentrations. These results suggested that wafer-scale NWA immunosensor will be useful for biosensing applications because their interface response is appropriate for detecting molecular binding events

Biography

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Construction and membrane optimization of a new all solid-state contact Fe(II) selective sensor

Tugba Ozer and **Ibrahim Isildak**
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Iron is the fourth abundant element on Earth's crust and it is present in various rock and soil minerals in the form of iron (II) and iron (III). Iron is a crucial element for both plants and animals since it plays an important role in several metabolic processes. Iron and its compounds interfere with natural sources as a pollutant through the discharge of wastes since they have been widely used as containers and pipelines all over the world. Consequently, it became important to detect and monitor iron levels selectively and sensitively for not only in biological samples but also in industrial and environmental samples. Potentiometric sensors have been developed by immobilizing a membrane matrix on all-solid-state contact. This technique has advantages such as sample monitoring without any reagent consumption and very short analysis time. The all-solid-state contact sensor was prepared in certain ratio of graphite-epoxy resin and then the surface of all-solid-state contact sensor was coated with the sensing membrane without an internal reference solution. Therefore, its potentiometric response became more stable and life-time of the sensor increased without any loss in its potentiometric characteristics. To the best of our knowledge, there is no report available in literature regarding the detection of Fe (II) using dithiocarbamate as a sensing material. In this study, a novel iron(II) ion selective sensor was developed and the electrode composition was optimized

Biography

Tugba Ozer is a Research and Teaching Assistant in Yildiz Technical University, Istanbul, Turkey. She has completed her MSc in Marmara University. She is a PhD student in Yildiz Technical University, Department of Bioengineering in Turkey. She has been a visiting PhD student in Department of Soil and Crop Science, Colorado State University. Her PhD research is focused on developing iron(II) and iron(III) selective sensors and applying them in soil and water samples. Her specialization areas are: electrochemistry, environmental chemistry, analytical chemistry, heavy metals, sensors, crop science, biopolymer, bioinformatics and systems biology

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Electrochemical nanobiosensor on Si3N4 nanowell array electrode

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In order for an electrochemical sensor to be adopted in a wide range of applications such as diagnosis in traditional medical, pharmaceutical, and healthcare settings, medical diagnosis in non-hospital setting (e.g., self-administered consumer diagnostics as pregnancy test or blood glucose monitoring), non-medical detection of biological and small molecule detection, companion diagnostics for pharmaceutical therapeutics; research applications where detection of small molecules are required, and other settings or circumstances where detection of biological molecules is needed, the electrochemical sensor must be sensitive, selective, easy to use, and readily available to users (i.e., able to manufacture scalably, in large quantities, and at a low cost). We have reported that Si3N4 nanowell array (NWA) can enhance electrochemical detection of molecular binding events by controlling the binding sites of the captured molecules. Here we'll report an immunosensor based on wafer-scale NWA for electrochemical detection of TNF-α. In order to develop NWA sensor through the cost-effective combination of high-throughput nanopattern, the NWA electrode was fabricated on glass wafer by krypton-fluoride (KrF) stepper semiconductor process. Finally, 12,500,000 ea nanowell with a 230 nm diameter having a pitch ratio (ratio between the diameter of the nanowell openings and the shortest distance between neighboring nanowells) of approximately 1:1 was fabricated on 4 x 2 mm2 substrate

Biography

YoungTae Seo is currently graduate student at Queens College of the City University of New York, USA.

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Highly sensitive toxicity evaluation using nanotechnology methodology

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The clinical need for an assay reliably predicting cardiovascular and cardiovascular-related symptoms such as lung injuries is immediate. These symptoms caused by inhalation of hazardous substances in the atmosphere are increasing rapidly and the current technology for evaluation of toxicity has been remained on simple analysis. Demand for evaluation of toxicity by using highly sensitive nanobiosensor is growing rapidly, but research and development levels are insufficient condition. Here we will present a reliable cardiac biomarker (Oxidized LDL, Fibrinogen, Adiponectin, 8-isoprostane) based electrochemical (EC) nanowell array sensor for evaluation of cardiac toxicity in human serum using nanotechnology methodology. EC techniques are rapid, reliable, and the resulting electric signals can be easily amplified and scaled down. These EC nanobiosensors have significantly advanced the biomolecular detection by increasing sensitivity, limit of detection (LOD), multi-targeting, and being label free, compared with conventional ELISA or HPLC. Especially, the nanosensors have extremely low volume on the order of atto-liters per well, and a total volume of approximately 32 femto-liters per array. This research will aid in the early diagnosis of cardiovascular disease, especially in patient throughout worldwide. We already reported that nanowell array electrode can enhance electrochemical detection of molecular binding events by controlling the binding sites of the captured molecules. Using nanowell biosensor based electrochemical analysis, we have detected biological macromolecules such as DNA, proteins or aptamers at extremely low concentrations. We will utilize evaluation of cardiac toxicity system by combining with (1) nanowell array electrode based on nanotechnology and (2) EC measurement.

Biography

YoungTae Seo is currently graduate student at Queens College of the City University of New York, USA.

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Muhammad Imran Amirzada, J Biotechnol Biomater 2017, 7:4 (Suppl)
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Recombinant human interleukin 24 reverses Adriamycin resistance in a human breast cancer cell line

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The major cause of multidrug resistance is over-expression of membrane P-glycoprotein (P-gp). We investigated the effect of recombinant human interleukin 24 (rhIL-24) on the Adriamycin (ADM)-resistant human breast cancer cell line MCF-7/ADM. Methods: The cytotoxicity of rhIL-24 and ADM was determined by 3-[4,5-dimethylthiazol-2-yl], 5-diphenyl tetrazolium bromide (MTT) assays. The expression of P-gp was assessed by confocal microscopy and Western blot analysis. Results: The IC50 values for rhIL-24 in MCF-7/wild-type and MCF-7/ADM cells were 0.17 and 14.6 mM, respectively. The IC50 value of Adriamycin in MCF-7/ADM cells decreased in a dose-dependent manner when rhIL-24 was used. The resistance modulating factor (RMF) was directly proportional to the dose of rhIL24. ADM accumulation increased while P-gp expression decreased at a low dose (4 mM) of rhIL24 in MCF-7/ADM cells. The expression of P-gp was decreased at 4 mM in confocal microscopy and western blot analysis. Conclusions: rhIL-24 circumvented the drug-resistance of MCF-7/ADM cells via activation of the transcription factor Stat 3. rhIl24 has potential to act as a P-gp inhibitor to reverse Adriamycin resistance in breast cancer.

Biography

Muhammad Imran Amirzada has completed his Master of Science from Liverpool John Moores University, UK and Ph.D from Jiangnan University, P. R. China. Currently he is serving as Assistant Professor of Pharmacy, COMSATS Institute of Information Technology, Abbottabad, Pakistan. He has published research papers in reputed journals related to Recombinant Human Therapeutic Protein production. He has successfully win a project from Higher Education Comission (HEC) Pakistan for Recombinant Human Interleukin 24 with Doxyrubicin formulation

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Khuat Huu Trung et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Exploitation of rice genome sequence and genetic diversity in Vietnamese rice landraces for research and breeding programs to cope with climate change

Khuat Huu Trung, Pham Thi Ly Thu and **Tran Dang Khanh**
Agricultural Genetics Institute, Hanoi, Vietnam

Rice (*Oryza sativa* L.) is a principle crop in Vietnam and plays an important role in economic activity in this country. However, rice growing areas and rice productivity are significantly reduced due to increasing adverse impacts from climate changes as emerging pathogens, drought and rising sea levels. Our attempts have made collaboration with The Genome Analysis Center-TGAC (Earlham Institute), the John Innes Centre (JIC) in UK to launch the collaborative projects which focused on re-sequencing some Vietnamese rice landraces. Some successful works have been made in investigation, data collection, building of native rice landraces of Vietnam (focus on good quality rice varieties with tolerance to biotic and biotic stresses); analyzing genetic diversity of native varieties in Vietnam at the molecular level, selected 36 elite rice varieties with high diversity for genome sequencing; coordinating full genome sequencing, building genotype databases; adding supplementary assessments of major morphological agronomy traits, establishing phenotype databases of the genome sequenced rice landraces; mapping SNPs; design to CAPS markers for research and breeding; launching the project website to manage and sharing genotype and phenotype database of rice; training researchers and staff in the field of genome sequence and genome-data mining of rice; published some research articles on the National and International Journals. In the second phase of collaboration, we have re-sequenced the genomes of more 600 Vietnamese native rice varieties and exploited their databases by applying bioinformatics pipelines to identify association of alleles with specific agronomic phenotypes traits of interest. The rapid identification of rice landraces those are tolerant and resilient to adverse conditions which will work towards alleviating the current challenges the country agriculture industry face and contribute to food security. Currently, a number of promising rice lines carrying specific/multiple genes, candidate genes of abiotic and biotic tolerances from rice genome sequence (salt tolerance, bacterial leaf blight, and plant brown hopper resistances) have been developed by molecular breeding programs and were sent for National Test and will be released to the farmers soon time.

Biography

Huu Trung is a Senior Researcher, working at Agricultural Genetics Insitute (AGI), Hanoi, Vietnam. He has expertise in field of genetics and crop breeding, molecular breeding as well as bioinformatic analysis. He is currently Deputy Director General of AGI

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Virginia Dinca-Balan et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Studies on the CTi an CAg biomaterials obtained by thermionic vacuum arc technique

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Metal containing DLC films with properties intermediate between DLC and metal carbides have been shown to improve the adhesion and the wear properties. Studies have shown that carbon thin films are suitable as surface coatings on biomedical devices. Doping of carbon with selective elements is an attractive method to enhance the biological and other properties of the thin film. In this work, Silver was chosen as the dopant because of its anti-bacterial properties. As the chemical interactions between Ag and C generally are very weak, and the interactions between the used transition metals and C are strong, it was expected that Ag to form a separate phase. This was observed as the nanocomposite films were consisting of Ag grains embedded in a nanocrystalline or nanocomposite matrix. Titanium carbide (TiC) hard films are considered as high technology materials due to their unique characteristics that have made them of particular interest in a wide variety of applications. TiC it is one of the tribological thin film materials originally developed to replace highly toxic hexavalent chromium (HCr) for prosthetic joints. CAg and CTi nanocomposites thin films were prepared on glass and silicon substrates by Thermionic Vacuum Arc method in a single-gun configuration. After the ignition of the plasma, the shuttle was removed, allowing homogeneous thickness and composition in the substrates over an area of 10 cm^2. Under certain operating electrical parameters, the plasma becomes stable and can be maintained for as long as the anode material is present. During the deposition, the substrate holder was not rotated. The substrate was kept at ground potential during the deposition of the metal plasma. The results showed that the films' hardness could be significantly increased, and the wear resistance as well. This is due to the microstructure of the films comprising nanocrystalline grains in an amorphous carbon matrix.

Biography

Virginia Dinca-Balan has completed her PhD from PhD School at Physics Faculty from Bucharest University Romania. She is an Assist Prof Dr. at "Ovidius" University of Constanta, Romania, Medicine Faculty on Biophysics and Medical Physics discipline. She published more than 22 papers in reputed international journals (e.q. *J Appl Phys, Contrib Plasma Phys, - Eur. Phys. J. D*) and three chapters in books at Wiley – VCH Publisher 2010, NOVA Publisher 2012 and IN TECH Publisher 2016

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Aranha Netto L et al., J Biotechnol Biomater 2017, 7:4 (Suppl)
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Plant cell and tissue cultures as experimental models for studies of coumarin and chlorogenic acid biosynthesis in *Mikania laevigata* (Asteraceae) and UHPLC-MS analysis

Aranha Netto L, Forato J, Andreazza N L, Sawaya A C H F and **Salvador M J**
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Statement of the Problem: *Mikania laevigata*, guaco, is commonly used as treatment for respiratory system diseases due to its bronchodilator effect, bioactivity assigned to coumarin. Plant cell cultures are very promising tolls to produce secondary metabolites *in vitro*. They allow a rapid cell proliferation, they are under strict controlled conditions and are easy to manipulate. This research aimed to investigate the use of tissue and cell cultures of *M. laevigata* as experimental models applied to the study of coumarin and chlorogenic acids biosynthesis in this plant.

Methodology & Theoretical Orientation: Explants obtained from young leaves were sterilized and maintained on MS medium supplemented with 30 g.L-1 of sucrose and sub-cultured every 30 days. After several tests with different proportions of hormones to determine their optimal concentrations, medium containing 4.52 µM of 2,4-dichlorophenoxyacetic acid and 26 µM 6-benzilaminopurina was established as the best condition for callus induction for this species. Following the successful establishment of the callus culture, cell suspensions were established by inoculating cells from friable calli into liquid MS medium. Analysis for identification and quantification of the phenylpropanoid biosynthetic pathway products were done by UHPLC-MS, using methodology described by Melo.

Findings: The analysis of the chemical profile of *M. laevigata* calli, after 15, 30 and 60 days of subculture, detected mainly coumarin, caffeoylquinic acid and dicaffeoylquinic acid. The highest concentration of coumarin was observed after 30 days, 0.12 ± 0.01 mg.g-1 dry mass. The highest quantification of caffeoylquinic acid was observed after 15 days, which contained 5.53 ± 1.29 mg.g-1 of dry mass, while dicaffeoylquinic acid had a higher concentration after 60 days, with 36.39 ± 4.58 mg.g-1 dry mass.

Conclusion: Tissue and cell cultures of *M. laevigata* are promising biological models for application in studies of biosynthesis of its bioactive secondary metabolites. However, to confirm their full potential, the continuity of the investigations is necessary

Biography

Aranha Netto L is currently a Master's student in the Plant Biology Program at the Institute of Biology - Unicamp. Her research investigates the use of plant cell and tissue cultures as biological models applied to the study of coumarin and chlorogenic acid biosynthesis in *Mikania glomerata* and *Mikania laevigate* (Asteraceae). She holds a Bachelor's degree in Biological Sciences from the State University of Campinas (2015), including a year studying at the University of East Anglia (UEA), England as part of the exchange program Science without borders. Also in England, she did a research placement at Inspiralis Ltd

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Mono- and dipeptide derivatives of 17β-Amino-5α-androstan-3β-ol

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Saturated and unsaturated 17 β-aminosteroids are used as intermediates to synthesize biologically active derivatives. Peptide analogs of aminosteroids with various physiological activities such as anti-tumor and anti-arrhythmic have been found, were prepared by adding amino acids to aminosteroids. Mono- and dipeptide derivatives of 17β-amino-5α-androstan-3β-ol were synthesized by N-acylation with N-protected amino acids (N-Cbz-L-Ala-Bt, N-Cbz-L-Val-Bt, N-Cbz-L-Phe-Bt and N-Cbz-L-Ala-L-Val-Bt) and their antiviral activity have been studied. Starting amine, which exhibited anti-arrhythmic activity, was prepared from steroidal saponin - tigogenin using the method developed by us. To conclude, N-protected (α-aminoacyl) benzotriazoles have been utilized in the successful N-acylation of steroidal amines. Studies of antiviral activities (NIAID) of synthesized mono- and dipeptide derivatives did not reveal significant activities

Biography

Nanuli Sh Nadaraia has completed her PhD from Mendeleev Moscow Chemical-Technological Institute. She is a Lead Research Scientist at Tbilisi State Medical University. Her field of interest is chemistry and synthesis of biologically active compounds. She is the author of more than 40 papers in reputed journals and has presentations at 50 international scientific conferences

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Biosurfactant produced by *Serratia spp*. for enhanced oil recovery

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Conventional hydrocarbons recovery from a heavy oil field is less than 40%; therefore, new technologies are needed to improve the recovery factor. Bio-surfactants (BS) are amphiphilic molecules produced by microorganisms, that change the surface tension (ST), the interfacial tension (IT) and also the wettability of a rock. Recently, the interest in the use and applications of BS has increased because they have low toxicity, and capability to work in drastic conditions (high temperature, salinity and pH). The aim of this work was to evaluate the use of a BS, produced by IMP-X strain, as an enhanced oil recovery process in porous media impregnated with heavy oil from a Mexican field. IMP-X strain was isolated from a hydrocarbon-contaminated soil and identified by 16S rRNA as Serratia sp (GenBank number HQ686060). This microorganism was able to produce BS using waste cooking oil. BS reduces ST up to 28.3 mN/m, IT with heavy oil up to 2.75 mN/m, and has critical micelle concentration of 750 ppm. BS in the porous media changes the wettability from preferential oil-wet to preferential water-wet (contact angle of the system with BS 57.6°), within these changes, mobilization of the oil increase. A first oil recovery experiment was carried out in a sand packed column using Ottawa rock impregnated with heavy oil of 15.3 °API (viscosity 258 cP @ 60°C) at 60°C. Water and BS were used for injections. The results show that an additional 16.5 % of oil recovery was achieved with respect to the water injection. A second oil recovery experiment was done using oil of 15°API (viscosity 146 cP @ 70°C) and in a Bedford-limestone core at reservoir conditions (70°C and 1,706 psig). Water and BS were used for sequential injections. The results showed an additional 10.1% of oil recovery achieved with BS.

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Microsensor arrays for the measurement of acute cellular responses to stimulation

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Near real time quasi-continuous measurements of low molecular weight signalling molecules and markers of metabolism enable quantitative characterisation of acute cellular responses to stimulants such as drugs, growth factors or pathogens. In combination with well-characterised intracellular pathway blockers and conventional cell biology, the mechanisms of early stage cellular responses can be quantitatively established. Whilst longer term responses are well-characterised from gene expression studies, immunohistochemistry and Western blotting, the initial cellular responses and their dynamics remain understudied. Electrochemical methods hold out the possibility of non-destructive repeated measurements provided the sensor chemistry can still function in the complex matrix and that typical healthy behaviour of the cells can be observed in the presence of the sensors. We have produced microfabricated sensor arrays comprised six or eight working electrodes as platform technology. Choice of operating potential allows selective quantification of O¬2, H2O2, and NO and, with further modification, pH, glucose and lactate. Key to getting meaningful biomedical results is quantitative data demonstration that valid electroanalytical results are achievable in the presence of surface active cell culture components. We have developed standardised protocols for quantifying the effects of medium of mass transport and electrode kinetics and, in combination with classical cell biology these have enabled rational biodevice development. Methods for the determination of nitric oxide release, O2 concentration, O2 consumption and pH will be presented along with the results of biomedical application these in two principal areas: (1) the early stages of angiogenesis, the growth of new blood vessels, a critical process in both wound healing and metastatic disease in cancer and (2) acute responses of macrophages to anthrax protective antigen, a key early stage in the development of the disease

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Influence of growth time and concentration of silver nitrate over the biosynthesis of silver nanoparticles using white rot fungi *Bjerkandera sp.* Anamorph R1

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Statement of the Problem: Currently, silver nanoparticles (AgNPs) are being used in areas such as medicine, catalysis, optics and bactericidal sensors; for this reason, the development of recent methodologies allows a more efficient production of AgNPs with better antimicrobial and antitoxic properties. The obtention of AgNPs by physical methods tend to produce low amounts of nanoparticles, while chemical methods are often dangerous and require the use of stabilizing agents. Biological synthesis from white rot fungi are an alternative to improve these processes and reduce the generation of harmful toxic wastes. This work studies how *Bjerkandera sp*. Anamorph R1 is affected both by growth time as well concentration of silver nitrate ($AgNO_3$) over AgNPs synthesis.

Methodology & Theoretical Orientation: The synthesis of AgNPs was carried out in two ways: bio-reduction of silver ions by proteins secreted in the culture broth, as well as, by absorption of silver atom on the mycelia-pellet; to do so, the fungus was grown for 3-8 days, then the mycelium was separated from the culture broth. Both fractions were mixed with AgNO3 (0.5, 1 and 1.5 mM) evaluated at different time (24h, 48, 72h, 96h, 120h and 144h).

Findings: The action of the capping proteins on the surface of the mycelium played a determining role in the reduction of the Ag+ ion to Ag0 nanoparticles producing a particle size that oscillated between 10-100 nm.

Conclusion & Significance: The operational conditions at which the incubated fungi were maintained improved both the adsorption of the silver ions on the surface of the mycelium and subsequent synthesis of AgNPs. The best synthetic properties were found at 1mM of $AgNO_3$concentration, growth time of 8 days, and incubation time of 144 hours

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Enhancement of fatty alcohols production with modified *Escherichia coli* strains

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Microbial synthesis of fatty alcohols from renewable resources has attracted increasing attentions. We designed a novel strategy for fatty alcohol production based on fatty acid starvation. For the first time, the deletion of acyl-ACP thioesterases coupled with overexpression of exogenous fatty acyl-ACP reductase were employed to enhance fatty alcohol production. Fatty alcohol titer increased about 58% while the accumulation of fatty acids concentration decreased 73%. In order to explore the effects of acyl-ACP thioesterase deletion on the biosynthesis of fatty alcohol, we performed whole-genome transcriptional analysis. Deletions of ldhA, pta and ackA from KLCB coupled with over expression of FAR were performed and resulted in strain MGL2. The highest OD600 were increased from 5.2 to 7.8. The fatty alcohol tilter was increased from 756 mg/L to 2024 mg/L. Fed-batch fermentations with MGL2 were performed in a 3-L Bioflo 110 fermentor using defined LB medium. Total fatty alcohol accumulation reached a maximum of 6.33 g/L after 50 h. At the same time point, OD600 reached 46. Two saturated fatty alcohols (C14:0 and C16:0) and two unsaturated ones (C16:1 and C18:1) are the major components. C14:0 (2.42 g/L) and C16:1 (1.81 g/L) are the two most abundant fatty alcohols, constituting 38.2% and 28.6% of total fatty alcohols, respectively. The percentage of unsaturated fatty alcohols was up to 36.5% of the total fatty alcohols. Notably, our best strain MGL2 produced 6.33 g fatty alcohols

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Effects of different seedling-raising substrates on physiological characters and grain yield for mechanized-transplanted rice

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Statement of the Problem: With the expansion area of the mechanized-transplanted rice, rice seedling-raising substrate becomes the development tendency because of its wide material sources, low production cost and strong compatible of mechanized transplanting techniques.

Methodology & Theoretical Orientation: In this study, light soilless substrate (LSS), mixed substrate (MS, containing 35% of the soil), and nutrient soil (control) were used to determine the effects of substrate characters on physiological characteristics and grain yield for mechanized-transplanted rice.

Findings: The result showed that bulk density for LSS and MS were 85.19% and 74.07% lower than the control, while the aeration porosity, water-holding porosity and their nutrient content were significantly higher than the control. Rice seedlings qualities for LSS and MS treatment showed advantages compared with that of the control treatment. The intertwining force for rice seedlings roots and the missing mechanized-transplanted rice seedlings for LSS and MS treatment were 4.17% and 4.32% lower than the control, respectively. After 7 days of mechanized-transplanted, rice root dry matter for LSS was 10.0% and 30.8% higher than the MS and the control treatment, shoot dry weight were higher by 7.8% and 25.7% than the MS and the control treatment, respectively. With more dry matter accumulation in rice seedling stage, the seedlings from LSS treatment revived quickly and tillers started earlier after transplanted in field. Rice grain yield for LSS and MS treatment was 5.30% and 6.14% higher than the control treatment, respectively.

Conclusion & Significance: Light soilless substrate is made of crops, straw, which is easy to be decomposed completely in soil. Rice grain yield from light soilless substrate was almost the same with the mixed substrate treatment, but was significant higher than that of control. With the obvious application advantages above, light soilless substrate was better for the production and application of seedling substrate

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Molecular cloning involving AAV-CXCL 12 gene

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The American Cancer Society reports that this year there will be an estimated 600, 920 deaths due to cancer in the United States. Current cancer research includes the use of biomarkers on the surface of cancer cells to distinguish the cancerous cells from normal body cells. Molecular cloning can enhance these biomarkers. Over the past thirty years, molecular cloning has progressed immensely. From digestion to plasmid insertion, the possibilities are endless. The AAV (Adeno Associated Virus) CXCL 12(C-X-C Motif Chemokine Ligand 12) is a Protein Coding gene that shows great promise with cloning and plasmid insertion. Our project aims to use this gene to bind tightly to biomarkers on the surface of cancer cells. However, before this optimal binding can occur, it is essential to know more about the AAV CXCL 12 Gene itself. For this reason, our project includes multiple gel electrophoresis assays, plasmid insertion/digestion assays, and PCR purification. From the results of these assays, the efficacy of AAV CXCL 12 to bind to cancer biomarkers will become clear. In particular, the cloning assay for the AAV CXCL 12 gene holds great potential, as it is possible to clone extraneous DNA into a different host. If extraneous DNA can be cloned into a different host, then there is the possibility of that DNA binding to a biomarker on a cancer cell.

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Use of white biotechnology for the production of dimerized human bone morphogenetic protein 2 in Bacillus subtilis

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Bone Morphogenetic Protein 2 (BMP2) has an important clinical application in the spine fusion and ortho/maxillofacial surgeries. Here we first report the secretory expression of recombinant human BMP2 from *Bacillus substilis* system. The mature domain of BMP2 gene (Accession no. KF250425) was amplified from pTz57R/BMP2 plasmid. Two constructs were designed; one with single BMP2 gene and the other having two mature BMP2 genes coupled with glycine serine rich linker to produce a dimer. Both the constructs were cloned into the pHT43 expression vector and sequence analyzed. For secretory expression analysis and optimization, the pHT43-BMP2-M and pHT43-BMP2-D vectors were transformed into two different strains of *Bacillus subtilis* i.e. SCK6 and WB600 respectively. Expression conditions like media and temperature were optimized and the maximum 35% and 25% secretory expression of monomer (~13kDa) and dimer (~25kDa) respectively was achieved in 2xYT medium at 30°C in SCK6 strain. The expression and dimeric nature of the BMP2 was confirmed by western blot and Native PAGE analysis. For the purification of recombinant protein, 200ml culture supernatant was freeze dried to 10ml, dialyzed against the Tris-Cl (pH 8.5) and Fast Protein Liquid Chromatography, Resource Q (6ml) column was run. The recombinant human BMP2 monomer and dimer were eluted at 0.9M and 0.6M NaCl respectively. The biological activity of both the monomer and dimer BMP2 (0, 50, 100, 200 and 400 ng/ml) and commercially available positive control were analyzed by alkaline phosphatase (ALP) assay on C2C12 cells. The results showed maximum ALP activity at 200ng/ml in a dose dependent manner. In conclusion, our results showed the recombinant production of biological active dimerized BMP2 from Bacillus subtilis in culture media which was not previously reported

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Combined therapeutic medical device and stem cells for regenerative nanomedicine

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In our group, we explore a new generation of smart living implants combining not only active therapeutics but also stem cells, as a novel strategy to regenerate stabilized cartilage and avoid prosthesis, by achieving regeneration of its subchondral bone foundation, requirement which is failing today in the clinic. In our group, a unique nanotechnology strategy is used to entrap, protect, and stabilize therapeutic agents into polymer coatings: nanoreservoirs, covering nanofibres of implantable nanofibrous membranes for bone and cartilage regeneration. Upon contact with cells, therapeutic agents become available through enzymatic degradation of the nanoreservoirs. As cells grow, divide, and infiltrate deeper into the porous membrane, they trigger slow and progressive release of therapeutic agents that, in turn, stimulate further cell proliferation. The nanoreservoirs technology enables to reduce the quantities of required therapeutic agent (compared to soaked membranes for instance) thereby reducing costs. Clinical trial: phase 1 Horizon 2020, (FR, UK, SP, SW) will be submitted. Feasibility and safety assessment of a therapeutic implant based on an active polymeric wound dressing and autologous mesenchymal stem cells derived from bone marrow for the treatment of femoral cartilage isolated lesions

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Mutated and wild type Gossypium universal stress protein-2 (GUSP-2) gene confers resistance to abiotic stresses in transgenic cotton plant

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Gossypium arboreum is considered to be a rich source of stress responsive genes and EST data base revealed that mostly of its genes are uncharacterized. The full length *Gossypium universal stress protein-2 (GUSP-2)* gene (510bp) was cloned in *E.coli*, Pichia pastoris and *Gossypium hirsutum*, characterized and point mutated at three positions separately at 352-354, Lysine-60 to proline (M1-usp-2) and 214-216, aspartic acid-26 to serine *(M2-usp-2)* & 145-147, Lysine-3 to proline *(M3-usp-2)* to study its role in abiotic stress tolerance. It was found that heterologus expression of one mutant *(M1-usp-2)* provided enhanced tolerance against salt and osmotic stresses, recombinant cells have higher growth up to 10-5dilution in spot assay as compared to *W-usp-2* (wild type *GUSP-2), M2-usp-2* and *M3-usp-2* genes. *M1-usp-2* in *Pichia pastoris* transcript profiling exhibited significant expression (7.1-fold) to salt and (9.7) and osmotic stresses. *M1-usp-2* gene was also found to enhance drought tolerance and significant expression (8.7) in CIM-496-*Gossypium* hirsutum transgenic plants. However, little tolerance against heat and cold stresses both in recombinant yeast and bacterial cells was observed. The results from our study concluded that activity of *GUSP-2* was enhanced in *M1-usp-2* but wipe out in *M2-usp-2* and *M3-usp-2* response remained almost parallel to W-usp-2. Further, it was predicted through in silico analysis that *M1-usp-2*, *W-usp-2* and *M3-usp-2* may be directly involved in stress tolerance or function as signaling molecule to activate the stress adaptive mechanism

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Scale up of enzymatic hydrolysis of bovine plasma protein for producing an antioxidant

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In modern society, there is an increasing awareness of the relationship between diet and health. Reason why the use of synthetic antioxidants in the food industry is regulated, due to the side effects on consumer health, which has created the trend of consumption of products of natural origin. One of the alternatives to satisfy this demand is the search for antioxidants from natural sources, such as the production of hydrolyzed antioxidants from bovine plasma, a fraction of the blood produced in the plants. In the present study, the response surface methodology was used to minimize the time to reach a degree of hydrolysis (GH) of 20%, in the enzymatic hydrolysis of bovine plasma, taking as factors the concentration of substrate, enzyme/substrate and the answers the time required to obtain a degree of hydrolysis of 20% and the antioxidant activity. The hydrolysis was carried out in a 1L reactor controlled by a Titrando 842 automatic titrator (Metrohm, Switzerland), with Alcalasa 2.4L, pH 9.0 and 61.5 ° C. The optimum conditions obtained in the 1L reactor were scaled to 5 L, in a BioFlo 310 reactor (New Brunswick, USA). The best conditions at 5L were 79 g/L of substrate, 22% of I/O, to obtain a time of 16.1 ± 0.7 min (GH 20%) and antioxidant capacity of 59.64%.

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Effects of 1-MCP on plant growth characteristics and spikelets development under salt stress in rice

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Statement of the Problem: Rice growth and development was badly affected by its salt sensitivity. It faces osmotic, ionic as well as hormonal (ethylene) stress due to salt stress. High ethylene production effects inferior and superior spikelets development in rice cultivars under salt stress. 1-Methylecyclopropne (1-MCP) is an excellent ethylene inhibitor in plants, which has been recently explored as an important ethylene inhibitor in rice. However, effects of 1-MCP in rice growth and development under salt stress is a novel study.

Methodology & Theoretical Orientation: Two rice cultivars, Liangyoupiejiu (LYP9, Indica) and Nipponbare (NPBA, Japonica, sensitive to salt stress) were grown in green house with three salt stress levels: 0 (Control, CK), 1.5 g NaCl/kg dry soil (Low Salt Stress, LS), and 4.5g NaCl/kg dry soil (Heavy Salt Stress, HS).

Findings: The results showed that 1-MCP significantly improved photosynthesis rate (Pn), transpiration rate (Tr), stomatal conductance (Gs), and SPAD values of flag leaf in both cultivars than no 1-MCP. It was higher effective in increasing grain weight and grain filling rate of LYP9 than NPBA under all salt levels. 1-MCP significantly inhibits ethylene production in inferior spikelets in LYP9 and NPBA than superior spikelets. Application of 1-MCP positively improved rice plant dry matter and improved grain yield and yield components for LYP9 and NPBA, especially for LYP9.

Conclusion & Significance: 1-MCP favorably improved plant growth characteristics and development of superior and inferior spikelets in rice under slat stress, LYP9 showed better performance than NPBA after application of 1-MCP under salt stress

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Effect of heavy metals on antioxidant activities on the growth and yield of potato irrigated with tannery effluent

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A wide range of environmental factors including heavy metals stress stimulates oxidative stress. So, antioxidant resistance mechanisms may provide a strategy to enhance metal tolerance by minimizing the toxic effects of reactive oxygen species (ROS) In the present investigation, the effects of heavy metals producing antioxidative defense systems i.e. Superoxide dismutase, Peroxidase and Catalase were studied in the potato plants grown in soil polluted with heavy metals (Cr, Cd, Pb). The results showed that the tannery effluent was highly toxic with high values of all pollution parameters (pH, TDS, NaCl %, ECe, bicarbonates, chlorides). Amount of heavy metals was also high in the effluent and increased with increasing the concentration of effluent. Amount of chromium was the maximum as compared to other heavy metals. Four concentrations of tannery effluent (0, 2, 4, 6 and 8%) were prepared with normal garden soil along with the two treatments of ascorbic acid i.e., 0 and 0.5mM. The maximum biomass was observed in the control and it was decreased with the increasing effluent concentration. The exposure of plants to the highest concentration (8%) reduces the height of shoot. Among the treatments, 0.5mM was observed to show the best tolerance and growth properties with respect to metals. The antioxidant enzyme activities were increased by metals stress. The present investigation revealed that the ascorbic acid treatment by foliar spray on the growing plants of potato could enhance the efficiency of antioxidants that significantly reduce the toxic effect of metals from tannery effluent amended soil. The antioxidative activity seems to be of primary importance for adaptive response of potato plants against ROS produced under environmental stress

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Nerve growth factor-loaded heparinized cationic solid lipid nanoparticles for regulating membrane charge of induced pluripotent stem cells during differentiation

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Nerve growth factor (NGF)-loaded heparinized cationic solid lipid nanoparticles (NGF-loaded HCSLNs) were developed using heparin-stearic acid conjugate, cacao butter, cholesterol, stearylamine (SA), and esterquat 1 (EQ 1). The effect of cationic lipids and lipid matrix composition on the particle size, particle structure, surface molecular composition, chemical structure, electrophoretic mobility, and zeta potential of HCSLNs was investigated. The effect of HCSLNs on the membrane charge of induced pluripotent stem cells (iPSCs) was also studied. The results indicated that the average diameter of HCSLNs was 90-240 nm and the particle size of HCSLNs with EQ 1 was smaller than that with SA. The zeta potential and electrophoresis analysis showed that HCSLNs with SA had a positively charged potential and HCSLNs with EQ 1 had a negatively charged potential at pH 7.4. The high-resolution transmission electron microscope confirmed the loading of NGF on the surface of HCSLNs. Differentiation of iPSCs using NGF-loaded HCSLNs with EQ 1 exhibited higher absolute values of the electrophoretic mobility and zeta potential than differentiation using NGF-loaded HCSLNs with SA. The immunochemical staining of neuronal nuclei revealed that NGF-loaded HCSLNs can be used for differentiation of iPSCs into neurons. NGF-loaded HCSLNs with EQ 1 had higher viability of iPSCs than NGF-loaded HCSLNs with SA. NGF-loaded HCSLNs with EQ 1 may be promising formulation to regulate the membrane charge of iPSCs during neuronal differentiation.

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