

Dear Participant,

On behalf of the IPFB2014 organizing committee, I am honored to welcome you to the 8th Conference Polymer and Fiber Biotechnology taking place at Braga, Portugal.

The designations of previous events indicated Textile Biotechnology (2000, Póvoa de Varzim, Portugal, 2002; Athens, Georgia, U.S.A; 2004, Graz, Austria; 2006, Seoul, Korea and 2007, Wuxi, China) and Textile and Polymer Biotechnology, (2009, Gent, Belgium and 2011, Milano, Italy) as main topics of discussion. The focus of this year's event has progressed from textiles to fibres and polymers, as the range of applications greatly widened to include medical, pharma, cosmetics, detergent and industrial applications.

Our expectation is that the IPFB2014 will provide a forum of ample scientific discussion and promotion of cooperation between academia and industry.

Sincerely,

Artur Cavaco-Paulo

Conference Chair

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Vadim Volkov

May 24th – Reception and official opening of the conference, 18:00

May 25th - Day 1

Chair: ARTUR CAVACO-PAULO

9:00 – 9:20 *GIANLUCA CIARDELLI - Politecnico di Torino, ITALY*
Biomimetic design of nanomaterials for cancer treatment

9:20 – 9:35 *ARTUR RIBEIRO - University of Minho, PORTUGAL*
Production of extracellular L-asparaginase: from bio-prospecting to the engineering of an antileukemic biopharmaceutical

9:35 – 9:50 *ANTONIO FRANCESKO - Universitat Politècnica de Catalunya, SPAIN*
Sonochemically generated bioconjugate nano/micro-capsules as non-viral vectors for efficient nucleic acid delivery

9:50 – 10:05 *CLARISSE RIBEIRO - University of Minho, PORTUGAL*
Nano and sub-micron PLLA surface roughness effect on osteoblast and fibroblast cell response

10:05– 10:20 *DANIELA CORREIA - University of Minho, PORTUGAL*
Tailoring poly(vinylidene fluoride) microstructure and dimensionality for tissue engineering applications

10:20 – 10:50 **COFFEE BREAK**

10:50 – 11:05 *DINA M. SILVA - University of Minho, PORTUGAL*
Potential of injectable dextrin-based hydrogel for biomedical applications

11:05 – 11:20 *DORIS SCHIFFER - ACIB GmbH, AUSTRIA*
Novel (bio)polymer based detection methods for wound infection and advanced wound management

11:20 – 11:35 *MARGARIDA FERNANDES - Universitat Politècnica de Catalunya, SPAIN*
Sonochemically processed cationic nanobiopolymers - efficient antimicrobials with membrane disturbant capacity

11:35 – 11:50 *PAOLA GIARDINA - University of Naples Federico II, ITALY*
Self-assembly propensity of Vmh2 hydrophobin from *Pleurotus ostreatus*

11:50 – 12:00 *VANESSA FERREIRA - University of Minho, PORTUGAL*
Silk Fibroin Nanoparticles for Drug Delivery purposes: Stabilization, Incorporation and Release Design

12:00 – 14:00

LUNCH

Chair: ANTONIO VENÂNCIO

14:00 – 14:15 *ANDREIA GOMES* - University of Minho, PORTUGAL
Folate-based nanobiodevices for integrated diagnosis/therapy targeting chronic inflammatory Diseases

14:15 – 14:30 *ALEXANDRA ROLLETT* - BOKU, AUSTRIA
Surface functionalization of HSA nanocapsules with folic acid and antibodies for targeted drug delivery

14:30 – 14:45 *PETYA PETKOVA* - Universitat Politècnica de Catalunya, SPAIN
Sonochemical formation and enzymatic stabilisation of functional hybrid nanocomposites

14:45 – 15:00 *RICARDO N. PEREIRA* - University of Minho, PORTUGAL
Production of Whey Proteins Nanosystems by Electro-Heating for Nutraceuticals Encapsulation

15:00 – 15:15 *SELESTINA GORGIEVA* - University of Maribor, SLOVENIA
The influence of physicochemical surface properties and micro-structuring on gelatin-scaffold biocompatibility

15:15 – 15:30 *MONICA BOFFITO* - Politecnico di Torino, ITALY
Thermosensitive PEG-based hydrogels for drug release and tissue engineering

15:30 – 15:45 *GIANLUCA CIARDELLI* - Politecnico di Torino, ITALY
Biomimetic mussel adhesive protein inspired coatings for biomedical applications

15:45 – 16:15

COFFEE BREAK

16:15 – 16:30 *HÉLDER SILVA* - University of Minho, PORTUGAL
Layer-by-layer deposition of biopolymers on curcumin nanoemulsions: an edible multi-layer system

16:30 – 16:45 *NUNO G. AZOIA* - University of Minho, PORTUGAL
Study of transdermal permeation of large molecules by coarse-grained molecular dynamics simulation

16:45 – 17:00 *TZANKO TZANOV* - Universitat Politècnica de Catalunya, SPAIN
Upgrading the co-streams of fish processing industries through nanoparticles formation for skin conditioning purposes

17:00 – 17:10 *CÉLIA CRUZ* - University of Minho, PORTUGAL
Effect of a peptide in cosmetic formulations for hair humidity-control

17:10 – 17:30 *KENZO KOIKE* - KAO, JAPAN
Hair research and hair care: New hair care technology development based on the evidences of fundamental hair research; hair coloring, damage care, and aging care

May 26th - Day 2

Chair: MIGUEL GAMA

9:00 – 9:20 *JOHAN SMETS – Procter & Gamble, BELGIUM*
New laundry detergent technology for fibre modification

9:20 – 9:35 *MOJCA BOŽIČ* - University of Maribor, SLOVENIA
Biochemical modification and functionalization of nanocellulose surfaces

9:35 – 9:50 *FERNANDO DOURADO* - University of Minho, PORTUGAL
Bacterial Nano Cellulose innovative Biopolymer in Research and Application

9:50 – 10:05 *OMARA A. EL SEOUD* - University of São Paulo, BRAZIL
Cellulose derivatization under homogeneous conditions: A multi-technique study on the acetylation of the biopolymer in binary mixtures of ionic liquid and molecular solvents

10:05 – 10:20 *SANDRA CERQUEIRA BARROS*, University of Minho, PORTUGAL
Thermo-sensitive chitosan-(hydroxypropyl)methyl cellulose hydrogels: swelling, thermal and morphologic behavior

10:20 – 10:50

COFFEE BREAK

10:50 – 11:05 *KAREN DE CLERCK* - Ghent University, BELGIUM
The moisture sorption behaviour in naturally coloured cotton fibres

11:05 – 11:20 *LENKA MARTINKOVA* - INOTEX, Ltd., CZECH REPUBLIC
Biotechnologies for Processing of Natural-based Polymers: Enzyme Treatment of PLA Fibers

11:20 – 11:35 *MANFRED ZINN* - HES-SO Valais-Wallis, SWITZERLAND
Bacterial synthesis of medium-chain-length polyhydroxyalkanoate homopolymers with improved material properties

11:35 – 11:45 *JOSE E. ANTUNES* - University of Minho, PORTUGAL
Keratin Molecular Dynamics Models

11:45 – 12:00 Poster Session

12:00 – 14:00

LUNCH

Chair: GEORG GUEBITZ

14:00 – 14:15 *JINSONG SHEN* - De Montfort University, UK
Novel finishing of protease treated wool

14:15 – 14:30 *QIANG WANG* - Jiangnan University, CHINA
Keratinase application in wool bio-antifelting

14:30 – 14:45 *VANJA KOKOL* - University of Maribor, SLOVENIA
The effect of specific and selective binding of newly-structured oligo-acyl-lysyl peptides on wool fibre, its antimicrobial activity and biocompatibility

14:45 – 15:00 *RUI PEREIRA* - University of Minho, PORTUGAL
Silk fibroin based multifunctional materials

15:00 – 15:15 *ROXANA-ELENA GHITESCU* - "Gheorghe Asachi" Technical University of Iasi, ROMANIA
Polyphenols encapsulation by electrospinning to obtain substrates with biological activity.

15:15 – 15:30 *GUOCHENG DU* - Jiangnan University, CHINA
Characterization and production of Polyvinyl alcohol (PVA)-degrading enzyme: Crystal structure analysis, heterologous overexpression, and applications

15:30 – 16:00 COFFEE BREAK

16:00 – 16:20 *CHEN, J.* - Jiangnan University, CHINA
Applications of bioscouring enzymes in the green dyeing: Recent advances and future prospects

16:20 – 16:35 *VERONIKA PERZ* - ACIB GmbH, AUSTRIA
Mechanistic insights into enzymatic hydrolysis of biodegradable polyesters

16:35 – 16:50 *MIGUEL GAMA* - University of Minho, PORTUGAL
Biomass saccharification: development of strategies for enzyme recycling

16:50 – 17:05 *CINZIA PEZZELLA* - University of Naples Federico II, ITALY
High Redox-Potential Oxidative Enzymes For Industrial Applications

Conference Dinner 20:00

May 27th - Day 3

Chair: JINSONG SHEN

9:00 – 9:20 *VINCENT NIERSTRASZ - University Borås, SWEDEN*
Surface modification and functionalisation of textile materials using digital inkjet

9:20 – 9:35 *BEHARY NEMESHWAREE - Univ Lille Nord de France, FRANCE*
Bioactivation of Nonwoven PET using immobilised β -galactosidase

9:35 – 9:50 *ENRIQUE HERRERO ACERO - BOKU, AUSTRIA*
Enzymatic functionalization of synthetic polymers

9:50 – 10:05 *PRAMOD AGRAWAL - Saxion Chair Smart Functional Materials Research centre Design & Technology, HOLLAND*
Biodegradation of diverse PET materials by polyester hydrolases from *Thermobifida fusca* and *Fusarium solani*

10:05 – 10:20 *ANNE VATERRODT - Universität Duisburg-Essen, GERMANY*
Novel antifouling surface functionalizations using polymeric zwitterions via adsorption/entrapment and layer-by-layer deposition

10:20 – 10:55 COFFEE BREAK

10:55 – 11:10 *BARBARA THALLINGER - BOKU, AUSTRIA*
Cellobiose Dehydrogenase – antimicrobial functionalization of polydimethylsiloxane

11:10 – 11:25 *KRISTINA IVANOVA - Universitat Politècnica de Catalunya, SPAIN*
Enzyme-based nanoparticles to inhibit bacterial biofilm formation in urinary catheters

11:25 – 11:40 *AHARON GEDANKEN - Bar-Ilan University, ISRAEL*
Antimicrobial Textiles for A Safer Hospital

11:40 – 11:55 *STEPHANIE FOLLONIER - HES-SO Valais-Wallis, SWITZERLAND*
Sustainable production of medium-chain-length polyhydroxyalkanoates from fruit pomace and waste frying oil.

11:55 – 12:05 *IDALINA GONÇALVES - University of Minho, PORTUGAL*
Laccase/ultrasound system for cotton bleaching – an ultrasonic pilot-scale reactor

12:05 – 14:00 LUNCH

Chair: TZANKO TZANOV

14:00 – 14:15 *KLAUS OPWIS* - Deutsches Textilforschungszentrum Nord-West GmbH, GERMANY

Recent Advances in the Immobilization of Bio-Catalysts on Textile Carriers

14:15 – 14:30 *CLÁUDIA BOTELHO* - University of Minho, PORTUGAL

Enzymatic polymerization of phenolic compounds.

14:30 – 14:45 *KATRIN GREIMEL* - BOKU, AUSTRIA

Evaluation of a new enzyme based method for the removal of coatings

14:45 – 14:55 *MADALENA MARTINS* - University of Minho, PORTUGAL

Interfacial stabilization of enzymes in microemulsions

14:55 – 15:10 *CARLOS DÍAZ BLANCO* - Universitat Politècnica de Catalunya, SPAIN

Development of a lignin-based adhesive for wool floor coverings using laccase and natural phenols

15:10 – 15:25 *DIANA RIVERA* - Universitat Politècnica de Catalunya, SPAIN

Rapeseed production industry's co-streams used as a raw material to develop value-added products: bioactivities and possible applications

15:25 – 16:00 COFFEE BREAK

16:00 – 16:15 *JIAJIA FU* - Jiangnan University, CHINA

Xylanase and cellulase aided bio-processing of bamboo

16:15 – 16:30 *JAN MAREK* – Inotex, CZECH REPUBLIC

Single step enzymatic post-harvest "Bio-retting" process and customisation of the fibre moisture/bio-resin retention

16:30 – 16:45 *JÜRGEN ANDREAUS* - FURB, Universidade Regional de Blumenau, BRAZIL

Oxidoreductases for the decolorization of reactive dye bath effluents and for the removal of unfixed dyes from reactive dyed cellulosic textiles

16:45 CLOSING

ORAL PRESENTATIONS



Production of extracellular L-asparaginase: from bioprospecting to the engineering of an antileukemic biopharmaceutical.

Artur Ribeiro^{1, 2}, André Moreni Lopes³, Attilio Converti⁴, Marcos Antonio de Oliveira⁵, Cristina Maria de Souza Motta⁶, Pérola de Oliveira Magalhães⁷, Jorge Gonzalo Farías Avendaño⁸, Priscila Gava Mazzola⁹, Carlota de Oliveira Rangel-Yagui¹⁰, Artur Manuel Cavaco-Paulo¹, Adalberto Pessoa Júnior³

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¹⁰ Department of Pharmacy, School of Pharmaceutical Sciences, University of São Paulo – FCF/USP.

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Abstract

The L-asparaginase (L-asparagine amino hydrolase, E.C.3.5.1.1) catalyzes the hydrolysis of L-asparagine into L-aspartic acid and ammonia. The effective depletion of L-asparagine results in cytotoxicity for leukemic cell. Therefore the enzyme has been a clinically acceptable anti-tumour agent for the effective treatment of acute lymphoblastic leukemia (ALL) and lymphosarcoma. L-asparaginase production using microbial system had attracted considerable attention, owing to the cost effective and eco friendly nature. A wide range of microorganisms such as filamentous fungi, yeasts and bacteria have proved to be the good sources of the enzyme L-asparaginase. Thus, in this review mainly focuses on the biochemical aspects of L-asparaginase production, aiming to comprehend the physiochemical characteristics, such as stability, bioavailability, toxicity, allergenic aspects, application, and enzyme properties and kinetics of recombinant enzyme production by fermentation. Processes central to these biochemical aspects, including fermentation of L-asparaginase producing organisms and downstream processing of the enzyme are also discussed.

Sonochemically generated bioconjugate nano/micro-capsules as non-viral vectors for efficient nucleic acid delivery.

A Francesko¹, MM Fernandes¹, T Tzanov¹

¹ Group of Molecular and Industrial Biotechnology, Depart. Chem. Eng., Universitat Politècnica de Catalunya, Terrassa, Spain

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Abstract

Viral vectors provide efficient intracellular delivery of genetic materials, necessary to achieve therapeutic effects for treatment of many diseases. However, safety issues due to the physiological responses are major barriers for their use.

Alternative synthetic carriers still fail to exhibit sufficient transfection capacities due to the poor interactions with cell membranes. Yet another prerequisite for efficient gene therapy is to combine the nucleic acids protection during the extracellular transit and trigger release once internalised. One strategy to avoid the premature drain and provide delivery on-demand is to use stimuli-responsive vehicles exploiting as a triggering mechanism the redox gradient between the extra- and intracellular compartments. In this work a one-step sonochemical technology was employed as a versatile route to generate a variety of bioconjugate nano/micro-capsules and simultaneously load nucleic acids. These capsules with the shell composed of redox-sensitive disulphide-bearing macromolecules, polysaccharide/amino acid conjugates simulating viral envelopes or specific peptide sequences for cellular recognition, effectively protected nucleic acids from human serum degradation.

Moreover, tailored delivery of nucleic acids was achieved via: i) enhanced and sustained release based on intracellular glutathione levels to reduce disulphides in the capsules shell, or ii) interactions of amino acids from the shell with cell membranes to promote transfection.

Nano and sub-micron PLLA surface roughness effect on osteoblast and fibroblast cell response

Clarisse Ribeiro^{1,2}, Vitor Sencadas^{1,3}, Anabela C Areias¹, F Miguel Gama^{1,4} and Senentxu Lanceros-Méndez^{1,2}

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Abstract

Poly(L-lactide) electrospun mats with random and aligned fiber orientation and films have been produced with degrees of crystallinity ranging from 0 up to nearly 50% [1]. The overall surface roughness is practically constant irrespective of the sampling areas (1 x 1 m to 20 x 20 m) for degrees of crystallinity below 30%, increasing for higher degrees of crystallinity for the larger sampling areas. Further, due to fiber confinement, surface roughness variations are smaller in electrospun mats. Variations in contact angle, particularly large for the electrospun fibers, are thus mainly attributed to micron-scale variations (e.g. fiber arrangement) and not to submicron variations [2-3]. Samples with 50% of crystallinity show the lowest osteoblast and the highest fibroblast proliferation. Therefore, it is verified that higher roughness promotes lower osteoblast adhesion and proliferation whereas fibroblast showed higher proliferation. The overall results indicate the relevant role of the sub-microenvironment variations associated to the microscale roughness, which is suggested to play a more relevant role than the nanoscale roughness in determining the different cell responses.

Acknowledgements: This work was supported by FEDER through the COMPETE Program and by the Portuguese Foundation for Science and Technology (FCT) in the framework of the Strategic Project PEST-C/FIS/UI607/2011 and PTDC/CTM-NAN/112574/2009. V.S. and C.R. thanks the FCT for the SFRH/BPD/63148/2009 and SFRH/BPD/90870/2012 grants, respectively.

The authors also thank funding from Matepro – Optimizing Materials and Processes, ref. NORTE-07-0124-FEDER-000037, co-funded by the “Programa Operacional Regional do Norte” (ON.2 – O Novo Norte), under the “Quadro de Referência Estratégico Nacional” (QREN), through the “Fundo Europeu de Desenvolvimento Regional” (FEDER).

Tailoring poly(vinylidene fluoride) microstructure and dimensionality for tissue engineering applications

DM Correia^{1,2}, R Goncalves^{1,2}, C Ribeiro^{1,3}, V Sencadas^{1,4}, G Botelho², JL Gomez Ribelles^{5,6}, S Lanceros-Mendez^{1,3}

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Abstract

Smart biomaterials play an important role for tissue engineering (TE) applications with the aim to develop functional substitutes for damaged tissue. In particular, electroactive polymers that generate electrical charges in response to mechanical strain may

be used for TE applications. It has been shown that varying piezoelectric electrical stimulation influences cell proliferation, differentiation and regeneration [1-4].

Poly(vinylidene fluoride), PVDF, is a semi-crystalline polymer with the strongest piezoelectric properties among polymers, high mechanical strength, thermal stability and chemical resistance [3]. PVDF has four crystalline structures (α , β , γ and δ), the β phase being the one with the largest electroactivity [1, 3].

PVDF and PVDF composites can be processed in various morphologies and dimensionalities such as particles, fibers, films and scaffolds, which can play in critical role in determining biomaterial-cell interaction.

This presentation will focus on the processing techniques and strategies to obtain different PVDF micro and nanostructures: nano and microspheres, fibers, films, membranes and three dimensional scaffolds. The influence of processing conditions on piezoelectric crystalline phase and sample crystallinity will be discussed. Finally, the effect of charge surface and sample morphology on MC3T3-E1 osteoblast cells attachment, spreading and response will be evaluated under static and dynamic conditions.

Acknowledgements: Work funded by FEDER funds through "COMPETE" and by FCT, project references PTDC/CTMNAN/112574/2009; PEST-C/FIS/UI607/2011; PESTC/QUI/UIO686/2011 and Matepro: NORTE-07-0124-FEDER-000037", co-funded by QREN and FEDER. The authors thank the COST Action MP1003, MP1206 and MP1301. D.M.Correia, R.Goncalves, C.Ribeiro and V.Sencadas thanks the FCT for the SFRH/BD/ 82411/2011, SFRH/BD/88397/2012, SFRH/BPD/90870/2012 and FRH/BPD/63148/2009 grants, respectively.

References:

- [1] Martins, P.M., et al., Effect of poling state and morphology of piezoelectric poly(vinylidene fluoride) membranes for skeletal muscle tissue engineering. *Rsc Advances*, 2013. 3(39): p. 17938-17944.
- [2] Ribeiro, C., et al., Fibrinectin adsorption and cell response on electroactive poly(vinylidene fluoride) films. *Biomedical Materials*, 2012. 7(3).
- [3] Ribeiro, C., et al., Enhanced proliferation of pre-osteoblastic cells by dynamic piezoelectric stimulation. *Rsc Advances*, 2012. 2(30): p. 11504-11509.
- [4] Martins, P., A.C. Lopes, and S. Lanceros-Mendez, Electroactive phases of poly(vinylidene fluoride): Determination, processing and applications. *Progress in Polymer Science*, (0).

Potential of injectable dextrin-based hydrogel for biomedical Applications

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Abstract

Bone tissue engineering is a very challenging and promising field, which handles with the limitations of bone regenerative capacity and the failure of current orthopedic implants [1].

This work describes the preparation and characterization of an injectable dextrin-based hydrogel (oDex) through dextrin oxidation followed by cross-linking with a dihydrazide [2]. In vitro and in vivo experiments allowed to conclude that this system can carry and stabilize cells, nanogels, Bonelike[®] granules [3] and other biomolecules. This is a promising biomaterial due to its biocompatibility, and potential to promote an adequate environment for bone regeneration, which was increased by the combined bioactive molecules. Its injectability allows a minimal invasive surgical procedure with decreased patient morbidity, lower infection risk and reduced scar formation. Furthermore, an adequate sterilization protocol for this kind of material was established.

The tight collaboration between University of Minho and Bioskin S.A. company, envisioning technology transfer and product valorization, has resulted in a published international patent of the product (WO2011070529A2) [4]. Currently, the submission of a request for the authorization for the clinical trials is being planned.

Acknowledgments: D.M.S. was supported by the grant SFRH/BD/64571/2009 from Fundação para a Ciência e Tecnologia (FCT), Portugal. We thank FCT funding through EuroNanoMedENMED/0002/2010.

References:

[1] M.M. Stevens, Mater Today, 11 (2008) 18-25. [2] M. Molinos, V. Carvalho, D.M. Silva, F.M. Gama, Biomacromolecules, 13 (2012) 517-527. [3] M.A. Lopes, F.J. Monteiro, J.D. Santos, Biomaterials, 20 (1999) 2085-2090. [4] M.C.M. Molinos, F.M.P.D. Gama, in: Patent number WO2011070529A2, Portugal, 2012.

Novel (bio)polymer based detection methods for wound infection and advanced wound management

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Abstract

Wound infection is a global problem affecting 5-10% of post-surgical wounds and 25% of chronic wounds. Based on the reaction of the human immune system on infections, the enzyme activities of human neutrophil elastase (HNE) and matrix metalloproteinases (MMPs) were directly monitored in wound fluids of affected patients. Infected wounds led to significant higher substrate conversion compared to non infected wound fluids. In addition, the gelatinolytic activity was investigated to develop a biopolymer based enzyme-responsive detection method. Upon incubation of dyed gelatin beads with infected wound fluids, an incubation for 30 minutes led to a clearly visible dye release.

Advanced wound healing was achieved with natural polyphenolic compounds, known for their antioxidant, antimicrobial, anti-inflammatory and consequently wound healing promoting properties. Therefore those phenolics were immobilized on biopolymers to achieve a partly inhibition of elevated enzymes present in wounds.

Allowing integration of sensors in bandage materials enzyme substrates were immobilized on various (bio)-polymers. These substrates were converted only by infected wound fluids, allowing on-line monitoring of wounds due to different colour stages of the bandage. The combination of these rapid and simple diagnostic methods provide a powerful instrument in consideration of early stage warning of wound and additional promoting wound healing.

Sonochemically processed cationic nanobiopolymers – efficient antimicrobials with membrane disturbant capacity

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Abstract

Bacterial infections are still a major healthcare concern worldwide, due to the rapid spread of antibiotic resistant bacteria. One strategy to manage the bacterial infections while avoiding the emergence of resistance implies specific targeting and disruption of bacteria membranes. This work evaluated the potential of nanostructured biopolymers, nanobiopolymers (NBs), to disrupt the bacteria cell walls and kill planktonic microorganisms. Chitosan and cellulose were chemically modified to synthesize conjugates with improved cationic character (thiolated chitosan and aminocellulose), prior to their processing into spherical NBs via a one-step sonochemical process. Their interactions with bacteria membrane were evaluated using two membrane models: Langmuir monolayers and liposome bilayers composed of a L- α -phosphatidylglycerol phospholipid extracted from *Escherichia coli*. NBs possessed improved membrane disturbing capacity in comparison to the non-processed conjugates, in a trend that was directly proportional to the NBs cationic charge. Whereas evidences showed that thiolated chitosan and aminocellulose interacted with the bacteria membrane through a carpet model, the NBs were found to induce larger surface defects and high local perturbation through a detergent model. Importantly, the degree of disruption caused by the conjugates and NBs correlated well with the antimicrobial capacity against *Escherichia coli*, killing selectively bacteria without imparting toxicity to human fibroblasts.

Self-assembly propensity of Vmh2 hydrophobin from *Pleurotus ostreatus*

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Abstract

Hydrophobins are a family of small fungal proteins that have unusual biophysical properties. They are amphipathic, highly surface-active, and self-assembling proteins. Class I hydrophobins form an extremely robust layer composed of amyloid-like fibrillar structures. Vmh2 is a class I hydrophobin secreted by the fungus *Pleurotus ostreatus*. The protein is soluble in solvents less polar than water, i.e. ethanol, or in aqueous buffers, but its solubility largely depends on the pH of solutions. Vmh2 in ethanol forms amyloid-like structures at $\text{pH} \geq 7$, while at these pHs it is soluble in aqueous buffers where it self-assembles at acidic pHs. These findings show that the protein solubility is strongly related to its charge. Fluorescent analyses in the presence of Thioflavin T indicated that the protein aggregates are amyloid-like and their amount increases when the protein concentration increases in buffer at pH 7. The presence of aggregates has been verified by Dynamic Light Scattering. It is worth noting the peculiarity of the Vmh2 behavior since the conversion into the amyloid-like assembled form is not triggered by migration to hydrophobic/hydrophilic interfaces as in the other known class I hydrophobins. Vmh2 is able to self-assemble on solid surfaces, forming a nanometric monolayer stable from the chemical point.

Silk Fibroin Nanoparticles for Drug Delivery purposes: Stabilization, Incorporation and Release Design

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Abstract

Silk Fibroin (SF) has been extensively studied for various applications due to its impressive mechanical properties and biocompatibility. Recently, SF based-particles have been proposed as controlled drug delivery systems.

A new and efficient method was developed to prepare SF nanoparticles (SF-NPs) by high pressure homogenization (HPH) emulsification, in oil-in-water emulsions (o/w). During the NPs production by HPH emulsification process, the secondary SF structure changed from random-coil conformation to a more stable structure, β -sheets. To improve even more the NPs stability over time the effect of various surfactants was studied, namely poloxamer 407, transcucol, tween 80 and sodium dodecyl sulfate, in which SF nanoemulsions with 1% of transcucol demonstrated lower diameters and better polydispersity values during the 4 weeks of evaluation.

The drug incorporation efficiency and release of SF-NPs was assessed using orange IV dye as model-drug. The influence of a human protease (human neutrophil elastase) on orange IV release profile was also evaluated. The encapsulation of orange IV effectively stabilized the size and size distribution of the SF-NPs over time, being evident the conformational change to β -sheets. SF-NPs encapsulated with orange IV had a formation and encapsulation efficiency of 67% and 91%, respectively, with a controlled release over time.

The stability and release profile induced by the SF-NPs enhances its potential for various applications, including biomedical.

Folate-based nanobiodevices for integrated diagnosis/therapy targeting chronic inflammatory Diseases

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Abstract

It is estimated that inflammatory diseases affect more than 80 million people worldwide and these numbers are expected to increase in the next 20 years. Disorders such as rheumatoid arthritis (RA) can shorten life span by 10 years and its treatment remains a challenge for the medical and scientific community. More efficient strategies are required in order to improve clinical benefit. Nano-enabled drug delivery systems aim to improve therapy of chronic inflammatory disorders by creating a new, highly specific and efficient strategy, with reduced treatment costs.

The consortium of NANOFOL FP7 project produced FBN (liposomal, protein-based and PLA (poly (L-lactic acid)) nanoparticles) with encapsulated anti-inflammatory drugs that were shown to be biologically active, non cytotoxic and capable of specifically targeting folate receptor (FR)-positive cells, in particular activated macrophages, mediators of chronic inflammation in RA. The NANOFOL nanobiodevices targeting activated macrophages may be an interesting theranostics solution, i.e., simultaneous diagnosis and treatment of the site of inflammation in RA patients. The production of a validated, stable, specific FBN with incorporated imaging agent and therapeutic agent (drugs or siRNAs) by the NANOFOL consortium will have many applications in all inflammatory diseases but may also extend to cancer treatment.

Surface functionalization of HSA nanocapsules with folic acid and antibodies for targeted drug delivery

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Abstract

Specific targeting of malignant cells can improve efficacy of drugs and prevent damage of healthy cells and tissues. Therefore it is essential to modify the surface of drug delivery systems such as nanoparticles to introduce receptor or ligand molecules, which can be recognized by target cells. Folic acid (FA) can be recognized by folate receptor beta (FR β) which is specifically expressed by chronically activated macrophages playing a key role in rheumatoid arthritis. Also antibodies (mAb) are often exploited to achieve targeting to malignant cells.

Here we established an ultrasound-based method for the non-toxic preparation of human serum albumin (HSA) nanocapsules where active substances can be encapsulated. We followed different strategies to modify the surface of HSA-nanocapsules. On the one hand chemical cross-linkers were used to achieve a site specific covalent linkage of FA and mAbs on the particle surface. While in another approach we established a new enzymatic method to produce an antibody-protein conjugate avoiding all kind of toxic chemicals.

Surface modification was monitored by CLSM, LC-MS/MS and SDS-PAGE. Furthermore it was demonstrated that folate based nanocapsules are able to target FR β positive macrophages. ELISA and FACS were used to demonstrate the binding ability of conjugated mAb to its antigen.

Sonochemical formation and enzymatic stabilisation of functional hybrid nanocomposites

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Abstract

Development of nanocomposites with enhanced functionalities for a wide range of biomedical applications is being extensively explored using new manufacturing routes. Among them, sonochemistry appears as an attractive method for designing such materials providing the rapid reaction rates, uniform shape and high purity of the prepared composites. Biopolymers are particularly attractive for sonochemical processing into nanoscale structures due to the high charge density, in addition to their biocompatibility and nontoxicity. Further successful utilisation of biopolymeric nanosystems, as e.g. drug delivery vehicles or antimicrobial agents, depends on their stability in physiological conditions, controlled release capacity of therapeutic cargoes, or ability to prevent microbial spreading.

For example, sustained release of therapeutics upon various physiological stimuli can be provided using cross-linked biopolymer carriers loaded with active molecules. In this study, hybrid nanocomposites comprising thiolated chitosan as a carrier and polyphenolic compounds as active agents were formulated via a one-step sonochemical technology and additionally stabilised by enzymatically induced cross-linking.

The release of phenolic therapeutics upon different stimuli, such as the pH change or intracellular glutathione levels was evaluated *in vitro*. Antibacterial and anti-inflammatory capacities of these non-invasive hybrid systems were also assessed.

Production of Whey Proteins Nanosystems by Electro-Heating for Nutraceuticals Encapsulation

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Abstract

Whey proteins are an excellent material to buildup GRAS nanosystems and develop forms of administration of bioactive compounds once they are widely used in formulated foods, are relatively inexpensive and have high nutritional value. The pathways for the production of whey protein nanosystems are controlled by protein concentration, pH, ionic strength, solvent medium and particularly by heating. Denaturation and aggregation of whey proteins by conventional heating methods can be used for the production of whey protein nanohydrogels.

The objective of this study is to induce thermal aggregation of a liquid dispersion of whey protein isolate (WPI) into a three dimensional network through an innovative treatment that combines heating and moderate electrical fields for encapsulation of vitamin B2. Nano-scale phenomena of whey protein aggregation and B2 encapsulation were assessed by nano-tracking analysis, dynamic light scattering and spectrofluorimetric techniques. Results show that the combination of heat and electric treatment has the potential to interfere with aggregation of whey proteins and thus with their particle size. Accurate control of electrical parameters during heating, either by varying electric field strength and the food product's electrical conductivity, provides significant encapsulation efficiencies of B2 in whey protein particles at different nanometer-scale range.

The influence of physicochemical surface properties and microstructuring on gelatin-scaffold biocompatibility

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Abstract

In vivo cellular response starts in minutes-scale contact with scaffold surface and is mainly guided by density and conformation of adsorbed plasma proteins, being adsorbed in a seconds-scale contact and thus being primary in a function of scaffold's physicochemical surface properties. As an answer on these phenomena, cells are restructuring on sub-cellular level, thus connecting their cytoskeletons to their microenvironment, being visualized as change in their morphology. However, beside of many studies dealing with this topic, the interactions between cell's and scaffold remains poorly understood.

In the contribution, new strategy for fabrication of 3D polymeric (i.e. gelatin-based) scaffold will be presented using combined temperature-controlled freeze-thawing and in-situ cross-linking cycles, enabling to modulate processing parameters and thus scaffolds final properties. In that respect, the fibroblast cell's response by means of their seeding density and morphology, on the scaffold with varying surface-related physicochemical properties (its charge type/quantity, availability and mobility of cell-recognition sequences, plasma-proteins adsorption kinetic profile, micro-to-nano structuring parameters), will be presented and discussed.

Thermosensitive PEG-based hydrogels for drug release and tissue engineering

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Abstract

Biodegradable thermogelling copolymers have a great potential in drug release and tissue engineering. These injectable materials can be implanted in the human body with minimallyinvasive surgery or be used in pharmaceutical formulations in combination with different drugs.

Thermogelling materials offer several advantages over chemical hydrogels: they do not require photo irradiation, organic solvents or crosslinking agents, and do not release heat during polymerization, avoiding denaturation of incorporated Active Pharmaceutical Ingredients and damage surrounding cells and tissues.

In this work, various formulations consisting of proprietary polyurethane copolymers have been developed and the sustained release of therapeutic agents from these matrices has been demonstrated. For instance a series of PEG based polyurethanes were synthesized and characterized. Different compositions and media (water, PBS, DMEM) were tested and formulations showing sol-gel transition close to physiological temperature were selected for the encapsulation of drugs and cells. Paclitaxel, one of the most potent anti-neoplastic drugs, was encapsulated in the gel, showing a controlled release up to 7 days. These results are expected to open up new perspectives in the treatment of localized tumors, such as prostate or brain tumors, where long-lasting therapy may be obtained by a single injection, reducing patient's discomfort and improved outcomes.

Biomimetic mussel adhesive protein inspired coatings for biomedical applications

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Abstract

Mussels and other marine organisms secrete protein-based adhesive materials for adherence to substrates. One of the unique structural features of mussel adhesive proteins (MAPs) is the presence of L-3,4-dihydroxyphenylalanine (DOPA), an amino acid that is believed to be responsible for both adhesive and crosslinking characteristics of MAPs. DOPA self-oxidation and polymerisation reactions carried out at a slightly alkaline pH were exploited to coat polymeric scaffolds (electrospun poly(lactic acid-co-glycolic acid), PLGA fibrous matrices) and prostheses (poly(propylene), PP hernia meshes), to confer them the ability to react with proteins for improved cell compatibility, as well as tissue adherence at the implant site. In both cases, functionalization parameters, such as DOPA solution concentration, treatment temperature and me, were selected on the basis of an accurate physicochemical (FTIR-ATR, Raman, static contact angle and XPS analyses) and morphological (SEM and AFM analyses) characterisation. DOPA coating was in the form of nano-aggregates on the substrate surface.

DOPA-functionalised PP meshes were then coated with a DOPA-functionalised gelatine freeze-dried sponge to obtain a bi-layered prosthesis with improved tissue adherence after implantation. DOPA-functionalised PLGA electrospun membranes (average diameter: $1.37 \pm 0.23 \mu\text{m}$, apparent porosity: $75.6 \pm 1.9 \%$) were successfully graEed with gelatine to improve cell response.

Layer-by-layer deposition of biopolymers on curcumin nanoemulsions: an edible multi-layer system

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Abstract

The present work aimed at preparing stable curcumin nanoemulsions and multilayer nanoemulsions as potential bioactive compounds for food formulations.

Nanoemulsion-based technology offers the methodologies for encapsulate, protect and control the release, while improving solubility and bioavailability of these compounds.

Curcumin nanoemulsions and multilayer nanoemulsions were prepared using high-pressure homogenization and electrostatic layer-by-layer deposition techniques, respectively. Chitosan was used to build the first and third layers, being the second layer formed by alginate. Size stability and zeta potential studies showed that both systems were stable during 60 days, obtaining hydrodynamic diameters of 80, 110 and 140 nm for the nanoemulsion alone, second and third layer, respectively. Size stability against pH was also evaluated, being both nanosystems stable between pH values of 2 to 12.

Curcumin release studies showed that only curcumin nanoemulsions allowed release of this compound; results clearly showed that the addition of biopolymers layers (multilayer nanoemulsions) reinforced the stability of these structures, avoiding curcumin release.

This work shows that it is possible to prepare multi-layer oil-in-water nanoemulsions through LbL technique using edible biopolymers and that this technology offers the potential to significantly improve solubility and bioavailability of bioactive compounds.

Study of transdermal permeation of large molecules by coarse-grained molecular dynamics simulation

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Abstract

Skin permeation of large hydrophilic molecules remains a challenge. The barrier function of mammalian skin is mainly attributed to the stratum corneum (SC), the outer protective layer, consisted by flat dead cells filled with keratin and surrounded by lipid bilayers. Within the SC, the lipid bilayers are the major responsible for the skin impermeability to relatively large compounds (molecular weight over 500 Da).

The stratum corneum and the skin permeation phenomena can be study by molecular dynamics simulation (MD). Lipid bilayers are studied by MD for a long time, but the most studied ones are cell membranes. There are a few studies on membranes resembling the SC lipids, and even fewer focused on skin permeation. We have developed a membrane model to be identical to the SC lipid bilayers. The composition of the membrane was based in previously reported values for young-normal skin samples and in accordance with previously reported simulations: ceramide-2, lignoceric acid, cholesterol and cholesterol sulphate. This model was used to study skin permeation of large molecular aggregates. The simulations were in accordance with experimental results, and were a valuable tool to understand the mechanism responsible for the transdermal permeation of such large aggregates.

Upgrading the co-streams of fish processing industries through nanoparticles formation for skin conditioning purposes

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Abstract

The appearance and mechanical function of human skin undergo profound changes with both increasing chronological age and cumulative exposure to external factors such as ultraviolet radiation and smoking. These factors induce premature appearance of wrinkles, reduce the elasticity of skin and promote the development of inflammatory skin disorders such as chronic wounds. This process is mainly associated with collagen damage as a result of the presence of free radicals and high levels of collagenases, elastases and myeloperoxidases (MPO). In this work, skin care formulations containing protein-based bioactive extracts from salmon fish backbones co-streams were developed. After proving their ability to inhibit elastase and MPO activity, as well as their antioxidant and antimicrobial potential and biocompatibility, the extracts were converted into stable bioactivities-carrying nanoparticles (NPs) by a short single-step sonochemical treatment allowing for production of affordable nano-sized carriers of bioactivities. The behaviour of the particles at the air-water interface of Langmuir monolayers of dipalmitoylphosphatidylcholine (DPPC) - a phospholipid present in human cells membrane, and their biocompatibility in contact with fibroblasts are prerequisites for their use as skin conditioning agents.

Effect of a peptide in cosmetic formulations for hair humidity-control

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Abstract

Humidity affects hair by changing the surface texture of the fiber. In higher humidity conditions, it is verified an increase of water uptake, which causes the swelling of the hair fibers. This leads to the extension of the hair cuticles and consequent increased friction between fibers, which causes static and an increase in the volume of hair tresses. However, these changes are distinct in different types of ethnic hair, where Caucasian Brown hair evidences a higher increase in hair tresses volume. We tested the application of several climate control formulations with and without a keratin-based peptide. The hair tresses treated with the formulations containing the peptide showed reduced volume change even after several hours of high humidity conditions. Due to its chemical nature, the peptide has affinity towards the hair fiber providing long-lasting moisture resistance and allowing its application in climate control formulations.

Hair research and hair care: New hair care technology development based on the evidences of fundamental hair research; hair coloring, damage care, and aging care.

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Abstract

There are various interests in human hair to be studied for example growth shape color aging and chemical effects. On the other hand there are technologies developed for hair care products to control the hair fibers e.g volume style and color. We Kao Corporation have studied on the properties of hair fibers and developed useful new hair care technologies. Our hair research started in 1980s by measurement of more than thousands of hair fibers from Japanese women were examined on their diameters and other properties. As a result it was found that hair thickness was relating to age and the tendency was different in men and women. After that we found t-flavanone a hair growth agent which is effective for hair thickness. In another case we have studied the lipids of hair composition and distribution 18-MEA is found to be important Recently MEA can be used as a hair conditioner. The relationship of Melanin and hair color was investigated. It is found that the color of hair is determined by the amounts of Eumelanin and Pheomelanin. Recently melanin coloring was developed by using melanin precursor. As shown above the fundamental hair research is very useful for the development of new.

New laundry detergent technology for fibre modification

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Abstract

Multicycle ageing has important consequences in both the appearance and cleanability of clothing. There are many causes of fabric integrity loss, but the move to new unit dose 'pod' detergents and cooler wash temperatures can preserve the appearance of clothes, with obvious benefits to sustainability and consumer value. Recent developments in laundry fibre modification technology including care cellulases, cleaning cellulases and polymers are working by different mechanisms to help keep clothes looking new for longer, but there are plenty of opportunities for further improvement in this space.

Biochemical modification and functionalization of nanocellulose surfaces

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Abstract

Cellulose nanofibers (CNFs) have been receiving a great importance in the last decade due to their light weight, high aspect ratios as well as being renewable, sustainable and biodegradable nanomaterial. Moreover, the chemical structure of the cellulose molecule enables the creation of new functional groups or even introducing new molecules, additionally governs the CNFs properties and broadens their application. In the contribution, three recently developed bio-catalytically induced strategies for surface modification and functionalization of CNFs will be presented as substrate-specific, ecologically-friendly and not-aggressive alternative to chemical approaches. Firstly, the modification of CNFs using laccase/TEMPO systems in combination with different additionally post-treated steps will be shown to enable the formation and tuning of aldehyde vs. carboxyl functional groups. Secondly, the phosphorylation of CNFs using hexokinase-mediated reaction, showing flame-resistance, highly ion-adsorbing and hydroxyapatite-growth induced properties will be presented. Finally, the hydrophobization/acylation of CNFs with acetic anhydride as acyl donor using lipase in organic solvent performed with/without of scCO₂ will be introduced.

Acknowledgment: The research leading to these results has been funded from the EU 7FP under the grant agreement NMP4-SL-2012-280519-NanoSelect.

Bacterial Nano Cellulose innovative Biopolymer in Research and Application

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Abstract

Bacterial cellulose (BC) is a nanofibrillar polymer produced by strains such as *Gluconacetobacter xylinus*. This biopolymer has high crystallinity, ultrafine fiber network, high tensile strength in the wet state, can be shaped into 3D structures during synthesis, and biocompatibility. These properties have attracted an ever-growing attention of both academia and industry, where studies demonstrate the use of BC in a wide range of applications such as biomedical and pharmaceutical industry, acoustic and filter membranes, biotechnological devices and in the food and paper industry.

This presentation aims to review the main features of BC, with focus on its modifications and potential applications. This will be followed by an overview of the main strategies concerning the largescale production of this biopolymer.

Following this review, examples concerning the main research activities of our research group will be presented. The FUNCARB FUNctional CARBohydrates Nanobiotechnology Group, integrates the Center of Biological Engineering (CEB) of Minho University, a part of the Associated Laboratory IBB. Our research group operates in the fields of Biotechnology and Biomedical Engineering, aiming at developing new biomaterials and tools for biomedical application, based on carbohydrates. Polysaccharides currently used in our group include bacterial cellulose, dextrin, hyaluronic acid, chitosan and mannan.

Cellulose derivatization under homogeneous conditions: A multi-technique study on the acetylation of the biopolymer in binary mixtures of ionic liquid and molecular solvents

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Abstract

We used conductivity to study the kinetics of acetylation of microcrystalline cellulose, MCC, by ethanoic anhydride in the presence of the ionic liquid, IL, 3-allyl-1-methylimidazolium chloride in dipolar aprotic solvents, DAS, N,N-dimethylacetamide, DMAC, and acetonitrile, MeCN. We explain the linear dependence of the third order rate constants on [IL] by assuming that the reactant is cellulose hydrogen-bonded to the IL. This is corroborated by kinetic data of the acetylation of cyclohexylmethanol (a model for the C6-OH of the anhydroglucose unit, AGU), FTIR, and conductivity data in absence, and presence of MCC. Cellulose acetylation is faster in IL/DMAC than in IL/MeCN. This is explained based on solvatochromic data (empirical polarity and basicity) and molecular dynamics simulations. Results of the latter indicated hydrogen-bond formation between the hydroxyl groups of the AGU, (Cl-) of the IL, and the dipole of the DMAC. Under identical experimental conditions, acetylation in IL/DMAC is faster than that in LiCl/DMAC (2.7 to 8 times), due to differences in the enthalpies and entropies of activation.

Acknowledgment: We thank TWAS and CNPq for fellowship to H. Nawaz, an undergraduate fellowship to T. A. Bioni, and a productivity fellowship to O. A. El Seoud, and FAPESP for financial support.

Thermo-sensitive chitosan-(hydroxypropyl)methyl cellulose hydrogels: swelling, thermal and morphologic behavior

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Abstract

Hydrogels are high water content materials prepared by polymer crosslinking that are able to deliver active species, such as therapeutic, antibacterial, antiperspirant and moisturising agents and fragrances. In recent years¹, several polymeric hydrogel systems have been reported based on both natural and synthetic polymers. Among the natural polymers, chitosan and cellulose-derived polymers have been extensively studied, due to their thermo-reversible properties². In this study, we develop physically cross-linked hydrogel blends based in chitosan (CH) and (hydroxypropyl)methyl cellulose (HPMC). These blends were prepared by two methodologies: solvent-casting and freeze-thaw techniques. In this study, it was evaluated the swelling, thermal (LCST, DSC and TGA), structural (FTIR-ATR and X-Ray) and morphological (SEM and AFM) properties of the developed hydrogel blends. The resulting thermosensitive blend polymers were designed to be potentially applied in textile substrates as carriers/delivers of scents, antiperspirant and moisturising substances.

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Acknowledgments:

The authors gratefully acknowledge the financial support of the Chemistry Centre at Minho University (Pest-C/QUI/UI0686/2011) and the Portuguese Foundation for Science and Technology for the Post-Doc grant ascribed to Sandra Cerqueira Barros (SFRH/BPD/85399/2012).

The moisture sorption behaviour in naturally coloured cotton fibres

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Abstract

The moisture sorption behavior of developing cotton fibers is studied by dynamic vapor sorption. Mature fibers show a typical sigmoidal isotherm, IUPAC type II, describing the adsorption on macroporous and non-porous adsorbents with a typical hysteresis. This is different from the type III isotherms exhibited by elongating fibers explained by the weak adsorbate–adsorbent interactions. The maximum sorption capacity clearly decreases throughout the fiber development. This decrease is very rapid during the elongation phase of the fibers, but declines beyond 25 days post anthesis (DPA). This transition corresponds to the time point where the secondary cell wall becomes dominant over the primary cell wall, confirmed with FT-IR. The study clearly elucidates the sorption mechanism during the elongation phase of the fiber to be different from the one during the secondary cell wall synthesis. This improved understanding is applied to the moisture sorption behavior of white and naturally colored cotton fibers. Dark brown and brown fibers show a higher sorption capacity compared to beige and white fibers. The differences in sorption capacity are found to be related to the maturity and crystallinity index of the fibers. In addition the monolayer and polylayer moisture content is analyzed using the Hailwood Horrobin model.

Biotechnologies for Processing of Natural-based Polymers: Enzyme Treatment of PLA Fibers

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Abstract

PLA as a degradable biopolymer produced from polysaccharide-based natural renewable resources by starch fermentation and polymerization is a sustainable raw material for compostable bioplastics and recyclable textiles. This aliphatic polyester is produced directly by polymerization of lactic acid or via its dimer - lactide. Due to specific properties – biocompatibility, moisture transport, UV-protection, enhanced thermostability and minimum smoke generation, the PLA is used for medical applications (implants, absorbable surgical suture), clothing (PPE, sportswear), furnishing (seat covers, bedlinen) and composites (bico-fibres, filtration, geo- and agrotexiles). Different technologies for finishing of PLA textiles were studied for barrier effects achievement and added value increase (flameproof, abrasion resistance improvement, optical brightening) incl. biotechnologies. In this work the influence of enzymatic processing of PLA and PLA/Viscose 50/50 twills for clothing application on their physical and physiological parameters has been studied using two commercial esterases (TEXAZYM PES, TEXAZYM CP). Increase of hydrophilicity and breathability, and liquid transport (MMT) enhancement were observed without impact on the strength or weight of fabrics. The enzymatic processing resulted also in surface resistivity decrease like at conventional polyester fibre. Influence of the enzymatic processing on dyeing behaviour of PLA (direct dyes) and PLA/Viscose blend (disperse and reactive dyes) was also evaluated.

Bacterial synthesis of medium-chain-length polyhydroxyalkanoate homopolymers with improved material properties

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Abstract

Polyhydroxyalkanoates have been identified to be valid alternatives for petrochemical plastics. However, in most cases their physico-chemical properties need to be enhanced by means of chemical or enzymatic derivatization, blending with other polymers or supplementation with additives, to fulfill the necessary requirements for industrial or biomedical applications. Monomeric unit composition, average polymer weight, as well as content of functional groups are the main parameters influencing material characteristics of medium-chain-length polyhydroxyalkanoates (mcl-PHAs). Typically, mcl-PHAs are co-polymers with weak mechanical properties and low melting temperatures (<60°C). In this study, homopolymers of mcl-PHA were biosynthesized under tailored growth conditions by *Pseudomonas putida* KTQQ20, a mutant of *P. putida* KT2442 deprived of the *phaG* gene and six genes encoding enzymes related to β -oxidation (Liu et al. *Metabolic Engineering* (2011) 13:11-17). With the establishment of a simple batch with initial addition of co-substrates, mcl-PHAs homopolymers consisting of C8 to C12 3-hydroxyalkanoates were produced. In contrast to co-polymers, homopolymers of mcl-PHA exhibited significantly higher melting temperatures, enhanced tensile strengths and Young's moduli, as well as a higher crystallinity. In conclusion, these new polymers are significantly easier to be processed than mcl-PHA copolymers.

Keratin Molecular Dynamics Models

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Abstract

The keratin is a key element of the hair, nails and skin in vertebrates. Understand the keratin features such as its assembling in the mentioned structures, its interaction with some compounds or mechanical properties is of great interest in the fight against some diseases or in the development and optimization of cosmetic products.

Molecular dynamics modelling is the only technique able to provide information at atomic and molecular level in a dynamic way, which can greatly help in the study of these features. However there are only a few studies using molecular dynamics simulations. This is likely to the non-existence of full length crystallographic structure models of keratin. In the few works published about keratin using molecular dynamics simulations the authors had to design and build the computational keratin model, to make the simulations of interest.

This work addresses the different keratin models developed, from its physicochemical properties to the correlation of the simulations results with experimental data. One big computational model of a truncated protofibril (8 chains of keratin), which was able to predict the increase of peptide absorption by hair shaft in response to alcohol based formulations, is also discussed.

Novel finishing of protease treated wool

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Abstract

Wool fabric has natural warmth and hydrophobic character which means it is an ideal choice for use in outerwear garments. The demand for environmentally friendly wool wet processes has greatly increased, for example AOX (absorbable organic halide)-free shrink-resist finishing for machine washable wool. Proteolytic enzymes have been suggested for incorporation in wool processing for improving scouring efficiency, handle properties and imparting shrink resistance.

The current research has undertaken enzymatic modification of wool fibres using protease for improvement of fabric shrink-resistance and creation of fabric surface patterning effect. Further novel finishing using sol-gel processing was applied on wool fabrics to enhance their functional properties. Sol-gel technology has become an important tool for producing nanoparticles and nanolayer coating on various materials. Depending on the precursors used in sol-gel synthesis, hybrid polymers can be made for fibre surface modification to achieve multifunctional properties of textile fabrics. This research aims to investigate sol-gel hybrid polymer preparation and its application process on the protease treated wool fabric to achieve combined shrink-resist, hydrophobic and antibacterial properties.

Keratinase application in wool bio-antifelting

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Abstract

As the outer scales of wool consist of keratin containing high amount of cystines and there is less degradation than general protease treatment. Keratinase can degrade keratin specifically thus the bio-antifelting of wool with keratinase might be a new choice to replace the traditional chlorinated anti-shrinkage finishing process. In this paper keratinase from *Bacillus subtilis* was applied in the wool processing aiming at hydrolyzing the disulfide bonds in wool scales thoroughly. The efficacy of the removal of keratin structure from the fiber surface and the properties of the keratinase-treated wool were evaluated. The results indicated that the area shrinkage of wool fabric treated with keratinase decreased to ca 5 which had reached machine-washable requirements. The dye uptake increased significantly and the strength loss was also acceptable Allwörden phenomena and SEM of wool showed that the removal of wool scales with keratinase was remarkable which could effectively degrade and remove scales. Amino acid analysis results further demonstrated that keratinase could break cystine disulfide bonds XPS analysis demonstrated that the elemental composition of wool surface was obviously changed by keratinase treatment. The combination of cutinase and keratinase in one bath and two bath was also investigated and positive results were reached.

The effect of specific and selective binding of newly-structured oligo-acyl-lysyl peptides on wool fibre, its antimicrobial activity and biocompatibility

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Abstract

In the last decade, wool keratin is studied as potential biomaterial to be used for human clinical applications as wound dressings and implantable devices, being related to both content of proteins having cell adhesion sequences, thus be a good substrate for fibroblast and osteoblast cells, as well as be biodegradable. However, as a protein fibre, the wool easily suffers degradation, skin irritation, and/or infection due to the generation and propagation of microorganisms under certain temperature and humidity. In the contribution, the effect of functionalization of the wool fibre surface with differently and specifically antimicrobial-active oligo-acyl-lysyl (OAK) peptides, being an alternative to generally used and by Food and Drug Administration (FDA) approved ϵ -poly-L-lysine (ϵ PL), using different coupling strategies will be presented. In addition, spin-labelled EPR spectroscopy will be shown to be highly precise analytical technique in evaluation of proteins coupling efficacy and chemistry. This work shows new ways in design of antimicrobially-active surface, thus offering the potential for the development of biocompatible medical and protective materials with targeted antimicrobial properties.

Acknowledgement The research was supported by the Ministry of Higher Education, Science and Technology and 7FP Era-Net Matera-Plus program (Antimicrob Peptides grant no. 3211-10-000369).

Silk fibroin based multifunctional materials

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Abstract

Silk fibroin is a commonly available natural biopolymer produced in specialized glands of arthropods, such as silkworms or spiders, scorpions, mites, bees and flies. This biopolymer has a long history of use in textile production and also as sutures or treatment of skin wounds. Silk fibroin has been increasingly explored in other areas of biomedical science where we can find a higher morphological diversification of silk biomaterials like films, electrospun fibers, 3D porous scaffolds or nanoparticles. In recent years it has been demonstrated that fibroin is an excellent material for active components in optical devices. This new application opens the way towards the development of multifunctional optoelectronic devices, which in perspective can be made fully biocompatible and eventually bioresorbable. Moreover, fibroin can be added to other biocomponents in order to modify the biomaterial properties leading to optimized and total different functions. These improvements can go from higher cell adhesion in tissue engineering or enhanced optical transparency, smoothness or flexibility in optoelectronic devices.

The tuning and completely understanding of silk fibers physicochemical properties and interaction with other elements are of crucial importance for the improvement of already existent silk-based materials and the basis for the development of new products.

Polyphenols encapsulation by electrospinning to obtain substrates with biological activity.

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Abstract

Electrospinning allows the production of nanofibers with encapsulated active ingredients. In the present work we explore the encapsulation of active polyphenolic species in polymeric matrices by solution and emulsion electrospinning. As such, various content of different poly-phenols (gallic acid, vanillic acid, catechin and syringic acid) loaded poly(2-hydroxyethyl methacrylate) nanofibers have been successfully fabricated. The morphological characterization by SEM allowed us to monitor the dependence between the diameters of fibers and the polyphenols content. Moreover, FTIR spectroscopy confirmed that the main interaction between polymer and polyphenols consist of hydrogen bonds and this leads to an increase in the fiber diameter when increasing the concentration of the active ingredient. The polyphenols re-release in-vitro was monitored by UV spectroscopy. The antioxidant properties of free and en-capsulated polyphenols, as well as their stability in time were determined by the DPPH method. We demonstrate here that the choice of matrix polymer and the fiber morphology are essential factors for tuning the polyphenol release profile.

Characterization and production of Polyvinyl alcohol (PVA)-degrading enzyme: Crystal structure analysis, heterologous overexpression, and applications

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Abstract

Polyvinyl alcohol (PVA), with its various desirable properties including tensile strength and thermostability, has found wide applications in many industrial processes such as fabric and paper manufacturing. The production of PVA is ever-increasing due to sustained demand. At the same time large quantities of consumed PVA have been poured into water systems. Although said to be non-toxic, PVA exhibits a strong surfactant activity that may cause a serious environmental problem. On the other hand, PVA is one of the few biodegradable polymers that can be processed by microbial assimilation. Recently, as the environmental threat posed by PVA usage continues to increase, microbial degradation of PVA is receiving much attention worldwide, and new PVA-degrading species continue to be identified. We expressed *Sphingopyxis* sp. 113P3 oxidized PVA (OPA) hydrolase (OPH) in *Pichia pastoris* and had the recombinant protein characterized biochemically. We further crystallized the protein. Subsequent determination of its structure by X-ray diffraction was carried out in parallel with the highly homologous *Pseudomonas* sp. VM15C OPH expressed in *Escherichia coli*. Based on the crystal structures, several mutants were constructed and characterized. The results not only help explain the catalytic mechanism but also encourage other modifications of OPH for industrial use.

Mechanistic insights into enzymatic hydrolysis of biodegradable polyesters

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Abstract

The demand and use of synthetic polymers is constantly rising. Polymers were initially developed to exhibit high stability and durability. These material features are leading now to big environmental problems. For that reason the environmental and legislative pressure to reduce polymer and packaging waste is increasing and there is a strong demand to find and improve polyesters that are not only biodegradable but also meet the requirements of expected material properties. Ecoflex[®] is such a biodegradable polyester which shows promising material features as well as it is compostable. Nevertheless, there is considerably less known about enzymatic hydrolysis of Ecoflex. Here, enzymatic hydrolysis of Ecoflex was mechanistically studied by using a variety of synthesized oligomeric and polymeric model substrates. Moreover, the substrate specificities of two different enzymes namely Cutinase 1 from *Thermobifida cellulositica* (Thc_Cut1) and a Cutinase from

Humicola insolens (HiC) were compared. Hydrolysis of the substrates was measured over time and reaction products were quantified by means of HPLC-MS analysis. Interestingly, the two enzymes show a distinct hydrolysis mechanisms for the modelsubstrates and for Ecoflex.

Biomass saccharification: development of strategies for enzyme recycling

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Abstract

In the present work the recycling of free enzymes after prehydrolysis and simultaneous saccharification and fermentation of pretreated wheat straw under a variety of conditions was investigated. It was found that a significant amount of active cellulase and glucosidase could be recovered by recycling the free cellulases the amount of free enzymes increase with its thermostability and hydrolytic efficiency. At 50° C normally regarded as an acceptable operational temperature for saccharification processes the enzymes significantly loses its activity and this thermal deactivation was independent of initial enzyme concentration used. The degree of cellulose conversion through a series of consecutive hydrolytic/recycling rounds dropped more substantially when low concentrations of cellulases were used. The hydrolysis yield and enzyme recycling efficiency in consecutive recycling rounds can be increased by using high enzyme loadings and moderate temperatures. Furthermore the recovery of cellulases from lignin lignocellulosic hydrolysates and cellulose by alkaline wash at pH 9 and 10 has been analysed.

High Redox-Potential Oxidative Enzymes For Industrial Applications

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Abstract

Laccase is an enzyme able to oxidise a broad spectrum of substrates using oxygen as co-substrate and producing water as by-product. Interest towards exploitation of laccase as a green tool for several applications is notable increasing in the last decades. Five *Pleurotus ostreatus* isoenzymes, representing a variegated group of laccases endowed with peculiar properties, have been purified from liquid culture broth and characterized. Two of them, POXC and POXA1b deserve particular attention for their applicative potentiality.

POXC, a canonical laccase, most abundantly produced by *P. ostreatus*. It shows a high redox potential (+0.74 V vs NHE) and is very effective in dye synthesis. POXA1b, a neutral blue laccase, very stable at alkaline pH. The enzyme shows a high redox potential (+0.65 V) and can be heterologously expressed at high levels. A collection of variants with improved features has been obtained through random mutagenesis. Among them, 1H6C is interesting for its improved redox potential (+0.77 V) together with the peculiar POXA1b stability at alkaline pHs. Case studies assessing the applicability of laccases as industrial biocatalysts in several sectors are presented. Results on laccase catalyzed bio-synthesis of textile dyes and of a conductive polymer, polyaniline, will be discussed in detail.

Surface modification and functionalisation of textile materials using digital inkjet

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Abstract

Digital inkjet technology not only allows deposition of color but also allows deposition of very small quantities of functional inks containing e.g. conductive materials, functional molecules, catalysts and biocatalysts at a precise location on a fabric and if required even at the same location. This enables the construction of complex functional or smart textile materials at low costs. Inkjet technology enables more flexible production, and increased productivity with improved ecological footprint (e.g. minimization of textile waste combined with reduced consumption of energy, water and chemicals) thereby stimulating product innovations.

Digital inkjet technology often requires adequate fabric preparation to inhibit dye migration a homogeneous pretreated fabric often ink specifically for a specific printhead and a specific (pretreated) substrate. Till today there is no generic understanding of the relations between ink, substrate and printhead. Our research focusses on the development of functional inks, containing enzymes or functional proteins, binding them to the substrate, the interfacial and rheological properties of the inks, as well as on the ink-substrate interaction.

Bioactivation of Nonwoven PET using immobilised β -galactosidase

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Abstract

This study focuses on the immobilization of β -galactosidase at PET non-woven pre-activated using different plasma treatments.

The results of enzyme immobilization by inclusion in a hydrogel fixed to textile fibers, showed that the best enzyme activity was recorded for the PET treated by atmospheric air plasma treatment and cross-linked with 0.25g /l of CaCl₂. In the case of immobilization by adsorption, PET nonwoven pre-activated by N₂/O₂ plasma yielded the highest enzyme activity. By comparing the rate of conversion of ONPG to ONP for both techniques, the adsorption method seems more interesting than the inclusion method which limits substrate and product diffusion. Moreover, enzyme immobilization by inclusion offers a poor operational stability which decreases readily to 20%. Enzyme immobilized by adsorption has an operational stability which is 3 times that of free enzyme.

Enzymatic functionalization of synthetic polymers

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Abstract

Polymers have a wide range of uses because their properties can be tune by proper selection of its monomers. However in for many purposes, although the bulk properties of the material are outstanding, the surface properties are lacking of desired reactivity or properties.

Current activation processes involve the use of concentrated acids/alkali solution or plasma approaches. Harsh chemical treatments lead to massive product loss (up to 15%), tensile strength decrease, as well as cratering on the surface. Plasma treatments are very energy demanding and unable to functionalize complex geometries. Enzymes have revealed as powerful catalyst able to overcome all these limitations. Certain hydrolases like lipases, cutinases or aryl amidases are able to partially hydrolase the upper most layers of polyesters, even including the most recalcitrant ones like polyethylene terephthalate (PET) or PA 6.6. The generation of new surface groups on the one hand dramatically increases the hydrophilicity and also presents a chance to graft new molecules. In addition we have improved the polymer- enzyme interaction via rationale design of the enzyme surface and by fusion of binding modules leading to increased adsorption as measured via Quarz Crystal Microbalance, and consequently improved hydrolytic activity based on soluble released products via HPLC-MS.

Biodegradation of diverse PET materials by polyester hydrolases from *Thermobifida fusca* and *Fusarium solani*

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Abstract

The thermophilic actinomycete *Thermobifida fusca* KW3 produces a carboxylesterase TfCa different from the previously reported cutinases. This microorganism is able to depolymerize amorphous PET films, semicrystalline PET fibres and cyclic PET trimers. A comparison has been made with *Fusarium solani* cutinase in terms of PET substrate degradation. Enzyme degraded products were detected using LC-MS and RP-HPLC analysis. The surface modifications of amorphous PET films caused by the two enzymes were additionally detected by scanning electron microscopy.

Terephthalic acid (TPA) and mono(2-hydroxyethyl) terephthalate (MHET) were identified as major hydrolytic products of the bacterial carboxylesterase, while the main product released by the fungal cutinase was MHET. Additionally the degradation of cyclic PET trimers by the carboxylesterase TfCa and the cutinases TfCutI and TfCutII produced by *T. fusca* KW3, was compared. Compared to the fungal cutinase that released more products from amorphous PET films, the carboxylesterase from *T. fusca* was more efficient in hydrolyzing semicrystalline PET fibres and cyclic PET trimers. This enzyme could be useful for the modification, degradation and recycling of synthetic aromatic polyesters and oligomers.

Novel antifouling surface functionalizations using polymeric zwitterions via adsorption/entrapment and layer-by-layer deposition

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Abstract

Nonspecific adsorption of proteins biofilm formation and associated risk of infection are big complications for numerous biomedical application. The most common type in hospitals is catheter-associated urinary tract infection.

In this research the surface of silicone catheters is modified by hydrophilic zwitterionic polymers to improve antibiofouling properties. To establish an effective functionalization strategy two surface-selective methods grafting via adsorption-entrapment and layer-by-layer deposition are explored.

For adsorption-entrapment using adsorption from aqueous modifier solution and reversible swelling of the silicone well defined amphiphilic diblock copolymers poly(dimethylsiloxane)-block-poly(sulfobetaine) were prepared via atom transfer radical polymerisation (ATRP). A poly(dimethylsiloxane) macro-initiator was reacted with 2-(dimethylamino)ethyl methacrylate. The thus obtained copolymer was sulfobetainized by reaction with 1,3-propanesultone.

For layer-by-layer deposition zwitterionic segments were integrated in a cationic polyelectrolyte which was then used in combination with anionic polyelectrolytes. By varying the fraction of zwitterions in the copolymer influences onto modification efficiency and resulting surface properties were investigated.

Syntheses and effects of varied modification conditions were characterized by NMR and ATR-IR-spectroscopy, elemental analysis GPC, static water contact angle, zeta potential and bacterial adhesion studies. The results demonstrate that both methods with suited copolymers and modification conditions lead to stable antifouling coatings on silicone surfaces which can be transferred.

Cellobiose Dehydrogenase – antimicrobial functionalization of polydimethylsiloxane

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Abstract

Polydimethylsiloxane (PDMS) is a polymer widely used in biomedical devices especially urinary catheters due to its favorable biocompatibility properties. Even though PDMS lowers the risk of infections due to its low surface tension and high hydrophobicity, catheters are usually colonized by microorganisms in most patients leading to urinary tract infections. In order to decrease the risk of infection, an antimicrobial enzyme, namely cellobiose dehydrogenase (CDH) was successfully grafted onto PDMS surface. The system is based on the ability of CDH to use oxygen as electron acceptor and different oligosaccharides (cellobiose) as electron donors to produce H₂O₂. Several approaches of immobilizing CDH on PDMS surface were exploited including surface activation using oxygen plasma followed by covalent linkage of CDH as well as layer by layer coating techniques. Success of the immobilization process was monitored by analyzing the change in the functional groups on the surfaces by FTIR measurements as well as measuring the ability of grafted CDH related to produce H₂O₂.

CDH was successfully immobilized on the surface of PDMS as evidenced by H₂O₂ production in the presence of cellobiose.

The CDH modified PDMS catheters could help to prevent current problems of microbial colonization and multidrug resistant bacteria associated with catheters.

Enzyme-based nanoparticles to inhibit bacterial biofilm formation in urinary catheters

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Abstract

In hostile environments bacteria organize into a complex biofilm structure that ensures their survival. The biofilm - dwelling bacteria, embedded in self - produced extracellular polymeric matrix (EPS), are resistant to antibiotics, causing difficult to treat infections. Biofilms formed on indwelling medical devices induce serious complications and are a global health concern. An attractive way to prevent biofilm formation is based on agents that avoid resistance development. Herein, enzyme - based nanoparticles (NPs) of acylase - a quorum quenching enzyme, and amylase - an EPS - degrading enzyme, were generated via one - step sonochemical process. The processed into NPs enzymes preserved their activity, while the NPs showed important for their stability narrow - sized distribution and increased negative charge. In vitro biofilm inhibition studies against *P. aeruginosa* and *S. aureus* demonstrated improved antibiofilm activity of the NPs in comparison to the free enzymes. Their membrane disturbing capacity assessed using a liposome bilayer membrane model revealed that the NPs possess bactericidal activity, in contrast to their counterparts in solution. It is believed that these NPs induce less evolutionary pressure on bacteria and may be used as an alternative to the conventional antibiotics therapy for counteracting biofilm occurrence on urinary catheters.

Antimicrobial textiles for a safer hospital

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Abstract

Sonochemical technique was used for the deposition of metal oxide CuO ZnO Nanoparticles (NPs) on various textiles. The coated fabrics killed efficiently 8 nosocomial bacteria. The good antibacterial properties were maintained even after 65 washing cycles in Hospital washing machines 75° and 92°C. Lately an experiment in which 25 patient were dressed in cotton-coated ZnO NPs and slept on beds whose sheets and pillow covers were made of the coated fabric were examined for their bacterial infection level as compared to patient that were using regular textiles. The results of this study will be presented in my talk. Recently we have sonochemically synthesized NPs of Cu_{0.89}Zn_{0.11}O and coated them also on textiles. These NPs have killed bacteria 10,000 to 100,000 times better than ZnO or CuO. We will discuss the reason for these excellent antibacterial properties. Finally the various inorganic metal oxides NPs are killing not only sensitive bacteria but also bacteria resistant to antibiotics¹. Examples will be presented.

References

¹ Eradication of Multi-Drug Resistant Bacteria by a Novel Zn-doped CuO Nanocomposite
Aharon Gedanken et al. Small In Press DOI

Sustainable production of medium-chain-length polyhydroxyalkanoates from fruit pomace and waste frying oil.

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Abstract

Medium-chain-length polyhydroxyalkanoates (mcl-PHAs) are biopolymers synthesized by *Pseudomonas* type strains. They are very versatile as their composition and the possibility of further functionalization depend on the carbon substrate used during bacterial fermentation. Therefore they are appealing alternatives to petrol-derived polymers but their break-through has been prevented by a high production cost. Thus we investigated whether food waste from the food industry, i. e. local and non-food competing fruit pomace and waste frying oil (WFO) could replace the costly sugars and fatty acids generally used.

First, the pomaces required an enzymatic hydrolysis step to convert polysaccharides (mainly cellulose) into fermentable monosaccharides: 47, 49 and 106 g/L glucose were recovered from the pomace of apricots, cherries and grapes, respectively.

Second, the presence of growth inhibitors was found in the pomace of cherries only but their concentration was sufficiently low to allow bioprocesses. Finally, we established that mcl-PHA could be efficiently synthesized during a 2-step fermentation with *Pseudomonas resinovorans* using hydrolyzed pomace as growth substrate and then WFO as mcl-PHA precursor. In particular, the pomace of Solaris grapes proved to be really promising as 21.3 g PHA/L pomace were produced compared to 1.4 g PHA/L pomace for apricots.

Laccase/ultrasound system for cotton bleaching – an ultrasonic pilot-scale reactor

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Abstract

This work exploited the bleaching efficiency of cotton fabrics using a combined laccase-hydrogen peroxide system assisted by ultrasound. The main goal was to reach the whiteness levels obtained by conventional treatments, reducing the amount of chemical and energy consumption. Laccase promoted the oxidation of flavonoids responsible for the natural color of the fabric. In addition, ultrasound energy enhanced the mass transfer and speed-up of bleaching reactions. Laboratory experiments demonstrated that the biobleaching process allowed higher whitening levels than those obtained by standard methods. Thus, an adjustment of different operational parameters such as hydrogen peroxide concentration, temperature and incubation time was possible. As result, comparing with conventional processes, the amount of hydrogen peroxide was reduced 50% as well as the energy consumption in terms of temperature (reduction of 40 °C) and processing time (reduction of 90 minutes). Further, a pilot reactor for the explored technology was scaled-up by adapting an existing dyeing machine with piezoelectric ultrasonic devices. The developed ultrasonic pilot-scale reactor contributes for a sustainable bleaching process with reduced environmental impact as well as offers a better performance for the finishing operations.

Recent Advances in the Immobilization of Bio-Catalysts on Textile Carriers

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Abstract

Low-cost textile fabrics made of polyester (PET), polyamide (PA) or cotton are alternative carrier materials for the immobilization of enzymes. In contrast to conventional carriers, fiber materials are considerably inexpensive. The flexible construction of fabrics enables reactor constructions of any geometry and a quick removal of the catalyst without any residues after the reaction. Moreover, their open structure guarantees an optimal substrate turn-over and the active surface is easily adjustable by the fiber diameter.

We have demonstrated successfully, that fabrics with a high enzyme load, a high relative activity and good permanence against enzyme desorption can be produced with low preparative and economic expense. Here, we report various methods for the permanent fixation of enzymes on fiber forming polymers such as photochemical grafting, the use of bifunctional anchor molecules, monomeric or polymeric cross-linking agents or specific enzyme modification for direct immobilization. In addition, we compare the strategies in terms of load, catalytic activity and reusability. Finally, we discuss the widespread scientific and commercial potential of our research in the growing field of “White Biotechnology”.

Enzymatic polymerization of phenolic compounds.

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Abstract

Phenolics are natural compounds which contains hydroxyl group bonded to aromatic hydrocarbon ring. These compounds can be divided into two categories depending on the oxidation state of the central pyran ring: flavonoids and non-flavonoids. It is important to highlight the excellent properties of the phenolic compounds, such as anti-inflammatory, antimicrobial, and antioxidant activity. It has been described that the oxidation of phenolic compounds enhances its antimicrobial properties since the resulting polymers can contribute to the microorganism's toxification. The polymerization reaction can be achieved by different biocatalysts such as oxidoreductases, namely laccases and peroxidases. Laccases use molecular oxygen as a co-substrate, being therefore the perfect co-substrate. Depending on the structure of the phenolic compound, namely the number of hydroxyl groups, the enzymatic polymerization can be performed via laccase or using laccase-mediator systems (LMSs). On the first reaction, the phenolic compound is oxidized and the resulting phenoxy radicals conduct to polymers formation through recombination processes. On the second method, the phenolic compound oxidation is mediated by redox species yielding the polyphenols formation. Any of these two routes allows the polymerization of phenolic products with enhanced properties.

Evaluation of a new enzyme based method for the removal of coatings

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Abstract

Polyesters are important components of coating materials including oxidatively drying alkyd resins and waterborne oil-free polyesters. These mentioned coatings are used in different fields like road marking, house and decorative paints, but also architectural paints.

Removal of coatings required in renovation processes is quite difficult and includes either harsh chemicals or techniques that tend to harm the surface beneath. Besides this, recycling of individual components is currently almost impossible. Enzymes could catalyze the hydrolysis of coatings into their individual components under environmentally friendly conditions.

Moreover, partial surface targeted hydrolysis could introduce new reactive groups allowing easier coating/grafting of further functional layers.

We designed model substrates resembling the structure of phthalic-acid based polyester coatings and we show that those coatings can be hydrolyzed with enzymes – especially the cutinase from *Humicola insolens* was able to hydrolyse both model substrates with varying complexity. Interestingly, alkyd resins were not only hydrolysed as liquid emulsion but indeed as dried film. The hydrolysis was followed using titration studies and the release products were analysed using HPLC-MS.

It could be shown, that enzymatic hydrolysis indeed has a potential for the removal of coating materials from surfaces, their recycling or their functionalization.

Interfacial stabilization of enzymes in microemulsions

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Abstract

One of the major constrains to the use of enzymes in industrial processes is their insufficient stability under processing conditions, namely high temperatures, presence of ultrasounds, among others. Herein, we investigated the use of oil-in-water proteinaceous (BSA) microemulsions as a novel methodology for the stabilization of laccase from ascomycete *Micelliophthora thermophila*. The immobilization of laccase onto the produced microemulsions benefitted its stability under ultrasonic conditions. The half life time of immobilized laccase was 2.4-fold higher (from 23 to 56 minutes) than laccase in the free form. This technique show promising potentialities for the stabilization of enzymes used onto a variety of processes, namely textile bleaching, surface hydrolysis, among others.

Development of a lignin-based adhesive for wool floor coverings using laccase and natural phenols

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Abstract

This work reports on the development of a lignin-based adhesive for wool floor coverings by using laccase enzyme as a tool for lignin functionalization with reactive quinone structures capable to further cross-link with amino groups from wool. In order to increase the phenolic content in lignin and thus its reactivity, the plant phenolics gallic and tannic acid were enzymatically copolymerized with lignin. In addition, the amino-phenolic compound dopamine was assayed as adhesive precursor based on its ability to autopolymerize in alkaline pH, mimicking the mussel adhesive proteins. The use of laccase from *Miceliophthora termophila* resulted advantageous over commonly employed fungal laccases (of higher redox potential), because of its ability to work under alkaline conditions, thus allowing carrying out the process at a pH that favours lignin solubilisation, phenolics' polymerization and Michael addition reactions. Polyethylene glycol in the adhesive formation served as an external plasticizer to overcome the intrinsic brittleness of lignin. The strength performance of the adhesive in terms of a pile withdrawal force was comparable to that of the conventional latex-based adhesive. The possibility to enzymatically generate an entirely natural adhesive for highly-priced wool floor coverings would improve the marketable value of lignin and the recyclability of the carpets.

Rapeseed production industry's co-streams used as a raw material to develop value-added products: bioactivities and possible applications

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Abstract

Processing oil plants such as rapeseed generates large amounts of by-products that are excellent feedstock for many valuable components such as proteins, phenolics, essential amino acids, and other health promoting substances. However, these agricultural co-streams are currently under-utilized, sold off at low price or converted into low-value products. In this study, rapeseed oil production by-products were hydrolysed into small biocompatible peptides using different proteolytic enzymes, and several activities related with cosmetics and biomedical applications were evaluated. The hydrolysates showed improved anti-oxidant, anti-aging and anti-inflammatory activity when compared to the non-treated samples, mainly due to the smaller size of the peptides obtained. This work shows the potential of using rapeseed oil industry by-products to develop value-added products for skin care formulations.

Xylanase and cellulase aided bio-processing of bamboo

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Abstract

Bamboo was treated by cellulases and xylanase in sequence under the acidic and neutral conditions. Different pretreatments were employed to strengthen enzymatic hydrolysis. It was found that the addition of cellulase made the substrates more accessible to xylanase thereby benefitted the hydrolysis of bamboo hemicellulose. The steam-high-temperature treatment facilitated the release of reducing sugar by enzymes. Considering the fact that the steam-high-temperature treatment prior to cellulase processing caused more cellulose hydrolysis which should be avoided in degumming, it is suggested to be launched after cellulase performance. HPLC test confirmed the yield of xylanase hydrolysis was enhanced by the implement of cellulase pretreatment. Significantly, the bamboo powers treated by cellulase and xylanase in sequence exhibited an increase from 3.35mg/L to 48.94mg/L (by acidic process) and 9.45 mg/L to 16.55mg/L (by neutral process) in xylose yield versus non-cellulase-treated powders, respectively. XRD spectra indicated the crystallinity of bamboo increased from 46% to 53% after enzymatic processing, which might due to the hydrolysis and removal of hemicellulose and amorphous cellulose. The present work strives to gain insight into the combined action of cellulase and xylanase in bamboo processing and paves the way toward further investigation on enzymatic degumming of bamboo.

Single step enzymatic post-harvest "Bio-retting" process and customisation of the fibre moisture/bio-resin retention

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Abstract

Principles of bio-economy build on the implementation of emerging bio-processing and search for new possible materials based on natural, renewable resources. Inherently the effectiveness, complex waste-less utilization of resources and cleaner production need to be respected.

This is a good opportunity for the extensional use of bast-fibrous resources in the area of new added value materials. Existing troubles with reproducibility and yield caused by changing weather conditions, limited utilization of waste fibrous part of linseed flax stalk and modifications of inherent properties of fibres to fill requirements for new – particularly technical applications require new processing methods. One of decisive parameters - the fibre water and/or processing liquors retention plays an important role by customisation of functionality and service-life of technical textiles. Compatibility of fibre can help by optimisation of processing conditions and affinity to the treatment systems like bio-resins. An optimal scenario is to realise these improvements by modern bio-based procedures to extend the bio-economy implementation.

The over mentioned requirements can be realised as the combined treatment boosting the field retting process (called "bio-retting") and facilitating the customised liquid retention of fibres. Process can be effectively carried out by field spraying using the selected INOTEX enzymes.

Oxidoreductases for the decolorization of reactive dye bath effluents and for the removal of unfixed dyes from reactive dyed cellulosic textiles

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Abstract

Despite of relatively high dyeing bath exhaustion and fixation of reactive textile dyes on cellulosic fibers, a still significant amount of dyes is lost to the waste water, and in order to obtain textiles with good washing fastness post-washing procedures to remove unfixed dyes are necessary. The colored effluents need to be adequately treated before being discharged to surface water or reused in the textile industry. A promising green and sustainable alternative to reduce the amount of washing water and decolorize effluents is the use of dye degrading enzymes such as laccases and peroxidases. In this work the decolorization of aqueous solutions of commonly used reactive textile dyes was investigated with different oxidoreductases in order to evaluate the potential of their use in postwashing of reactive dyed cellulose textiles and in the decolorization of dyeing bath effluents for repeated reuse of the process water. Decolorization degree depended on the dye structure, the used enzyme and the specific conditions. Afterwashing of reactive dyed cellulosic fabrics was found to be effective and a potential alternative to traditional wash-off procedures. Enzymatic decolorization of dyeing bath effluents permitted repeated reuse of the process water and a significant water economy.

POSTERS



Eco-design innovative methods for fabric finishing

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Abstract

In order to reduce the environmental impacts generated by conventional fabric finishing, eco-design processes for innovative functional textile surfaces are explored and evaluated with respect to functionality and life cycle analysis (LCA). The project investigates resource efficient technologies and renewable materials. Specifically, jettability of proteins such as lysozyme and sericin using digital inkjet technology for imparting antimicrobial properties to textiles is explored.

For the development of functional jettable proteins the rheological properties are characterized. Shear viscosity data of protein solutions at different concentrations and different molecular weight at shear rates between 100 and ~ 35.000 s⁻¹ are measured, so as to investigate shear thinning behavior. The considered inkjet fluid is jetted onto synthetics using piezoelectric printhead. After deposition of the protein to the textile, the surface properties are studied using surface tension and zeta potential measurements.

Future research will focus on the functionality hence performing antibacterial tests of the textile treated with lysozyme or sericin and the amount of protein adsorbed to the fabric. Furthermore, the effect of plasma treatment on protein adsorption and antimicrobial activity will be studied.

Development of zeolites with metal cations for use in bio-polymer matrices with antimicrobial properties.

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Abstract

Zeolites are microporous aluminosilicates with the ability to allow the transfer of matter from within to the surrounding environment. This controlled release ability as well as its heat stability were used to study the possibility of using this modified material with antibacterial agents and biopolymer matrices for use in water containment rafts in order to maintain its conditions.

For this, the modification of different zeolites with different solutions (silver, copper and zinc nitrate) and pH for various ions necessary for ion exchange, and hence for encapsulation was studied. Following testing, zeolites treated with silver nitrate 0.07 M at pH = 7, were those that showed best antimicrobial properties. These zeolites were inserted into a matrix of a bio-polymer produced from sugar cane ethanol in varying proportions (1%, 5%, 10% and 15% by weight) to study both antimicrobial and mechanical behavior. After the results, it was concluded that it is necessary to find a balance between antimicrobial activity and mechanical properties, as increasing the amount of zeolite, antimicrobial capacity increases but also the tensile strength of the biomaterial.

Textiles modified with chitosan hydrogels for dermocosmetic applications

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Abstract

Hydrogels are hydrophilic three-dimensional crosslinked networks, which can adsorb and retain a large amount of water without dissolving. Hydrogels can be stimuli-sensitive and their swelling behaviour can change in response to external parameters, resulting in the release of entrapped active molecules in a controlled manner [1,2]. In this work, hydrogels were obtained by mixing a chitosan solution (1% w/w) with a genipin solution (0.05% w/w) [3], where cosmetic actives were incorporated. This loaded chitosan hydrogels were incorporated to polyamide fabrics. The functionalized fabric was characterized by different techniques: weight gain, cryo-scanning electron microscopy and thermoporosimetry, confirming the presence of hydrogel on polyamide fabric. The functional properties of hydrogels were characterized by release studies based on the dialysis bag method, and the functional properties of the coated textile were tested by a user panel. The results show that fabrics coated with hydrogel have improved their water absorption ability. Moreover, active molecules have been successfully incorporated in the hydrogels, and their release has been studied. The release rate of cosmetic molecules loaded in chitosan hydrogels was slower when comparing with the release from an aqueous solution. The fabrics functionalized with these chitosan hydrogels have a beneficial effect on the skin.

DYES4EVER, Demonstration of cyclodextrin techniques in treatment of waste water in textile industry to recover and reuse textile dyes.

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Abstract

DYES4EVER project intends to resolve one of the main pollution problems facing the textile industry the loss of unfixed dyes which colour the water. The project aims at using cyclodextrins (CDs) to recover these dyes so that they may be reused as raw material in other dyeing processes which will avoid the withdrawal of those chemicals used to clean the water of unfixed dye. This means that the water can be reused. To demonstrate the feasibility of the process a trichromy of used colorants was selected: direct and scattered reactants and its CD complexation using aqueous media in the absence and presence of increasing concentrations of CDs where reaction medium containing excess of each dye was studied. Subsequently the samples were sonicated for 1h, filtered and evaluated spectrophotometrically to 80% ethanol in the maximum absorption for each dye. The result was that all the dyes studied in CDs complex greater or lesser extent depending on the dye resulting in minimal increases from 2.6-fold solubility to maximum increases of 16.2-fold solubility. We are currently working on the study of modified CDs to increase complexing capacity and then try wastewater. This project has been funded by the LIFE program call 2012.

Dyeing with reactive dyestuffs using effluent treated by UV/H₂O₂

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Abstract

Five different dyeings were made with reuse water obtained from effluent after UV/H₂O₂ treatment. Before and after treatments, the concentration of sodium chloride, the absorbance and the amount of total organic carbon were monitored. All rates of decolorization were above 92% and the removal of total organic carbon was above 94% in all treatments. Compared with the same dyeings made with deionized water, the total deviation (ΔE^*) between the colors did not exceed 1.05. Currently, for a production of five dyeings of one kilogram each, 80 dm³ of water is consumed and an equal volume of effluent is generated. The same dyeings made by the process proposed in this study would consume the same amount of water but without effluent discharge containing high amounts of organic matter and high values of absorbance.

Discolorization of Textile Effluent: A Comparative Study between Treatment by *Pleurotus ostreatus* immobilized in support material and Homogeneous Photocatalysis UV/H₂O₂

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Abstract

This study compared two alternative treatments for textile effluent from dyeing of cotton with the dyestuff C.I. Blue 222. Biological process with *Pleurotus ostreatus* and the advanced oxidation process (H₂O₂/UV), were assessed by absorptiometry using a spectrophotometry. The results of both were promising. Biological treatment showed good rate of discoloration in a short time, staying above 50% in 48 h whereas treatment by homogeneous photocatalysis showed discoloration of 92% in 1h 45min, demonstrating that both processes can be an alternative for treatment of textile effluents with this type of dyestuff.

Determination of toxicity of textile effluent treated by electron sheaf, by physical- chemical process and by homogeneous photocatalysis process

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Abstract

The toxicity to organisms *Daphnia similis* and *Vibrio fischeri* was observed in textile effluent obtained from dyeing of cotton with the dyestuff C.I. Reactive Blue 222. Rates in raw wastewater and treated effluent samples by electron sheaf, by physical-chemical process and homogeneous photocatalysis UV/H₂O₂ process were evaluated. For *Daphnia similis*, samples irradiated with electron sheaf had no significant improvement in reduction of toxicity, these results also show relative uniformity of EC50 % which remained between 12.94 % (lower dosage = 0.5kGy) to 14.36 % (= highest dosage 20kGy). These values are close to the EC50 % of the raw wastewater (11.66%). The photocatalysis and physical-chemical treatments obtained similar results. For *Vibrio fischeri*, assay of crude sample showed higher sensitivity compared to *Daphnia similis*. The results obtained with irradiation presented similar toxicity to *Daphnia similis* and there were no significant improvement in the reduction of toxicity at different doses. Treatments for photocatalysis and physical-chemical obtained better results in the reduction of toxicity compared to gross and irradiated sample.

Study of the Influence of the Steam on the Thermofixation of Printing with Reactive Dyes on Cotton

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Abstract

This study compared two methods of printing thermofixation (with and without steam) on cotton fabric printed with dyestuff CI Reactive Blue 160, CI Reactive Red 120 and CI Reactive Yellow 84. The difference in coloristic intensity (K/S) between processes was 0.34 for yellow, 0.24 for red and 2.59 for blue. The assessment of water fastness test showed equal values for the three dyestuff studied, demonstrating that the dry thermofixation process can be an alternative to colors that are developed with these dyes, an important alternative for small industries in Brazil.

Heterologous expression and protein engineering of alkaline pectinase, an important bioscouring enzyme

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Abstract

The present work aims to construct a robust recombinant *Bacillus subtilis* to achieve secretory production of alkaline polygalacturonate lyase (PGL). First, 6 signal peptides (amyX, bpr, vpr, yvgO, wapA and nprE) were screened with a semi-rational approach and comparatively investigated their effects on the production of PGL. The signal peptide bpr directed efficient PGL secretory expression and increased PGL titer to 313.7 U mL⁻¹. By optimizing and applying strong promoter P43 and Shine_Dalgarno sequence, higher titer of 446.3 U mL⁻¹ PGL was achieved. Finally, the capacity of the recombinant *B. subtilis* WB43CB was evaluated with a fed-batch strategy in 3 L fermentor. The PGL titer reached 632.6 U mL⁻¹ with a productivity of 17.6 U mL⁻¹ h⁻¹, which was the highest secretory production of PGL by the *B. subtilis* system. The recombinant *B. subtilis* strain WB43CB constructed in the present work has great potential in production of alkaline PGL.

Structure-based molecular engineering of alkaline amylase for improved oxidative stability and catalysis efficiency

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Abstract

High oxidative stability and catalytic efficiency are required for the alkaline α -amylases to keep the enzymatic performance under the harsh conditions in detergent industries. In this work, we attempted to significantly improve both the oxidative stability and catalytic efficiency of an alkaline α -amylase from *Alkalimonas amylolytica* by engineering the five oxidation-prone methionine residues around the catalytic domain via a systematic approach. Specifically, based on the tertiary structure analysis, five methionines (Met 145, Met 214, Met 229, Met 247 and Met 317) were individually substituted with oxidation-resistant threonine, isoleucine and alanine, respectively. Among the created 15 mutants, 7 mutants M145A, M145I, M214A, M229A, M229T, M247T and M317I showed significantly enhanced oxidative stability or catalytic efficiency. These 8 positive mutants (M145A, M145I, M214A, M229A, M229T, M247T, M247L and M317I) were used to conduct the second round of combinational mutations. Among the constructed 85 mutants, the mutant M145I-214A-229T-247T-317I showed a 5.4-fold increase in oxidative stability and a 3.0-fold increase in catalytic efficiency. Interestingly, the specific activity, alkaline stability and thermal stability of this mutant were also increased. The increase of salt bridge and hydrogen bonds around the catalytic domain contributed to the significantly improved catalytic efficiency and stability.

Enzyme-catalysed modification of poly(ethersulfone) membranes

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Abstract

Poly(ethersulfone) (PES) is the thermoplastic material of choice for the manufacture of ultrafiltration and microfiltration membranes, due to its structural and chemical stability. Unfortunately, the separation performance of PES membranes often deteriorates because of membrane fouling, which is attributed to the intrinsic hydrophobic character of this material. Therefore, introduction of different polar functional groups to the PES membrane surfaces has been reported in literature by incorporation of hydrophilic polymer through blending, coating, and radiation induced-grafting. Although successful to some extent, these methods only offer random control over the resulting surface structure and may be environmentally adverse. This study presents the first successful example of enzyme-catalyzed modification of PES membranes. Various phenolic acids were coupled to the membrane at room temperature using laccase in aqueous medium, and the resulting surfaces/polymers were investigated by XPS, FT-IR, ¹H-NMR, SEM, TGA, DSC and TMA. The effect of modification on both membrane flux and repellence of proteins, polysaccharides, polyphenols and *Listeria* cells were determined. Based on our results, it is clear that it is not just hydrophilicity of the surface that influences adsorption of foulants.

Influence of the 3-hydroxyvalerate content on the hydrophilic properties of the surface of poly (3-hydroxybutyrate and poly (3-hydroxybutyrate-co-3-hydroxyvalerate) films partially hydrolyzed with cutinase TfCut2 from *Thermobifida fusca*.

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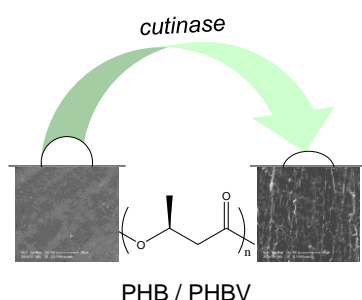
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Abstract

The treatment of the surface of poly (3-hydroxybutyrate) (PHB) and poly (3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV) films with the cutinase TfCut2 from *Thermobifida fusca* KW3 resulted in the decrease of the water contact angle from 90° to 36 ° of the PHB films and from 93°- 98° to 50°- 60° of the PHBV films with a 3-hydroxyvalerate content ranging from 6 to 9 mol %, respectively. A linear dependence up to 10 mol % was found between the 3-hydroxyvalerate content of the PHBV and the resulting water contact angle decrease caused by the partial hydrolysis of the surface of the films by the cutinase. PHBV films with a 3-hydroxyvalerate content of 6 mol % showed the highest gain in hydrophilicity of the surface following treatment with the enzyme.

KEYWORDS. Surface treatment, biopolymer, cutinase, polyhydroxyalkanoate, poly (3-hydroxybutyrate, poly (3-hydroxybutyrate-co-3-hydroxyvalerate) , hydrophilic properties.



Desizing and bleaching of cotton fabric with glucose oxidases

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Abstract

Glucose oxidases (GOD) catalyse the oxidation of β -D-glucose and simultaneously produce hydrogen peroxide (HP) which can be used for bleaching of textile fabrics. In previous research a sizing agent was decomposed by various starch degrading enzymes to gain the needed glucose for further HP production by GOD. Our experiments showed that HP was also formed only with GOD preparations in the absence of starch degrading enzymes. For this reason, the production of HP with GOD through the sizing agent and from glucose was compared. Experiments with various concentrations of enzyme, glucose and sizing agent were prepared at pH 4.6 and 50 °C. During 2 h 0,04 mol/l of HP was produced from glucose and 0.024 mol/l through the sizing agent. In the second stage, a one-bath HP production and bleaching was performed with TAED. The pH was set to 7.5 to activate the bleaching process and continued at 50 °C. The achieved WI values of bleached samples with TAED and produced HP either from glucose or through the sizing agent were very similar, around 60. Finally, experiments were also made with increasing bleaching temperature. At the optimal bleaching temperature of 60 °C the WI value of 68 was achieved.

The influence of PLLA and PDLA architectures and their end groups on stereocomplexation.

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Abstract

Recently, our research group have shown that stereocomplex of PLLA and PDLA containing end groups able to self-assembly precipitates from 1,4-dioxane solution in the form of spherical microspheres. Moreover, when the PLA stereocomplexation mixture consisted of equimolar amounts of six arm star-shaped PLLA and the linear PDLA-IL with ionic liquid end groups, spontaneous colloidal crystallization of microspheres was observed. In this work, we have expanded the combined effect of the influence of end group nature and PLA architecture on the morphology of stereocomplexes. Four different mixtures of star-shaped PLLA with linear PDLA were prepared: six arm star-shaped PLLA and the linear PDLA-IL; six arm star-shaped PLLA with COOH end groups and the linear PDLA-IL; six arm star-shaped PLLA and the linear PDLA ureidopyrimidinone (UPy) end groups; star-shaped PLLA with COOH end groups and the linear PDLA-UPy. The stereocomplexation was confirmed by the ATR-FTIR spectroscopy and DSC analysis. The morphology was investigated by the scanning electron microscopy (SEM) and atomic force microscopy (AFM).

The present work was financed by the project of National Science Centre nr: 2013/09/B/ST5/03616

Composite PLA/PP nonwoven for potential filtration applications

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Abstract

Introduction: Polypropylene (PP) nonwovens are well known high-performance filtration materials. However polypropylene is very resistant to degradation that causes problems with utilization of polypropylene materials after use. To solve this problem various modifiers can be added to polypropylene matrix, e.g. biodegradable polymers.

Materials and Methods: Composite PLA/PP nonwoven was produced from 1:1 wt. mixture of polymer granulates according to melt-blow technique. DSC analysis, physicomechanical parameters analysis and susceptibility to hydrolytic degradation were studied for obtained nonwoven.

Results and Discussion: PLA/PP composite nonwoven was produced using optimal for PLA temperature profile – for PP these temperatures were low. Hence elementary fibres diameter of PLA/PP nonwoven was higher comparing to nonwovens made from PP and PLA alone. Nevertheless filtration properties of PLA/PP nonwoven, compared to PP and PLA nonwovens, were not much worse and strength parameters were considerably better.

PLA/PP composite nonwoven was more susceptible to hydrolysis in alkaline then in neutral media. Content of PLA provides over 50% mass loss after 6 weeks of hydrolysis in alkaline media.

PLA/PP composite nonwoven is predicted for filtration applications. Improved ability to degradation allows for its better utilization.

Acknowledgment: Study financed by Polish Ministry of Science within statutory research works in 2013.

Study on melt-blown PLA and composite PLA/P(3HB) nonwovens

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Abstract

Introduction: Currently quite a lot research works focus on replacing traditional polymers by biodegradable ones to improve ecological aspects of polymeric materials. Poly(lactic acid) (PLA) is a very popular biodegradable polymer and poly(3-hydroxybutyrate) (P(3HB)) of bacterial origin is also of the increasing interest.

Materials and Methods: PLA and composite PLA (85% wt.)/P(3HB) (15% wt.) nonwovens were produced using melt-blow technique. DSC analysis, physicomechanical parameters analysis and susceptibility to hydrolytic degradation were studied for obtained nonwovens.

Results and Discussion: PLA/P(3HB) composite nonwoven was produced using optimal for PLA temperature profile – nevertheless temperatures were very high for P(3HB) with middle P(3HB) content it was possible to obtain nonwoven of good parameters. DSC analysis indicated that processing changed thermograms of both polymers. One of the reasons was thermal decomposition of polymers.

Addition of P(3HB) to PLA generally downgraded physicomechanical properties of nonwoven, especially strength parameters. However properties of PLA/P(3HB) composite nonwoven were much better than properties of nonwoven made from P(3HB) alone.

Both PLA and PLA/P(3HB) nonwovens were more susceptible to hydrolysis in alkaline media. PLA nonwoven hydrolyzed faster in alkaline media and PLA/P(3HB) nonwoven – in neutral media.

Acknowledgment: Study financed by Polish Ministry of Science within statutory research works in 2013.

Bio-active polyester textiles protecting against UV

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Abstract

Bio-active polyester textiles protecting against UV abstract Nanotechnology and textile bio-processing allow to obtain functional fabrics possessing new properties unattainable by conventional technologies In this area functional nanoparticles most often inorganic as well as polymer composites with such nanoparticles are applied These issues are the subject area of our research team studies The aim of presented studies was the development of new materials composed from nano-structural non-toxic components based on metal oxides providing antibacterial and UV barrier properties Polyester fabrics were the tested material They were subjected to bio-treatment applying esterase-based preparation The obtained effect of enhanced hydrophilic properties of polyester fibre surface was the result of hydrolysis of ester bonds and formation of reactive groups OH COOH Described research stage allowed to effectively deposit layers based on acrylic resins containing oxide hybrids particles ZnO-SiO₂ or CuO-SiO₂ micrometric size onto polyester fabric surface Fiber wettability SEM and FTIR analyses confirmed purposefulness of surface bio-modification of polyester fibres which increased the quantity of attached modifiers Final products textile-polymer composites exhibit good bacteriostatic bacteriocidal activity AATCC Test Method 100-2004 JIS L 1902 for selected gram positive and gram negative bacteria as well as good barrier properties against UV UPF 40 Studies financed.

Racemization as a Side Reaction in Organocatalytic Polymerization of L,L-lactide.

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Abstract

Usage of polylactide in industry is increasing nowadays, but some areas such as medical or electronic applications requires high purity and absence of heavy metal atoms (e.g. tin, aluminium). During a search of new metal-free catalysts, which have efficient catalytic properties both guanidine derivatives and strong amide bases have been revealed. They include 1,8-diazabicyclo[5.4.0]-undec-7-ene (DBU) or 1,5,7-Triazabicyclo[4.4.0]dec-5-ene (TBD)¹. However, literature reports have never described problems of side reactions. In our laboratory racemization side reaction, accompanying polymerization process, was observed in the presence of DBU. In polylactide chemistry racemization plays important role. Even 5% of the D-lactide derived repeating units within poly(L-lactide) chain leads to the change of material crystallization process. Crystallization degree of materials determine its' properties such as mechanical properties or (bio)degradability².

In the present work the DBU has been used as a catalytic and/or racemization agent. Poly(L-lactide) was treated by a DBU in otherwise identical conditions resembling the propagation step. The final product was characterized by SEC, MALDI-ToF, 1H and 13C NMR, and polarimetry. The results provide explanation where the racemization is located.

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² R.Séguéla, at all; *Macromolecules* 2011, 44, 4961

Use of filamentous fungi for improving electricity production and textile dye treatment in a microbial fuel cell

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Abstract

Urban and industrial wastewaters have received an increased interest towards energy harvesting using microbial fuel cells (MFC). The combined use of microbial anode oxidizing organic substrates and enzymatic cathode reducing oxygen is a promising new approach for the simultaneous treatment of wastewater and generation of electricity. In this context, our study evaluated the performance of a two-chambered MFC operated with three laccase producing strains of filamentous fungi (Ff), immobilized on the cathodic compartment and filled up with simulated textile dye effluent (TDE) and urban wastewater in the anodic compartment. The result indicated a rapid TDE decolourisation (>86 % within 72 h). Electrochemical monitoring of the MFC during TDE decolourisation indicated power density (>35 mW m², control 3,61) and laccase activity (989.6 U l⁻¹) in the presence of *Pleurotus ostreatus* on the cathodic compartment. Considering the initial COD value of 464 ± 20 mg.l⁻¹, the organic removal in the anodic compartment after 20 days of MFC operation was 90.2%. Final toxicity measurements in the TDE treated indicated a much lower impact when compared to the original TDE. These are the initial studies to select Ff as models for MFC application and further adaptation for wastewater treatment and bioelectricity generation.

Nerve Tissue Engineering Using Blends of Polyhydroxyalkanoates

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Abstract

PHAs are a family of linear polyesters synthesised by a variety of bacterial species. They can be produced from renewable carbon sources; they are easily degradable; biocompatible and exhibit thermoplastic and elastomeric properties. PHAs are potential materials to be used in the manufacturing of nerve guidance conduits to assist axonal regeneration. Properties such as: controllable surface erosion; variability in material properties, lower acidity of by-products after degradation and longer stability compared to their synthetic counterparts are of special interest in this field. The main objective of this work is to develop an advanced nerve conduit made from novel biopolymers that have not been used in nerve tissue engineering. A key objective is the identification of an ideal PHA or PHA blend with the desired material properties and degradation rate for optimal nerve regeneration. PHA films and their blends were characterised with respect to their mechanical and thermal properties. Biocompatibility studies were carried out using neuronal cells which were grown on the polymer films and growth evaluated by immunolabeling and confocal microscopy. The PHA blends with the most suitable properties for nerve tissue engineering will be used for the manufacturing of innovative designs of nerve guidance conduits in the future.

Antibacterial electrospun poly(vinyl alcohol)/enzymatic synthesized poly(catechol) nanofibrous membranes

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Abstract

An enzymatic synthesized poly(catechol) (PC) nanofibrous antibacterial membrane-containing poly(vinyl alcohol) (PVA) was produced by electrospinning using a microfiltration poly(vinylidene fluoride) (PVDF) as basal membrane, for potential applications in water ultrafiltration. Nanofibres were obtained from solutions of 20 % (w/w) PVA dissolved in a PC solution obtained from a 50 mM of catechol with a 5 mL/L of *Trametes versicolor* laccase reacted overnight at 50 °C in acetate buffer at pH5. The membranes were further cross-linked by glutaraldehyde (GA) to maintain its morphology. Blended nanofibres have shown a smooth morphology, no relevant beads formation and diameters between 50 and 120 nm. PC electrospun membranes were characterized by complete dynamical and mechanical (DMA), thermogravimetry (TGA) and differential scanning calorimetry (DSC) analysis showing relevant conformational changes in the PVA side groups attributed to hydrogen bond assemblies and high thermic stability and high residual mass. The antimicrobial activity results show almost a total growth inhibition in *S. aureus* (92%) and a slight inhibition in *E.coli* (13%) in the poly(catechol) containing membranes. The ultrafiltration test in a dead-end high-pressure (10 bar) cell using a reactive dye have shown a maximum rejection of 85% after 5 cycles with have an average flux rate of 70 L/m²*h.

Surface functionalization through a fungal hydrophobin for proteomic applications

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Abstract

Hydrophobins are a large family of proteins produced by fungi, able to self-assemble into amphipilic membranes. The hydrophobin Vmh2 from *Pleurotus ostreatus* forms stable nanometric bio-layer on a wide range of surfaces, changing their wettability and enabling these surfaces to bind other proteins or enzymes. This innovative biological functionalization is a feasible strategy for the fabrication of new classes of hybrid devices, for biosensing and proteomic research. Vmh2 hydrophobin layers have been tested for coating MALDI sample plates with the aim to develop a lab-on-plate platform focused on proteomic applications. Immobilization of peptides, as well as intact proteins, has been verified by using MALDI-TOF spectrometer. The stable interaction between Vmh2 layer and peptides/proteins has been exploited performing on-plate washing obtaining good quality MALDI spectra of complex samples without pre-treatment. Moreover the ability of Vmh2 layer to retain enzymes in their active form has been also verified. In these experiments a protease (trypsin) has been adsorbed on Vmh2 layer, and its ability to hydrolyze intact proteins has been evaluated. Results showed that the immobilized enzyme was active and able to perform the complete hydrolysis of substrate quickly than free enzyme.

New systems for polyhydroxyalkanoates production from related carbon sources: valorisation of waste materials.

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Abstract

Polyhydroxyalkanoates (PHAs) are microbial polyesters produced as intracellular energy-reserve granules by a large variety of microorganisms. Thanks to their biodegradability and biocompatibility, PHA are considered a “green” alternative for conventional petroleum-based plastics, finding applications in various fields. The interest towards PHA is also encouraged by the prospect to adjust their properties by modifying their monomeric composition. In particular, the concentration of medium-length chain (mcl) precursors in PHA copolymer has been shown to play an important role, affecting several properties, e.g. melting temperature, crystallinity etc. Application of PHA based materials is however hampered by the costs related to the C-source required for microbial growth. The use of waste materials as low-cost C-sources meets a twofold purpose: reducing PHA production costs and facilitating waste disposal.

In this work, the performances of a newly isolated class IV PHA biosynthetic operon from *Bacillus cereus* in directing biosynthesis of PHA copolymers, have been tested through its heterologous expression in *E. coli*. Lipid-containing waste materials have been chosen as related C-sources in order to drive the biosynthetic pathway towards the incorporation of mcl-precursors. Genetic engineering of the operon allowed to give some hints on the role played by each biosynthetic gene in determining polymer composition.

Magnetolectric electrospun fibers for biomedical applications

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Abstract

Magnetolectric (ME) materials are scientifically and technologically interesting due to their large potential on advanced applications. ME materials have the property of varying the electrical polarization with an applied magnetic field and/or of inducing variations in the magnetization with an applied electrical field. Body tissues including bone, skeletal muscle and neurons, among others, are subject to electrical and/or electromechanical solicitations during their functional activity. In this sense, novel strategies based on electroactive materials are being design to developed suitable active scaffolds. Piezoelectric films and fiber mats have been successfully used for osteoblast, fibroblast and myoblast cell cultures [1-3]. On the other hand, the related magnetolectric effect has not been explored despite its obvious advantages related to the magnetic activation of the electromechanical response. This work reports on the development of multiferroic CoFe₂O₄/poly(vinylidene fluoride) fibers mats for tissue engineering applications. Electrospun fiber mats have been prepared under different electrospinning conditions with increasing magnetostrictive CoFe₂O₄ nanoparticle loading and fully physical-chemical characterized. The polymer electroactive phase content is larger than 80% for all samples leading to a suitable magnetolectric response, which increases with filler concentration. Further, the cytotoxicity of the fiber mats was assessed together with their suitability for tissue engineering applications.

Acknowledgements: This work was supported by FEDER through the COMPETE Program and by the Portuguese Foundation for Science and Technology (FCT) in the framework of the Strategic Project PEST-C/FIS/UI607/2011 and the project PTDC/CTM-NAN/112574/2009. The authors also thank funding from Matepro –Optimizing Materials and Processes”, ref. NORTE-07-0124-FEDER-000037”, co-funded by the “Programa Operacional Regional do Norte” (ON.2 – O Novo Norte), under the “Quadro de Referência Estratégico Nacional” (QREN), through the “Fundo Europeu de Desenvolvimento Regional” (FEDER). R. Gonçalves, D. M. Correia, C. Ribeiro, P. Martins and V. Sencadas acknowledge support from FCT scholarships SFRH/BD/88397/2012, SFRH/BD/82411/2011, SFRH/BPD/90870/2012, SFRH/BPD/96227/2013, SFRH/BPD/6495 8/2009.

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Nanoemulsion-based photoprotective and antioxidant systems

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Abstract

An increase in public awareness about the harmful effects caused by UV radiation on the skin has resulted in an increased interest in sunscreen products. In the present work, nanoemulsions O/W containing a blend of sunscreens that confer broad-spectrum protection and antioxidant extracts were developed. Tween[®] 80 and Span[®] 80, non-ionic surfactants, were used in the formulations conferring at least four month stability to the nanoemulsions, that were prepared using an ultrasonic processor and the stability evaluation of the formulas were performed using Zetasizer Nano ZS. Mansur's method (1986) and the Labsphere[®] essays were performed for the determination of SPF (solar protection factor) and the results obtained were quite different for both methods, SPF 13.5 and SPF 25, respectively. These results could be explained due to the similarity that Labsphere[®] plates have to the skin surface while Mansur's method requires a solubilization of the formula before analysis. The photoprotective formulations were irradiated using a solar simulator and the formulations containing pomegranate extract presented 4 h stability while formulations without the extract presented 2 h stability (One-Way ANOVA, $p < 0.05$). The developed formulations with sunscreens and pomegranate extract showed a great potential to be applied as sunscreens formulations with antioxidant activity.

***In vitro* and *in vivo* biological analysis of cellulose-based scaffolds for bone tissue engineering**

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Abstract

Polymer based three-dimensional scaffolds are rapidly growing materials for bone regeneration procedures due to their surface chemistry, controllable biodegradability and mechanical strength. The polymers are usually modified in order to manipulate on their biodegradability *in vivo* or reinforced with bioactive materials to enhance osteoconductive properties.

The aim of this study was to evaluate biological properties *in vitro* and *in vivo* of cellulose-based scaffolds for bone tissue engineering. Porous cellulose/hydroxyapatite and carboxymethylated cellulose/hydroxyapatite scaffolds were fabricated by freeze-drying method. The rabbit mesenchymal stem cells were used to assess their cytotoxicity. Cells displayed a good ability to interact with the different tested scaffolds. Highly porous scaffolds were implanted subcutaneously in the back of mice. Animals were sacrificed after 14, 28, 56, 84 days of implantation. After macroscopic inspection cellulose/HA scaffold revealed its biological properties while carboxymethylated one showed immune response. It was concluded that unmodified cellulose/HA composite material is a better choice for bone tissue engineering applications.

Cell adhesion and proliferation of skeletal muscle cells on piezoelectric poly(vinylidene fluoride) membranes

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Abstract

Several body tissues, including bone and muscle, are subjected to electromechanical solicitations during their functional activity [1-3]. Thus, the use of electroactive polymers as active scaffolds shows innovative large potential for tissue engineering applications as it offers functional resemblance to biological clues [2]. In particular, piezoelectric polymers have shown suitability for tissue engineering due to their ability to vary surface charge when a mechanical load is applied [4] and their possibility to be processed in form of films, porous 2D and 3D membranes and scaffolds and fiber mats. The influence of poling state and morphology (film or fiber morphology) of piezoelectric poly(vinylidene fluoride) (PVDF) on the adhesion and morphology of myoblast cells was studied. Non-poled, “poled +” and “poled-” β -PVDF films were prepared by solvent casting followed by corona poling. Further, random and aligned electrospun β -PVDF fiber mats were also prepared. It is demonstrated that negatively charged surfaces improve cell adhesion and proliferation and that the directional growth of the myoblast cells can be achieved by culturing the cell on aligned fibers. Therefore, the potential application of electroactive materials for muscle regeneration is demonstrated.

Characterization of k-carrageenan/Locust bean gum-based films with b-carotene emulsion

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Abstract

New bio-based materials have been exploited to develop biodegradable and edible films as an effort to extend shelf life and improve quality of food while reducing packaging waste. The objective of this study was to investigate physicochemical properties of k-carrageenan/locust bean gum (k-car/LBG) films with different b-carotene emulsion concentrations. To prepare oil-in-water emulsions, b-carotene (0.03% v/v) was dissolved in medium-chain triglycerides (MCTs), and the solution was mixed (1:9 v/v) with a pectin solution (3% w/v) as emulsifier. Film forming solutions were prepared by adding b-carotene emulsion (0-3% w/w) into the k-car/LBG solution (40/60% w/w) with 0.3% (w/v) of glycerol. Films with different b-carotene concentrations were characterized in terms of optical, mechanical and barrier properties and compared with control films without b-carotene.

The results suggested that mechanical, physical and barrier properties of k-car/LBG films were influenced by the presence of b-carotene. Results showed that addition of b-carotene to the k-car/LBG films studied resulted in significant decrease ($p < 0.05$) in water vapour transmission rate values. Film opacity values (ranging from 4.9 to 12.5 %) increased when b-carotene was incorporated to the film.

Therefore, b-carotene emulsions have potential to be used as a natural additive on k-car/LBG films, particularly in the food packaging industry.

Immobilization of Laccases and its application in the production of medium-density fiberboards (MDF)

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Abstract

Immobilization on a solid surface is a convenient method to reuse enzymes; furthermore it is possible to enhance its activity and stability (Zimmermann et al., 2011). Enzyme immobilization is already applied in food and biomedicine (Sarioglu et al., 2001; Shimiri et al., 1983)

In our present DFG (Deutsche Forschungsgemeinschaft) project, different Laccase-Mediator-Systems (LMS) with immobilized laccases will be used for the production of medium-density fiberboards (MDF). A LMS activates the lignin on the wood fiber surface, which leads to a polymerization of the activated lignin molecules. Conventional binders for MDF-production are usually containing formaldehyde, which was declared as carcinogenic (IARC, 2004) and its application is consequently discussed. Euring et al. (2011, 2013) showed that it is possible to produce MDF with a LMS, which fulfills most of the required European standards for wood-based panels. As a new innovation the application of immobilized laccase in the MDF-production is supposed to be more efficient and economic. Different immobilization methods were tested, including entrapment in alginate-gelatin mixed gel, covalent binding on chitosan and covalent binding on Sepabeads EC-EP3.

Apart from the immobilization of laccases, the project also includes the optimization of the LMS by using new mediators, which is presented by A. Kirsch.

Optimization of Laccase-Mediator-Systems (LMS) for the production of medium-density fiberboards (MDF)

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Abstract

Prices of petrochemical resins will increase very probably (FAZ, 2008). For this reason the wood-panel industry has considered environmental friendly alternatives such as enzymatic binder systems for MDF-production. One possibility is to use laccase which is able to activate the surface lignin of wood fibers. Euring et al. (2011, 2013) produced MDF with Laccase-Mediator-System (LMS) which fulfilled mostly required European standards for wood-based panels. Mediators additionally work as redox-molecules between laccase and the wood fibers surface lignin. Consequently using LMS for the production of MDF seems to be a suitable alternative.

Our present DFG (Deutsche Forschungsgemeinschaft) project focuses on the optimization of LMS by using new mediators and immobilized laccases for MDF-production. The project is divided into two sub-objectives. This abstract refers to the part "Optimization of Laccase-Mediator-Systems for the production of medium-density fiberboards". Therefore oxygen consumption rates of the LMS were determined to find potential mediators. At the same time MDF were produced in pilot-plant scale and the physical-mechanical properties of the MDF were tested. The other part "Immobilization of laccases and its application in the production of medium-density fiberboards (MDF)" is presented by B. Fredrich.

Spectroscopic on-Line monitoring and stopped-flow kinetic analysis of dye degradation by laccase/mediator systems

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Abstract

The laccase catalyzed transformation of the acid dye Indigo Carmine (CI Acid Blue 74) was studied using various redox mediators: violuric acid (VIO), 2,2,6,6-tetramethyl-1-piperidinyloxy (TEMPO), 1-hydroxybenzotriazole (HOBT), and 2,2-azinobis-(3-ethylbenzothiazoline-6-disulfonic acid diammonium salt (ABTS). Inline UV/Vis and IR spectroscopy was employed to monitor the decolorization in real-time during batch decolorization. ABTS was the most effective mediator followed by TEMPO. Stopped flow kinetics was employed to study the initial phase of dye degradation in more detail. While the batch decolorization experiments suggested zero-order rate laws for dye transformation at an early stage, the more accurate stopped-flow kinetic experiments revealed that the rate laws for the initial phase were actually more complicated. Different pH optima for dye decolorization were found for the laccase catalyzed reaction (pH 3.5) and for the oxidation brought about by the isolated ABTS radical cation (pH 6.7).

Detoxification of polyphenolic rich polymers with laccase and cellobiose dehydrogenase systems

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Abstract

The combination of a laccase-hydroxybenzotriazole HBT mediator system with/without cellobiose dehydrogenase CDH or an additional Fenton reaction step for the elimination and/or detoxification of phenolic compounds in dry olive mill residues DOR and liquid olive mill wastewaters OMW was evaluated. The laccase-HBT-CDH and laccase-HBT-CDH-Fenton system were the most effective removing at least 69 and 72 of phenolic compounds from a total of 698 and 683 mg in OMW and DOR respectively in 12h. The efficient removal of phenolic compounds was also accompanied by 80 reduction in biochemical oxygen demand BOD and chemical oxygen demand COD in both DOR and OMW. Microbial community analysis using Single-Strand Conformation Polymorphism SSCP gels showed that biogas reactors supplemented with untreated and laccase-HBT-CDH-Fenton treated DOR and OMW strongly inhibited growth of microorganisms. In contrast the laccase-HBT and laccase-HBT-CDH pretreated OMW and DOR were detoxified as evidenced by SSCP analysis which also indicated a distinct sensitivity of the individual members of the anaerobic population towards the toxicants. Further although the laccase-HBT-CDH-Fenton system was effective in bleaching and removing phenolic compounds in both OMW and DOR it was not able to support methane production. However laccase-HBT and laccase-HBT-CDH indeed supported biogas production.

An investigation into enzyme processing technology to generate textile surface patterning

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Abstract

Enzyme processing in the production of textiles has increased rapidly in recent years. The use of enzymes offers an alternative to traditional wet textile processing methods. The use of enzyme technologies in many industrial textile wet processing methods is proving to be advantageous due to the elimination of adverse effects on the environment caused by harsh chemical treatments commonly used in the textile sector.

The research presented explores undeveloped opportunities to use enzyme processing as a creative tool. The study reports on an investigation into the use of protease to develop a novel technique of creating decorative surface patterning on a wool/polyester blend fabric. Optimum processing parameters were determined to achieve coloured effects on the fabric. Information gained from the analysis of controlled studies identified opportunities for exploitation during the creative phase. This stimulated new ideas for design generation. Resist methods inspired by traditional resist techniques provided a language through which designs could be communicated. These methods entailed using the physical properties of the fabric combined with compression to restrict enzyme accessibility to selected areas of the fabric to generate surface design.

Investigations concluded that localised enzyme treatments enabled selective removal of the wool fibre component from the blend producing effective colour contrast. It demonstrated that controlled enzymatic processing could effectively remove wool fibres from the blend to impart successful surface patterning, similar to discharge type surface effects.

CTAB-assisted Hydrothermal Synthesis Of Zinc Phthalocyanine Nanospheres and its Use as a Mimic Enzymic Photocatalyst for Dye Degradation under Visible-Light

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Abstract

The phthalocyanines metal complexes are one of the most promising mimic enzymic photocatalyst for wastewater treatment. Here we report the fabrication of a well dispersed zinc phthalocyanine (ZnPc) nanospheres with high photocatalytic ability by a facile one-step hydrothermal process aided by the surfactant CTAB. The obtained products were characterized by FESEM, TEM, XRD, FTIR, N₂ adsorption-desorption and UV-vis spectrum analysis. It is shown that the introduction of CTAB was crucial for the formation of small sized ZnPc and the enhancement of its photocatalytic activity. Without CTAB, the morphology of ZnPc is with a shape of microrods mixed with microballs. Instead, the morphology of CTAB-assisted ZnPc turns out to be a nanosphere with a diameter of about 100 nm of fine surface wrinkle edge structure. The result of the visible-light photocatalytic activity has shown the degradation of 100% Rhodamine B (RhB) dye under CTAB-assisted ZnPc. However, the degradation of RhB dyes under ZnPc without forming by CTAB was 42%, approximately. The possible formation mechanism for ZnPc nanospheres was also proposed based on the evolution of morphology as a function of reaction time, which turn out to be a micelles self-assembly growing process. The mechanism of mimic enzymic photocatalysis was also studied.

Enzymatic phosphorylation of silk fibroins: a platform for the production of biocompatible, cell-static, materials

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Abstract

Silks are natural protein polymers produced by insects¹. Silk heavy chain of *B.mori* is primarily composed of hydrophobic, $-(\text{Ala-Gly})_n-$ β -sheet crystalline domains³. Based on silk biocompatibility, biodegradability and strength, different materials were developed^{4, 5}. Silk offers a stabilizing environment for incorporated proteins and molecules⁶. Silk properties can be controlled via structure manipulation^{8,9}, by coupling molecules^{11,12} of biological significance; its Tyr and Ser residues can be modified^{13,14}. Once incorporated into a protein, the phosphate group establishes hydrogen bonds that affect intra- and inter-molecular interactions¹⁶. Phosphorylation is stable under physiological conditions¹⁷, thus directing the formation and reorganization of protein networks. Curiously, using phosphorylation for protein functionalization is largely unexplored¹⁴. Significant research is devoted to bio-inspired materials with various cell-differentiating²⁰ and cell-supporting^{21,22} features. However, little attention is paid to develop cell-static bio-materials. Such materials do not promote cell growth. That can be achieved by lowering the probability of cell attachment to the material, via creation of negatively charged material surface²³.

The goal of this study was to produce bio-compatible materials with the cell-static properties by phosphorylation. Silk solutions were made to cast films of variable pH and phosphorylated content. Obtained materials were tested and a dependency between amount of phosphorylation and bio-chemical properties confirmed.

Potential of human γ D-crystallin for hair damage repair: insights into the mechanical properties and biocompatibility

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Abstract

There is great interest in the development of new hair care products that facilitate repair and prevent adverse effects on the capillary structure. The objective of this work was to use the human eye γ D-crystallin for the development of a new strategy to physically 'repair' chemically damaged hair based on the ability of proteins belonging to the superfamily characterized structurally by the Greek key motif to be involved in the coating of specific structures.

The wild type and the mutant crystallins were able to restore and improve the mechanical properties of overbleached hair to values higher than the ones displayed by virgin, unaltered hair. Both proteins deposit and strongly bind to the damaged hair, and were capable to penetrate into the fibre cortex. Moreover, none of the crystallins displayed a toxic effect in fibroblasts for all the range of tested concentrations upon 72 h of exposure. The active aggregation process of mutant crystalline induced an inflammatory response in fibroblasts in the first 24 h of contact. In contrast contact with wild type crystallin did not lead to significant inflammation.

The γ D-crystallin proved to be an excellent candidate for a new restorative hair care product, opening new perspectives for other proteins belonging to the superfamily characterized structurally by the Greek key motif to be used in hair cosmetics.

Preparation and characterization of protein nanospheres prepared by high pressure homogenization

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Abstract

Protein nanospheres have a huge potential as drug carriers due to their proven biocompatibility and biodegradability. Nanospheres can be used for controlled drug release, thus reducing dosage and frequency of drug administration, and therefore of treatment associated toxicity. In this study, nanospheres of bovine serum albumin (BSA) were produced by subjecting a biphasic system, consisting of an aqueous protein solution and an organic solvent, to high pressure homogenization. The effect of BSA concentration and ratio of protein solution/oil was studied in order to determine how these factors affect particle properties. Size, polydispersity and stability of the nanospheres were analyzed with a dynamic light scattering instrument and their morphology was examined by microscopy. As these nanospheres are intended for intravenous injection, size is a key factor. The obtained nanospheres presented adequate small sizes and polydispersity, and demonstrated stability over time. Hence, these proteinaceous particles exhibit characteristics compatible with a potential application as drug delivery systems.

Characterization of peptides as a membrane anchor in liposomes for the incorporation of ligands for specific targeting

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Abstract

Liposomes have received considerable scrutiny as possible vehicles for drug delivery due to properties such as sustained release, increased drug stability, ability to overcome drug resistance and targeting of specific tissues. Phospholipids and cholesterol have been previously characterized as membrane anchors for the incorporation of ligand targeting into liposomal vesicles. We report a new peptide as membrane anchor into liposomes, derived from the pulmonary surfactant-associated protein D (SP-D). Pulmonary surfactant, a lipoprotein complex, was originally described for its essential role in reducing surface tension at the air–liquid interface of the lung. The knowledge that mammalian pulmonary surfactant proteins can, in nature, promote self-assembly of phospholipids towards almost zero potential interfaces, lead to the assumption that fragments or models representing those proteins could recognize and interact with liposomal phospholipids. Detailed characterization of interaction of the new peptide with lipid bilayer of liposomes was performed, in order to evaluate their application as membrane anchor for the incorporation of ligand targeting into liposomal vesicles.

Delivery of cytarabine by pegylated liposomes for efficient, long-term anticancer effects

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Abstract

The cytosine arabinoside cytarabine is an effective marine-derived antineoplastic agent for the treatment of acute myelogenous and lymphocytic leukemias. As this nucleoside antimetabolite is an S-phase-specific drug, prolonged exposure of cells to toxic concentrations is critical to achieve maximum biological effect. The activity of cytarabine is nevertheless decreased by its rapid deamination to the biologically inactive metabolite uracil arabinoside. This rapid degradation process is the reason for the ongoing search for efficient formulations and derivatives of cytarabine that cannot be deaminated and exhibited better pharmacokinetic parameters. In the present study, pegylated liposomes were modified for intended prolonged delivery of cytarabine and tested for improved cytotoxic and cytostatic effect in different human cancer lines.

Time-dependent effect of tamoxifen on melanogenesis in normal human melanocytes

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Abstract

In medical literature, occasional case reports describe gray hair re-pigmentation in patients after administration of certain drugs, such as tamoxifen, supporting the possibility of reversing pigmentation loss associated with ageing. This work aimed to study, *in vitro*, the effect on melanin production in primary human melanocytes of tamoxifen, an antagonist of the estrogen receptor in breast tissue, and of its most bioactive derivative, 4-hydroxy-tamoxifen.

Adult normal human epidermal melanocytes (NHEM) were exposed to physiological concentrations of tamoxifen and 4-hydroxy-tamoxifen for 72 hours.

The results showed that tamoxifen and 4HO-tamoxifen treatments promoted melanin extrusion. The transcript levels of genes coding for premelanosome protein and melan-A, directly related to skin and hair pigmentation, showed an increased tendency upon tamoxifen and 4-hydroxy-tamoxifen treatment. Induction of catalase gene expression in NHEM points towards a promelanogenic effect mediated by reactive oxygen species.

According to the results, these compounds seem to act as melanogenesis stimulators at a molecular level. Our data suggests that SERMs might be a new tool for increasing melanogenesis and might be of great interest for topical formulations in cosmetic industry.

Development of therapeutic and cosmetic formulations based on sardine-based products

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Abstract

Sardine is one of the most common fish of the Portuguese coast and has important nutritional features. Sardine oil is also a source of nutrients with proven benefits for human health, being rich in polyunsaturated fatty acids (PUFAs) [1]. Several studies show that there is a direct link between a diet enriched in omega-3 and the prevention of diseases such as cardiovascular disease, inflammatory conditions such as rheumatoid arthritis or asthma, mental disorders and prevention of various types of cancer [2].

The aim of this work was to characterize in a systematic way the potential protective role of sardine oil and derived PUFAs. To evaluate the antioxidant and anti-inflammatory effect of sardine oil and PUFAs, human fibroblasts (BJ-5ta), human melanocytes (A375) and human keratinocytes (NCTC2544) were used. Cell viability was affected for concentrations higher than 8mg/ml for sardine oil and higher than 0.1mg/ml for PUFAs. However and regarding PUFAs, melanocytes revealed a higher susceptibility. With the lowest tested concentrations, sardine-based compounds promoted cell proliferation and protected cells from induced oxidative stress, with higher protection conferred by PUFAs. These results open the opportunity to develop new therapeutic and cosmetic applications based on sardine-derived compounds. Their incorporation in topical creams may contribute to a better treatment of inflammation and in the prevention of skin aging.

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Development and characterization of PLA nanoparticles as carriers for topical delivery

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Abstract

Nanoparticles are seen today as one of the best approaches for the delivery of drugs into the skin. Poly (Lactic Acid) (PLA) is biocompatible and biodegradable and already approved for clinical use. Thus, this work aimed to study the effect of several parameters on the properties of PLA nanoparticles (PLA-NPs) intended for topical delivery. The yield of nanoparticles formation and entrapment efficiencies of lipophilic and hydrophilic model compounds in PLA-NPs were assessed. We evaluated the effects of mechanical stirring, solvent composition and presence of tri-bloc polymers on the protocol for the production of PLA-NPs. The best protocol provided a monodispersed population of non-cytotoxic spherical particles of ≈ 150 nm and a yield of nanoparticles formation of $\approx 90\%$. This formulation also proved to be efficient in the encapsulation of lipophilic and hydrophilic model compounds ($>80\%$). The best protocol for the production of PLA-NPs includes a nanoprecipitation step, which is easily up scalable.