ISSN:2335-8653

15th International Scientific Conference

THE VITAL NATURE SIGN

May 20th-21st, 2021

Kaunas, Lithuania

ABSTRACT BOOK

ISSN:2335-8653
ONLINE ISSN: 2335-8718

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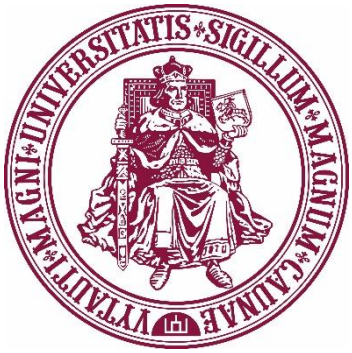

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

15th International Scientific Conference "The Vital Nature Sign" Program

Thursday, 20th of May 2021

9.00-9.15 (time zone GMT+3) **Opening of the conference** *Prof. Habil. Dr. Audrius Maruška (chairman of the conference)*

Oral presentations. *Chairpersons:* *Prof. Ona Ragažinskienė; Prof. Audrius Maruška*

9.15-9.30 **New Functional Aspects of Enzyme-Assisted Extraction of Plant Material**
K. Rafińska, A. Krakowska-Sieprawska, J. Walczak-Skierska, A. Kiełbasa, P. Pomastowski, B. Buszewski

9.30-9.45 **Ecotoxicological and Genotoxic Assessment in *Lemna minor* L. and *Spirodela polyrhiza* L. Plants Exposed to Dimethyl Phthalate (DMP)**
Fabrizio Pietrini, Valentina Iannilli, Laura Passatore, Serena Carloni, Giulia Sciacca, Massimo Zacchini

9.45-10.00 **Combined Enzyme Assisted - Supercritical Fluid Extraction with Flash Chromatography**
Aneta Krakowska-Sieprawska, Katarzyna Rafińska, Madalena Ligor, Justyna Walczak-Skierska, Bogusław Buszewski

10.00-10.15 **Developing Mass Spectrometry Based Single Cell Proteomics**
Ákos Végvári

10.15-10.30 **Baltic Sea Macro-Algae Characteristics Including Microbial Profile and Trace Elements Content**
Ernesta Tolpeznikaite, Modestas Ruzauskas, Renata Pilkaityte, Vadims Bartkevics, Paulina Zavistanaviciute, Vytaute Starkute, Vita Lele, Egle Zokaityte, Erika Mozuriene, Romas Ruibys, Dovile Klupsaite, Elena Bartkiene

10.30-10.50 ***Coffee break***

10.50-11.05 **ZnO Tetrapod Synthesis and Application in Electrochemistry: Effect of Morphology**
Mindaugas Ilickas, Agnė Šulčiūtė, Simas Račkauskas

11.05-11.20 **Statistical Analysis of Characteristics of Thermoluminescent Dosimeters**
Lijana Lileikyte, Benas Gabrielis Urbonavicius

11.20-11.35 **Investigation of Dose Sensitivity in Irradiated Polymer Gels Containing Water Co-solvents**
Ignas Pikas, Mantvydas Merkis, Diana Adlienė

11.35-11.50 **Biological Activity of Trivalent Chromium**
Sylwia Terpilowska

11.50-12.05 **Integration of Ultrasound Treatment into Valorization of Flaxseed Press Cake to Produce Functionalized Hydrogels**
K. Trakšelytė-Rupšienė, J. Arūnaite, G. Juodeikienė, D. Žadeikė, V. Jakštas, J. Bernatonienė, L. Ivanauskas, E. Bartkienė, V. Lėlė, P. Viškelis

12.05-12.35 ***Lunch break***

Poster session I. *Chairpersons:* *Dr. Mantas Stankevičius, MSc. Kristina Bimbiraitė-Survilienė*

12.35 **Endophytic *Bacillus cereus* Group Bacteria Isolates Promote *Nicotiana tabacum* L. Shoot Growth *in Vitro***
Elena Andriūnaitė, Inga Tamošiūnė, Monika Aleksandravičiūtė, Danas Baniulis

12.42 **Recovery of Total Phenolic Content of the Flower Extract of *Opuntia ficus***
Fatiha Brahmi, Redha Sellami, Sabrina Mehdi, Hayette Haddadi-Ghemghar, Khodir Madani, Lila Boulekbache

12.50 **Evaluation of Antibacterial Activity and pH Change in Herbal Drinks Fermented by Kombucha Culture**
Milda Bulotaitė, Vilma Kaškonienė, Rūta Mickienė, Audrius Maruška

12.57 (time zone GMT+3) **Anthocyanin Profiles in Fruits of Lithuanian Heirloom Apple Cultivars**
Aurita Butkevičiūtė, Mindaugas Liaudanskas, Darius Kviklys, Dalia Gelvonauskienė, Valdimaras Janulis

13.05 **Enhancement of Transungual Delivery of Amorolfine *ex vivo***
Moonsun Choi, Indrė Šveikauskaitė-Radučienė

13.12 **Correlations Between the Different Chocolate Samples Overall Acceptability and Emotions Induced by Chocolate Samples Taste, Smell and Package**
Mantvydas Dapšas, Aiste Valionyte, Gabija Sutkute, Rusne Janulyte, Greta Kotinskaite, Simonas Trunce, Karolina Siriakovaite, Alma Baltrusaityte, Juste Alisauskaite, Aura Kaminskaite, Konstancija Vaiginyte, Agne Stankaityte, Dominyka Baltutyte, Vita Lele, Vytaute Starkute, Paulina Zavistanaviciute, Egle Zokaityte, Elena Bartkiene

13.20 **Chemical Properties of Biologically Active Compounds of The Medicinal Plant *Potentilla fruticosa* L.**
Patricija Dieninytė, Ona Ragažinskienė, Liudas Ivanauskas, Elena Juodkienė

13.27 **Activity of New Sunitinib Derivatives on Colon Cancer Cell Migration in Normoxia and Hypoxia**
Ugnė Endriulaitytė, Elias Maccioni, Vilma Petrikaitė

13.35 **Investigation on Intracellular Compounds and Glutathione Effect on Cell Viability Post Electroporation**
Gintarė Gerbenytė, Baltramiejus Jakštys, Saulius Šatkauskas

13.42 **Development of Blue Phosphorescent Organic Light-Emitting Diodes Using Novel Exciton-Blocking/Hole Transporting Materials**
Ghasemi Melika, Dmytro Volyniuk, Monika Cekaviciute, Jonas Keruckas, Ronit Sebastine Bernard, Viktorija Andruleviciene, Oleksandr Bezvikonnyi, Juozas Vidas Grazulevicius

13.50 **SDS-PAGE Gel Electrophoresis of Proteins Extracted from *Glycyrrhiza glabra* (L.) Roots**
G. Gražulytė, N. Savickienė, O. Ragažinskienė, G. Balčiūnaitė-Murzienė

13.57 **Optimisation of Extraction Conditions for Phenolic Compounds from *Cirsium vulgare* Leaves**
U. Griškevičienė, M. Marksa, A. Ževžikovienė, A. Ževžikovas, L. Ivanauskas

14.05-14.20 ***Coffee break***

Poster session II. ***Chairpersons:*** *Dr. Rūta Mickienė, Dr. Vima Kaškonienė*

14.20 **Characterization of Bioactive Compounds Extracted from By-products of *Matricaria recutita* Essential Oil Distillation Processes with Potential Use as Cosmetics Ingredients**
Ilva Nakurte, Marta Berga, Liene Kienkas, Laura Pastare, Mārtiņš Borodušķis, Anna Ramata-Stunda

14.27 **Quality Evaluation of the Semisolid Formulations with Bee Products**
Matas Inkėnas, Kristina Ramanauskiene, Asta M. Inkeniene, Monika Stanciauskaite, Aurimas Galkontas

14.35 **Influence of Culture Conditions and Methods of Sample Preparation on the Level of Bacterial Identification Using MALDI-TOF MS Technique**
Daria Janiszewska, Małgorzata Szultka-Młyńska, Michał Złoch, Paweł Pomastowski, Bogusław Buszewski

14.42 **Tetrahydrocarbazolyl- and 2-Phenylindolyl-Substituted Benzophenone Compounds for Optoelectronic Devices**
Lukas Graibus, Rasa Keruckiene, Jonas Keruckas, Ronit Sebastine Bernard, Egle Jatautiene, Jurate Simokaitiene, Dmytro Volyniuk, Juozas Vidas Grazulevicius

14.50 **Effect of Caffeic Acid Phenethyl Ester on Lactate Dehydrogenase Activity After Rat Kidney Ischemia/reperfusion *in Vivo***
Justina Kamarauskaite, Rasa Baniene, Sonata Trumbeckaite

14.57 (time zone GMT+3) **Chemical Composition of Nanovesicles Produced by Fruit Cells of Lingonberies (*Vaccinium vitis-idaea* L.)**
Greta Kanapeckaitė, Lina Raudonė

15.05 **The Expression of SR Protein Kinases in Brain Cell Lines**
Ignė Kanopaitė, Aušrinė Bajoriūnaitė, Eglė Jakubauskienė

15.05-15.15 ***Coffee break***

15.15 **Functionality of Oak (*Quercus* spp.) Parts and Possible Prospects for Their Usage: a Review**
Aurelija Kondratavičiūtė, Vilma Kaškonienė, Audrius Maruška

15.22 **Natural Anti-Estrogens Augment Proline Oxidase-Dependent Apoptosis in Breast Cancer Cells**
Sylwia Lewoniewska, Łukasz Szoka, Jerzy Pałka

15.30 **Amperometric Glucose Biosensor Based on Reduced Graphene Oxide and Polyaniline Nanofibers**
Viktorija Lisyte, Gabija Agasarjanaite, Anton Popov, Asta Kausaite-Minkstimiene, Almira Ramanaviciene

15.37 **Synthesis and Studies of Thioxanthone Based Derivatives Exhibiting TADF for New Generation OLED's**
Simas Macionis, Nasiri Sohrab, Dalius Gudeika, Dmytro Volyniuk, Juozas V. Grazulevicius

15.45 **LC-MS-based Quantitative Analysis of the Selected Metabolites in Aqueous Humor from Patients with Rhegmatogenous Retinal Detachment**
Magdalena Misiura, Diana A. Dmuchowska, Karolina Pietrowska, Emil T. Grochowski, Paweł Kraśnicki, Zofia Mariak, Adam Krętowski, Michał Ciborowski, Wojciech Miltyk

15.52 **Synthesis and Properties of Derivatives of 3,6-Disubstituted Naphthlimide**
Naveen Masimukku, Dalius Gudeika, Melika Ghasemi, Malek Mahmoudi Sharabiani, Dmytro Volyniuk, Juozas V. Grazulevicius

16.00 **The Microbiome Identification of Urine Samples Using the MALDI Technique**
Ewelina Maślak, Michał Złoch, Fernanda Monedeiro, Paweł Pomastowski, Bogusław Buszewski

16.07 **Production of Cyclodextrins and Their Application in Enantiomer Separations**
Ernesta Lisauskaitė, Robertas Sičiovas, Audrius S. Maruška

16.15 **Biological Degradation of Creosote Contaminated Soil from a Phytoremediation and Mycoremediation**
Guoda Kolkatovaitė, Rūta Mickienė, Vilma Kaškonienė, Audrius S. Maruška

16.22 **An Impact of Drying and Freezing to Biological Activity of Medicinal Plants and Spices**
Rugilė Karklytė, Paulius Kaškonas, Vilma Kaškonienė, Audrius S. Maruška

16.30 **The Technology and Quality Evaluation of Low pH Value Gel with Glycolic Acid and Cellulose Polymers**
Viktorija Juozapaitytė, Gailutė Drakšienė

16.37 **Fluctuations in Concentrations of N and P Compounds in Marvelė River: a Case Study of Urban Pollution**
Giedrė Kacienė, Daria Hordina

16.45 **Rab14 Endosome Targeting to the Intercellular Bridge Regulates Cytokinesis**
Paulius Gibieža, Rytis Prekeris

16.52 **Interfacing the Rachets to the Fluidic System for the Separation of Macromolecules**
Audrius Maruška, Tomas Drevinskas, Mantas Stankevičius, Kristina Bimbiraitė-Survilienė, Vilma Kaškonienė, Olga Kornyšova, Linas Jonušauskas, Jonas Artūras Jagelavičius, Staffan Nilsson

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15th International Scientific Conference "The Vital Nature Sign" Program

Friday, 21st of May 2021

Oral presentations. ***Chairpersons:*** *Dr. Nicola Tiso; Dr. Paulius Ruzgys*

9.00-9.15 (time zone GMT+3) **Evaluation of Antibacterial and Antioxidant Activities of Enzymatically Hydrolyzed Bee Pollen**
Vaida Adaškevičiūtė, Vilma Kaškonienė, Lukas Asanavičius, Audrius S. Maruška

9.15-9.30 **Olive Tree Pruning as a Renewable Source of Natural Bioactive Compounds**
Enrica Donati, Marco Mazzonna, Zeineb Aturki, Isabella Nicoletti, Anatoly P. Sobolev

9.30-9.45 **Influence of Genetic Origin on the Early Development of Scots Pine (*Pinus sylvestris*) Seedlings and their Interaction with Bacteria from the Genera *Paenibacillus* and *Pseudomonas***
Milana Augustauskaitė, Jonas Žiauka

9.45-10.00 ***In Vitro* Investigation of Probiotic Properties of Strains Belonging to *Lactobacillus* and *Bifidobacterium* Genera**
Jolita Jagelavičiūtė, Dalia Čižeikienė

10.00-10.15 **New Combination of Antimicrobial Lactic Acid Bacteria Strains for More Sustainable Livestock Production**
Laurynas Vadopalas, Modestas Ruzauskas, Vita Lele, Vytaute Starkute, Paulina Zavistanaviciute, Egle Zokaityte, Vadims Bartkevics, Sarunas Badaras, Dovile Klupsaite, Erika Mozuriene, Agila Dauksiene, Sonata Sidlauskiene, Romas Gruzauskas, Elena Bartkiene

10.15-10.30 **Influence of *Fusarium* Fungi Residing in Dicotyledonous Weeds on the Wheat Diseases**
Neringa Matelionienė, Skaidrė Supronienė, Gražina Kadžienė, Evelina Zavtrikovienė

10.30-10.45 ***Coffee break***

10.45-11.00 **The Use of Food Industry By-products for Technological Microorganisms Biomass Production**
Paulina Zavistanaviciute, Vytaute Starkute, Egle Zokaityte, Vita Lele, Vita Lele, Pranas Viskelis, Jurga Bernatoniene, Elena Bartkiene

11.00-11.15 **Additional Value Beverages Developed from Technological Functionalized Dairy, Cereal, and Fruit/Berry Industries By-Products**
Egle Zokaityte, Vita Lele, Vytaute Starkute, Paulina Zavistanaviciute, Darius Cernauskas, Dovile Klupsaite, Modestas Ruzauskas, Juste Alisauskaite, Alma Baltrusaityte, Mantvydas Dapsas, Karolina Siriakovaite, Simonas Trunce, Raquel P. F. Guiné, Pranas Viskelis, Vesta Steibliene, Elena Bartkiene

11.15-11.30 **IFN-β Enhances Cell Death in HCT116 and SW620 Cells and Their Chemoresistant Sublines**
L. Sudeikis, I. Juškaitė

11.30-11.45 **Serpin B5 Localization in the Chemoresistant Colorectal Carcinoma Cells**
B. Aleksandravičiūtė, V. Žitkutė

12.00-12.15 **The Role of Phospholipids in Prostate Cancer Development**
Magdalena Buszewska-Forajta, Paweł Pomastowski, Justyna Walczak-Skierska, Marcin Markuszewski, Marcin Matuszewski, Michał J. Markuszewski, Bogusław Buszewski

12.15-12.30 **Ozone Biomonitoring Using Vascular Plants in Cities**
Valentinas Černiauskas, Valda Araminienė

12.30-13.00 ***Lunch break***

Poster session I. ***Chairpersons:*** *Dr. Mantas Stankevičius, MSc. Kristina Bimbiraitė-Survilienė*

13.00 **MALDI Imaging of Honeybee Larvae**
Eliza Matuszewska, Szymon Plewa, Dagmara Pietkiewicz, Jan Matysiak

13.07 (time zone GMT+3) **Biologically Active Compounds of *Salvia officinalis* L. and Their Practical Application in Cosmetic Practice**
Laura Mažuolytė, Ona Ragažinskienė

13.14 **Hemagglutinating Activity of Proteins from *Kirchneriella* Sp. Schmidle Biomass**
Evelina Noreikaitė, Gabrielė Balčiūnaitė-Murzienė, Judita Koreivienė, Zoja Miknienė, Nijolė Savickienė

13.21 **Acridan Derivatives Exhibiting Aggregation Induced Enhancement of Thermally Activated Delayed Fluorescence for Organic Light Emitting Diodes**
Saimonas Pakstys, Viktorija Andruleviciene, Monika Cekaviciute, Jonas Keruckas, Ronit Sebastine Bernard, Juozas Vidas Grazulevicius

13.29 **Protein Quantification by Lowry Microassay and Haemagglutinating Activity Determination of Proteins from Cyanobacteria Biomass in Different Animal RBC Suspensions**
Neda Pašukonytė, Nijolė Savickienė, Jūratė Kasperovičienė, Zoja Miknienė, Gabrielė Balčiūnaitė-Murzienė

13.36 **Comparison of Hemagglutination with Blood Samples from Different Species of Animals**
Agata Paurytė, Jūratė Karosienė, Zoja Mikniene, Nijolė Savickienė, Gabrielė Balčiūnaitė-Murzienė

13.43 **Influence of Antibiotics on Bacterial Biofilm Formation**
Katarzyna Pauter, Viorica Railean-Plugaru, Paweł Pomastowski, Małgorzata Szultka-Młyńska, Bogusław Buszewski

13.50 **Investigation of Dependence of Phenolic and Volatile Compounds Composition, Antioxidant Activity and Energetic Properties of *Artemisia dubia* Wall. on Nitrogen Fertilization**
Vilmantas Pedišius, Audrius Maruška, Mantas Stankevičius, Irena Vaškevičienė

13.57 **Matrix-assisted Laser Desorption/Ionization Mass Spectrometry Imaging of Lipids in Ovarian Tumor Tissue – A Pilot Study**
Szymon Plewa, Dagmara Pietkiewicz, Agnieszka Horała, Piotr Jasiński, Ewa Nowak-Markwitz, Jan Matysiak

14.04 **The Influence of Different Conditions on The Formation of Zn-Lactoferrin Complexes**
Oleksandra Pryshchepa, Gulyaim Sagandykova, Katarzyna Rafińska, Joanna Rudnicka, Paweł Pomastowski, Myroslav Sprynskyy, Bogusław Buszewski

14.11 **Investigation of Intracellular Protein Release After Electroporation**
Leta Remeikytė, Baltramiejus Jakštys, Saulius Šatkauskas

14.18 **New Approaches in Functionalization of Cow's Milk Proteins**
Agnieszka Rodzik, Paweł Pomastowski, Viorica Railean-Plugaru, Bogusław Buszewski

14.25 ***Leonurus* L. Genus and Its Biologically Active Compounds for Therapeutical Purpose**
Ingrida Romaškaitė, Ona Ragažinskienė

14.25-14.35 ***Coffee break***

Poster session II. ***Chairpersons:*** *Dr. Vilma Kaškonienė, Dr. Benas G. Urbonavičius*

14.35 **Investigation of Phenolic Compounds and Antioxidant Activity of Brazilian Green and Red Propolis Preparations**
Raminta Rupeikaitė, Mindaugas Liaudanskas, Vaidotas Žvikas, Sonata Trumbeckaitė

14.42 **Influence of Short-Term Hypoxia on Alternative Pre-mRNA Splicing**
Karina Šapovalovaitė, Aistė Semionovaitė, Laurynas Vilys, Arvydas Kanopka

14.49 **Evaluation of the Composition of Phenolic Compounds in *Artemisia abrotanum* L. During Different Vegetation Stages**
Sandra Saunoriūtė, Ona Ragažinskienė, Liudas Ivanauskas, Lina Raudonė, Mindaugas Marksa

14.56 (time zone GMT+3) **The Impact of PTBP1 on Alternative Splicing of MAPT Pre-mRNA Under Normoxic and Hypoxic Conditions**
Justas Šidiškis, Kostas Sivickis, Laurynas Vilys, Eglė Jakubauskienė

15.03 **New Organic Materials Exhibiting Thermally Activated Delayed Fluorescence and their Application in Organic Light-Emitting Diodes**
Uliana Tsiko, Saimonas Pakštys, Dmytro Volyniuk, Jonas Keruckas, Ronit Sebastine Bernard, Viktorija Andrulevičienė, Juozas Vidas Grazulevicius

15.10 **Antimicrobial Activity of Lactic Acid Bacteria Immobilised in the Rice Bran-Lingonberry-Based Gel-Type Food Matrix**
Ruta Vaitkeviciene, Daiva Zadeike, Elena Bartkiene, Vita Lele, Grazina Juodeikiene, Valdas Jakstas

15.17 **Formation and Investigation of Fuchsine and Malachite Green Films and It's Possible Use for Low Dose Dosimetry Application**
Džiugilė Valiukevičiūtė, Judita Puišo

15.25 **Sacubitril/Valsartan Metabolism in HUVEC Cells**
Ugnė Venckytė, Ieva Eitminavičiūtė, Vaiva Lesauskaitė, Diana Žaliaduonytė, Audrius Pukalskas, Vaidotas Žvikas, Valdas Jakštas, Agnė Giedraitienė, Vacis Tatarūnas

15.25-15.40 ***Coffee break***

15.40 **Antiradical, Reducing, and Chelating Activities of Phenolic Fractions from *Vaccinium vitis-idaea* L. Fruits**
Gabrielė Vilkickytė, Lina Raudonė

15.47 **Evaluation of Hemagglutination Activity of Proteins Isolated from *Helianthus tuberosus* (L.) Tubers**
Živilė Zagorajevaitė, Ona Ragažinskienė, Gabrielė Balčiūnaitė-Murzienė, Zoja Miknienė, Nijolė Savickienė

15.52 **Synthesis and Properties Investigation of New Organic Semiconductors with Efficient Hole Transfer**
Emilija Zdanavičiūtė, Audrius Bučinskas, Jūratė Simokaitienė, Juozas V. Gražulevičius

16.00 **The Morphology of Laminated Cellulose/Alginate Biocomposites**
Aušrinė Žiūkaitė, Monika Strykaitė and Jonas Damašius

16.07 **Fermentation Possibilities of Biodegradable Organic Waste by *Saccharomyces bayanus***
Radvilė Žižytė, Vilma Kaškonienė, Lukas Asanavičius, Mantas Stankevičius, Audrius Maruška

16.15 **The Comparison of Surface Structures and Chemical Composition in Cocoons of Leeches (*Hirudinea*) and Planarias (*Turbellaria*)**
J. Rutkauskaitė-Sucilienė, I. Šatkauskienė

16.22 **The Influence of Bystander Effect on Viability of Untreated Cells After pDNA Electrotransfer, Irreversible Electroporation or Electrotransfer of Anticancer Drug Bleomycin**
Neringa Barauskaitė, Paulius Ruzgys, Saulius Šatkauskas

16.30 **Phytochemical Composition of Black Pepper (*Piper nigrum* L.), White Pepper (*Piper album* L.) and Fragrant Pepper (*Pimenta dioica* L.) Essential Oils**
Rugilė Narbutaitė, Mantas Stankevičius, Audrius Maruška

16.37 **Technology and Assessment of Orodispersible Granules**
Ieva Baškytė, Gailutė Drakšienė

16.44 **Biotechnologies for the Microbe-Mediated Remediation of Expended Railway Crossties: an Integrative Approach**
A. Maruška, N. Tiso, J. Mikašauskaitė-Tiso, T. Drevinskas, M. Stankevičius, R. Mickienė, V. Kaškonienė, K. Bimbiraitė-Survilienė, G. Dūda, V. Tumosaitė, O. Ragažinskienė, V. Snieškienė, A. Stankevičienė, D. Levišauskas, T. Tekorius, P. Kaškonas, C. Polcaro, E. Galli, E. Donati, M. Zacchini, O. Kornyšova, S. Grigiškis, I. Kiminaitė, A. Baliasnyj

16.50 **Closing of conference**

ISSN:2335-8653
ONLINE ISSN: 2335-8718

ORAL PRESENTATIONS

ISSN:2335-8653
ONLINE ISSN: 2335-8718

POSTER PRESENTATIONS

ISSN:2335-8653
ONLINE ISSN: 2335-8718

ORAL PRESENTATIONS

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Evaluation of Antibacterial and Antioxidant Activities of Enzymatically Hydrolyzed Bee Pollen

Vaida Adaškevičiūtė, Vilma Kaškonienė, Lukas Asanavičius, Audrius S. Maruška

Instrumental Analysis Open Access Centre, Faculty of Natural Sciences, Vytautas Magnus University, Kaunas, Lithuania

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Abstract

Bee pollen is one of the most nutritious apitherapeutic products. This natural product cannot be digested in the human body due to its cell wall and a lot of valuable compounds accumulated in bee pollen are not absorbed. In order to obtain not only higher bioactivity, higher diversity of compounds from bee collected pollen, but also to improve bioavailability, additional processing of the raw material is proposed: to ferment using lactic acid bacteria or to hydrolyze using selective enzymes [1, 2]. The aim of this study was to evaluate the impact of enzymatic hydrolysis on antioxidant and antibacterial activities of bee collected pollen.

Nine bee pollen samples from Spain, Italy, Netherlands, Sweden, Poland, Lithuania, Denmark, Malta and Slovakia were hydrolyzed using *Viscozyme L*, *Clara-diastase*, lipase, protease, amyloglucosidase and cellulase enzymes. Total phenolic content, total flavonoid content and antioxidant activity were determined using spectrophotometric methods [3]. Also, oxidation-reduction potential was analyzed [4]. Antibacterial activity against *S. aureus*, *L. monocytogenes*, *S. enteritidis* and *S. typhimurium* was evaluated using agar well diffusion method. Moreover, an interaction between bee pollen extracts and antimicrobial drugs, ceftazidime pentahydrate, erythromycin, oxytetracycline dihydrate, ciprofloxacin, was analyzed [5].

The results showed that the enzymatic hydrolysis increases total phenolic content (by 1.3-2.5 times), total flavonoid content (by 1.2-1.9 times) and antioxidant activity (by 1.2-2.1 times) of bee collected pollen. Samples hydrolyzed by cellulase enzyme showed significantly higher ($p \leq 0.05$) oxidation-reduction potential after hydrolyzation. Overall, hydrolysis with enzymes improved bee pollen antibacterial activity (by 1.4-3.2 times) and had a positive effect on interaction of raw material extracts with antibiotics. It was determined that extracts of hydrolyzed bee pollen samples and antimicrobial drugs have synergistic effect on all studied pathogenic bacteria strains.

Keywords: Bee pollen, enzymatic hydrolysis, antibacterial activity, antioxidant activity, synergistic effect.

Acknowledgements: This project was financed by Research Council of Lithuania project No. 09.3.3-LMT-K-712-16-0030.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Olive Tree Pruning as a Renewable Source of Natural Bioactive Compounds

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Abstract

Nowadays, the management of agri-food by-products and wastes is arousing growing concern. Modern agricultural practices are wasteful, producing billions of tons of wastes a year. Therefore, the transition towards a circular economy is desirable also in view of the increasing food demand. This transition requires a departure from the traditional linear production models, in which a large amount of wastes is generated from natural resources in addition to valuable products. On the contrary, the circular economy is aimed at developing a model that avoids a negative impact on the environment, decreasing the use of natural resources and reducing waste production [1]. An interesting approach from the economic and environmental points of view to obtain this economic model is the reuse of agro-food by-products to achieve added value products and energy. In particular, residual biomass derived from the olive-oil industry represents a renewable source available for use in further applications [2]. The present work evaluates the suitability, as a source of natural bioactive compounds, of olive tree pruning collected in an olive grove placed in an area located on the outskirts of Rome. Indeed, it is well known the presence in the olive leaves and branches of interesting biologically active substances that may be employed in the food industry as food additives or as nutraceutical ingredients, in order to obtain products with enhanced nutritional value, potential health benefits, longer shelf-life and good organoleptic properties. A multimetodological analytical approach was applied to characterize the extracts of the olive tree pruning. Phytoextracts chemical profile was examined by NMR spectroscopy and the most abundant bioactive compounds were determined by means of liquid chromatography (HPLC-ESI-MS, nano-LC).

Keywords: Circular economy, olive tree pruning, bioactive compounds.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Developing Mass Spectrometry Based Single Cell Proteomics

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Abstract

Single cell proteomics (SCP) has the potential to address several biologically important, yet poorly understood questions at the level of individual cells. Many of the cellular mechanisms resulted in treatments of low molecular weight drug compounds have been accessed in bulk analyses, identifying multiple target proteins and pathways by comparison of expression proteomes under various conditions and at time points. However, certain protein quantities may be normalized when a heterogenous population of cells are investigated in bulk.

To understand the effects of drug treatments at single cell level, we designed experiments to generate global proteomes profiles and compared to bulk data. Using well characterized drug compounds with known targets we hypothesized that SC and bulk data should positively correlate and expected to observe additional benefit of individual cellular profiles.

Using the well characterized cellular model challenged with various drug molecules helped to accomplish an entire workflow to isolate single cells, prepare protein samples, acquire and analyze quantitative data. SCP profiles were obtained during an optimized sample preparation and data acquisition workflow, based on the original SCoPE approach [1] employing tandem mass tag (TMT) labeling strategy and carrier. The results indicated that the correlations between single cells and bulk experiment were established proving the validity of the experimental design. Since some regulated proteins were expressed at medium to low levels, their detection was challenging and often were not possible in SC datasets, which impaired the correlation with "classic" bulk data. For this reason, we have introduced a built-in bulk design that was found to be a more relevant.

With this set of experiments, we could confirm that the effect of drug treatments was achievably captured at SC level. Provided the target proteins were detectable, their expressions could be reliably determined, which may pave a new path for chemical proteomics at the single cell level.

Keywords: Single cell proteomics, mass spectrometry, tandem mass tag, chemical proteomics.

Acknowledgements: The project was financially supported by the Swedish Research Council (VR-2018-05766).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Influence of Genetic Origin on the Early Development of Scots Pine (*Pinus sylvestris*) Seedlings and their Interaction with Bacteria from the Genera *Paenibacillus* and *Pseudomonas*

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Abstract

Scots pine (*Pinus sylvestris*) is one of the most common trees in Northern Europe and its wood is in demand and commercially important. Many pines suffer from the pathogenic fungus *Heterobasidion annosum* which damages roots and trunk. It is therefore necessary both to select genetically superior, more resistant pine families and to search for novel biotechnological tools that could suppress fungal infection [1]. One of the possibilities would be to use plant-associated bacteria, capable to suppress the development of phytopathogenic fungi [2]. In this context, it is important to test if selected bacterial candidates are themselves not harmful for pines. The bacteria taken for this research were *Paenibacillus* sp. and *Pseudomonas* sp. that were originally isolated from the tissue cultures of hybrid aspen. The aim of this study was to investigate the early development and interaction of different Scots pine (*Pinus sylvestris*) families with *Pseudomonas* sp. and *Paenibacillus* sp. bacteria *in vitro* and *ex vitro*. Two groups of pine genetic families were investigated: five families were taken from the genetic reserve of potentially resistant pines in Vaišvydava and five were randomly selected families from Jonava plantation. The seeds from these families were surface-disinfected and planted *in vitro* on the nutrient medium inoculated with either *Paenibacillus* sp. or *Pseudomonas* sp. Also, the effect of bacteria on pines during their *ex vitro* adaptation stage was investigated. The results showed that *Pseudomonas* sp. had a negative effect on root growth in seven of the ten tested pine families and, in three of them, the shoot growth was also impaired. *Paenibacillus* sp. only had negative impact on root growth in the pine family No. 2J; however, in another family (No. 12), this bacteria even caused the development of longer shoots. In turn, *ex vitro* results showed that *Pseudomonas* sp. had the strongest negative effect on the seedling development in the family No. 2J. This family was the only one affected negatively also by *Paenibacillus* sp. In conclusion, *Paenibacillus* sp. was found to be well-tolerated by most of the tested pine families.

Keywords: Bacteria, *in vitro*, inoculation, pine, seedling.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

ZnO Tetrapod Synthesis and Application in Electrochemistry: Effect of Morphology

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Abstract

Zinc oxide (ZnO) is a direct wide band gap (3.37 eV) semiconductor material with a large exciton binding energy of 60 meV [1]. The tetrapod form consists of four arms interconnected by a central core at angles ~105° to 110° with respect to each other. Their specific 3D morphology provides easy access to nanoscale features [2]. ZnO nanostructures in different sizes, nanoparticles and nanorods were compared in order to detect how their morphology influenced their electrochemical properties. The continuous synthesis (combustion) method [1] is used to synthesize ZnO tetrapods with high yield possibilities. Obtained ZnO tetrapods were separated into two fractions of different scale by centrifugation. Various techniques of characterization have been applied, including scanning and transmissions of electron microscopy (SEM and TEM), X-ray powder diffraction, BET and UV-Vis spectroscopy, which can be used to classify structural properties of different ZnO nanostructure morphologies. Electrochemical analysis show ZnO tetrapod highest active area of 0.095 cm^2 and the lowest peak separation of 61.7 mV which is close to theoretical value. Electrical properties due to their packaging and electrochemical features, there is a link between pores in different ZnO nanostructures. The thorough study of ZnO nanostructures could benefit future electrochemical and biosensing applications.

Keywords: ZnO, tetrapods, electrodes, electrochemistry, particle separation, synthesis.

Acknowledgements: This research was funded by the European Regional Development Fund, "Targeted research in the area of smart specialization" measure, Project 01.2.2-LMT-K-718-02-0011.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

In Vitro Investigation of Probiotic Properties of Strains Belonging to *Lactobacillus* and *Bifidobacterium* Genera

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Abstract

Probiotics have been defined by The Food Agricultural Organization/World Health Organization as "live microorganisms which when administered in adequate amounts confer a health benefit on the host". Probiotic bacteria have to be safe, survive in the gastrointestinal tract conditions and possess beneficial properties [1]. Food may maintain the survival of probiotic bacteria during passage through the gut by the buffering properties [2]. Interest in the functional probiotic foods strongly increased during recent years. Selection of the proper strains with strong probiotic properties is an essential factor for novel functional product development with high nutritional value and acceptance.

This study was aimed to evaluate *in vitro* probiotic properties of commercially available strains belonging to *Lactobacillus* and *Bifidobacterium* genera.

The 9 *Lactobacillus* and 5 *Bifidobacterium* strains were characterized *in vitro* for their probiotic characteristics such as resistance to bile salts and low pH, antibiotic sensitivity by gradient diffusion using MIC Test Strips, autoaggregation and coaggregation assay with *Escherichia coli* DSM 27503 and antioxidant activity by DPPH and ABTS radical scavenging assay.

The results demonstrated that tested probiotic properties varied among the different strain. Most of the tested strains were able to tolerate 0.3% bile salts for 4 h and pH 3 for 4 h, while in pH 2 for 4 h survived 8 of 14 strains. None of the strains were resistant to ampicillin. All tested strains exhibited a high percentage of autoaggregation and coaggregation with *E. coli* DSM 27503. *L. reuteri* DSM 20015 and *L. paracasei* subsp. *paracasei* DSM 4905 exhibited the highest DPPH radical scavenging activity, while *B. longum* subsp. *infantis* DSM 20088 exhibited the highest ABTS radical scavenging activity. Results suggest *L. paracasei* subsp. *paracasei* DSM 20020, *B. pseudolongum* DSM 20099, *B. animalis* DSM 20105 as probiotic candidates can be used in the development of functional food as food ingredients.

Keywords: *Lactobacillus* spp., *Bifidobacterium* spp., probiotic properties.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Combined Enzyme assisted - Supercritical Fluid Extraction with Flash Chromatography

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Abstract

Plants are basically an unlimited source of biologically active compounds and the products obtained from them are increasingly used not only in the pharmaceutical and cosmetic industries, but also in the food industry. The largest group of bioactive compounds in plants are polyphenols, which exhibit antioxidant and antibacterial properties, thanks to which they can be used as natural preservatives in cosmetic and food products. They can increase their microbiological safety and extend their shelf life.

The presence of low concentrations of bioactive compounds in plants forces modern science to take steps to develop an effective methodology for isolating and increasing the concentration of these compounds. Due to the fact that the bioactive compounds present in the plant material are a multi-component matrix, the purification process is necessary for further identification. One technique used to purify and fractionate compounds from natural products is low pressure column chromatography (flash chromatography).

The aim of the research was to develop a method of fractionating plant extracts. The research material consisted of extracts from alfalfa (*Medicago sativa* L.) leaves obtained by enzyme assisted - supercritical fluid extraction (EA-SFE). In order to develop a flash chromatography method, the obtained extracts were first subjected to thin layer chromatography (TLC) analysis. For this purpose, were prepared charts that allow a direct selection of an appropriate eluting solvents based on the various concentration of formic acid in mobile phase mixtures applied for the separation of polyphenols from alfalfa extracts. Due to the high concentration of biological active compounds in analysed extracts some nonpolar mixtures of organic solvents typically use for the separation of flavonoids were compared. The established pre-separation conditions on TLC plates were transferred to flash chromatography. The obtained fractions were analyzed for specific groups of bioactive compounds using supercritical fluid chromatography (SFC).

Flash chromatography is a very valuable technique in the field of studying natural compounds because it provides a fast and economical way to separate the major components of complex plant extracts. The use of a combination of supercritical CO_2 extraction in conjunction with flash chromatography allowed to improve the selectivity of the extraction.

Keywords: EA-SFE, polyphenols, flash chromatography**,** SFC.

Acknowledgements: This study was supported by PRELUDIUM 15 (No. 2018/29/N/ST4/01180) from National Science Center, Poland.

MSc. Aneta Krakowska-Sieprawska, Dr. Katarzyna Rafińska and Prof. Bogusław Buszewski are members of Toruń Center of Excellence “Towards Personalized Medicine” operating under Excellence Initiative-Research University.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Statistical Analysis of Characteristics of Thermoluminescent Dosimeters

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Abstract

Thermoluminescent dosimeters are one of the most popular dosimetric methods used in the field of radiation safety and patient dosimetry. Patient dosimetry is particularly important for optimizing the radiotherapy and diagnostic procedures. To be able to use TLDs for high accuracy measurements, evaluation of Element Calibration Coefficient and its statistical distribution in a regulated batch is pertinent. Grading of dosimeters based on statistical parameters is of high value and such grading is done in this research.

In this experiment 400 units of TLD-100 were used. Dosimeters were calibrated with linear accelerator Varian Clinac DMX at [0.5–5] Gy range, these doses are used in radiotherapy procedures. 40 dosimeters were assigned to each experimental point. Selected photons energy – 6 MeV.

The Rialto thermoluminescent dosimeter scanner was used to prepare and scan the dosimeters. After calibration procedure, the obtained Element Calibration Coefficients were analyzed. The highest standard deviation is 0.1704 and the lowest is 0.3870. After 2σ rule application, the highest standard deviation is 0.087 and the lowest is 0.0342. The lowest value of standard deviation are at the same calibration dose – 0.5 Gy in both cases.

It can be shown that the coefficients from 0.829 to 1.123 are distributed relatively evenly after evaluating the initial coefficients. 23.5 percent of TLDs were rejected after the application of the 2σ rule and the coefficients range that are distributed relatively evenly is [0.928; 1.097].

Keywords: Thermoluminescence, dosimetry, absorbed dose, statistical analysis.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Influence of Fusarium Fungi Residing in Dicotyledonous Weeds on the Wheat Diseases

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Abstract

Growing of wheat is one of the most profitable agricultural business in Lithuania. To ensure the quality of the grain and protect it from common wheat diseases, researchers are being carried out to identify possible sources of infection and their impact on the wheat diseases. *Fusarium* fungi cause wheat and other cereal roots rots and head blight. *Fusarium* head blight (FHB) reduces the yield of the grain, decreases the quality, contaminates the grain with mycotoxins, decreases the germination of the seeds and the vitality of the seedlings [1]. *Fusarium graminearum* residing in common dicotyledonous weeds is known to be as the source of these infection. The aim of this research is to investigate the presence of other *Fusarium* spp. in dicotyledonous weeds as alternative host plants and to assess their impact on wheat diseases.

The identification of *Fusarium* fungi using classical microbiological and molecular methods has shown that the main species associated with FHB, like *F. graminearum, F. avenaceum* and *F. culmorum*, were detected in the internal tissues of all dicotyledonous weed species obtained from three different field, without showing any externally visible signs of infection. Nevertheless, the most common species was *F. equiseti* (91-98%), which is known as weak wheat pathogen, the second species was *F. culmorum* (60-70%). Pathogenicity of *Fusarium* spp. to spring wheat was the highest when wheat heads were inoculated with *F. culmorum* (99.4%) and *F. graminearum* (98.8%) species, while *F. equiseti* caused the wheat head blight only at 1.5% of severity.

Keywords: Alternative source of infection, broad-leaved weed, Fusarium head blight, host plant, wheat.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Investigation of Dose Sensitivity in Irradiated Polymer Gels Containing Water Co-solvents

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Abstract

Radiation treatment of oncological diseases is based on delivery of relevant ionizing radiation dose to the target according to the prepared patient specific dose plan. The outcome of the treatment procedure to high extent depends on the accuracy of dose delivery which can be achieved performing *in vitro* dose distribution measurements in patient pre-treatment stage. Gel dosimetry is the only method among many other different dose measurement methods, which combines 3D phantom and dosimeter features in one allowing for 3D dose distribution measurements within the volume.

Dose gels usually contain sensitive to ionizing radiation monomer, cross-linker and oxygen scavenger and are tending to polymerize upon irradiation to high energy photons or other particles recording volumetric shape (tumor) of the irradiated target. Gelatine or agarose is used as a scaffold for accommodation of polymerized structures. Low sensitivity is one of the disadvantages of dose gel. It could be improved using higher concentrations of cross-linker however in many cases its low solubility in water sets the limits for concentrations. To overcome the problem solvents other than water can be used, thus contributing to more effective polymerization of the gels.

The aim of this work was to investigate dose sensitivity of gels, containing N-(isobutoxymethyl) acrylamide, BIS-Acrylamide, tetrakis, gelatin and water (poly-acrylamide gels) by substituting some part of water in the gel with other solvents (glycerol, acetone, isopropyl alcohol) and enhancing concentration of cross-linker BIS acrylamide.

Polymer gels were prepared following the procedure developed by our research group and irradiated to different doses with 6MeV photons in linear accelerator CLINAC (Varian). Optical properties of experimental samples before and after the irradiation were analyzed using UV-VIS spectrophotometer “OCEAN OPTICS USB 4000”. It is shown that the application of acetone as co-solvent for cross-linker allows for gel’s dose sensitivity enhancement from 0.09 to 0.19 for the standard concentration (3%) of BIS acrylamide in the gel. Further enhancement of dose sensitivity in irradiated gels containing >3% concentrations of BIS acrylamide is discussed and the results with application of other in water co- solvents are provided in this work.

Keywords: Ionizing radiation, polymer gel dosimetry, dose sensitivity.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

New Functional Aspects of Enzyme-Assisted Extraction of Plant Material

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Abstract

Extraction of plant material using enzymes has gained great popularity in recent years. This approach has many advantages such as lower solvent consumption, shorter extraction times and milder conditions. Cell walls are made up of many different polymers, and their enzymatic hydrolysis can release their short chains or monomers, to the solvent during extraction. However, most of research focuses on the selection of conditions that guarantee the effectiveness of obtaining biologically active compounds. They correlate the quality of obtained extract only with the content of compounds of interest. Nevertheless, changes in the structure of the material and their consequences on the extraction process as well as the quality of the obtained extracts are not taken into account. Therefore, the aim of the study was to analyze the impact of enzymatic hydrolysis on structure of plant material and quality of extracts. For research, the most common enzymes were used e.g., cellulase, pectinase, protease and kemzyme.

FTIR spectroscopy, optical and transmission electron microscopy were used for evaluation of the level of plant cell wall hydrolysis. MALDI-TOF-MS analysis of extracts obtained from hydrolyzed plant material showed that they differ significantly in relation to the presence of interfering substances with high molecular masses. Similar, a significant difference was seen in the amount of proteins and reducing sugars released during hydrolysis. DLS analysis was used for size characterization of particles released from plant material. As a result, we could discuss the usefulness of various enzymes in obtaining high-quality extracts with the highest possible content of biologically active compounds and the lowest possible presence of interfering compounds. Extracts obtained from material hydrolyzed by pectinase was characterized by the highest level of flavonoids and phenolic acids and at the same the lowest presence of interfering substances in the range 1-3 kDa. Moreover, FTIR analysis allowed to assign characteristic bands to the hydrolyzed plant material and in the future this technique can be used for the fast and unexpensive control of enzymatic processes.

Keywords: Enzymatic-assisted extraction, biologically active compounds, transmission electron microscopy, plant material.

Acknowledgements: This work was supported by the National Science Center, Poland (MINIATURA o nr 2018/02/X/ST4/0135). B.B., K.R. and A.K.S. are members of Toruń Center of Excellence "Towards Personalized Medicine" operating under Excellence Initiative-Research University.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

IFN-β Enhances Cell Death in HCT116 and SW620 Cells and Their Chemoresistant Sublines

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Abstract

Colorectal cancer (CRC) is one of the most frequent malignancies worldwide, being the second most common cause of cancer death in Europe. Depending on the cancer progression, a surgical removal of the tumour can be performed, other times, chemotherapic agents such as FOLFOX – a combination of 5-fluorouracil (5-FU), leucovorin and oxaliplatin (OxaPt) are used to battle the disease. Unfortunately, intrinsic or acquired chemoresistance is understood as a major reason for therapy failure, with consequent tumor growth and spreading ultimately leading to the patient's premature death. The ultimate function of chemoresistance is to ensure the survival of malignant cells through continuous adaptation within an organism, therefore, the nature and spectrum of cell-survival strategies in CRC represent a highly significant target of scientific inquiry.

Interferons (IFNs) are a family of signaling molecules (cytokines) that are divided into three classes: (Type I, Type II, Type III). Different types of interferons exert various functions in the cell: they can activate immune cells, such as natural killer cells and macrophages, increase host defenses by up-regulating antigen presentation. Type I interferons also possess a wide range of potential anti-tumor effects: inhibition of cell proliferation, induction of cell cycle arrest and cell death.

The aim of this study was to investigate the potential chemosensitising effects of type I interferon – β (IFN-β) in HCT116 cells and their 5-fluorouracil and oxaliplatin resistant sublines HCT116/FU and HCT116/OXA, respectively. Another point of interest was to elucidate the effects of IFN-β on cell survival in metastatic colorectal carcinoma SW620 cells. Our data indicate that IFN-β is able to sensitise human colorectal carcinoma cells to chemotherapeutic agents, leading to a decrease in cell survival *in vitro*. In addition, we evaluated expression of proteins involved in apoptotic and necroptotic death pathways.

Keywords: Colorectal cancer, interferon-β, 5-fluorouracil, oxaliplatin, chemoresistance.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Biological Activity of Trivalent Chromium

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Abstract

Chromium belongs to the group of trace elements. Chromium occurs naturally in soil, water and air. It is present in human and animal organisms. Chromium exists in valence stages range from -2 to +6. However, two major oxidation states: trivalent Cr(III) and hexavalent Cr(VI) are dominant. Cr(VI) enters the cells *via* sulfate and phosphate anionic transporters. Inside the cells Cr(VI) is reduced to Cr(III) [1]. Chromium(III) penetrates cell membrane by endocytosis, diffusion or phagocytosis [1,2]. Chromium(III) can be transported inside cells *via* the transferrin receptor. Cr(III) has been recommended as an essential element involved in many processes in the body. Trivalent chromium is involved in insulin receptor activation and is required for maintaining normal glucose tolerance. However, chromium(III) is able to produce radicals *via* the Haber-Weiss reaction. Radicals $O_2^{\bullet -}$ and $^{\bullet}OH$ produced in these reactions interacts with cell molecules: DNA, proteins, lipids and other. What is more, decrease of CAT, SOD and glutathione peroxidase activity was observed [3]. Destruction of cell organelles, i.e. cell membrane, mitochondria, lysosomes, nucleus, spindle apparatus, as well as disorders of cell metabolism, i.e. respiratory and proliferation processes may be induced by reactive oxygen species induced by trivalent chromium.

Cr(III) compounds can bind directly to DNA *in vitro*, forming Cr-DNA adducts and DNA-DNA crosslinks [4]. Moreover, after chromium(III) treatment DNA decreases in the G0/G1, while it increases in the G2/M phase. It can be concluded that chromium(III) arrests cell cycle in the G2/M phase. Moreover, aneuploidy peak (subG1) of DNA content was observed [5]. This peak may represent cells that escaped mitotic arrest and which replicate as multinuclear cells without dividing. Giant, multinuclear and apoptotic cells observed in microscopic analyses are the effect of this process [6].Chromium(III) increases the frequency of mutations: substitutions, transversions, deletions and insertions, base–pair substitution [6]. All these changes lead to apoptosis. The increase of caspase 3, 6 and 9 was observed after chromium(III) treatment. Additionally, Bax induction, while Bcl-2 inactivation were observed.

Chromium(III) interacts with other microelements. Chromium(III) and molybdenum(III) or chromium(III) and nickel(II) show antagonistic effect – chromium(III) protects from nickel(II) or molybdenum(III) toxicity. Moreover, chromium(III) and iron(III) show antagonistic effect in toxicity.

Keywords: Chromium, cell metabolism, toxicity, oxidative stress.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Baltic Sea Macro-Algae Characteristics Including Microbial Profile and Trace Elements Content

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Abstract

Macro-algae are natural marine ecosystem components, which showing chemical composition desirable for it possible application in various industry areas [1], and the most common macro-algae, which can be used for food / feed purposes, in the Baltic Sea region are *Ulva intestinalis*, *Cladophora rupestris* and *Furcellaria lumbricalis* [2]. The aim of this study was to evaluate possibility to apply fermentation for *U. intestinalis*, *C. rupestris* and *F. lumbricalis* using antimicrobial properties possessing *Lactobacillus plantarum* LUHS135 strain, without fermentable substrate additional pre-treatment. To evaluate effectiveness of the used treatment, analysis of the microbial profile, antimicrobial and antioxidant characteristics, and trace elements concentration were performed. Fermentation with LUHS135 strain by 1.6 times reduced *C. rupestris* pH, however, *U. intestinalis* and *F. lumbricalis* pH after 12 hours of fermentation remained similar. Metagenomic analysis showed that the algae can be contaminated with pathogenic microorganisms, which remain after fermentation, despite that technological microorganism possessing antimicrobial properties was used. However, synergic mechanism of algae and LUHS135 combination showed broader spectrum of pathogens inhibition. Also, the health claims associated with algae products must be based on sufficient evidence of algae chemical safety parameters, from which heavy metals are very important. Finally, in this study tested algae showed antimicrobial potential, therefore, algae extracts preparation could be a possible way to use this material, because, during the extracts preparation, bio- and chemical safety can be ensured.

Keywords: Baltic Sea macro-algae, microbial profile, antimicrobial activity, trace elements.

Acknowledgments: The authors gratefully acknowledge the COST Action 18101 SOURDOMICS – Sourdough biotechnology network towards novel, healthier and sustainable food and bioprocesses. COST is a funding agency for research and innovation networks.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Integration of Ultrasound Treatment into Valorization of Flaxseed Press Cake to Produce Functionalized Hydrogels

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Abstract

This research is dedicated to study the effect of ultrasonication for flaxseed dietary fibre solubilization and functionalized hydrogels production. In the initial stage of the experiment, dry fractionation was applied for flaxseeds to produce dietary fibre-enriched products with an average total dietary fibre content of 26 %. Ultrasonication of dietary fibre-enriched flaxseed fraction at 850 kHz and different intensities (0.9 W/cm^2, 1.3 W/cm^2, 2.0 W/cm^2) had a significant effect on flaxseed dietary fibre by increasing the content of soluble fibre on average by 8.40 % (pH 7). Solubilised flaxseed fibre was more suitable medium for lactic acid bacteria (LAB) than without the pre-treatment. After 30h of US treated flaxseed fibre fermentation with *Lactobacillus brevis* and *Lactobacillus plantarum* the LAB viable count was found to be greater on average by 19.8% and 13.0%, respectively, compared to the control sample. Additionally, ultrasound has been tested for hydrated gel production. Ultrasonication of aqueous suspensions increased gel viscosity by an average of 33.18 % (after 24 hours of cooling) and the maximum effect was achieved using the highest intensity (2.0 W/cm^2). The optimal texture of the gels was obtained by using 40 % of flaxseed press cake and 60 % of blackcurrant press cake. Thus, ultrasonication intensify the process of hydrogel formation, which could apply for antimicrobial LAB immobilization and enrichment with phytochemicals of blackcurrant press cake.

Keywords: Plant-based by-products, ultrasound treatment, dietary fibre, gel coatings.

Acknowledgments: The authors gratefully acknowledge the COST Action CA18101 “Sourdough biotechnology network towards novel, healthier and sustainable food and bioprocesses”. The research is funded by The European Regional Development Fund by the program “Research Projects Implemented by the World-class Researcher Groups” (Grant No. 01.2.2-LMT-K-718).

ISSN:2335-8653
ONLINE ISSN: 2335-8718

New Combination of Antimicrobial Lactic Acid Bacteria Strains for More Sustainable Livestock Production

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Abstract

Considering that the world population is growing, it is crucial to ensure increased agricultural industry production in a more sustainable and eco-friendly manner [1]. According to the prognosis for the near future, the global expectation for livestock products demand will double [2]. The aim of this study was to apply newly isolated antimicrobial characteristic possessing lactic acid bacteria (LAB) starters for local stock (rapeseed meal) fermentation and to evaluate the influence of changing from an extruded soya to biomodified local stock in a feed recipe on piglets' fecal microbiota, health parameters, growth performance, and ammonia emission. In addition, biomodified rapeseed meal characteristics (acidity and microbiological) were analyzed. At the beginning of the *in vivo* experiment, the microbial profiles in both piglet groups were very similar: the highest prevalence was *Prevotella* (34.6–38.2%) and *Lactobacillus* (24.3–29.7%). However, changing from an extruded soya to fermented rapeseed meal in the feed recipe led to desirable changes in piglets' fecal microbiota. There was a more than four-fold higher *Lactobacillus* count compared to the control group. Furthermore, there was significantly lower ammonia emission (20.6% reduction) in the treated group section. Finally, by changing from extruded soya to less expensive rapeseed meal and applying a fermentation model with selected LAB combination, piglets were fed without any undesirable changes in health and growth performance, as well as in a more sustainable manner.

Keywords: Lactic acid bacteria; fermentation; feed; piglets; microbiota; ammonia emission.

Acknowledgments: The authors gratefully acknowledge the COST Action CA18101 "SOURDOugh biotechnology network toward novel, healthier and sustainable food and bIoproCesseS" and the EUREKA Network Project E!13309 "SUSFEETECH" (Nr. 01.2.2-MITA-K-702-05-0001).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Ecotoxicological and Genotoxic Assessment in *Lemna minor* L. and *Spirodela polyrhiza* L. Plants Exposed to Dimethyl Phthalate (DMP)

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Abstract

Phthalates represent a class of compounds of very high concern for their ubiquity and increasing production worldwide, being largely used as plasticizers and in personal care products. For their recognized toxicity on living organisms, they were included in the list of priority pollutants by USEPA and put under restriction by EU Commission. Adverse effects of the exposure to these compounds have been clearly reported in humans (hypertension, diabetes and pregnancy loss among others) and in animals (reproductive and developmental toxicities) [1]. Contrarily, few studies dealing with the effects of phthalates on plants are reported in literature. In particular, there is a lack of information on genotoxicity and physiological toxicity in aquatic macrophytes. Duckweeds (among which *Lemna* and *Spirodela*) are recognized as biological organisms able to assess the environmental quality of the aquatic ecosystem (bioindicators). For this purpose, a specific toxicity test using these plant species has been officially set up (OECD n. 221) [2]. According to these guidelines, a 7-day test was performed in multi-well plate in growth chamber by exposing selected fronds of *Lemna minor* and *Spirodela polyrhiza* to different concentrations of dimethyl phthalate (DMP) in Hoagland nutrient solution. By using the Eco-Tox Photosystem Tool (ETPT) [3] and Comet assay [4], toxicity effects at morphological (biometric indicators), physiological (pigment content and photosynthetic performances) and molecular (DNA damage) level were observed, particularly at the highest concentration tested. These results described for the first time the adverse effects exerted by DMP on aquatic plants, contributing to highlight the environmental risk associated to the presence of this compound in the aquatic ecosystem.

Keywords: Aquatic plants, bioindicators, chlorophyll fluorescence, Comet assay, duckweed, emerging contaminants.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Use of Food Industry By-products for Technological Microorganisms Biomass Production

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Abstract

In this study, for lactic acid bacteria (LAB) biomass production and stabilization several technological solutions were tested. For LAB biomass growth (I) acid whey (enriched with 2.5% glucose, 2% yeast extract and 0,5% saccharide), and (II) potato juice combination with apple juice production by-products were tested. After 48 h of fermentation, LAB biomass was lyophilized (-40 °C) and spray dried (+150 °C). Viable LAB count, before and after dehydration, as well as during the storage at different temperature, was evaluated. It was established that acid whey enriched with 0.5% glucose, 2.0% yeast extract, and 0.5% saccharide is suitable substrate for *L. plantarum* and *L. paracasei* biomass preparation and stabilization during the spray drying (+150 °C), lyophilization (-40°C), and storage: in spray-dried nano capsules the viable *L. plantarum* and *L. paracasei* cell concentration was 7.52±0.03 and 7.12±0.03 log10 CFU/g, respectively; in lyophilized biomass, the viable *L. plantarum* and *L. paracasei* cell concentration was 8.45±0.06 and 8.25±0.03 log10 CFU/g, respectively, established. After lyophilization (-40 °C) and spray drying (+150 °C) the viable cell concentration in fermented with *P. acidilactici* and *P. pentosaceus* potato juice powder was 9.18 ± 0.09 and 9.04 ± 0.07 log10 CFU/g, respectively, and in apple by-products 8.03 ± 0.04 and 7.03 ± 0.03 log10 CFU/g, respectively. The highest stability after 12 months of storage at 20 °C temperature by *L. paracasei* and *P. acidilactici* lyophilized in acid whey and potato juice (6.16±0.03 and 7.00±0.04 log10 CFU/g, respectively), were found. According to obtained results, food industry by-products can be alternative substrates for LAB biomass growth and dried powders can be used in food/feed industry as the technological microorganisms' starter.

Keywords: By-products, encapsulation, lyophilization, lactic acid bacteria, sustainability.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Additional Value Beverages Developed from Technological Functionalized Dairy, Cereal, and Fruit/Berry Industries By-Products

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Abstract

The aim of this study was to develop additional value beverages in a sustainable manner by using for it formulation food processing by-products. The main components used for the study of added-value beverages were fermented milk permeate, extruded and fermented wheat bran (WB), and different fruit/berry by-products (FBB). The characterisation of the amounts of bioactive components was based on the overall acceptability and emotions induced in consumers by the tested beverages. The functional properties of the developed beverages have been proven by evaluating their antimicrobial and antioxidant properties, as well as the promising amount of LAB during storage. Provided changes in extruded and fermented WB were determined: fermentation reduced sugar concentration and pH samples with the predominant lactic acid isomer L (+). Also, the viable amount of LAB substrate was greater than 6.0 log10 CFU/g, and no enterobacteria persisted. FBB showing required antimicrobial activity: Shepherd inhibited – 2, sea buckthorn –3, blueberries –5, and raspberries–7 pathogens from the 10 tested. Comparing different groups of beverages produced using different types of FBB, in almost all cases, increasing the amount of FBB improved the overall acceptability. Comparison of the samples showed that most FBB improved the total phenolic compounds (TPC) content (9.0% on average). A relatively high positive correlation was found between TPC and antioxidant activity (r = 0.9919). The study concludes that the newly developed nutraceutical beverages were admissible to consumers, induced positive emotions, and had desirable antimicrobial and antioxidant properties, while they were produced in a sustainable and environmentally friendly method.

Keywords: Beverages, milk permeate, wheat bran, fruit/berry by-products, antimicrobial properties, antioxidant properties, overall acceptability, emotions induced for consumers.

Acknowledgments: The authors gratefully acknowledge the EUREKA Network Project E!13309 "SUSFEETECH" (No. 01.2.2-MITA-K-702-05-0001) and COST Action 18101 SOURDOMICS—Sourdough biotechnology network towards novel, healthier and sustainable food and bioprocesses (https://sourdomics.com/; https://www.cost.eu/ actions/CA18101/).

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Serpin B5 Localization in the Chemoresistant Colorectal Carcinoma Cells

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Abstract

Colorectal cancer is the second by the highest mortality rate cancer worldwide. In the first disease stages it could be removed surgically, but in more developed – chemotherapeutic treatment is required. The combination of 5-fluorouracil (5-FU) and oxaliplatin (OxaPt) is commonly used in a chemotherapy. Although the DNA damage caused by these drugs leads to the programmed cell death – apoptosis, the treatment often becomes complicated because of the intrinsic or acquired chemoresistance, caused by various molecular mechanisms. One of the chemoresistance mechanisms could be altered gene expression of serine protease inhibitors (serpins). Serpin B5 can be extracellular and intracellular protein localized in nucleus, mitochondria or attached to plasma membrane. The molecular functions are determined by localization and it is closely related to cell adhesion, metastasis formation, regulation of apoptosis. It is known that during mitochondrial stress, serpin B5 is transported to the mitochondria, where it competes with cytochrome C for binding to a cardiolipin and induces apoptosis by causing release of cytochrome C into the cytosol[1]. Furthermore, serpin B5 nuclear localization is at least partially dependent on the altered epidermis growth factor receptor (EGFR) signalling pathway[2]. Nuclear serpin B5 isoform acts as transcription factor by binding to chromatin and modulating several genes expression. Moreover, serpin B5 expression is induced by transcription factor p53, which is mostly known as the tumour suppressor and the main player inducing apoptosis.

In this study, we have examined serpin B5 cellular localization in chemotherapeutic drug sensitive colorectal carcinoma cells and compared it with serpin B5 localization in 5-FU or OxaPt chemoresistant colorectal carcinoma cells.

Keywords: Colorectal cancer, serpin B5 localization, chemoresistance.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Role of Phospholipids in Prostate Cancer Development

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Abstract

Prostate cancer (PCa) is one of the leading cancer death in man's world. Its diagnosis is found to be problematic due to the asymptomatic development and morphological diversity. Moreover, the mechanism is still undiscovered. Screening diagnosis is mostly based on determination of prostate specific antigen (PSA), which is found to be tissue but not specific PCa factor. For this reason, new tools are sought for sensitive diagnosis. One of the alternative and is lipidomics performed with the use of mass spectrometry.

The main goal of the study was to perform lipidomic profiling of prostate tissue with the use of two imaging methods: matrix-assisted laser desorption ionization with time-of-flight mass spectrometer and electrospray ionization with triple quadrupole mass spectrometer. For this purpose, profiles of phospholipids were determined in tissue samples obtained from PCa patient (n=40) and non-cancer controls (n=40). Finally, we determined few classes of compounds, namely phosphatidylcholines (PCs), lysophosphatidylcholines (LPCs), sphingomyelins (SMs), and phosphatidylethanolamines (PEs).

The obtained results were analysed with the use of univariate (Mann–Whitney *U*-test) and multivariate statistical analysis (principal component analysis, correlation analysis, Volcano plot, Artificial Neural Network and Random Forest algorithm). Statistical evaluation provided the knowledge about the phospholipids alteration linked with PCa progression. Based on results, PC 16:0/16:1, PC 16:0/18:2, PC 18:0/22:5, PC 18:1/18:2, PC 18:1/20:0, PC 18:1/20:4 and SM d18:1/24:0, were assigned as most discriminative features.

Keywords: Metabolomics**,** prostate cancer, phospholipids; MALDI-ToF/MS; ESI-ToF/MS, GC-MS

Acknowledgments: The study was funded by the National Science Center (NCN, Poland; Grant No. UMO-2016/21/D/ST4/03730) received by Magdalena Buszewska-Forajta.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Ozone Biomonitoring Using Vascular Plants in Cities

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Abstract

Ground-level ozone (O_3) is a strong oxidant that, at elevated concentrations, can negatively affect growth, carbon allocation, phenology and physiological functions of plants. One of the methods for monitoring of ozone (O_3) and its potential harmful effects is biological monitoring (biomonitoring), by observing quantitative changes in plants physically present in a specific environment. O_3 biomonitoring can be active (observing changes in plants transplanted to another area) or passive (observing changes in plants growth in their natural habitat). There is a high variation of O_3 sensitivity within species, cultivars/varieties, and genotypes/biotypes, suggesting that not all plants are suitable for biomonitoring. Several biomonitoring programs exist. For instance, in 1985, the International Co-operative Programme on Assessment and Monitoring of Air Pollution Effects on Forests (ICP Forests) was established. One of the aims of this network is biomonitoring of O_3 all over the Europe. A limitation is that all these networks have different methodologies. O_3 biomonitoring with homogeneous methodology might be a powerful tool for application worldwide. The aim of this work is to overview different methodologies and suggests the best solution for O_3 biomonitoring in cities.

O_3 biomonitoring could be also incorporated within new-generation biomonitoring projects of citizen science.

Keywords: Biomonitoring, ozone, vascular plants, citizen science.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

POSTER PRESENTATIONS

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Endophytic *Bacillus cereus* Group Bacteria Isolates Promote *Nicotiana tabacum* L. Shoot Growth *in Vitro*

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Abstract

In vitro plant tissue culture is protected from common environmental stressors but still faces different unfavorable conditions such as mechanical damage, osmotic shock, or phytohormone imbalance that could be detrimental to culture viability, growth efficiency, and genetic stability [1, 2]. Often *in vitro* plant cultures have been considered axenic, but recent studies revealed a presence of diverse endophytic bacteria community whose composition depends on explant origin and cultivation conditions [3]. Engineering of the endophytic microbiome of *in vitro* plant tissues has the potential to improve their acclimation to stress induced by the *in vitro* conditions [4]. Therefore, the aim of this study was to identify tobacco endophytic bacteria isolates capable of promoting biomass accumulation of *in vitro* tobacco shoots. Forty-nine endophytic bacteria isolates were obtained from greenhouse-grown tobacco plant shoots and were identified as *Bacillus sp.* and *Pseudomonas sp*. Twenty-one of the isolates were used to study endophytic bacteria effect on tobacco shoot growth *in vitro*. The shoots were inoculated with bacterial suspension and shoot fresh weight was assessed after 3 weeks of co-cultivation. Isolates belonging to the *Bacillus cereus* group, *B. mobilis* (Nt.3.2), *B. mycoides* (Nt.10.1), B. *thuringiensis* (Nt.18, Nt.20.2) promoted shoot growth 11% to 21%. Interestingly, fresh weight of shoots inoculated with isolates of the same species, such as *B. mobilis* (Nt.14.2), *B. mycoides* (Nt.25, Nt.12.1), and *B. thuringiensis* (Nt.37), was reduced 4% to 7% or the bacteria had no significant effect on shoot growth. Inoculation with the remaining isolates of *B. aryabhattai*, *B. marisflavi*, *B. simplex* and *P. koreensis* had no significant effect on biomass accumulation or was detrimental to tobacco shoot growth. The results suggest that isolates with a contrasting effect on shoot growth represent the capability of multiple bacterial strains to establish different interactions with the host. The isolates of the *Bacillus cereus* group with shoot growth-promoting properties have a potential application to improve the growth of plant tissue culture *in vitro*, and further studies based on their interaction with plant and host specificity would aid practical implementation.

Keywords: *Bacillus* sp., endophytic bacteria, microbiome engineering, plant stress.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Recovery of Total Phenolic Content of the Flower Extract of *Opuntia ficus* indica

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Abstract

In this work, 3 variables were employed in order to optimize the recovery of total phenolic content (TPC) in the extract of *Opuntia ficus indica* flower (OFIF). It is about the solvent concentration which varies from 20 to 80% of ethanol, the time ranging from 60 to 360 min and the liquid-to-solid ratio from 10 to 60 mL/g, and maceration was used as extraction method. The best TPC (35.22 mg Gallic Acid Equivalent/g Dry Weight (DW)) was found using ethanol at 35.2% during 60 min with a ratio of 1g/39.25 mL. Using these conditions, the total flavonoid and betalain contents of the extract obtained were 1.97±0.01 mg Quercetin Equivalent/g DW and 0.63 ± 0.01 mg/g of DW, respectively. Furthermore, the antioxidant capacities of this extract were 971.6±23.9, 1.47±0.5 and 3.45±0.1 mg/g Trolox® Equivalent in DPPH, ferric reducing power and molybdate assays, respectively.

Keywords: Total phenolic content, recovery, *Opuntia ficus indica* flower, antioxidant activity

Acknowledgments: This investigation was endorsed by a allowance from the Ministry of High Education and Scientific Research of Algeria. We thank everyone who contributed to the realization of the current work.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Evaluation of Antibacterial Activity and pH Change in Herbal Drinks Fermented by Kombucha Culture

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Abstract

Kombucha is unique because of symbiotic culture of bacteria and yeast (SCOBY), which carries out the fermentation process [1]. Traditional kombucha is obtained by fermenting sweetened black tea of *Camellia sinensis* L. Other plants and fruits (e.g., winter savory, peppermint, elderberry, quince) also can be used for fermentation with kombucha culture [2]. According to the scientific literature, SCOBY contains *Acetobacter* spp., *Gluconobacter* spp., *Lactobacillus* spp., and *Dekkera* spp., *Saccharomyces* spp., and others [1]. During fermentation process, various organic acids are formed e.g., acetic, D-glucuronic, gluconic, lactic, citric, oxalic acids. Possibly due to these acids and low pH the kombucha drink has antimicrobial properties against *Staphylococcus aureus*, *Salmonella typhimurium*, *Escherichia coli* and various other pathogens and may be very healthful to maintain human health [2]. The aim of this study was to evaluate antibacterial activity and pH change of kombucha beverage prepared from different herbs after 1, 2, 3 and 8 weeks of fermentation. Sweetened water infusions from lemon balm (*Melissa officinalis* L.), nettle (*Urtica dioica* L.), fireweed (*Chamaenerion angustifolium* L.), rose hips (*Rosa canina* L.), Ceylon black tea (*Camellia sinensis* L.) and their mixture were selected for fermentation process. Antibacterial activity of fermented drinks was determined against gram-positive *Bacillus mojavensis* and *Bacillus megaterium*, gram-negative *Azotobacter vinelandii* using agar well diffusion method [3]. The pH of prepared drinks was determined with the use of a calibrated pH-meter. During 8 weeks of fermentation, the pH values reduced from 7.98±0.01 to 2.07±0.01. The highest and the lowest pH values were found in nettle tea before fermentation and in fireweed kombucha after 8 weeks of fermentation, respectively. Antibacterial activity of kombucha samples ranged from 1.0±0.0 mg/ml to 39.1±0.1 mg/ml (expressed as acetic acid equivalents) in all samples in 8 weeks fermented drinks. The highest antibacterial activity was determined against *Bacillus mojavensis* in the nettle kombucha after 8 weeks of fermentation. Antibacterial activity was not observed after 1 week of fermentation. This study showed that there is no statistically significant difference between pH value and herbal tea type, but there is statistically significant difference between pH and fermentation duration. The same tendency is with antibacterial activity.

Keywords: Antibacterial activity, kombucha, herbals.

Acknowledgments: Financial support from Research Council of Lithuania project No. 09.3.3-LMT-K-712-22-0104 is acknowledged.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Anthocyanin Profiles in Fruits of Lithuanian Heirloom Apple Cultivars

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Abstract

The heirloom apple cultivars, which chemical composition has not been studied are replacing by the cultivars of the commercial apple grown in the industrial gardens. Therefore, is significant to determine the anthocyanins composition variability by old cultivars of the apple, which have long cultivated traditions in Lithuania. The aim of the study was to investigate variability of the qualitative and quantitative composition of anthocyanins in the samples of heirloom apple cultivars.

The study included 22 heirloom apple cultivars, of which 21 (except for 'Golden russet') are included in the List of the National Plant Genetic Resources. During the analysis, 2.5 g of lyophilizate powder (exact weight) was weighed, added to 25 mL of 2% hydrochloric acid solution in 70% (v/v) ethanol and was extracted in a ultrasonic bath at room temperature for 20 min. The variability in the qualitative and quantitative content of anthocyanins was determined by the validated UPLC-PDA method described by Vilkickyte et al. [1].

The sum of the identified anthocyanins in the samples of heirloom apple cultivars included in the collection of the Lithuanian heritage of genetic resources varied from 5.78±0.01 μg/g DW to 85.43±0.53 μg/g DW. The highest amounts of sum of the identified anthocyanins (85.43±0.53 μg/g DW) was determined in fruit samples of the 'Panemunės baltasis' apple cultivar, the lowest (5.78±0.01 μg/g DW) 'Beržininkų ananasinis' apple cultivar. The anthocyanins were identified and quantified in the analyzed fruit samples of heirloom apple cultivars grown in Lithuania: cyanidin-3-O-galactoside, cyanidin-3-O-glucoside, cyanidin-3-O-arabinoside, and cyanidine. Cyanidin-3-O-galactoside was the predominant anthocyanins and accounted for 93.4% of the total amount of the identified and quantified anthocyanins the apple samples. The highest amount of cyanidin-3-O-galactoside (80.48±0.12 μg/g DW) was found in fruit samples of the 'Panemunės baltasis' apple cultivar.

According to their amount in fruit samples of heirloom apple cultivars grown in Lithuanian orchards, the compounds of the anthocyanins can be arranged in the following order: cyanidin-3-O-galactoside >cyanidine >cyanidin-3-O-glucoside >cyanidin-3-O-arabinoside. The results of our study will enable a wider cultivation of heirloom apple cultivars in gardens and collections and will help consumers to obtain and use apples with a known chemical composition of anthocyanins, which determine the use of apples in the healthy food chain and the development of innovative food products.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Enhancement of Transungual Delivery of Amorolfine *ex vivo*

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Abstract

Transungual drug delivery is defined as system involving drug transport through the nail to obtain targeted drug delivery for treating nail disease such as onychomycosis [1]. However, topical therapy of onychomycosis is considered highly challenging due to hard nail plate structure [2]. Therefore, chemical and physical enhancement methods may be used to improve the transungual delivery of antifungal agents.

The aim of this study was to evaluate chemical and physical enhancers' impact on transungual delivery of amorolfine. To achieve this goal, human nail clippings were used as human nail model membrane, while "wetted cotton ball" mimic nail bed system. Different concentrations of various chemicals were incorporated as enhancers into nail lacquers formulations. To determine effect of non-invasive physical enhancement method, ultrasound was applied. Quantitative analysis of penetrated antifungal agent was performed by UPLC.

It was determined that film-forming polymer type had influence on accumulation of amorolfine. A film-forming polymer Eudragit E100 had statistically significant influence on accumulation of amorolfine in acceptor phase. While, after ultrasound applying, nail lacquers with Eudragit RL had shown a significant influence on accumulation of amorolfine in acceptor phase. The obtained results allowed to evaluate that urea with thioglycolic acid nail lacquer is the most suitable chemical enhancer to improve the transungual delivery of amorolfine to acceptor phase. It was determined that simultaneous used ultrasound had increased accumulation rates of amorolfine in acceptor phase.

The current study demonstrated that more efficient amorolfine hydrochloride transungual delivery could be achieved when physical and chemical enhancers were used in combination.

Keywords: Onychomycosis, amorolfine hydrochloride, chemical enhancer, physical enhancer, nail lacquer.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Correlations Between the Different Chocolate Samples Overall Acceptability and Emotions Induced by Chocolate Samples Taste, Smell and Package

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Abstract

The aim of this study was to evaluate correlations between the different chocolate (Choc) samples overall acceptability (OA) and emotions (EM) induced for consumers by Choc taste, smell, and package. Three different Choc sample groups were tested. All Choc were originally packed, and first of all, EM induced by the package were evaluated. During the second step of the experiment, EM induced by the Choc samples smell were evaluated. Third, analysis of the Choc OA was performed, as well as EM induced by Choc taste were fixed. The OA of the Choc samples was carried out using a 10-point scale, ranging from 0 (extremely dislike) to 10 (extremely like). Also, the Choc samples were tested by applying *FaceReader 5* software (Noldus Information Technology, Wageningen, The Netherlands), scaling the 8 EM patterns (neutral, happy, sad, angry, surprised, scared, disgusted, contempt). The correlation coefficients were calculated using the statistical package SPSS for Windows (v15.0, SPSS, Chicago, Illinois, USA). The results were recognised as statistically significant at $p \leq 0.05$. It was found that in comparing different Choc samples OA, significant differences were not established (on average, OA of the Choc samples was 9.8 points). However, in comparing induced EM by the different Choc, the highest "happy" EM was fixed by tedting Choc No. 2 taste, smell and packaging. Other analysed samples (No. 1 and No. 3) induced EM "happy" intensity was by 1.2 and 1.5%, respectively, lower for the Choc taste, by 3.3 and 4.8%, respectively, lower for the Choc smell, and by 2.6 and 6.6%, respectively, lower for the Choc packaging. Correlations between EM induced by Choc smell and overall acceptability showed that by increasing Choc OA, intensiveness of EM "angry" was decreased ($r = -0.374$). However, weak positive correlations between Choc OA and EM "sad" and "contempt" were found ($r = -0.319$ and $r = -0.315$, respectively). Finally, it can be stated that the fixing of the EM induced for consumers is very promising method, which showed higher sensibility than OA standard method. However, for the final statement, more research should be performed, as well as more samples and more judges must be involved.

Keywords: Chocolate, overall acceptability, emotions, smell and taste.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Chemical Properties of Biologically Active Compounds of The Medicinal Plant *Potentilla fruticosa L.*

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Abstract

The World Health Organization states that medicinal (aromatic) plants (MAPs) and herbal medicines are equivalent to synthetic ones used to improve health [3]. Research has shown that the biologically active substances accumulated in plants - glycosides, alkaloids, essential oils, vitamins, yeasts - have a pharmacological effect. Herbal medicines have a milder pharmacological effect on the body, cause fewer side effects on the body than the synthetic ones. Shrubby cinquefoil preparations have antimicrobial, antiviral, antioxidant, astringent, anti-diarrheal and antihaemorrhagic effects [1].

A medicinal plant of the *Potentilla fruticosa* L. *Rosaceae* Juss. (which chemical properties have not been sufficiently studied yet) family was selected as the study object.

The aim of the study - to perform a comparative analysis of the scientific data on the accumulation of biologically active compounds, their chemical composition and the quantity and quality of medicinal plant raw material and pharmacological effects.

In order to study chemical properties and pharmacognostic effects of medicinal plants, the evaluation of research data presented in literature sources and databases.

Potentilla fruticosa L. is a strongly branched shrub, up to 150 cm tall. Blooms from May to November. Fruit - ovoid, russet brown achene [2].

Plant distribution area: Eastern Siberia, Far East, Mongolia, Northern China, Japan; Europe, Western Siberia, Central and Southeast Asia, North America.

It does not grow on its own in Lithuania, it has been introduced and researched since 1981 in Scientific Sector of Medicinal and Aromatic Plants, Scientific Department, Botanical Garden of Vytautas Magnus University [2].

The raw material *Potentillae fruticosae folium cum flore* contains polyphenols: flavonoids (167.0-327.0 mg%), phenolcarboxylic acids (caffeic, ferulic, sinapic, ellagic, coumaric, chlorogenic; up to 0,40 %), yeasts (10.0-14.0 mg%), chlorophyll (a + b) (up to 200.0 mg%), carotenoids, vitamin C (up to 150.0 mg%) [1].

Potentillae fruticosae folium cum flore is characterized by a variety of biologically active compounds that have a wide range of pharmacological effects on the body. Research is underway to support new technologies for the production of herbal preparations.

Keywords: Medicinal (aromatic) plants, *Potentilla fruticosa* L., biologically active compounds.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Activity of New Sunitinib Derivatives on Colon Cancer Cell Migration in Normoxia and Hypoxia

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Abstract

Despite the progress in cancer treatment, there is still a need for advanced and more effective anticancer agents. Sunitinib is one of the kinase inhibitors which is used to treat kidney and gastrointestinal cancers after failure to the first-choice treatments [1]. No specific drugs have been developed to inhibit metastasis, and only a few drugs used to treat cancer have antimetastatic activity [2]. There are some evidences that kinase inhibitors could inhibit cancer cell migration [3]. As well as migration, the microenvironment is very important for the treatment effectiveness. Hypoxia is one of the main tumor microenvironment characteristics, which is important for drug efficiency [4]. Therefore, the aim of our research was to evaluate the antimigratory activity of new sunitinib derivatives synthesized at Cagliari University (Italy) in normoxia and hypoxia conditions.

In our study we used two human colon cancer cell lines HT-29 and HCT116. Single cell migration assay was used to evaluate compound effect on cell migration. Compounds were used at 10% of their established EC_{50} values. Activity of tested compounds was compared to the clinically used drug sunitinib.

In normoxia conditions, tested compounds 1, 6, 7 and sunitinib statistically significantly stronger reduced the migration of cells from both cancer cell lines compared to control. The most active one was compound 7. It inhibited HT-29 cell migration by 2.3 times and HCT116 cell migration by 1.5 times compared to control. Also, compound 7 was 1.2 times more active against HT-29 cells and possessed a similar activity as sunitinib on HCT116 cell migration in normoxia. In hypoxia conditions, there were no significant differences between the activity of compounds and control groups.

In conclusion, sunitinib derivative 7 is more effective than sunitinib *in vitro* in normoxia conditions for HT-29 line cells and could be developed further as anticancer agent against colon cancer.

Keywords: Sunitinib derivatives, colon cancer, single cell migration.

Acknowledgments: This project has received funding from European Social Fund (project No. 09.3.3-LMT-K-712-22-0102) under grant agreement with the Research Council of Lithuania (LMTLT).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Investigation on Intracellular Compounds and Glutathione Effect on Cell Viability Post Electroporation

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Abstract

Electroporation (EP) is - a process, which using high-voltage (HV) short (ns, μs or ms) electrical pulses induce alterations in the cells plasma membrane that facilitates nonselective molecular transfer into and from the cells [1].

Using very high-voltage or to many electric field pulses cause cell death by irreversible electroporation (IrEP) which hence is divided into programmed cell death (prolonged cell death) or cell death due to the loss of plasma membrane integrity (instant cell death) [2]. However, it is still not well understood the fact why and how cells die in prolonged cell death manner. Importantly, it was proved that molecules such as proteins either RNA or DNA can leak out from the cells through the electroporated plasma membrane. Here we hypothesize that the loss of intracellular biomolecules may be one of the reasons that the viability of electroporated cells decreases [3]. The aim of this study was to investigate whether the loss of the cells may be diminished by electroporating cells in the EP supernatant (EP SN) that was derived from irreversibly electroporated cells or in presence of natural antioxidant glutathione.

For experiments, we used Chinese hamster ovary (CHO) and a breast cancer (4T1) cell lines. Electroporation was performed by using 9 high voltage (HV) 1200 V/cm electric field pulses of a duration 100 μs repeating at 1 Hz. EP SN was made from CHO or 4T1 cell lines. Cells were electroporated in the presence or absence of glutathione or in EP SN from CHO or 4T1 cells lines. The viability of cells was assayed 24 and 48 hours after the experiment by Flow cytometry, MTT assay and 6 days after experiment by colony formation test for assessment of the final cell viability.

Results have revealed that the addition of glutathione alone into EP medium before EP was capable of increasing the viability of CHO cells post electroporation by 50 %. It is important to note that the cell viability was increases from 40 % - 60% if cells were electroporated EP SN medium in no matter from which cell line EP SN was obtained.

In conclusion, our results demonstrate that the viability of cells can be increased by adding glutathione into EP medium or electroporating cells in EP medium supplemented with intracellular compounds obtained from irreversibly electroporated cells. Our findings suggest that increased intracellular levels of ROS and the loss of vital intracellular compounds play a substantial role in inducing cell death after application of pulsed electric fields so called electroporation.

Keywords: Irreversible electroporation, cell viability, electroporation supernatant, glutathione.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Development of Blue Phosphorescent Organic Light-Emitting Diodes Using Novel Exciton-Blocking/Hole Transporting Materials

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Abstract

Modern displays and lightings can be developed using organic light-emitting diodes (OLEDs). Searching of new materials with improved properties such as charge mobility or exciton-blocking ability can lead to further enhancements of OLED performance. In this work, six carbazole derivatives containing different number of methoxy substituents were developed and characterized towards OLED applications [1]. Compounds showed very high triplet (T1) energies (2.85-3.05 eV) were found in all cases, exhibiting continuous- but small variations upon methoxy substitutions. Ionization potentials of compounds deduced from photoelectron emission spectra were in the wide range from 5.29 to 5.98 eV. They demonstrated hole-mobility values ranging between ca. 8.5×10^{-7}-2.8×10^{-3} cm^2/Vs at electric fields of 0.5×10^5-5.5×10^5 V/cm according to time of flight (TOF) measurements. These novel compounds were efficiently used as multifunctional hole-transporting, exciton-blocking and electron-blocking materials in blue phosphorescent organic light-emitting devices due to their quite high T1 energy levels and high hole mobilities. Fabricated devices showed high maximum current-, power-, and external quantum efficiencies of 47.6 cd/A, 36 lm/W, and 21.1%, respectively. These compounds have also high potential to be used in OLEDs as host- and for exciton blocking materials.

Keywords: Hole mobility, exciton blocking, electroluminescence, organic light-emitting diode.

Acknowledgments: This project has received funding from European Regional Development Fund (project No 01.2.2-LMT-K-718-01-0015) under grant agreement with the Research Council of Lithuania (LMTLT).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

SDS-PAGE Gel Electrophoresis of Proteins Extracted from *Glycyrrhiza glabra* (L.) Roots

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Abstract

Glycyrrhiza glabra (L.) is a perennial, herbaceous, legume plant and it belongs to the *Fabaceae* family. Roots of this herb have numerous biologically active constituents: saponins, flavonoids, phenolic compounds, tannins, carbohydrates, vitamins, amino acids and other substances. Due to its abundant composition, licorice is widely used in medicine [1] (e. g. 2% licorice roots extract, which contains 20% of 18β-glycyrrhetinic acid, is effective in the treatment of atopic dermatitis [2]). Moreover, *G. glabra* roots have antibacterial, antimicrobial, anti-inflammatory, antioxidant, antiviral, antimalarial and other important properties. Approved, that various amino acids have been found in the roots of licorice: serine, histidine, aspartic acid, glycine, tyrosine, threonine, glutamate, isoleucine, leucine, valine, proline, lysine, phenylalanine [3]. Proline has been found predominantly in fresh plant roots [4]. In this research, protein molecular mass was evaluated by SDS-PAGE. Materials: fresh *Glycyrrhiza glabra* (L.) roots collected in Vytautas Magnus University Botanical garden. Methods: protein extraction was made using phosphate-buffered saline (PBS, prepared of 7.6 mM Na_2HPO_4, 2.3 mM NaH_2PO_4, 0.15 mM NaCl, pH=7.4); protein precipitation using ammonium sulphate; resuspension using PBS, pH=7.4; spectrophotometric (at 660 nm wavelength) protein determination by Lowry method; sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE), using 10% polyacrylamide as the separating gel. Molecular mass of determined proteins was about 10-15 kDa. SDS-PAGE gel electrophoresis was the suitable method for identification molecular mass of protein extracted from *Glycyrrhiza glabra* (L.) roots.

Keywords: *Glycyrrhiza glabra*, licorice, gel electrophoresis, protein purification, protein extraction.

Acknowledgments: The author is grateful to Prof., PhD of Biomedical Science N. Savickienė, Doctor of Pharmacy G. Balčiūnaitė-Murzienė and Habil. Dr. of Biology, Doctor of Pharmacy Ona Ragažinskienė for their help and support.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Optimisation of Extraction Conditions for Phenolic Compounds from *Cirsium vulgare* Leaves

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Abstract

Bull thistle (*Cirsium vulgare*) is a species of perennial herb in the family *Asteraceae*. They have a self-supporting growth form. The main secondary metabolites of *Cirsium* species were reported to be flavonoids, tannins, sterols, triterpenes and also phenolic acids [1]. Bull thistle as a medical plant has not previously been used in modern medicine, but it was used in folk medicine. Nowadays, there is some studies about this herb hepatoprotective effect. Studies shows that extracts made from bull thistle acute liver damage in rat liver cells. Hexane extracts made form bull thistle totally prevented the acute TC-induced liver damage [2]. Some studies identify antioxidant activity of Cirsium vulgare ethanolic and methanolic extracts. Studies shows that these extracts could be used for both antibacterial and anti – cancer activity or even more in the future [3]. The aim of our research was to select the most appropriate extraction conditions and determine the main phenolic compounds in *Cirsium vulgare* extracts. The extraction time and extrahent were recorded during the study. The object of research was bull thistle leaves. Plant leaves were harvested and dried, and then extracts were prepared using various extraction media. Different types of extraction methods such as maceration, ultrasound – assisted analysis and thermal hydrolysis under reflux were used for the determination of phenolic compounds. The best results were achieved with 50% (v/v) ethanol, because of its stability and best extraction of phenolic compounds. During HPLC-PDA analysis five main phenolic compounds such as rutin, apigenin–7–O–glucoside, hyperoside, isoquercitrin and chlorogenic acid were found in extracts. The total average amount of phenolic compounds varied from 0.0526 mg/g to 13.098 mg/g. Most phenolic compounds were detected in the samples, which were obtained throughout thermal hydrolysis under reflux for 1.5 h.

Keywords: *Cirsium vulgare*, HPLC – PDA, phenolic compounds, extraction.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Characterization of Bioactive Compounds Extracted from By-products of *Matricaria recutita* Essential Oil Distillation Processes with Potential Use as Cosmetics Ingredients

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Abstract

Increasing consumer demand for herbal health products, has led medicinal plant growers to not only offer raw material of medicinal plants, but to also put into industrial cycle production essential oils from their own biologically grown aromatic plants. For example, industrial extraction of essential from chamomile by steam distillation, resulting in 99.9% of the so-called herbal waste remaining from the green mass used in the process. This large volume of by-product generated during distillation is of growing concern. Management of herbal waste products are sparsely described. Valorization of residues from agro-industrial obtained by-products is an opportunity to obtain profit in a sustainable way. Interest in bioactive compounds obtainable from natural sources has increased considerably in recent years mainly from the industry of food, medicine, and cosmetics. Cosmetic industry is interested in polyphenols as they can be applied to promote various aspects of skin health. This highly diverse group of compounds has diverse biological activities as well - UV protection, anti-oxidant and anti-inflammatory activity, regulation of skin microbiome, skin extracellular matrix production [1,2]. The objective of this research was to characterize bioactive compounds for potential use in cosmetics extracted from by-products of *Matricaria recutita* essential oil distillation process. A hot water extraction was used to obtain the extract of bioactive compounds after the atmospheric steam distillation. The distillation was followed by extraction at 85ºC for 45 minutes. Chemical characterization was performed using high performance liquid chromatography – time of flight mass spectrometry. In total seventy-five compounds were detected in the extracts, mainly apigenin, quercetin and kaempferol derivatives, compounds with high antioxidant potential. Cytotoxicity assays demonstrated a high safety of the extract. Proof for UV-protecting, proliferation stimulating and antioxidative activities was obtained in *in vitro* tests using skin cell cultures. The results obtained confirm that extracts from by-products of *Matricaria recutita* essential oil distillation processes are a good source of valuable biologically active substances with a high potential to be used as skin protecting and regenerating ingredients in cosmetics and medical devices.

Keywords: *Matricaria recutita*, by-products, skin protection, skin regeneration.

Acknowledgments: The work has been supported by ERDF project 1.1.1.1/19/A/075 "Biorefinery approach for the development of bioactive cosmetic ingredients from by-products of medicinal plant processing and plant cell cultivation".

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Quality Evaluation of the Semisolid Formulations with Bee Products

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Abstract

Dermatological products of natural origin attract more and more attention nowadays, so researchers in the scientific community seek to design stable products to meet those requirements. The most important thing is to develop such a system which would have natural base. As the main ingredients for modeling semisolid preparations there were beeswax, olive oil, propolis and honey. The experimental studies showed that the soft extract of propolis contains the largest amounts of phenolic compounds, and it is suitable to form the stable semisolid systems. The following aim was formed: to model semisolid formulations with bee products and evaluate their quality by applying a microbiological activity and biopharmaceutical research *in vitro*. At the time of the organoleptic evaluation various sensory characteristics of formulations, such as easy spreadable, hard washable, no phase separation, yellow colour, pleasant odour and pH values (4.00-6.5) were estimated. The release test was performed using the modified vertical type diffusion cell. The ointment sample (1.00 g) was placed into the cell with a hydrophilic dialysis membrane, temperature of acceptor medium was maintained at 37±0.2 °C. The samples from the acceptor solution were taken at 1, 2, 4, 6 h and were replaced with the same volume of fresh acceptor solution. The samples were analyzed by high performance liquid chromatography (HPLC) methods. Resistance to preparations from natural materials was examined in Mueller-Hinton agar with standard cultures of *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Escherichia coli*, *Enterrococcus faecalis*, *Proteus mirabilis*, *Bacillus subtilis*, *Bacillus cereus*, and *Candida albicans*. The research has established that the composition of bases has an impact on their stability, the releases of the active substances and acceptable organoleptic properties. The experiment demonstrated that the increase of the quantity of wax cause the lower release of phenolic compounds from ointment. The studies have found that the formulations have antimicrobial effects.

Keywords: Yellow beeswax, honey, propolis, *in vitro* release, antimicrobial activity.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Influence of Culture Conditions and Methods of Sample Preparation on the Level of Bacterial Identification Using MALDI-TOF MS Technique

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Abstract

Microorganisms inhabit all known ecosystems: from the natural environment to the human body. Among them, there are many pathogenic species that cause infections that threaten human health and life. Fast and accurate identification of bacteria in clinical microbiology is extremely important, among others, when implementing and individual antibiotic therapy. One of the most frequently used methods for the identification of microorganisms is the MALDI-TOF MS technique. The accuracy of identification is greatly influenced not only by the method of analysis used, but also by the selection of conditions for bacterial culture and sample preparation. This study investigated three factors that may affect the quality of microorganism identification. The first was the type of growing medium. Solid media (BHI, MHA and selective universals: BCP, VRE) and liquid media (TSB and BD BACTEC) were used. The second variable was the incubation time (4, 6, 24 h), and the third was the method of preparing bacterial extracts for MALDI-TOF MS analysis (standard method according to the manufacturer's protocol and the method using the Sepsityper kit). The obtained results indicate that the dyes contained in the selection media may deteriorate the quality of mass spectra. By analyzing the effect of liquid microbiological medium, the incubation time and the method of preparation of protein extracts for MALDI-TOF MS analysis, it can be concluded that a 4-hour incubation of bacteria allows for the correct identification of pathogens at the level of species. The results show, however, that the optimal incubation time for pathogenic bacterial strains is 6 hours on the liquid TSB medium. The most effective method of preparing bacterial extracts turned out to be the standard method performed according to the manufacturer's protocol. The conducted research suggests the possibility of faster and more accurate microbiological diagnostics using different growth media and shorter incubation time of bacteria.

Keywords: Bacteria identification, MALDI-TOF MS, matrix-assisted laser desorption/ionization, mass spectra.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Tetrahydrocarbazolyl- and 2-Phenylindolyl-Substituted Benzophenone Compounds for Optoelectronic Devices

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Abstract

In order to improve the efficiency of organic light emitting diodes and other optoelectronic devices, it is necessary to look for new structures of electroactive organic compounds with a set of required properties. Previous studies show that bipolar benzophenone based compounds with various donor moieties exhibit useful properties such as bipolar charge transport, triplet-triplet annihilation, thermally activated delayed fluorescence or aggregation-induced emission enhancement.

Two electroactive bipolar benzophenone derivatives were designed and synthesized. Rarely used tetrahydrocarbazole and 2-phenylindole were chosen as donor moieties. Nucleophilic aromatic substitution reaction of 4,4′-difluorobenzophenone and 1,2,3,4-tetrahydrocarbazole or 2-phenylindole was carried out under the action of potassium tert-butoxide in dimethylsulfoxide to obtain the target compounds. The yields of the products were 66-68%. Differential scanning calorimetry showed that tetrahydrocarbazole-based benzophenone compound formed molecular glass. Glass transition temperature was of 82 ºC. Both of the derivatives showed high triplet energy values exceeding 2.8 eV. The ionization potentials estimated by electron photoemission method in air were of 5.54 and 5.53 eV. The derivative with 2-phenylindolyl-substituent exhibited aggregation induced emission enhancement.

Keywords: Tetrahydrocarbazolyl, phenylindolyl, benzophenone, bipolar, organic light emitting diode.

Acknowledgments: this project has received funding from European Regional Development Fund (project No 01.2.2-LMT-K-718-01-0015) under grant agreement with the Research Council of Lithuania (LMTLT).

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Effect of Caffeic Acid Phenethyl Ester on Lactate Dehydrogenase Activity After Rat Kidney Ischemia/reperfusion *in Vivo*

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Abstract

Lactate dehydrogenase (also called lactic acid dehydrogenase, or LDH) is an enzyme found in almost all body tissues. Although LDH is abundant in tissue cells, blood levels of the enzyme are normally low. However, when tissues are damaged by injury or disease, they release more LDH into the bloodstream. Kidney ischemia/reperfusion injury is a condition that can cause increased LDH in the blood [1]. Our previous studies revealed, that kidney ischemia/reperfusion *in vivo* suppress mitochondrial function and causes the damage of mitochondrial outer membrane and release of cytochrome c [2]. For improving of ischemia/reperfusion tolerance, recently a lot of attention intended for natural compounds. Whereas caffeic acid phenethyl ester (CAPE), a natural phenolic chemical compound, an active component of propolis extract and of the various plants with broad spectrum of biological activities (anti-inflammatory, antioxidant, immunomodulating, cadioprotective, neuroprotective and others [3]) partially protects mitochondria from ischemia/reperfusion damage [4]. The aim of this study was to investigate the effect of CAPE on the changes in LDH activity in *in vivo* ischemia/reperfusion rat kidney model.

During the experiment rats were injected into the tail vein with CAPE (22 mg/kg, 90 min prior to ischemia/reperfusion). Warm renal ischemia (20 min, 30 min, 40 min and 60 min) in rats was induced by the clips around renal artery. Then the clips were removed and reperfusion was performed for 30 minutes. Cytosolic fractions from rat's kidneys were isolated by the method of differential centrifugation. Lactate dehydrogenase (LDH) activity in cytosolic fractions was measured spectrophotometrically according NADH oxidation rate at 340 nm.

Our results demonstrate that ischemia (20-60 min)/reperfusion 30 min induced damage of kidney cells and as consequence of that glycolysis enzyme LDH was released into bloodstream. Statistically significant result was after ischemia 40 min/reperfusion 30 min, when LDH activity decreased by 29% as compared to control group ($p<0.05$). CAPE (22 mg/kg) protected kidney after ischemia 20 min, 30 min, 40 min /reperfusion 30 min and completely restored the activity of LDH ($p<0.05$).

Kidney ischemia/reperfusion leads to LDH decrease in the cytosolic fractions. CAPE protected kidney from release of LDH and from necrotic cell death and might be beneficial as promising antioxidant agent.

Keywords: LDH, kidney, ischemia/reperfusion, CAPE, antioxidant, cell death.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Chemical Composition of Nanovesicles Produced by Fruit Cells of Lingonberies (*Vaccinium vitis-idaea L.)*

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Abstract

Lingonberry (*Vaccinium vitis-idaea L.*) fruits are widely used in the human diet and traditional medicine due to nutritional and health improvement benefits. This is because of their richness in bioactive phenolic compounds. The object of this research were nanovesicles produced by fruit cells of lingonberies (*Vaccinium vitis-idaea L.*) The aim was to measure total quantity of phenolic and proanthocyanidins compounds which were evaluated by HPLC method, also to determine antioxidant activity amount by applying spectrophotometric analysis methods: FRAP and ABTS. HPLC results showed 25 different compounds. The sum of identified phenolic compounds was 78.37 mg/g. The most abundant phenolic compounds were benzoic acid (21.46±5.44 mg/ml), catechin (14.416±2.41 mg/ml), following by procyanidin B1, B3 and cyanidin-3-galactoside (7.66±1.53, 5.74±1.18, and 4.64±0.57 mg/ml, respectively). FRAP showed average free radical scavenging capacity in nanovesicles produced by fruit cells of lingonberries samples as 798.41±70.49 μmol/l, while average antioxidant activity using ABTS method was 1041.17±60.57 μmol/l. Our results suggest that nanovesicles produced by fruit cells of lingonberries (*Vaccinium vitis-idaea L.*) are promising health therapeutic candidates in the pharmaceutical industries because of phenolics, and further research of composition and activity is required.

Keywords: Lingonberry, phenols, antioxidant.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Expression of SR Protein Kinases in Brain Cell Lines

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Abstract

Alternative splicing that produces multiple mRNAs from a single gene is an important mechanism in the expansion of proteome diversity in eukaryotic cells [1]. Especially alternative splicing contributes to cells' adaptability to altered cellular microenvironment – hypoxia [2]. Hypoxia is known to induce cell survival and promote the initiation and the progression of many human diseases, including neurodegenerative disorders [3].

The splicing process is carried out by the spliceosome and numerous splicing factors, including the serine and arginine-rich (SR) proteins [3]. A key event is the phosphorylation of SR proteins by multiple kinases that regulates their subcellular localization and interactions with target transcripts and other proteins. Four members of the CLK kinases that phosphorylate SR proteins significantly contribute to alternative splicing regulation [4, 5]. In this study, we have investigated the influence of hypoxia on the expression of CLK1-4 kinases in two different brain cell lines.

Keywords: Hypoxia, alternative splicing, CLK1-4 kinases.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Functionality of Oak (*Quercus* spp.) Parts and Possible Prospects for Their Usage: a Review

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Abstract

Oaks (*Quercus* spp.) are perennial, widespread throughout the Northern Hemisphere trees of whose certain parts can have a positive impact on human health and other areas like food, cosmetics. At these times, *Quercus* species are becoming reliable objects for fundamental studies due to the biological activities of their various parts (bark, leaves and acorns). It is known that the properties of *Quercus* bark have been used in ancient traditional medicine to treat human burns, infections, and gastrointestinal diseases [1]. Nowadays, studies show that oak bark extracts can be used as an antifungal agent and as a potential medicament against certain cancer types [2]. In addition, studies show that some species show antidiabetic activity and can cause improvement in energy, fatigue, and stress levels [3]. Oak fruits – acorns have a lot of potential in the field of pharmacy, as many studies demonstrate antioxidant and antibacterial activities of various *Quercus* species, as well as anti-inflammatory, antifungal or antiviral activities. Even more promising results are being achieved in area of antiproliferative activity; the suppression effect of acorns extract in cancer cells proliferation through induction of early apoptosis was determined [4]. Furthermore, studies have assessed acorns influence on anti-obesity effect: acorns extracts can reduce lipids droplet size and gradual gathering of hepatic lipids [5]. Oak leaves, like other oak parts, have bioactive components that provides them basic properties, such as: antioxidant, antibacterial, antifungal and anti-inflammatory activities [6, 7]. More studies currently underway on the various medical uses of *Quercus* spp. leaves and their potential with feasibility in finding new applications for treatments. In general, extracts obtained from different parts of the *Quercus* spp. are rich in phenolic compounds like tannins. Due to the large amount of biocompounds, biological activities of *Quercus* spp. namely antioxidant, antibacterial, anticancer, etc., should be investigated in order to find the key to diseases prevention. Thus, oaks are not only valuable as a source of wood products, but also as a potential pharmacological source in various health fields.

Keywords: *Quercus*, acorn, bark, phenolic compounds, tannins, flavonoids, antioxidant activity, antibacterial activity.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Natural Anti-Estrogens Augment Proline Oxidase-Dependent Apoptosis in Breast Cancer Cells

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Abstract

Stimulation of estrogen receptors is accompanied by increase in cell proliferation; therefore anti-estrogen therapy has found application in the treatment of some breast cancers [1]. However, the mechanism of anti-estrogens function on cell growth and apoptosis is unknown. It has been considered that underlying mechanism may involve proline oxidase (POX) [2]. The enzyme degrades proline yielding reactive oxygen species (ROS) inducing apoptosis or ATP promoting survival. The effect of certain plant anti-estrogens (genistein, equol and biochanin A) on POX-dependent apoptosis was studied in MCF-7 wild type (MCF-7^{WT}) and MCF-7 POX silenced (MCF-7^{-POX}) breast cancer cells. Among studied anti-estrogens, Biochanin A showed a strong cytotoxic effect in MCF-7^{WT} cells. When POX was up-regulated by PPAR-gamma agonist, troglitazone (TGZ), the cytotoxicity of Biochanin A was significantly increased, particularly in MCF-7^{WT} cells, indicating synergism in the action of both compounds. It was corroborated by increase in the expression of apoptotic markers, cleaved caspase-3, cleaved caspase-9 and p53, particularly in MCF-7^{WT}, suggesting important role of POX in this process. The data suggest that TGZ in combination with anti-estrogen, *e.g.*, Biochanin A could be considered as an approach to experimental therapy for breast cancer.

Keywords: Troglitazone, anti-estrogens, biochanin A, breast cancer.

Acknowledgments: Publication financed under the project № POWR.03.02.00-00-I051/16 from European Union funds, PO WER 2014-2020, grant № 06/IMSD/G/2019.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Amperometric Glucose Biosensor Based on Reduced Graphene Oxide and Polyaniline Nanofibers

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Abstract

Glucose is a main source of energy for cellular activity in the living body. It is very important to maintain a proper concentration of glucose in the blood [1,2]. The huge concentration of glucose in the blood eventually causes damage to many tissues in the body, including heart, eyes, kidneys, and nerves leading painful and life-threatening health complications [3]. Therefore, simple, cheap, and accurate monitoring of the blood glucose level is very important [4]. Electrochemical biosensors are the perfect candidates for this role.

Use of nanomaterials for fabrication of electrochemical biosensors allows to improve its analytical characteristics. For instance, conducting polymers are especially suitable for enzyme immobilization ensuring its activity [5]. Various carbon based materials such as reduced graphene oxide (rGO) allow to improve electron transfer increasing sensitivity of fabricated biosensors [6].

In this work, performance of electrochemical biosensor based on graphite rod working electrode modified with polyaniline (PANI) nanofibers, rGO, and Nafion dispersion and glucose oxidase was evaluated. PANI nanofibers and rGO were characterized by different methods. It was found that optimal ratio of PANI and rGO is equal to 1:10. The developed glucose biosensor was characterized by a wide linear range (from 0.5 to 50 mM), low limit of detection (0.089 mM), good selectivity, reproducibility, and stability. Therefore, the developed biosensor is suitable for glucose determination in human serum.

Keywords: Reduced graphene oxide; polyaniline nanostructures; electrochemical glucose biosensor.

Acknowledgments: This research was funded by a grant (No. S-LU-20-11) from the Research Council of Lithuania.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Synthesis and Studies of Thioxanthone Based Derivatives Exhibiting TADF for New Generation OLED's

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Abstract

Since the first publication of a practical light emitting diode (OLED) was reported in late 1980s, it has been widely researched topic in organic electronics [1]. A lot of progress has been made in this field of research since then and definitions such as thermally activated delayed fluorescence (TADF), have made their way into this technology, and have become one of key factors in pushing internal quantum efficiency of OLEDs up to 100% of their theoretical value [2]. Due to this phenomenon, it is possible to achieve relatively low energy gap (ΔE_{ST} = <100 meV), which allows for both singlet and triplet harvesting.

In 2018 a study was published, describing synthesis and applications of two different organic molecules containing electron accepting thioxanthenone moieties. The devices fabricated from these materials exhibited fairly high external quantum efficiencies ranging up to 21.5%. Moreover, these materials also demonstrated aggregation-induced emission enhancement (AIEE) and TADF capabilities [3].

In this work, using donor-acceptor molecular concept, four new bipolar materials were synthesized in a two-step synthesis, and their electrochemical, photophysical and thermal characteristics were tested. Thioxanthenone was used as an electron accepting moiety, while combining it with four different electron donating moieties. Synthesized materials possessed TADF and AIEE capabilities in addition to room temperature phosphorescence. Highest external quantum efficiency of OLED's fabricated using these materials reached up to 8%, additionally, synthesized materials showed fairly high thermal stability with their $T_{-5\%}$ reaching up to 366 ºC.

Keywords: TADF, OLED, host, phosphorescence.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

LC-MS-based Quantitative Analysis of the Selected Metabolites in Aqueous Humor from Patients with Rhegmatogenous Retinal Detachment

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Abstract

The retina is a multilayer of neurons aiming in converting photons into neural signals. One of the retinal diseases is rhegmatogenous retinal detachment (RRD). RRD starts when the vitreous pulls the retina creating a tear [1] which results in ischemia and degradation of photoreceptors and may affect the anterior segment of the eye in form of uveitis and hypotony [2]. Aqueous humor (AH) fills the anterior segment of the eye and nourishes the cornea and the lens [3]. The project aim was to evaluate the concentration of the selected metabolites in the AH and compare its composition from patients with short- and long-standing RRD. The AH samples were collected from 28 patients undergoing pars plana vitrectomy with simultaneous lens exchange (12 patients with RRD < 21 days, 8 patients with RRD > 30 days, and 8 patients with epiretinal membrane as controls). The analysis was performed using liquid chromatography coupled to a quadrupole time of flight mass spectrometer. For statistical analysis, a *t*-Student test was applied and $p<0.05$ was considered significant. We quantified 13 metabolites related to the Krebs cycle (α-ketoglutarate, malate, fumarate, citrate, succinate, oxaloacetate, glyoxylate), proline cycle (proline, glutamine, glutamate, pyroglutamate), urea cycle (arginine), alanine, and glycine in AH samples. LC-MS-based metabolomics analysis revealed that the concentrations of fumarate, oxaloacetate, malate, pyroglutamate, arginine, glycine, glutamine, and proline remained unchanged between RRD groups vs. control. The concentrations of α-ketoglutarate, glyoxylate, and alanine were significantly decreased in the group of RRD < 21 days in comparison to controls ($p=0.010$, $p=0.015$, $p=0.002$, respectively), while that of citrate increased ($p=0.012$). The concentration of succinate was decreased in the group of RRD > 30 days compared to controls ($p=0.010$). We did not observe any significant differences between short- and long-standing RRD. Implementation of targeted metabolomics provided a better insight into the impact of RRD on the anterior segment of the eye. However, further study needs to be performed on a larger cohort to help in patient stratification based on disease state or prediction of postoperative complications.

Keywords: LC-MS, metabolomics, retinal detachment, aqueous humor.

Acknowledgement: The project was financed within the framework of the Polish Ministry of Science and Higher Education program "Strategy of Excellence - the University of Research" in the years 2018 - 2019 project no 0017/SDU/2018/18 the amount of funding PLN 690 000. The project has been accomplished during doctoral studies under the project № POWR.03.02.00-00-I051/16 co-funded from European Union funds, POWER 2014-2020.

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Synthesis and Properties of Derivatives of 3,6-Disubstituted Naphthlimide

ISSN:2335-8653
ONLINE ISSN: 2335-8718

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Abstract

Third generation of organic electroluminescent materials known as thermally activated delayed fluorescent (TADF) materials, have many advantages when compared to the conventional emitters used in organic light-emitting diodes (OLEDs), such as 100% internal quantum efficiency, absence of heavy metals [1]. TADF materials considered to be the most competitive organic light-emitting materials. They have great application prospects in the field of OLEDs. So far, low-molar-mass TADF materials have achieved high photoluminescence quantum yield and full-color range of red, green, and blue [2]. In this presentation, we report on three electron accepting 1,8-naphthalimide-based compounds combined with three different donors, i.e. phenoxazine, di-tertbutyl phenothiazine and 9,9-dimethy-9,10-dihydroacridine. The synthesis scheme comprised three steps such as bromination, imidization and Buchwald-Hartwig coupling reactions. The target materials were identified by nuclear magnetic resonance spectroscopy and mass spectrometry. The yield of the target compounds ranged from 54 to 76%. The glass transition temperatures of the compounds ranged from 157 °C to 210 °C. The 5% weight loss temperatures ranged from 320 °C to 480 °C. The compound with acridine moieties attached at 3,6 positions of naphthalimide unit exhibited photoluminescence decays up to 1 millisecond and exhibited TADF.

Keywords: TADF, OLED, 1,8-naphthalimide.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Microbiome Identification of Urine Samples Using the MALDI Technique

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Abstract

Human urine as a quite non-invasive biological matrix, plays an important role in clinical diagnostics. Physicians have examined urinary samples from patients to diagnose various disorders for centuries. Urine culture is one of the basic laboratory tests used to identify the causes of undesirable urinary tract symptoms. Urine culture is helpful in making a diagnosis and initiating therapeutic therapy. This test involves transferring a sample of the patient's urine to a special culture medium, the composition of which allows the multiplication and growth of bacteria. The main aim of the study was to identify the microbiome of 186 urine samples collected from patients suffering from genitourinary cancer and to correlate the obtained results with the stage of treatment of patients with radiotherapy. Identification of the grown bacterial colonies was performed using the matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI TOF-MS) technique. MALDI has become an easy-to-use, fast and highly sensitive pathogen identification method for routine microbiology laboratories. The principle of this method is based on the generation and analysis of the unique protein profile of the microorganism and its comparison with that included in the reference spectrum database. As a result, received over 450 bacterial isolates representing 33 different species – 3 G(-) and 30 G(+). The most frequently isolated strains were: *Staphylococcus haemolyticus* (isolated from 19% of samples), *Staphylococcus epidermidis* (18%), *Staphylococcus hominis* (16%), *Enterococcus faecalis* (13%) and *Micrococcus luteus* (10%). The Fisher test - carried out to compare the primary and middle urine streams - showed that only the presence of *Enterococcus faecalis* and *Corynebacterium accolens* were statistically significant. The amount of bacterial species found in urine depended on the stage of treatment. Before and at the end of radiotherapy lower number of bacteria was detected. One month after radiotherapy an increase in the number of isolated bacteria was observed. The number of bacterial species in urine did not correlate with the other parameters obtained from blood. Performed studies proved that the radiotherapy affects the urinary microbiome.

Keywords: Urine, genitourinary cancer, bacteria identification, MALDI, matrix-assisted laser desorption/ionization, radiotherapy.

Acknowledgments: This study was supported by OPUS 19 project No. 2020/37/B/ST4/02136 from National Science Centre.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

MALDI Imaging of honeybee larvae

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Abstract

Insects, including honeybees (*Apis mellifera*) in some cultures, are eaten as a delicacy. They are a rich source of proteins and other nutrients. Thus, potentially, bee larvae may be used for the conventional production of protein for direct human or livestock consumption. However, in order to make sure that bee larvae are safe for eating, their composition must be carefully studied. In addition to examining the overall content, the spatial distribution of nutrients and other molecules in the larvae's body should also be analysed to check that all parts are entirely safe for consumption.

Therefore, in this study, we analysed *Apis mellifera* larvae samples collected from a beehive localised in west-central Poland. Frozen larvae were cut in a cryostat and vacuum-dried. Then, an α-Cyano-4-hydroxycinnamic acid (HCCA) matrix solution was automatically applied into the prepared samples using the *ImagePrep* (Bruker) matrix deposition station. For the proteomic spatial analysis, a MALDI-TOF (matrix-assisted laser desorption/ionisation-time of flight) mass spectrometer, working in a linear-positive mode in an m/z range of 2-20 kDa, was used. The obtained spectra were analysed in flexImaging (Bruker) software.

The analyses allowed for the selection of m/z features differentiating between sections of the honeybee larvae's body. This pilot study highlights the great potential of the MALDI Imaging technique in the detailed characterisation of insect's body composition, along with the spatial distribution of the proteins and peptides. In addition to increasing the safety of using insects as food, this research may be the first step towards better understanding the physiological processes occurring in the bee larvae's body.

Keywords: MALDI Imaging, honeybee larvae, proteomics, mass spectrometry.

Acknowledgements: This project received financial support from the Polish National Science Centre (grant number 2016/23/D/NZ7/03949).

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Biologically Active Compounds of *Salvia Officinalis* L. and Their Practical Application in Cosmetic Practice

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Abstract

In order to solve the problem of increasing the diversity of medicinal plants and biologically active compounds and their application in cosmetic practice, is being studied *Salvia officinalis* L. – perennial, medicinal (aromatic) plant of *Lamiaceae* Martinov family [1].

The aim of study was to analyze scientific articles and research data of *Salvia officinalis* L. and evaluate the possibilities of their scientific and practical uses in cosmetic practice.

In order to study chemical properties and pharmacognostic effects of medicinal plants, the evaluation of research data presented in literature sources and databases (NCBI, PubMed, Gbif).

Salvia officinalis L. are analyzed by spectrometric method for total phenolic compounds, total flavonoids and total DPPH radical scavenging activity. Its essential oils are analyzed by GC-MS [3,4].

In previous studies was analyzed *Salvia officinalis* L. secondary metabolites such as alkaloids, carbohydrate, fatty acids, glycosidic derivatives, phenolic compounds (coumarins, flavonoids, tannins), poly acetylenes, steroids [2]. *S. officinalis* is an essential oil accumulating plant [4]. Essential oil is accumulated in the overground raw material. It has been determined that *S. officinalis* growing in different countries is dominated by monoterpenes such as camphor and 1,8-cineole [2] The predominant compounds were identified α-thujone, β-thujone, viridiflorol, β-caryophyllene, α-humulene [2]. *S. officinalis* essential oil have possessed various biological activities including antimicrobial, antioxidant, anti-cancer, antimutagenic, anti-inflammatory, choleretic, cognitive and memory-enhancing effect [1].

It appears that there are a number of biologically active compounds explored in *Salvia officinalis* L. essential oil and various combinations and numerous medicinal properties of its extract, oil, and leaves demand further and more studies on the other useful and unknown properties of this multipurpose plant.

Keywords: Medicinal (aromatic) plants, *Salvia officinalis* L., biologically active compounds cosmetic practice.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Hemagglutinating Activity of Proteins from *Kirchneriella Sp.* Schmidle Biomass

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Abstract

It is known that algae are rich sources of structurally biologically active metabolites, such as fatty acids, polysaccharides and proteins. Lectins are proteins of non-immune origin which bind specifically and reversibly to carbohydrates. The erythrocytes hemagglutination activity of lectins is a major attribute of these proteins and is used routinely for their detection and characterization [1].

Lyophilized biomass of green algae *Kirchneriella sp.* Schmidle, was received from the Nature Research Centre, Laboratory of Algology and Microbial Ecology. At first lyophilized green algae biomass was swelled in the purified water, protein extraction was carried out using 0.2 M NaOH solution; protein precipitation using mixture of 13.3% trichloroacetic acid (TCA) and 0.2% β-mercaptoethanol (β-ME) in acetone; protein pellets were washed with 100% acetone and washing was performed 3 times; proteins were resuspended in phosphate buffer saline (PBS) pH 7.4. The protein content was determined by spectrophotometric Lowry method. The rabbit, alpaca and goat blood erythrocytes treated with enzyme trypsin. 50 µl of PBS (pH 7.4) were added to the wells of a flat-bottomed enzyme plate. 50 µl of protein extract and 50 µl of blood erythrocytes suspension added to the wells, gently mixed, covered and 60 min. incubated at 22 °C [2-5].

Hemagglutination activity was observed only in rabbit blood erythrocytes suspension with protein concentration – 0.829±0.232 µg.

Proteins extracted from *Kirchneriella sp.* Schmidle biomass showed positive hemagglutination activity with rabbit erythrocytes suspension, but negative result was with alpaca and goat erythrocytes suspensions.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Acridan Derivatives Exhibiting Aggregation Induced Enhancement of Thermally Activated Delayed Fluorescence for Organic Light Emitting Diodes

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Abstract

The thermally activated delayed fluorescence (TADF) materials have been recently attracted significant interest in the field of organic optoelectronics. [1] TADF based organic light emitting diodes (OLEDs) can achieve 100% internal quantum efficiency, which can be attained due to harvesting of triplet excitons through the reverse intersystem crossing (RISC). A very small singlet–triplet energy gap (ΔE_{ST}) is essential parameter to achieve efficient TADF effect via RISC. The small ΔE_{ST} can be enhanced using compounds with spatially separated donor (D) and acceptor (A) units. [2] The different organic polymorphs of TADF compounds in solid-sate, exhibiting the effect of aggregation induced emission enhancement (AIEE), demonstrate different TADF efficiency because of changes in ΔE_{ST}. [3] Moreover, intra- or intermolecular phenomena may induce changes of colour in multi-colour emitters. [4]

In this research new D–A and D–A–D compounds composed of perfluorobiphenyl acceptor and 9,9-dimethyl-9,10-dihydr-acridine or 2,7-di-*tert*-butyl-9,9-dimethyl-9,10-dihydroacridine donor moieties were synthesized and investigated as TADF, AIEE and colour-changing materials. The properties of target compounds were studied using thermal, optical, photophysical, electrochemical, photoelectrical and charge-transport measurements. Considerably differences of the solid-state emission properties were observed for the compounds depending on the presence or absence of *tert*-butyl groups. The most efficient emitters were used for the preparation of either hosted or non-hosted light emitting layers of blue and sky-blue TADF OLEDs. Maximum external quantum efficiencies of 6.6 and 16.3% were achieved for non-doped and doped devices, respectively.

Keywords: Aggregation induced emission enhancement, thermally activated delayed fluorescence, organic light emitting diode, donor, acceptor.

Acknowledgments: This project has received funding from European Regional Development Fund (project No 01.2.2-LMT-K-718-01-0015) under grant agreement with the Research Council of Lithuania (LMTLT).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Protein Quantification by Lowry Microassay and Haemagglutinating Activity Determination of Proteins from Cyanobacteria Biomass in Different Animal RBC Suspensions

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Abstract

Lectins are widely distributed in nature and play a crucial role in cell-cell or even cell-matrix interactions as recognition molecules. They are useful as practical detection and identification tools for carbohydrate structures and cell typing as they can distinguish carbohydrate structure differences [1]. In comparison to land plant and animal lectins, which are well portrayed, there is little information on the properties of algal lectins. In the last two decades, the number of confined and characterized algal and cyanobacterial lectins has increased [2]. Purpose of the research was to determine protein quantity from cyanobacteria biomass collected in lake Simnas by micro Lowry assay and test extracted proteins for their hemagglutinating activity.

The cyanobacterial scum biomass from lake Simnas (Alytus district) used in this study contained 84% *Microcystis* and 12% *Dolichospermum* species. Samples were generously provided by the Nature Research Centre, Laboratory of Algology and Microbial Ecology. 15 mL of wet cyanobacteria biomass extraction was carried out in ultrasonic bath for 60 min. The biomass was homogenised by adjusting pH to 13 with 1.0 M NaOH solution. The sample was cooled and centrifuged, the supernatant was drained and the precipitate was used for further experiments. Proteins were precipitated by -20 ºC 13.3% TCA solution in acetone, containing 0.2% β-mercaptoethanol (β-ME). The sample was incubated at -20 °C for 16 h. After incubation, the sample was centrifuged and the precipitated proteins were washed with acetone and 0.2% β-ME. After 3 washes protein pellets were resuspended in 1 ml of PBS buffer, pH 7.4. Before testing haemagglutinating activity, cyanobacteria proteins quantification carried by micro Lowry assay [3]. Results showed protein concentration 4923.5±133.643 µg/ml.

Hemagglutination assay was carried out by the method described by [4]. Activity was evaluated visually after 60 min. incubation at 22 °C [4].

Haemagglutination activity possibly was observed just in rabbit‘s blood by protein concentrations from 15.385±0.418 µg to 30.772±0.835 µg. There were no haemagglutinating activity detected in goat and alpaca’s blood suspension.

Protein fragments extracted from cyanobacterial biomass, which was collected in lake Simnas showed affirmative hemagglutination activity with rabbit RBC suspension. Further research is required.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Comparison of Hemagglutination with Blood Samples from Different Species of Animals

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Abstract

Microalgae (*Cyanophyta*) are thought to be known as a promising source of glycoproteins – lectins. A hemagglutination test shows lectins that are present in the tested samples. The aim of the study was to determine the hemagglutination activity with a suspension of erythrocytes from rabbit, alpaca and goat blood [1].

Blue-green (*Cyanophyta*) biomass obtained from the Nature Research Center, Laboratory of Algology. Protein biomass was sonicated using an ultrasound bath and centrifugated at 8500 rpm, 4 ºC. The supernatant was processed with 13.3% trichloroacetic acid, 0.2% β-mercaptoethanol. Following protein pellets were washed with 100 % acetone to remove additional impurities from the precipitate. The hemagglutination activity was tested with different animal blood samples kindly provided by the LSMU Veterinary academy. A suspension of rabbit, alpaca and goat erythrocytes was prepared by a method described by [2]. Hemagglutination activity was detected by adding 25 µl of PBS buffer to the microplate wells. Then 25 µl of protein extract is poured to the first well and serially diluted. Lastly, 25 µl of erythrocyte suspension was added to each well. Hemagglutination activity was evaluated microscopically after 60 min incubation [3].

The range of protein amount that caused hemagglutination in rabbit blood was from 0.52±0.02 µg to 66.18±2.70 µg. The amount of protein that caused hemagglutination with alpaca blood was 66.18±2.70 µg.

The microalgae (*Cyanophyta*) biomass studied is thought to contain lectins due to a positive agglutination test with rabbit and alpaca blood erythrocyte suspension. There is a necessity for further tests to confirm this hypothesis.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Influence of Antibiotics on Bacterial Biofilm Formation

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Abstract

Microorganisms can exist in two basic forms - as single cells (dispersed, slow floating) and in the form of a biofilm (organized form formed by cells settled on solid, natural or artificial surfaces). Biofilm formation is a multi-stage phenomenon that depends on the properties of the microorganisms that form it and the type of colonized materials [1]. One of the conditions for the formation of a monolayer of bacterial cells on a given surface is to overcome electrostatic repulsion forces by attracting hydrophobic interactions. According to literature data, bacterial cells are usually endowed with a negative surface electric charge, the value of which depends, among others from the strain, chemical structure and surface of the cell. The surface electric charge of cells affects the speed of their movement in the electric field, as well as the value of the so-called electrokinetic potential characterizing the double electrical layer around the bacterial cell [2]. The ability of bacteria to form a biofilm is of crucial due to both its protective and pathogenic functions. Therefore, a desirable alternative to systemic antibiotic therapy is undoubtedly topical antibiotic therapy in order to prevent formation of biofilm and protecting the patient from the side effects of often long-term antibiotic administration. The complexity of the biofilm structure and its high resistance to antibiotics are the causes of difficulties encountered during therapy of bacterial infections associated with biofilm. Expanding knowledge of the formation, functioning and structure of biofilm are the basis for seeking alternative methods on prevention and treatment of infections associated with its occurrence.

Therefore, this work aimed to investigate the effect of selected antibiotics on biofilm formation by model *Bacillus tequilensis sp*ecies.

Keywords: Antibiotic drugs, antibiotic resistance, biofilm, capillary electrophoresis, MALDI TOF-MS.

Acknowledgments: This work was financially supported by the National Science Center as part of the project Preludium 19 No. 2020/37/N/ST4/02358 (2021–2023).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Investigation of Dependence of Phenolic and Volatile Compounds Composition, Antioxidant Activity and Energetic Properties of *Artemisia dubia* Wall. on Nitrogen Fertilization

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Abstract

The secondary metabolites produced by *Artemisia dubia* WALL. (eng. perennial mugworth) exert significant antibacterial, insecticidic, allelopathic, pharmacological and toxicological effects and their efficient antioxidant activity has lead these compounds being used as nutraceuticals, which, according to World Health Organizations Twelfth General Programme of Work [1], is an equally important aspect, since it not only concerns with the purely medical aspects of illness, but with the determinants of ill health and the promotion of health. The plant is also a prosperous energetic plant, obtaining significant heating value and large growth rate [2]. The aim of the research is to investigate *A. dubia* Wall. methanol/water extraction fractions phytochemical properties and components as well as the solid phase residue heating value before and after the extraction. In this research *A. dubia* Wall. was harvested from 3 different areas (no fertilization, N90 and N180) on 2018 July. Extraction was performed using distilled water and 3 concentrations of methanol: 50%, 75% and 100%. Phytochemical capabilities were measured using spectrophotometric analysis methods according to Kaškonienė et al. [3]. Lower calorific value (LHV) of the dry basis for the solutions was measured by an IKA C5000 calorimeter in accordance with adiabatic method for automated bomb calorimetry [4]. Largest total phenol content was determined in the non-fertilized and N90 fertilized 50 % methanol extracts, respectively 95.3 RE mg/g, 96.3 RE mg/g. Same with total flavonoid content with values of 58.4 RE mg/g for non-fertilized and 57.2 RE mg/g with N90 fertilized extracts. Small concentrations of both polar and nonpolar fractions are required to trigger noticeable changes in antioxidant activity, ranging from 190 – 215 RE mg/g. Camphor, camphene, chrysanthenone and eucalyptol were identified in *A. dubia* WALL. using GC-MS and a computer-supported spectral library. 7.13 – 9.00% of ash content was found in non-fertilized plant material, while in rinsed biomass ash content dropped by 1-2 %. Calorific value of untreated plant material decreased insignificantly compared to rinsed-in-solvent one.

Keywords: Phytochemistry, *Artemisia dubia*, solid phase extraction (SPE), GC-MS, Folin-Ciocalteu, aluminium chloride, DPPH, lower calorific value.

Acknowledgments: This project has received funding from the Research Council of Lithuania (LMTLT), grant No. 09.3.3-LMT-K-712-22-0316.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Matrix-assisted Laser Desorption/Ionization Mass Spectrometry Imaging of Lipids in Ovarian Tumor Tissue – A Pilot Study

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Abstract

Methods used so far in traditional biomarkers discovery studies result in the loss of crucial histological information and significant differences that might be observed only as a localized target concentration. Thus, a technique such as MALDI-MSI (matrix-assisted laser desorption/ionization mass spectrometry imaging) that enables visualization of the biomolecules may be helpful for a proper interpretation of the processes ongoing within the tissue – at the disease site. In recent years, some lipid compounds have been proposed as potential cancer biomarkers.

Since its introduction, mass spectrometry imaging (MSI) has proven to be a powerful tool for studying lipids distribution in tissue samples. Moreover, this technology is helpful for simultaneous detection and visualization of a wide range of compounds with their local concentration within the analyzed tissue section.

The presented research aimed to investigate lipid compounds distribution in an ovarian tumor tissue section using the MALDI-MSI approach. The 2,5-Dihydroxybenzoic acid (DHB) matrix was applied onto the analyzed tissue section using a matrix deposition device "ImagePrep" (Bruker). The analysis was carried out using UltrafleXtreme MALDI-TOF/TOF (Bruker) mass spectrometer under the control of flexControl and flexImaging software (Bruker).

The proposed methodology made it possible to visualize a significant differentiation of potential lipid compounds in the analyzed tissue section of ovarian fibrothecoma. This study proves the utility of the MALDI-MSI approach in lipidomic studies as well as the contribution to a better understanding of a human disease's molecular background. However, because of the complexity of the lipidomic analysis, further in-depth studies are needed.

The presented technique provides valuable complementary information to histology for lipidomic studies of cancer tissue and tumor classification in clinical and research cancer applications as it adds the specificity of mass spectrometry to detailed spatial information.

Keywords: MALDI-MSI, mass spectrometry, ovarian tumors, fibrothecoma.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Influence of Different Conditions on The Formation of Zn-Lactoferrin Complexes

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Abstract

Lately, more attention is paid to the connection of nutrition with health. Thus, the concept of "functional food" has appeared, which supposedly provide biologically active substances (e.g. vitamins or microelements) to organism [1]. In the production of functional food, the utilization of naturally occurred substances is emphasized. Proteins and their derivates have beneficial effect on the functioning of the organism [2] and milk proteins attracts much attention in food supplements production [1]. Zinc deficiency is an issue of the worldwide importance [3], while the utilization of whey proteins has shown to promote the absorption of zinc and iron [4]. Lactoferrin (LTF) is a whey protein, which possesses one of the widest spectrum of biological functions. LTF belongs to metalloproteins which has two specific metal-binding sites in its structure. LTF has the highest affinity to the Fe^{3+} and supposed to take part in its homeostasis, but also can interact with other metals such as Zn, Mg, Cu, etc. [5]. Moreover, it was shown that it can bind much more Zn^{2+} which can be predicted by specific metal-binding sites [6]. Hence, the aim of the study was to investigate the influence of the synthesis parameters on the efficiency of Zn^{2+} adsorption by bovine LTF. Such complexes can be utilized as a food supplement. The synthesis was performed with utilization of different amount of Zn^{2+} at different pH. Multiple analytical techniques were utilized during the studies, namely SDS-PAGE coupled with MALDI-TOF/TOF MS, ATR-FTIR, Raman, ICP-MS and electron microscopy. Additionally, the obtained complexes were subjected to the assessment of cytotoxic properties.

Keywords: Bovine lactoferrin, zinc-lactoferrin complexes, biologically active substances.

Acknowledgments: This work was financially supported in the framework of the project "Advanced Biocomposites for Tomorrow's Economy BIOG-NET" (FNP POIR.04.04.00-00-1792/18-00). Oleksandra Pryshchepa, Paweł Pomastowski, Katarzyna Rafińska and Bogusław Buszewski are members of Toruń Center of Excellence "Towards Personalized Medicine".

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Investigation of Intracellular Protein Release After Electroporation

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Abstract

The electroporation method can be applied to gene transfection, drug delivery to a cell or tissue, cancer treatment, or extraction of molecules from cells. However, if the electroporation is too strong, the cells may lose viability. The exact reason of cells dying after electroporation has not yet been elucidated. However, it is speculated that cells after electroporation perish due to the loss of intracellular macromolecules that are vital. The aim of this work was to evaluate the dynamics of protein release from cells under different electric pulse parameters. Noncancerous CHO and cancerous 4T1 cells were electroporated with either 1 or 9 high voltage (HV) pulses with field strength of 0.6 kV/cm to 4.8 kV/cm in increments of 0,6 kV/cm and duration of 100 μs repeating at 1 Hz. The BCA assay was exploited to determine the total protein amount released from cells in comparison to nonelectroporated cells. The results showed that the release of proteins from electroporated cells depends on the strength and number of the pulses. The release of proteins was more evident from cancerous 4T1 cells. Interestingly, the highest amount of intracellular proteins extracted from cells did not exceed 60% and did not correspond the number or strength of the electric pulses.

Keywords: Electroporation, protein release, extraction, intracellular proteins, BCA.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

New Approaches in Functionalization of Cow's Milk Proteins

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Abstract

Cow's milk plays an important role in the human diet from the first days of our lives. Milk products provide the body with many valuable nutrients/bioactive components such as high-quality proteins, carbohydrates present as lactose, mineral salts (calcium and phosphorus compounds) or vitamins [1]. Milk consists of two main groups of proteins: casein and whey proteins.

From the point of view of biological activity of proteins, metals play an important role. The result of metal-protein interactions can be metalloproteins, metallocomplexes, nanoparticles and nanocomposites thus affecting the biological activity of proteins [2]. Determining these changes is significant because proteins have many functions in the human body and are used in many industries. The following factors are critical for evaluating changes caused by interactions with metals: (a) the formation of new binding sites that determine the protein's interactions with other ligands, (b) changes in protein structure, (c) interacting groups that allow the nature and thus strength of the interaction to be studied, and (d) possible aggregation of the protein [3].

In the present work, physicochemical characteristics of milk proteins and the uptake process of metal ions immobilized to them were carried out. The characterization of proteins by spectrometric and spectroscopic methods and the kinetics of immobilization of metal cations onto milk proteins will allow to formulate conclusions on the course and mechanisms of complex formation. The understanding of the processes and mechanisms of metal-protein binding creates potential application opportunities.

Keywords: Protein milk, caseins, interactions of metal-protein.

Acknowledgments: This work was financially supported by National Science Centre in frame of Opus 14 project No. 2017/27/B/ST4/02628 (2018-2021).
Paweł Pomastowski and Bogusław Buszewski are members of Toruń Center of Excellence "Towards Personalized Medicine" operating under Excellence Initiative-Research University.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Leonurus L. Genus and Its Biologically Active Compounds for Therapeutical Purpose

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Abstract

In order to solve the problem of increasing the diversity of medicinal plants and biologically active compounds and their application in medicine, pharmacy, is being studied *Leonurus cardiaca* L. and *Leonurus japonicus* Houtt. – perennial, medicinal (aromatic) plants of *Lamiaceae* Martinov family [1,2].

The aim of study was to analyze scientific articles and research data of *Leonurus* L. genus plants and evaluate the possibilities of their scientific and practical uses in the world and Lithuania.

In order to study introduction, pharmacognostic and chemical properties of medicinal plants, the evaluation of research data presented in literature sources and databases (PubMed, ReaserchGate, NCBI) were performed using the methodology of systematic theoretical analysis.

Leonurus cardiaca L. are included in Russian (1968), Great Britain (1992) and European pharmacopoeia (2008) or *Leonurus japonicus* Houtt. in the Chinese Pharmacopoeia [3].

Leonurus L. is widespread in South Asia, Central and Eastern Europe, Scandinavia, China, Mongolia, as well as an invasive plant in America and Africa. In previous studies was analysed *Leonurus cardiaca* L. and *Leonurus japonicus* Houtt. secondary metabolites such as essential oils, phenylpropanoids, flavonoids, phenolic acids, sterols and tannins [4,5]. Pharmacological studies have confirmed by them antibacterial, antioxidant, anti-inflammatory and analgesic activity, as well as its effects on the heart and the circulatory system [6,7].

Since 1933 *Leonurus cardiaca* L. amd since 2013 *Leonurus japonicus* Houtt. introduction and phytochemical studies are carried out in Lithuania for therapeutic and prophylactic purposes.

Keywords: Medicinal plant, *Leonurus cardiaca* L., *Leonurus japonicus* L., introduction.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Investigation of Phenolic Compounds and Antioxidant Activity of Brazilian Green and Red Propolis Preparations

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Abstract

Propolis exhibits a broad spectrum of biological activities because of great diversity of chemical components. As Brazil is one of the main producers of propolis (especially green type) in the world, it is useful to compare different commercial Brazilian propolis preparations composition and antioxidant effects. The aim of our study was to determine phenolic composition of ethanolic and water extracts of Brazilian green and red propolis using UV-VIS spectrophotometric and UHPLC methods and to evaluate the antioxidant activity *in vitro* by FRAP, CUPRAC, DPPH, ABTS. Four 10% propolis extracts were analysed: two ethanolic green propolis extracts (EBG), one water green propolis extract (WBG) and one ethanolic red propolis extract (EBR).

The highest total amounts of phenolic compounds, flavonoids and hydroxycinnamic acid derivatives were evaluated in one of the EBG (21,4±0,93 mg/ml; 13,8±1,25 mg/ml; 2,82±0,32 mg/ml, $p < 0,05$, respectively). This sample showed the highest antioxidant activity which was determined using CUPRAC, FRAP, ABTS methods (336,04 mmol TE/l; 71,67±5,78 mmol TE/l; 382,63±17,42 mmol TE/l, $p < 0,05$, respectively). The highest antiradical activity (measured by DPPH method) showed both EBG (18,68±0,033 mmol TE/l; 18,27±0,14 mmol TE/l, $p < 0,05$, respectively). 19 individual compounds were identified in the tested extracts by UHPLC. EBG had different chemical compositions depending on the botanical sources: in one of them dominated flavonoids apigenin, hyperoside, isorhamnetin, pinocembrin and phenolic acids (p-coumaric acid, 3,4-dihydroxybenzoic acid) and in other – flavonoids (galangin, kaempherol, isoliquiritigenin, liquiritigenin, pinocembrin). In WBG were more phenolic acids than flavonoids (galangin, liquiritigenin, kaempherol). In EBR dominated flavonoids (formononetin, liquiritigenin, galangin) and phenolic acids. Biochanin A was detected in all preparations, the highest amount was found in EBR. The greatest total amount of identified phenolic compounds (14661 μg/ml) was detected in EBG but other manufacturer's EBG had the lowest amount.

In conclusion, our results showed that commercial Brazilian extracts possess antioxidant (antiradical and reducing) activities, and phytochemical composition, as well as antioxidant activity, varies among the preparations. This variability may be due to the different geographical location of propolis, collection season or technological differences. Despite that, ethanolic Brazilian propolis extracts are a good source of natural antioxidant compounds which are valuable for diseases prevention and future research.

Keywords: Propolis, phenolic compounds, antioxidant activity.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Influence of short-term hypoxia on alternative pre-mRNA splicing

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Abstract

All living organisms respond to, and defend against, environmental stresses. These include temperature shock, oxygen shock (hypoxia), nutrient deprivation and DNA damage. One of the ways for cells adapt to altered environmental conditions are changes in pre-mRNA splicing. It is an important mRNA maturation process of eukaryotic cells, during which introns are excised and exons are joined. Alternative splicing is a fundamental mRNA processing event that explains how a high biologic complexity is achieved from a limited number of genes. By allowing each gene to encode several polypeptide variants, alternative splicing largely expands the coding capacities of the genetic information [1]. The splicing process is carried out by the spliceosome, a complex macromolecular machinery, composed of five small nuclear ribonucleoprotein particles (U1, U2, U4, U5 and U6 snRNPs) and more than 200 auxiliary proteins [2]. Up to 95% of all human genes are alternatively spliced producing RNA isoforms that code for functionally distinct proteins [3, 4]. Only limited data is available on how short-term hypoxia, as a type of cell stress, affects the formation of mRNA isoforms. n this study we have investigated how pre-mRNAs of the genes, previously identified as chronic hypoxia-dependent, are influenced by short-term hypoxic stress by analyzing changes in splicing patterns.

Keywords: Hypoxia, alternative splicing, cell stress.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Evaluation of The Composition of Phenolic Compounds in *Artemisia abrotanum* L. During Different Vegetation Stages

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Abstract

In order to solve the problem of increasing the diversity of medicinal plants and biologically active compounds and their application in medicine, pharmacy, is being studied *Artemisia* L. genus [1]. Previous studies have shown that the *Artemisia abrotanum* L. accumulate high amounts of phenolic compounds (anthocyanins, phenolic acids, flavan-3-ols, flavonols), organic acids and mineral substances [2]. *A. abrotanum* essential oil have anti-inflammatory, expectorant, spasmolytic, antiseptic and antimicrobial effects [3]. The object of investigation was *A. abrotanum* a perennial medicinal, aromatic semi-shrub of *Asteraceae* (Bercht. & J. Presl) family.

The aim of this study was to determine the composition and content of phenolic compounds in ethanolic extracts of *Artemisia abrotanum* L. raw material during different vegetation stages.

The raw material of *Artemisia abrotanum* L. were collected during different vegetation stages in the Spice-Milliferous Plants Collection of Medicinal Plants of the Scientific Sector of Medicinal and Aromatic Plants, Scientific Department of Botanical Garden at Vytautas Magnus University in 2019. The samples were extracted with 70 % (v/v) ethanol in an ultrasonic bath for 30 minutes. The analysis of phenolic compounds was performed on the basis of high-performance liquid chromatography method.

During the study, the following glycosides of the flavonol group were identified: luteolin-7-rutinoside, luteolin-7-glicoside, luteolin-3-rutinoside, isorhamnetin-3-rutinoside, rutin. In addition, compounds of phenolic acids group were identified, such as chlorogenic acid, caffeic acid, neochlorogenic acid, 4-O-caffeoylquinic acid, 3,4-di-caffeoylquinic acid, 3,5-di-caffeoylquinic acid and 4,5-di-caffeoylquinic acid. Of all identified compounds chlorogenic acid predominated during all vegetation stages.

In conclusion, the results of this study will provide new data about the composition and content of phenolic compounds of *Artemisia abrotanum* L. in Lithuania. The highest concentration of chlorogenic acid (217.07±0.16 mg/g) was detected at the flower bud development stage and the lowest amount (85.41±0.02 mg/g) was during the end of flowering vegetation stage.

Keywords: *Artemisia abrotanum* L., phenolic compounds, vegetation stages.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Impact of PTBP1 on Alternative Splicing of MAPT Pre-mRNA Under Normoxic and Hypoxic Conditions

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Abstract

Hypoxia refers to a condition in which oxygen is limited and is defined as a decrease in the oxygen supply to a level insufficient to maintain cellular function [1,2]. Among the key responses to oxygen deprivation is the changes in alternative splicing which is critical for adaptation to altered cellular microenvironment. Alternative pre-mRNA splicing represents the exclusion or inclusion of different exons and/or intron sequences within the mature mRNA sequence which results in different structure and function of a protein [3,4]. It is catalysed by the spliceosome, a multiprotein complex comprised of five snRNPs and numerous proteins [5]. It is thought that the alternative splicing of Tau protein, the main causing factor of brain pathology encoded by the *MAPT* gene, is influenced by the oxygen level [6]. In this study, we have examined the impact of PTBP1 on the alternative splicing of neurodegenerative disease-related MAPT pre-mRNA in the environments of normal and reduced oxygen level.

Keywords: Hypoxia, alternative splicing, splicing factors, neurodegeneration.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

New Organic Materials Exhibiting Thermally Activated Delayed Fluorescence and their Application in Organic Light-Emitting Diodes

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Abstract

In recent years, great attention has been paid to organic materials exhibiting thermally activated delayed fluorescence (TADF) due to their potential application in organic light-emitting diodes (OLEDs) [1]. Theoretically, OLEDs based on TADF emitters can achieve 100% internal quantum efficiency via efficient up-conversion from non-radiative triplet state to radiative singlet state. Therefore, these compounds are promising materials as emitters for the fabrication highly efficient OLEDs [2]. TADF emitters usually have donor-acceptor or donor-acceptor-donor structures [3]. In this work, new donor-substituted derivatives of pyrimidine were designed, synthesized, investigated and used for fabrication of OLEDs.

The target compounds containing pyrimidine electron-withdrawing scaffold and variety of electron-donating moieties such as carbazole, tert-butylcarbazole, methoxy carbazole were synthesized and investigated. The films of compounds containing carbazole and tert-butylcarbazole donor units exhibited emission in sky-blue region with maxima at 487 nm, whereas the film of methoxy carbazole-containing compound emitted light in green region with intensity maximum at 542 nm.

Compounds having carbazole and tert-butylcarbazole moieties showed aggregation induced emission enhancement and all the compounds showed TADF. OLEDs were fabricated in order to test the synthesized compounds as TADF emitters. Sky-blue devices based on the layers of compounds containing carbazole and tert-butylcarbazole units showed external quantum efficiency (EQE), reaching 12.8 and 5.1% of respectively. Along with maintaining spectral characteristics, high EQE with the maximal value of ca. 14% were achieved by using 1,3-bis(N-carbazolyl)benzene) as a host.

Keywords: Pyrimidine, carbazole, thermally activated delayed fluorescence, organic light-emitting diodes.

Acknowledgments: This project has received funding from European Regional Development Fund (project No 01.2.2-LMT-K-718-01-0015) under grant agreement with the Research Council of Lithuania (LMTLT).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Antimicrobial Activity of Lactic Acid Bacteria Immobilised in the Rice Bran-Lingonberry-Based Gel-Type Food Matrix

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Abstract

Rice bran is processing by-products enabling the industrialized production of functional ingredient for nutraceuticals or functional foods, thereby contributing to human health, which contains various nutrients such as protein, fat, carbohydrate, minerals, and vitamins, making it a very useful food source. The objective of the study was to develop rice bran (RB) and lingonberry press cake (LPC) based chewy-type nutraceuticals with encapsulated in rice resistant starch (RS) cells of *Lactobacillus brevis* LUHS173 and *Pediococcus acidilactici* LUHS236. A high intensity ultrasound (US) (37 kHz, power output 70 W) was used to structure RB-LPC matrix at optimal conditions for enhanced production of resistant starch (RS) and soluble dietary fibre (SDF). The effect of RB/LPC ratio on lactic acid bacteria against indicator strains (*Pseudomonas aeruginosa, Escherichia coli, Salmonella typhimurium, Bacillus cereus, Staphylococcus aureus, Listeria monocytogenes*) was analysed. The antimicrobial activity of cells (LUHS173 and LUHS236) immobilised in the RB-LPC matrix, stabilised with sodium alginate and xanthan gum, was analysed over a two-week period. The addition of LPC up to 30–50 % in the RB fermentation medium ambiguously affected the viability and antimicrobial activity of LAB. The highest viability and the highest antimicrobial activity was shown at RB/LPC ratio of 0.5–0.9 for *L. brevis* and at 0.9–0.8 for *P. pentosaceus*. RB substrate supplemented with LP (20–50 g/100 g d.w.) enhanced the antimicrobial activity of tested LAB against *Escherichia coli, Salmonella typhimurium*, and *Staphylococcus aureus* for *L. brevis*, and against *Bacillus cereus* and *Staphylococcus aureus* for *P. acidilactici* depending on fermentation time. The study showed that RB and LPC can be promising ingredients for the production of functional food with specific properties. However, the properties of the RB and LPC materials are also very important, as it was established that they have a significant influence on the antimicrobial activity of the product.

Keywords: Resistant starch, ultrasound, lactic acid bacteria, antimicrobial activity, viability.

Acknowledgments: This research is funded by the European Social Fund under the measure No. 09.3.3-LMT-K-712 "Development of scientific competence of scientists, other researchers and students through practical scientific activities" of the European Funds Investment Operational Program for 2014-2020.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Formation and Investigation of Fuchsine and Malachite Green Films and It's Possible Use for Low Dose Dosimetry Application

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Abstract

In the last decades, the use of dyes and polymers in the form of films and their applications as dosimeters for reporting dose against gamma, electron beam and X-ray radiation in industrial and medicine applications. Dyed polyvinyl alcohol (PVA) films shows promise as a stable dosimeter for low-dose dosimetry processing. Fuchsine acid cyanide (FAC) and malachite green dyes diffuse in PVA, forming a colorless sample, which developed a color with the increase in absorbed doses. We a new dosimetry system formulation for low-dose dosimetry applications at the room temperature. The effect of dose rate, irradiation temperature, film thickness and dye intensity were found not to influence the response. The effects of shelf life and the post-irradiation storage in darkness and indirect daylight conditions on dosimetry performance were discussed.

Keywords: PVA, Fuchsine, malachite green, X-ray irradiation.

Acknowledgments: this work was partly supported by Department of Physics of Kaunas University of Technology and Kaunas and Oncological Hospital of Kaunas Clinics, Radiotherapy Department.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Sacubitril/Valsartan Metabolism in HUVEC Cells

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Abstract

A new combination drug sacubitril/valsartan has recently been introduced in clinical practice for the treatment of heart failure. The drug represents a new class of therapeutics called angiotensin receptor neprilysin inhibitors (ARNI). This drug improves endothelial cell function, however, till now there are only few studies which explored the metabolism of sacubitril/valsartan in endothelial cells. Thus, the aim of this pilot study was to determine metabolites in human endothelial cells after treatment with sacubitril/valsartan.

The human umbilical vein endothelial (HUVEC) cells were cultivated in a six-well plate. Cells were affected with seven different concentrations of sacubitril/valsartan (0.05 μM, 0.1 μM, 0.25 μM, 0.5 μM, 1 μM, 2.5 μM, 5 μM). Sacubitril/valsartan solutions were obtained by dissolving the drug in 0.9% of NaCl in water. Screening for metabolites was performed in HUVEC cell lysates. Lysates were obtained by treating the cells with lysis buffer. Metabolites in a collected cell media were identified using liquid chromatography-quadrupole time-of-flight mass spectrometry (UPLC-QTOF-MS) method. Quantification of the formed compounds in cell media and HUVEC cell lysates were performed by ultra-efficient liquid chromatography-triple quadrupole mass spectrometry (UPLC-TQD-MS) technique.

The main active metabolites of the studied drugs (sacubitril, valsartan, active metabolite of sacubitril – sacubitrilat (M1)) were determined. The concentrations of these compounds in cell lysate were increasing with higher drug concentrations in cell media. A new compound, 13-docosenamide, a potential HUVEC cell metabolite was detected in both cell lysates and media. However, 13-docosenamide peak area in cell lysates was twice lower in comparison to the cell media.

Results showed that sacubitril/valsartan may undergo metabolism in HUVECs. Also, a new metabolite, related to the treatment with this combination drug, 13-docosenamide, was identified.

Keywords: Prodrug, metabolites, HUVEC cells, sacubitril/valsartan.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Antiradical, Reducing, and Chelating Activities of Phenolic Fractions from *Vaccinium vitis-idaea L.* Fruits

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Abstract

Vaccinium vitis-idaea L. (lingonberry) fruits are considered to be a good source of phenolic compounds, which have a high potential for pharmaceutical development due to their versatile antioxidant activity [1,2]. However, the contribution of particular phenolics to the antioxidant activity of lingonberry fruits has not been extensively studied directly. Therefore, this research aimed at the fractionation of phenolics from lingonberry fruits and the evaluation of their antioxidant activity.

Powder of crude dry extracts of lingonberry fruits (marked F1) was applied to a glass column (3×60cm) packed with already equilibrated Sephadex LH-20 and eluted successively with one column volume of water to remove non-phenolic lingonberry constituents, as sugars (waste), four volumes of acidified 20% methanol (to obtain F2), four volumes of 50% ethanol (to obtain F3) and two volumes of 70% acetone (to obtain F4). For each fraction phenolic composition was determined by the validated HPLC-PDA method and radical scavenging, reducing, and chelating activities were investigated, using spectrophotometrical ABTS, FRAP, and FIC assays, respectively. Results of antioxidant activity assays were expressed as micromolar of Trolox or EDTA equivalents per gram of dry weight of fractions (μM TE/g DW and μM EDTA/g DW, respectively).

The radical scavenging activity assessed by ABTS assay ranged between 1751.4 and 7152.7 μM TE/g DW, with the lowest and highest values determined in the crude extract (F1) and fraction with predominant compounds of proanthocyanidins (F4), respectively. In FRAP assay, F3 fraction with prevailing compounds of catechins and flavonols, surpassed all other fractions by the greatest reducing activity of 5404.8 μM TE/g DW, followed by F4 and fraction F2, enriched with anthocyanins. FIC assay showed a similar trend to results of ABTS and FRAP assays. The highest chelating activity (177.6 μM EDTA/g DW) was expressed in the fraction of F4, and the lowest one, approx. three times lower, in F1. Antioxidant activity of fractions according to average values of all antioxidant assays was in the following ascending order: F1 < F2 < F3< F4.

The purified fraction, obtained by the last fractionation step, possessed the greatest antioxidant activity. Thus, predominant compounds of this fraction (proanthocyanidins) can be considered to be antioxidant activity markers of lingonberry fruits.

Keywords: *Vaccinium vitis-idaea*, fruits, phenolic compounds, antioxidant activity.

Acknowledgments: This research received no external funding.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Evaluation of Hemagglutination Activity of Proteins Isolated from *Helianthus tuberosus* (L.) Tubers

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Abstract

Helianthus tuberosus (L.) also known as sunchoke, sunroot, or topinambur is a herbaceous plant of the *Asteraceae* family native to North America. Although because of the high content of fructans in tubers it is grown primarily for its edible purposes, this raw material among other biologically active substances has a high biological value of proteins and balanced amino acids composition. Lectins are a type of protein with a specific binding affinity for a carbohydrate component. Moreover, they can agglutinate cells and precipitate glycoconjugates. Accordingly, they are known as hemagglutination agents. In this research, we evaluated hemagglutination activity of proteins isolated from *Helianthus tuberosus* (L.) tubers [1,2].

Fresh *Helianthus tuberosus* (L.) tubers collected in Vytautas Magnus University Botanical garden. Protein extraction was made using phosphate buffer solution (PBS, 7.6 mM Na_2HPO_4, 2.3 mM NaH_2PO_4, 0.15 mM NaCl, pH=7.4); precipitation using ammonium sulphate; resuspension using phosphate buffer saline (PBS) pH=7.4; spectrophotometric protein determination by Lowry method (at 660 nm and 750 nm); hemagglutination assay was carried out in a microplate following a twofold serial dilution method with rabbit, alpaca and goat blood erythrocytes suspensions.

Hemagglutination activity was observed by protein concentrations from 18.336±0.002 μg/ml to 36.671±0.004 μg/ml in rabbit blood erythrocytes suspension. Neither in alpaca red blood cell suspension nor goat erythrocytes suspension hemagglutination was positive.

Proteins isolated from fresh *Helianthus tuberosus* (L.) tubers collected in Vytautas Magnus University Botanical garden revealed positive hemagglutination activity with rabbit erythrocyte suspension.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Synthesis and Properties Investigation of New Organic Semiconductors with Efficient Hole Transfer

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Abstract

Organic semiconductors are widely used in optoelectronics, organic light-emitting diodes, transistors, and solar cells. Over the last three decades, numerous approaches to material synthesis and device design have been implemented for improving organic semiconductors performance. Organic semiconductors are superior to inorganic devices because they do not use toxic metals such as cadmium, indium, gallium. Also, the synthesis of organic materials is more flexible, efficient, and less expensive. These are the main reasons why organic semiconductors are widely researched in the scientific community and effectively applied in the display and lighting industry. [1] One of the rapidly evolving technologies where organic semiconductors are used is organic light-emitting diodes. The efficiency of the OLED device depends not only on the structure of organic molecules used in the emission layer and the efficient formation/dissociation of the excitons but also on balanced charge transportation. In consequence, the hole transporting layer ensures the efficient transportation of holes to the light-emitting layer and blocking of electrons, which are passing the emission layer from the cathode. This project aims to synthesize three new hole-transporting materials using benzonitrile as an acceptor and modified carbazole fragments as donors and compare their properties.

By modifying the carbazole moiety through C-3, C-6 or C-2, C-7 position hydrogens with methoxy- groups and/or tertbutyl- derivatives we aim to increase the number of intermolecular bonding (hydrogen bonds) which should improve the morphological characteristics, optimize properties, and improve the electrochemical stability of the compounds. [2] The main aspects of the synthesis and characterization of new compounds as well as comparative study of their photophysical, optical, thermal, electrochemical, charge transporting properties will be reported in the presentation.

Keywords: OLED, carbazole, hole transfer.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Morphology of Laminated Cellulose/Alginate Biocomposites

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Abstract

In the last decade the attention for food packaging and its effect to environment has increased. Bio-based films and coatings became a promising alternative to replace petroleum-based materials, which are non-biodegradable [1]. Materials widely used for developing bio-based packaging are polysaccharides (alginates, chitosan, pectin, etc.), lipids (sunflower oil, sesame oil, etc.), proteins (whey protein, soy protein, etc.), etc [2]. Thus, alginate is a promising bio-material in developing biocomoposites for food packaging. It could be cross-linked by using divalent calcium cations to form calcium alginate which possess different properties [3]. So, the main objective of this research was to investigate sodium alginate and calcium alginate laminated cellulose biocomposites.

Laminated cellulose/alginate biocomposites were produced by solvent casting method. Glycerol was used as plasticizer. The morphology of biocomposites were analyzed by using optical microscope and scanning electron microscope (SEM). Also, mechanical characteristics were evaluated of prepared cellulose/alginate biocomposites.

Obtained results showed that plasticizer is important in developing laminated cellulose/alginate biocomposites with improved mechanical characteristics. It was observed, that alginate concentration from 1,25 to 2,25 % is not suitable for laminating cellulose because it does not cover all cellulose fibers. Mechanical characteristics revealed that calcium alginate is the most suitable for food packaging development. To conclude, laminated calcium alginate/cellulose biocomposites with plasticizer glycerol have a potential for developing food packaging.

Keywords: Cellulose, alginate, biocomposite, food packaging.

Acknowledgments: This research is funded by the SC company ‘Birstono mineraliniai vandenys‘ and Ko to the supported activity ‘Development of hydrophobic cellulose bio-composites and investigation of their properties for liquid packaging development‘ by contract No. 19-197/SV9-2590 under measure ‘Inocekiai‘ No.01.2.1-MITA-T-851-02-0116.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Fermentation Possibilities of Biodegradable Organic Waste by *Saccharomyces bayanus*

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Abstract

Bioethanol is a promising renewable and sustainable liquid fuel for tackling today's global energy crisis and the worsening environment quality [1]. During fermentation, sucrose-containing raw materials, starches, and lignocellulosic biomass can be converted into bioethanol. Biologically active substances can be obtained by various extractions and/or fractionations [2, 3]. Therefore, with a proper, intelligent processing, waste can be a potential source of energy, fuel and value-added products.

The aim of this research was to compare the yield of bioethanol and other alcohols after fermentation of different biodegradable organic waste by *Saccharomyces bayanus*. Fermentation of different raw materials (food waste, cigarette butts and fallen leaves) were selected for this study. *Saccharomyces bayanus* yeast were used during fermentation. Biodegradable waste was fermented in two ways: without additional treatment of the raw materials and using enzymatic hydrolysis using cellulase. Quantitative analysis of bioethanol and other alcohols was performed after fermentation of biodegradable waste by gas chromatograph with a flame ionization detector.

Pectinases, cellulases or other enzymes degrade polysaccharides into dextrins, disaccharides or monosaccharides, which later are fermented by yeast to bioethanol. In this study, the concentration of ethanol by direct fermentation is 1.1 times lower compared to enzymatic hydrolysis. The highest alcohol concentration was determined after enzymatic treatment with *Saccharomyces bayanus* yeasts. The largest amounts bioethanol producing feedstock was estimated in food waste, where ethanol concentration reached 1.17%, while the lowest alcohol content was determined in cigarette butts (0.52%). Literature data shows, that *Saccharomyces bayanus* strains contain more glycerol, succinic acid, acetaldehyde, SO_2 and less acetic acid, malic acid and ethyl acetate compared to *Saccharomyces cerevisiae* [4]. This study shows that *Saccharomyces bayanus* can be applied for fermentation of biodegradable waste, however further studies about optimization of raw material/ enzymes/yeast ratio would be valuable.

Keywords: Biodegradable waste, bioethanol, fermentation, gas chromatography, *Saccharomyces bayanus*.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Comparison of Surface Structures and Chemical Composition in Cocoons of Leeches (*Hirudinea*) and Planarias (*Turbellaria*)

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Abstract

Leeches and planarias are common freshwater invertebrates. They are hermaphrodites that lay eggs in cocoons, that protect future offspring from adverse environmental conditions. Cocoons of freshwater worms are secreted and assembled under water, different from terrestrial cocoons of majority Arthropods.

Data on cocoons chemical composition are still scarce.

SEM-EDX is still rarely used in biology, although it has great potential. Energy-dispersive X-ray spectroscopy (EDX) is a surface analytical technique. This method is used to determine the local chemical composition and the map of chemical elements distribution on the surface of the research object. When a sample is irradiated by an electron beam under high vacuum conditions in a scanning electron microscope (SEM), characteristic X-rays are generated [1].

In this study, SEM-EDX have been applied to analyse the surface structure of cocoons of different species of worms. The samples were not treated with any solutions (except alcohol 70% used for fixation) that to avoid change composition of elements *in vivo*.

Cocoons of five leech species (i.e., *Erpobdella octoculata*, *Erpobdella testacea*, *Erpobdella sp*., *Piscicola geometra*, and *Haemopis sanguisuga*) and Planaria sp. were examined.

Data were collected for each element after analysing surfaces of different cocoons. EDX analysis shows the presence of four main elements (oxygen, carbon, sulphur, aluminium) in all samples. The trace elements (phosphorus, nitrogen, silicon, calcium, sodium, chlorine, magnesium, copper) were found only in some samples. The chlorine was detected in cocoon of planaria only.

Keywords: SEM-EDX, cocoon, leech, planaria.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Influence of Bystander Effect on Viability of Untreated Cells After pDNA Electrotransfer, Irreversible Electroporation or Electrotransfer of Anticancer Drug Bleomycin

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Abstract

Electroporation (EP) is a method that is used to induce temporal increase of cell membrane permeability for hydrophilic molecules. Electroporation occurs when cells are affected with electric field at high intensity, hence increasing transmembrane potential to a critical value. This, in turn, trigger the formation of hydrophilic pores that can act as a bridge between cytosol and extracellular matrix for various hydrophilic substances, such as anticancer drug bleomycin (BLM). After entering the affected cell, BLM triggers its mechanism of action by generation of reactive oxygen species (ROS), resulting in multiple genomic DNA breaks that may lead to cell death. This process is applied to clinics as an anticancer therapy called electrochemotherapy. Electroporation can also be applied for irreversible electroporation (cell death as a result of too high electric field) and the process of plasmid DNA (pDNA) intracellular electrotransfer (electrotransfection).

Another physical method for initiation of cell death is ionizing irradiation. It is known that irradiated cells at lethal doses does secrete signalling molecules, that affect adjacent unirradiated cells. This phenomenon is called “Bystander effect”. Although, the Bystander effect is well-known in radiotherapy, it has not been researched in the process of electrochemotherapy. Therefore, the aim of this study is to evaluate the influence of Bystander effect on viability of untreated cells after pDNA electrotransfer, irreversible electroporation or electrotransfer of anticancer drug Bleomycin.

The *in vitro* study presented here is performed using CHO-K1 cells. The electrotransfer of the anticancer drug BLM was achieved using a single electrical pulse of 1400 V/cm amplitude and 100 μs duration. Irreversible electroporation was achieved using a single electrical pulse with an amplitude of 2800 V/cm and a duration of 100 μs. Electrotransfer of the pDNA was achieved using 3 electrical pulses of 1400 V/cm amplitude and 100 μs duration. The cells after electroporation were incubated in a 24-well plate in a 0.2 ml DMEM growth medium for 24 hours. After incubation, the culture medium is collected and centrifuged twice. Finally, Bystander effect was produced by applying the collected medium to untreated cells. The efficiency of Bystander effect was assessed by evaluating cell viability using cell colony formation assay.

We found that Bystander effect was highly pronounced after electrotransfer of anticancer drug BLM. No Bystander effect was found after pDNA electrotransfer. However, the medium from irreversibly treated cells enhanced the viability of neighbouring cells.

Keywords: Electroporation, Bystander effect, pDNA electrotransfer, Bleomycin electrotransfer, irreversible electroporation.

Acknowledgements: Research funded by European Union Funds Investmens in Lithuania. Project number: 09.3.3-LMT-K-712-22-0309. Project manager: Vytautas Magnus University.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Phytochemical Composition of Black Pepper (*Piper nigrum* L.), White Pepper (*Piper album* L.) and Fragrant Pepper (*Pimenta dioica* L.) Essential Oils

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Abstract

The benefits of spices used daily for human health are also noticeable in traditional medicine and modern studies. Different species of pepper are characterized by an abundance of essential oils, in which valuable compounds accumulate. The main phytochemical compounds are found in peppers 3-karen, D-limonene, α-copaene, α-caryophylene, β-caryophylene and piperine [1]. Piperine has been shown to have a chemoprevention and antioxidant value and has a protective effect against radiation [2]. Black pepper (*Piper nigrum* L.), white pepper (*Piper album* L.) and fragrant pepper (*Pimenta dioica* L.), essential oils was prepared by hydrodistillation and liquid extraction methods. Gas chromatography with a mass spectrometer is commonly used to identify phytochemical compounds. The main compounds of black pepper (*Piper nigrum* L.), essential oil are 3-karen (25.99%), D-limonene (22.17%), caryophylene (13.28%), β-pinene (12.75%). The main compounds of fragrant pepper (*Pimenta dioica* L.) are caryophylene (63.11%), β-mircene (11.54%), eugenol (8.52%) and eucalyptol (6.96%). White pepper (*Piper album* L.), essential oil main compounds are 3-karen (26.22%), caryophylene (16.42%), D-limonene (17.83%) and β-pinene (11.66%).

Keywords: Black pepper, white pepper, essential oils, phytochemical compounds.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Production of Cyclodextrins and Their Application in Enantiomer Separations

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Abstract

Cyclodextrins are natural substances, truncated cone-shaped cyclic oligomers, which are obtained by enzymatic decomposition of one of the most important polysaccharides in nature - starch, which is broken down by alcaliphylic bacteria. In the Instrumental Analysis Open Access Center, such alcaliphillic bacteria were previously isolated from potato growth substrate [1]. Typical natural cyclodextrins contain six, seven, or eight glucose units and are labelled as α-, β-, and γ-cyclodextrins, respectively. The structure of cyclodextrins allows the formation of inclusion complexes because they have a hydrophobic cavity and hydrophilic rims called guest-host complexes. The complexes are used for dissolving lipophilic compounds, transporting such pharmaceuticals in the body. Cyclodextrins are used in many fields: medical, pharmaceutical, food, cosmetic, and biotechnology industries. In pharmacy, cyclodextrins are widely used in capillary electrophoresis and chromatographic methods. Cyclodextrins are used as chiral selectors in capillary electrophoresis and widely used as a stationary phase in chromatography. Also, a method of chiral gas chromatography was performed, where stationary phases were produced on the basis of cyclodextrins, when derivatized liquid or cyclodextrins dissolved in polyriloxanes were coated on the wall of capillary column as the stationary phase. Their wide applicability and good thermal stability allowed the separation of enantiomers. It depended on the shape and size of the analytes. Since each glycosidic residue consists of five chiral carbon atoms, the complexation is selective for enantiomers. The phases had the highest selectivity based on inclusive interactions [2]. In capillary electrophoresis, cyclodextrins have been the most widely used chiral selectors, a method designed to study weak, non-covalent supramolecular interactions [3]. Interestingly, thread cyclodextrins form mechanical supramolecular structures called as rotaxane. Polyrotaxanes were applied in capillary electrochromatography to impart a charge to the stationary phase. Although thread cyclodextrins have demonstrated chiral selectivity in the separation of enantiomers [4]. This is one of the practical applications of such supramolecular structures in the solid state. In 2016 the Nobel Prize in Chemistry was awarded to Jean-Pierre Sauvage, Sir J. Fraser Stoddart and Bernard L. Feringa for the practical use of solutions of similar superstructures as molecular machines, motors. In our further studies, the chromatographic stationary phases of cyclodextrins will be used to separate candidate racemic drugs.

Keywords: Cyclodextrins, chiral separations, chromatography, capillary electrophoresis, alcalihpilic bacteria, rotaxanes.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Biological Degradation of Creosote Contaminated Soil from a Phytoremediation and Mycoremediation

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Abstract

The wood industry uses more preservatives than any other industry worldwide. Creosote – the major chemicals employed for this purpose with serious potential health risks. Soil impregnated with creosote contain high concentrations of polycyclic aromatic hydrocarbons. Bioremediation - phyto and mycoremediatio represents one means by which these sites may be restored condition. Phytoremediation and mycoremediation are examined to identify development, limitations and perspectives for optimal utilization on creosote-contaminated soils. The process can be done by biological organisms such as bacteria, fungi, plants. The myco- and phytoremediation process is also important in making free or remediate the wastes in organic matter pollutant. In biological remediation metabolic processes release certain enzymes, biosurfactants, and microbial products that convert these hazardous pollutants to less toxic ones. According to the literature data there are selected number of plant species (*Medicago sativa L., Lolium perenne L., Salix viminalis, Trifolium pratense* and *etc.*) which have been found to be promising candidates for phytoremediation of such persistent pollutants as PAHs and could be used as an alternative technique to reduce creosote levels in soils [1,2].

Keywords: Wood, bioremediation, phytoremediation, mycoremediation, creosote.

Acknowledgements: The study was financed by the Research Council of Lithuania (project No. 01.2.2-LMT-K-718-01-0074 (REMTECH)).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

An Impact of Drying and Freezing to Biological Activity of Medicinal Plants and Spices

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Abstract

Drying is one of the most popular methods in medicinal plant raw material treatment ensuring availability of medicinal herbs throughout the year. Drying has been found to reduce the biological activity of medicinal herbs [1-3], however, different drying technologies, that rely in drying temperature, convection, etc.) may have different impact on the loss of the biological activity. Refrigeration is another preservation way of raw plant material, but it mostly dominates among fruits and vegetables rather than medicinal herbs. There are few studies in the scientific literature on freezing of medicinal herbs, which have revealed an increase of biologically active compounds [4], what could be explained by the fact that the cold breaks down the plant cells and improves the release of plant metabolites.

Various plants or spices, depending on their phytochemical composition, may react to drying and freezing differently, so it is important to investigate the effect. During the conference there will be discussed about benefits of preparing herbs tinctures, syrups, and other medicinal products from fresh and frozen material.

Keywords: Drying, freezing, medicinal plants, bioactive compounds.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

The Technology and Quality Evaluation of Low pH Value Gel with Glycolic Acid and Cellulose Polymers

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Abstract

Glycolic acid belongs to the group of alpha hydroxy acids (AHA) and it is widely used in topical pharmaceuticals and cosmetics. It is used for treatment of acne vulgaris, xerosis, seborrheic keratoses, irregular pigmentation, wrinkles and seborrheic skin. Glycolic acid leads to exfoliation, thickens epidermis and dermis, improves the skin's water barrier properties and augments the skin turgor. The formulation of stable, low pH, viscous gel formulations with desirable aesthetics and textures is often challenging. The remaining challenge in formulating with glycolic acid is balancing the efficacy and stability at formulation pH level of 3.5. Cellulose polymers hydroxyethylcellulose and hydroxypropylmethylcellulose are stable at pH levels from 2 to 12 so it is suitable for low pH gel formulations. Produced low pH gels is often in a low viscosity, so it is used synthetic and natural materials to increase the viscosity. Pectin is natural and safe thickener for cosmetical gels and it is suitable for pH levels from 2 to 6.

Twelve low pH value gels compositions with 2.0% glycolic acid, different concentrations of hydroxyethylcellulose (HEC) and hydroxypropylmethylcellulose (HPMC) (3.0, 4.0, 5.0 %) and the excipient pectin (0.5%) were produced. The pH level was chosen at 3.5 to lower the risk of glycolic acid side effects. Composition of gels was created based on analysis of scientific literature. Produced gels quality and changes over time are determined by the centrifugation, freeze – thaw tests, texture analysis, sensory analysis, viscosity tests.

The results of the study concluded that the addition of 0,5% natural excipient pectin and adding higher amount (4.0% and 5.0%) hydroxyethylcellulose and hydroxypropylmethylcellulose polymers remarkably and statistically significant ($p<0.05$) increased the viscosity compared to lower (3.0%) concentration of these cellulose polymers gels without pectin. The texture analysis revealed that the texture parameters – firmness, consistency, cohesiveness and index of viscosity correlate with gel composition and texture parameters are statistically significant ($p<0.05$) higher in gels with 0,5% concentration of natural excipient pectin and higher concentration of hydroxyethylcellulose and hydroxypropylmethylcellulose (4.0% and 5.0%) comparing with gels without pectin and with lower cellulose polymers concentration (3.0%). Centrifugation, freeze – thaw test and sensory analysis showed that all low pH gels were physically stable both – with and without pectin.

The results of the study provided that cellulose polymers (hydroxyethylcellulose and hydroxypropylmethylcellulose) in concentrations from 3.0% to 5.0% are suitable for the production of physically stable low pH gels with 2,0% glycolic acid. Studies have shown that the addition of the natural excipient pectin improves the physical, texture and stability characteristics of gels with glycolic acid and cellulose polymers. All gels with cellulose polymers with pectin remained stable throughout the study period.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Technology and Assessment of Orodispersible Granules

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Abstract

Oral route is still widely accepted route but have a common drawback of difficulty in swallowing**.** Therefore, there is a growing interest in new orodispersible formulations, such as orally disintegrating granules, in the scientific literature. It is an innovative and user-friendly dosage form that fast disintegrate in the mouth without water. The active substance is quick absorbed in the oral cavity, pharynx or esophagus and also enters the stomach together with saliva, hence orodispersible granules are used when a rapid therapeutic effect is required, such as during allergic reactions. Besides, dosage forms is easy and convenient to use for pediatrics, geriatrics, mentally disabled, bed-ridden patients who have difficulty in swallowing or for patients who are travelling and do not have access to water.

Orally disintegrating granules produced by simple and innovative moisture-activated dry granulation method using different amount of microcrystalline cellulose and polyvinylpyrrolidone. The technological properties of the produced granules are evaluated and the quality is determined with the tests specified in European pharmacopoeia: uniformity of content, disintegration time.

The best technological properties were in orodispersible granules consisting of 17.5% microcrystalline cellulose and 17.5% polyvinylpyrrolidone: flowability 6.89±0.41 g/s., angle of repose 25.67±0.7º, Carrʾs index 9.09±0.677 %, Hausner coefficient 1.100±0.06 – these granules corresponded to excellent flowability. The flowability of the other granules was good. Based on the results, the highest percentage of particles obtained corresponded to a particle size of 450-560 μm. The content uniformity test showed that the amount of active substance in all granules complied with the permissible limits 85-115%. Disintegration test showed that using moisture-activated dry granulation method all granules dissintegrates in 1 min: preparation No 1 – 23.71±0.47 s., preparation No 2 – 26.52±0.48 s., preparation No 3 – 30.09±0.95 s., preparation No 4 – 32.38±0.47 s., preparation No 5 – 38.10±0.55 s.

The results of the study provided that the moisture-activated dry granulation method is suitable for the production of orally disintegrating granules which rapidly disintegrating in the oral cavity. Furthermore, microcrystalline cellulose and polyvinylpyrrolidone improve the technological properties of the orally disintegrating granules.

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Interfacing the Rachets to the Fluidic System for the Separation of Macromolecules

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Abstract

Separation techniques such as capillary electrophoresis or liquid chromatography are the leading choices for analyzing complex biosamples. On the other hand, these methods require rigorous sample preparation, and as a result, analytical throughput declines in practical routine applications.

In this work, we present an interface of a 3D-fabricated rachet with the fluidic system and capacitively coupled contactless conductivity detector. The device is intended for uninterrupted separation of the molecules of a biosample. This realization of the macromolecular rachets was demonstrated with three flow-through channels providing different fractions. Each fraction is monitored using a capacitively coupled contactless conductivity detector. Also, there is a capability to couple a dedicated fraction channel to the capillary electrophoresis instrument. Such hyphenation can be used for monitoring purposes in biotechnological applications.

Interfacing, fabrication, operation peculiatrities, and preliminary results will be discussed.

Acknowledgment

Research Council of Lithuania financed this project, the project No. S-MIP-19-60.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Biotechnologies for the Microbe-Mediated Remediation of Expended Railway Mrossties: an Integrative Approach

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Abstract

Creosote is a widely used biocide for wood preservation obtained from distillation of hard coal tar. It contains several hundreds of different chemicals such as aromatic hydrocarbons, including polycyclic aromatic hydrocarbons (PAHs) and alkylated PAHs.

The usage of microbe-mediated technologies [1,2] for the remediation of waste materials containing creosote attracts growing interest as eco-friendly and economically valuable alternative to the conventional methods.

The aim of this research focuses on the development and scale-up of technologies for the biological decontamination of expended railway wooden crossties. It encompasses the study of the stress effects on the ligninolytic enzymes secretion of *Basidiomycetes*, in particular due to heat shock and cold shock, the influence of the inoculum format, soil and soil pre-treatment method on the bioremediation efficiency, the antagonistic and synergistic effects of the interactions in the microbial community and the investigation on the large scale effects on the overall process.

Keywords: Railway crossties, creosote, bioremediation, PAHs.

Acknowledgements: The study was financed by the Research Council of Lithuania (project No. 01.2.2-LMT-K-718-01-0074 (REMTECH)).

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ISSN:2335-8653
ONLINE ISSN: 2335-8718

Fluctuations in Concentrations of N and P Compounds in Marvelė River: a Case Study of Urban Pollution

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Abstract

The aim of this study was to evaluate the pollution of the urban stream Marvele, by investigating the seasonal and spatial distribution of the most important nutrients, i.e. ammonium (NH_4^+-N), nitrates (NO_3^--N) and phosphates (PO_4^{3-}-P) in water of the river. Marvelė stream is a small 1st type river (catchment area 21,1 km^2), left tributary of Nemunas. The valley of the river is situated in both agricultural (upper course), forested (middle course) or densely populated (lower course) areas. Two outfalls discharge surface runoff water from densely populated areas (Aleksotas and other districts) to Marvelė river near the Paupelio street and to a right tributary of Marvelė. As the river flows through residential, green and recreation areas of the city, pollution of Marvelė is a serious problem of Kaunas. Samples of water in the course of the Marvelė river and it's tributary were collected in February (winter season) and June (summer season). The surface runoff outfall (SRO) in the tributary had stronger effect than the SRO near the Paupelio street. We observed an increased concentrations of NO_3^--N (28%, $p<0.05$) in winter and PO_4^{3-}-P (12%, $p<0.05$) in summer bellow the SRO in the tributary. However, pollution from other sources seems to be more important in Marvelė river, as the highest concentrations of investigated nutrients were mostly found not bellow the SROs, but in other sections of the river basin. The highest concentrations of $PO_4^{3—}$P and NH_4^+-N were found in all channel of the tributary. The average concentrations of $PO_4^{3—}$P and NH_4^+-N in the tributary were 0,27 mg/l and 12,7 mg/l in summer. Concentrations in winter were from 7- to 8-fold lower, however in both seasons this segment of Marvelė basin were attributed to bad or very bad ecological status class. Whereas the highest concentrations of nitrates were found in winter in the upper and middle courses and the mouth of the river, reaching 0,69 mg/l, 0,61 mg/l and 0,51 mg/l, respectively. The ecological status class of Marvelė river in these sections were very bad. The increase of nitrates content in the upper course of the river were most possibly induced by nitrates wash off from agricultural fields, whereas an increase of $NO_3^—$N near the mouth implies natural purification of water by nitrification of ammonium, which get into the river in the tributary. The reasons of high concentrations of $PO_4^{3—}$P and NH_4^+-N in the tributary is unclear, however the most possible reason is household sewages. The most probable source of pollution in this case is urbanized residential areas of Kaunas (Kazliškiai, Tirkiliškiai, Yliškės).

Keywords: Urban stream pollution, phosphates, ammonium, nitrates, surface runoff outfalls.

ISSN:2335-8653
ONLINE ISSN: 2335-8718

Rab14 Endosome Targeting to the Intercellular Bridge Regulates Cytokinesis

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Abstract

Intracellular membrane transport is a major factor controlling cellular metabolism and survival. Vesicular delivery of membranes and various proteins to the specific cellular sites and organelles at different phases of the cell cycle, as well as during the performance of different cell functions, is essential for the maintenance of normal cell homeostasis. One of the most important events during cell lifetime is cell division, which culminates with an event known as abscission. Abscission is a complex cellular process that is prerequisite for mitotic division and defines upon successful physical separation of two newly formed daughter cells. It is now well-established that coordinated and localized changes in microtubule dynamics and actin cytoskeleton rearrangement are vital for cytokinetic ring formation, as well as for the establishment of the abscission site. In this regard, Rab GTPases are amongst the most important regulators of endocytic membrane transport in eukaryotic cells. Actin cytoskeleton reorganization during abscission would not be possible without the interplay between Rab11- and Rab35-containing endosomes and their effector proteins, whose roles in regulating endocytic pathways at the cleavage furrow have now been studied extensively. By employing various Cell Biology and Molecular Biology techniques we down-regulated Rab14 and performed different functional assays, which allowed us to identify Rab14 as a novel regulator of cytokinesis. We demonstrated that depletion of Rab14 causes either cytokinesis failure or significantly prolongs division time. We showed that Rab14 contributes to the efficiency of recruiting Rab11-endosomes to the intercellular bridge microtubules and that Rab14 knockout leads to inhibition of actin clearance at the abscission site. Collectively, our data identified Rab14 as a regulator of actin depolymerisation and endosome targeting during cytokinesis.

Keywords: Rab14, endosomes, intercellular bridge, actin, cell division, cytokinesis.