

*The 6<sup>th</sup> International Conference*

*New Trends on*

*Sensing - Monitoring - Telediagnosis*

*for Life Sciences*

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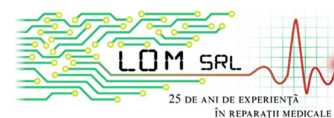


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## Topics

### *(Tele)diagnosis, (Tele)monitoring and (Tele)management in Medicine*

- Analytical and bioanalytical methods for screening and diagnosis in medicine
- Telemedicine and e-Health
- E-health and education
- Endocrine disruptors: advances in assessing environmental health risk and human health
- Environmental pollution and health- sensing, (tele)monitoring and modelling of environmental risk factors
- Personalized medicine
- Biological therapies in the treatment of medical conditions
- Social innovations to improve the quality of life and well-being of elder people
- Assessment of disease susceptibility and diagnosis
- Improving health information, data exploitation and providing an evidence base for health policies and regulation
- (Tele)diagnosis and (tele)management of environmental factors related disorders
- Impacts of poor health on economic output and productivity
- Approaching melotherapy through online sources/remote methods
- Improving health promotion, lifelong physical activity and disease prevention
- Make business a key partner in promoting health and preventing disease
- Physiotherapy – a way to increase the quality and sustainability of patient's lives
- Bioeconomy for health
- Social responsibility for health
- Health and care systems during and after COVID-19 pandemic

### *New Trends in Nutritional Sciences and Food Control*

- Analytical and bioanalytical methods for screening and diagnosis in medicine
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- Endocrine disruptors: advances in assessing environmental health risk and human health
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- Make business a key partner in promoting health and preventing disease
- Physiotherapy – a way to increase the quality and sustainability of patient's lives
- Bioeconomy for health
- Social responsibility for health
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### *New Trends in (Bio)engineering Sciences Applied in Life Sciences*

- Innovative smart healthcare and bio-medical systems: artificial intelligence, intelligent computing and connected technologies
- Assistive technologies to the needs of the elderly, disabled and chronic disease patients
- Materials for health
- New (bio)materials used in medical and electronic fields
- Progress in 3D bioprinting technology for tissue/organ regenerative engineering
- Nanotechnology for life sciences
- Personalized electronic tools for effective virtual rehabilitation
- Built environment, urbanization and chronic non-communicable disease
- Hospital hygiene through engineering and ICT solutions.
- Building materials impact on human health
- Occupant safety and health in construction
- Monitoring the indoor air quality in buildings
- Electronic medical devices
- Image, data and signal processing in life sciences
- Computing and simulation in life sciences
- Medical equipment and instruments

## CONTENT

<b>PL.1.</b>	<b>Optical biosensors for diagnostics of infectious viral disease: a recent update</b> Jean-Louis MARTY (1,2) 1 – Sensbiotech, Ceret, France; 2 – Université de Perpignan, Perpignan Cedex, France	<b>18</b>
<b>PL.2.</b>	<b>Biosensing across continents: from advanced materials development to field deployable devices</b> Silvana ANDREESCU Clarkson University Potsdam, USA	<b>19</b>

## S1. (Tele)diagnosis, (Tele)monitoring and (Tele)management in Medicine

<b>S1-KN.1.</b>	<b>Towards point of use systems for fast and on-site detection</b> Mihaela TERTIŞ, Oana HOSU, Andreea CERNAT, Bogdan FEIER, Cecilia CRISTEA Analytical Chemistry Department, Faculty of Pharmacy, “Iuliu Haţieganu” University of Medicine and Pharmacy, Cluj Napoca, Romania	<b>21</b>
<b>S1-KN.2.</b>	<b>Long-term treatment with chloroquine for healthy aging in animal model</b> Aurel POPA WAGNER (1,2) 1-Center for Clinical and Experimental Medicine, University of Medicine and Pharmacy, Craiova, Romania; 2-Essen University Medical School, Germany	<b>22</b>
<b>S1-KN3</b>	<b>Portable Surface Plasmon Resonance system for blood marker analysis supporting rapid point-of-care diagnostics and treatment monitorin</b> Cristina POLONSCHII (1), Arian OLOUMI (1,2), Mihnea ROSU-HAMZESCU (1), Sorin DAVID (1), Eugen GHEORGHIU (1,2) 1-International Centre of Biodynamics, Bucharest, Romania 2-University of Bucharest, Bucharest, Romania	<b>23</b>
<b>S1-O1</b>	<b>Use of wearable devices for health and fatigue monitoring</b> Horia Alexandru MODRAN (1), Doru URSUTIU (1,2), Cornel SAMOILĂ (1,3), Lilia ALJIHMANI (4), Khalid QARAQE (4) 1 - Transilvania University of Braşov, Brasov, Romania 2 - Romanian Academy of Scientists, Bucharest, Romania 3 - Romanian Academy of Technical Sciences, Bucharest, Romania 4 - Texas A&M University at Qatar, Doha, Qatar	<b>24</b>
<b>S1-O2</b>	<b>Studies of determination of urinary bioactive metabolites</b> Ligia CHELMEA, Mihaela BADEA, Ioan SCÂRNECIU Transilvania University of Braşov, Faculty of Medicine, Brasov, Romania	<b>25</b>

<b>S1-03</b>	<b>Myo-inositol - never-ending resource in patients with polycystic ovary syndrome (PCOS)</b> Elena VASILICA, Laura Elena GAMAN “Carol Davila” University of Medicine and Pharmacy, Faculty of Medicine, Bucharest, Romania	<b>26</b>
<b>S1-04</b>	<b>Clinical and paraclinical evaluations of deficiency rickets</b> Maria TOTAN (1,2), Oana-Raluca ANTONESCU (1), Ionela MANIU (3,4), Elisabeta ANTONESCU (1,5) 1- Lucian Blaga University of Sibiu, Faculty of Medicine, Sibiu, Romania; 2- Clinical Pediatric Hospital, Clinical Laboratory, Sibiu, Romania; 3- Lucian Blaga University of Sibiu, Faculty of Sciences, Mathematics and Informatics Department, Research Center in Informatics and Information Technology, Sibiu, Romania 4- Pediatric Clinical Hospital Sibiu, Research team, Sibiu, Romania 5- County Clinical Emergency Hospital, Sibiu, Romania	<b>27</b>
<b>S1-05</b>	<b>Drug safety for elderly people – ethical, psychological and pragmatic aspects</b> Liliana ROGOZEA, Mihaela BADEA, Angela REPANOVICI, Claudiu COMAN, Maria Cristina BULARCA, Daniela POPA, Eleonora DINU, Florin LEAŞU Transilvania University of Brasov, Braşov, Romania	<b>29</b>
<b>S1-06</b>	<b>Early puberty – clinical and paraclinical aspects</b> M.A.VLAD (1,2), B.M.PASCU (1), N.LACATUS (1), M.D.HAIDAUTU (1), D.A.TUTOVEANU (1), A.CIMPEAN (2) 1-INSMC Alessandrescu-Rusescu, Pediatrics Department, Bucharest, Romania 2-Faculty of Biology, Bucharest University, Bucharest, Romania	<b>30</b>
<b>S1-07</b>	<b>Towards an integrated automatic platform for rapid determination of antibiotic susceptibility of target bacteria</b> Sorin DAVID (1), Ionela-Cristina PETCU (1,2), Ioana-Cristina CERNAT (1,2), Cristina POLONSCHII (1), Raluca-Elena MUNTEANU (1,3), Mihaela GHEORGHIU (1,3), Dumitru BRATU (1), Daniela TUDOR (1,3), Eugen GHEORGHIU (1,3) 1-International Centre of Biodynamics, Bucharest, Romania; 2-University Politehnica of Bucharest, Bucharest, Romania; 3-University of Bucharest, Bucharest, Romania	<b>31</b>
<b>S1-08</b>	<b>EVALUATION OF OXIDATIVE STRESS IN THE GRANULOMATOUS DISEASE</b> C.E.DELIA <sup>1,2</sup> , M.G.TOMA <sup>1</sup> , A.V.COCHINO <sup>1</sup> , A.GHITA <sup>1</sup> <sup>1</sup> National Institute for Mother and Child Health Alessandrescu-Rusescu, Pediatric Component; <sup>2</sup> Faculty of Biology, Bucharest University	<b>32</b>
<b>S1-09</b>	<b>Lipid peroxidation and oxidative stress</b> Mihaela HORHOCEA (ŞTEFAN) (1,2), Gabriela MARIN-ŞTEFAN (1,2), Laurenţiu NEDELICU (1,2), Mihaela BADEA (1) 1- Faculty of Medicine, University of Transilvania Braşov, România 2- County Clinical Hospital of Brasov, Brasov, Romania	<b>33</b>
<b>S1-010</b>	<b>Vitamin D status in the pediatric population in the pandemic context</b> M.A.VLAD (1,2), C.E.DELIA (1,2), M.D.HAIDAUTU (1), N.LACATUS (1), M.G.TOMA (1)	<b>34</b>



	1- NIMCH Alessandrescu-Rusescu, Pediatric Department, Bucharest, Romania 2- Faculty of Biology, Bucharest University, Bucharest, Romania	
<b>S1-O11</b>	<b>Telemedicine for older people. Advantages and disadvantages</b> Sergiu Costin CIUPERCĂ Faculty of Medicine, University of Transilvania Braşov, România	<b>35</b>
<b>S1-P1</b>	<b>Strategies to address the non-specific adsorption in electrochemical aptasensors for clinically relevant proteins</b> Roberta Maria BANCUI, Alina VASILESCU, Cristina POLONSCHII International Centre of Biodynamics, 1B Intrarea Portocalelor, Bucharest, Romania	<b>6</b>
<b>S1-P2</b>	<b>The behavior of the medical staff and of the students from the Faculty of Medicine regarding the use of the mobile phone</b> Cristina ADOCHIŢE (1), Mihaela IDOMIR (1), Sarah Costinaş (1), Marius MOGA (1), Liliana ROGOZEA (1), Daniela NECULOIU (1), Laura Floroian (1), Catalin VITELARU (2), Anca C. PARAU (2), Adrian E. KISS (2), Iulian PANA (2), Mihaela DINU (2), Lidia R. CONSTANTIN (2), Alina VLADESCU (2,3), Mihaela BADEA (1) 1- Faculty of Medicine, University of Transilvania Braşov, România; 2- National Institute for Research and Development in Optoelectronics INOE 2000, Magurele, Romania; 3- Research Center for Physical Materials Science and Composite Materials, Research School of Chemistry & Applied Biomedical Sciences, Tomsk Polytechnic University, Tomsk, Russia	<b>37</b>
<b>S1-P3</b>	<b>Vitamin D deficiency in diabetes</b> Gabriela MARIN-STEFAN (1,2), Mihaela HORHOCEA (STEFAN) (1,2), Mihaela BADEA (1) 1- Faculty of Medicine, University of Transilvania Braşov, România 2- County Clinical Hospital of Brasov, Brasov, Romania	<b>38</b>
<b>S1-P4</b>	<b>Vitamin D levels and pathology association in hospitalized patients</b> Sandica N. BUCURICA (1,2), Alexandra HOZA (2), Monica GRIGORE (2), Mihaela PAVALEAN (2), Mariana JINGA (1,2), Florentina IONITA-RADU (1,2), Laura GAMAN (2), Ioana PRODAN (2) 1- Carol Davila University of Medicine and Pharmacy, Bucharest, Romania 2- Central Emergency Military Hospital Bucharest, Romania	<b>39</b>

## S2. New Trends in Nutritional Sciences and Food Control

<b>S2-KN1</b>	<b>Botanical extracts as new food as new food additives: are they really new?</b> Patrizia RESTANI (1,2), Chiara DI LORENZO (1,2), Corinne BANI (1), Francesca COLOMBO (1) 1 - Dept. Pharmacological and Biomolecular Sciences, Università degli Studi di Milano, Milan, Italy 2 - CRC "Innovation for Well-Being and Environment", Università degli Studi di Milano, Milan, Italy	<b>41</b>
<b>S2-KN2</b>	<b>Rapid and sensitive tools for enhanced chemical safety of food</b> G. ISTAMBOULIE, C. AYMARD, S. BEN AISSA, G. CATANANTE, T. NOGUER 1-Université de Perpignan Via Domitia, Biocapteurs-Analyse-Environnement, Perpignan, France 2-Laboratoire de Biodiversité et Biotechnologies Microbiennes, Sorbonne Universités (UPMC) Paris 6 et CNRS Observatoire Océanologique, Banyuls-sur-Mer, France	<b>42</b>
<b>S2-O1</b>	<b>Electroanalytical method coupled with chemometry for detection of virgin olive oil adulteration</b> Constantin APETREI (1), Elisabeta-Irina GEANĂ (2), Irina Mirela APETREI (3) 1- Faculty of Sciences and Environment, Dunarea de Jos University of Galati, Galati, Romania. 2- National Research and Development Institute for Cryogenics and Isotopic Technologies - ICSI Rm. Valcea, Ramnicu Valcea, Romania. 3- Faculty of Medicine and Pharmacy, Dunarea de Jos University of Galati, Galati, Romania	<b>43</b>
<b>S2-O2</b>	<b>Develop a series of ultra-sensitive detection methods for mycotoxins</b> Cheng YANG, Shengnan CUI, Xiaolin CHU, Fathimath ABBAS, Yaqi ZHANG, Shengnan DONG, Yue WANG, Yingying SUN State Key Laboratory of Fine Chemicals, Department of Chemistry, School of Chemical Engineering, Dalian University of Technology, Dalian, China	<b>44</b>
<b>S2-O3</b>	<b>Sustainable approach for functional fermented product development</b> Cristina POPOVICI (1), Gjore NAKOV (2,3), Jasmina LUKINAC (4), Marko JUKIĆ (4) 1-Technical University of Moldova, Faculty of Food Technology, Chisinau, Republic of Moldova; 2-Agricultural Academy, Institute of Cryobiology and Food Technologies, Sofia Bulgaria 3-Technical University of Sofia, College of Sliven, Sliven, Bulgaria 4-Josip Juraj Strossmayer University of Osijek, Faculty of Food Technology, Osijek Croatia	<b>45</b>
<b>S2-O4</b>	<b>Harmonized in vitro digestion and Ussing chamber to investigate the effects of polyphenols on the intestinal physiology</b> Marco TRETOLA, Giuseppe BEE, Frigga DOHME-MEIER, Paolo SILACCI 1-University of Milan, Lodi, Italy 2-Agroscope, Posieux, Switzerland	<b>46</b>

<b>S2-05</b>	<b>Cider yeasts associated with Hardanger cider during fermentation process</b> Urban ČESNIK (1), Mitja MARTELANC (1), Branka MOZETIČ VODOPIVEC (1), Ingunn OVSTHUS (2), Lorena BUTINAR (1) 1- Wine Research Centre, University of Nova Gorica, Nova Gorica, Slovenia 2 - NIBIO Ullensvang, 5781 Lofthus, Norway	<b>47</b>
<b>S2-06</b>	<b>New educational trends in European teenagers obesity management</b> Monica TARCEA, Roxana MARTIN-HADMAŞ, Irina MATRAN, George VRAPCEA, Anișoara POP Department of Community Nutrition and Food Safety, G.E. Palade University of Medicine, Pharmacy, Science and Technology of Targu Mures, Romania	<b>49</b>
<b>S2-07</b>	<b>Invasive knotweed species as a source of bioactivities</b> Lea POGACNIK (1), Rui SILVA (2) 1- Biotechnical Faculty, University of Ljubljana, Slovenia 2- Universidade de Lisboa, Lisbon, Portugal	<b>50</b>
<b>S2-08</b>	<b>Use trend and attitude of young people towards food supplements: comparison between surveys performed in 2011 and 2021</b> Matteo LODA (1), Chiara DI LORENZO (1), Francesca COLOMBO (1), Corinne BANI (1), Patrizia RESTANI (1,2), Mihaela BADEA (3) 1- Department of Pharmacological and Biomolecular Sciences, Università degli Studi di Milano, Milano, Italy 2- Centri di Ricerca Coordinata (CRC) "Innovation for Well-Being and Environment", Università degli Studi di Milano, Milano, Italy 3- Faculty of Medicine, Transilvania University of Brasov, Romania	<b>51</b>
<b>S2-09</b>	<b>Screening grapes for infection by Botrytis cinerea: detection of spores versus the evaluation of laccase activity</b> Alina VASILESCU (1), Elena BRINDUSE (2), Szilvester GASPARGASPAR (1), Monica POTARA (3), Andreea Catalina LULEA (1), Roberta Maria BANCUI (1), Robert RUGINESCU (4), Cristina PURCAREA (4) 1- International Centre of Biodynamics, Bucharest, Romania 2- Institute for Research and Development for Vine and Wine, Valea Calugareasca, Romania 3- Department of Microbiology, Institute of Biology Bucharest of the Romanian Academy, Bucharest, Romania 4- Babes-Bolyai University, Nanobiophotonics and Laser Microspectroscopy Center, Interdisciplinary Research Institute in Bio-Nano-Sciences, Cluj-Napoca, Romania	<b>52</b>
<b>S2-010</b>	<b>Research on the use of Saccharina latissima seaweed as an innovative ingredient in the bakery industry</b> Liviu GACEU (1), Mihaela BADEA (1), Michael BANTLE (2), Cristina ADOCHITE (1), Oana Bianca OPREA (1) 1 – Transilvania University of Brasov, Brasov, Romania 2 – Sintef Energy Research, Norway	<b>54</b>
<b>S2-011</b>	<b>Insect material as innovative feed ingredient in animal nutrition</b> Matteo OTTOBONI, Marco TRETOLA, Federica CHELI, Luciano PINOTTI	<b>56</b>

	Department Veterinary Medicine and Animal Sciences, University of Milan, Italy	
<b>S2-O12</b>	<b>PlantMolecularTasteDB: a versatile tool in designing fortified food</b> Dorin DRAGOŞ (1,2), Mădălina PETRAN (3), Cristiana GRĂDINARU (3), Marilena GILCĂ (3) 1-Department of Medical Semiology, Faculty of Medicine, Carol Davila University of Medicine and Pharmacy, Bucharest, Romania; 2- 1st Internal Medicine Clinic, University Emergency Hospital Bucharest, Carol Davila University of Medicine and Pharmacy, Bucharest, Romania; 3-Department of Functional Sciences, Biochemistry, Faculty of Medicine, Carol Davila University of Medicine and Pharmacy, Bucharest, Romania	<b>57</b>
<b>S2-O13</b>	<b>Pigmented cereals and gluten related diseases</b> Francesca COLOMBO (1), Stefano PIAZZA (1), Corinne BANI (1), Giulia MARTINELLI (1), Enrico SANGIOVANNI (1), Patrizia RESTANI (1,2), Mario DELL'AGLI (1), Chiara DI LORENZO (1) 1-Department of Pharmacological and Biomolecular Sciences, Università degli Studi di Milano, Milano, Italy 2-Centri di Ricerca Coordinata (CRC) "Innovation for Well-Being and Environment", Università degli Studi di Milano, Milano, Italy	<b>58</b>
<b>S2-O14</b>	<b>Nutritional status in Crohn's disease</b> Ruxandra CIOARCA-NEDELCU Biochemistry Department of Medicine and Pharmacy University "Carol Davila", Bucharest, Romania	<b>59</b>
<b>S2-O15</b>	<b>Food - the history of a society</b> Serena Lucia CIOCAZAN Faculty of Medicine, Transilvania University of Braşov, Braşov, Romania	<b>60</b>
<b>S2-P1</b>	<b>Phenolic profile and antioxidant activity of different corn and rice varieties</b> Corinne BANI (1), Chiara DI LORENZO (1), Patrizia RESTANI (1,2), Francesca MERCOGLIANO (3), Francesca COLOMBO (1) 1- Department of Pharmacological and Biomolecular Sciences, Università degli Studi di Milano, Milano, Italy 2- Centro di Ricerca Coordinata "Innovation for Well-Being and Environment" (CRC), Università degli Studi di Milano, Milano, Italy 3- Dr. in Safety Assessment of Xenobiotics and Biotechnological Products, Università degli Studi di Milano, Milano, Italy	<b>61</b>
<b>S2-P2</b>	<b>Electrochemical determination of hydroxytyrosol in extravirgin olive oils using screen-printed electrodes modified with single wall carbon nanotubes and tyrosinase</b> Alexandra Virginia BOUNEGRU, Constantin APETREI "Dunarea de Jos" University of Galati, Faculty of Sciences and Environment, Galaţi, Romania	<b>62</b>
<b>S2-P3</b>	<b>Consumer perceptions of functional foods with antioxidant and anticancer potential</b> Ecaterina ROBU, Elena SERGHEEVA, Cristina POPOVICI Technical University of Moldova, Faculty of Food Technology, Chisinau, Republic of Moldova	<b>63</b>

<b>S2-P4</b>	<b>Facile specific detection of acetaldehyde in air samples using electrochemical enzyme biosensors</b> Alina VASILESCU (1), Victoria Oana PAUN (2), Roberta Maria BANCUI (1), Sorin DAVID (1), Georgiana NECULA-PETRAREANU (2), Cristina PURCAREA (2), Petru EPURE (3) 1- International Centre of Biodynamics, Bucharest, Romania; 2 - Department of Microbiology, Institute of Biology Bucharest of the Romanian Academy, Bucharest, Romania 3 - EPI SISTEM SRL, Sacele, Brasov, Romania	<b>64</b>
<b>S2-P5</b>	<b>Consumer perceptions of functional foods for type 2 diabetes in Romania and Republic of Moldova</b> Elena SERGHEEVA (1), Mihaela GEICU-CRISTEA (2), Florentina MATEI (2), Mona Elena POPA (2), Elisabeta Elena POPA (2), Ecaterina ROBU (1), Cristina POPOVICI (1) 1-Technical University of Moldova, Faculty of Food Technology, Chisinau, Republic of Moldova 2-University of Agronomic Sciences and Veterinary Medicine of Bucharest, Faculty of Biotechnology, Bucharest, Romania	<b>66</b>

### S3. New Trends in (Bio)engineering Sciences Applied in Life Sciences

<b>S3- KN1</b>	<b>In situ remote monitoring of novel biodegradable cardiovascular stents - design &amp; validation</b> Lilia ALJIHMANI (1), Hasan ABBAS (2), Bilal MANSOOR (3), Khalid QARAQE (1) 1 - Electrical and Computer Engineering, Texas A&M University at Qatar, Education City, Doha, Qatar 2 - James Watt School of Engineering, University of Glasgow, Glasgow, United Kingdom 3 - Mechanical Engineering, Texas A&M University at Qatar, Education City, Doha, Qatar	<b>68</b>
<b>S3-KN2</b>	<b>Synthesis and characterization of nanostructured silver based antibacterial coatings on protective polymer foils of touch screen devices</b> Catalin VITELARU (1), Anca C. PARAU (1), Adrian E. KISS (1), Iulian PANA (1), Mihaela DINU (1), Lidia R. CONSTANTIN (1), Alina VLADESCU (1,2), Lavinia E. TONOFREI (3), Cristina S. ADOCHITE (4), Sarah COSTINAS (4), Liliana ROGOZEA (4), Marius MOGA (4), Mihaela E. IDOMIR (4), Mihaela BADEA (4) 1- National Institute for Research and Development in Optoelectronics INOE 2000, Magurele, Romania; 2- Research Center for Physical Materials Science and Composite Materials, Research School of Chemistry & Applied Biomedical Sciences, Tomsk Polytechnic University, Tomsk, Russia 3- ATS Novus SRL, Bucharest, Romania	<b>69</b>

	4- Faculty of Medicine, Transylvania University of Brasov, Brasov, Romania	
<b>S3-O1</b>	<b>Development of nanotechnologies and their impact on public health</b> Cristina ADOCHIȚE, Mihaela BADEA Faculty of Medicine, University of Transilvania Brasov, România	<b>70</b>
<b>S3-O2</b>	<b>Functional Cell Sorting and Cellular Spectroscopy: New technologies for the study of cellular heterogeneity</b> Catalin CHIMEREL University of Exeter, United Kingdom	<b>71</b>
<b>S3-O3</b>	<b>Defining protein function and activity associated to different pathologies by combining molecular, cellular, biochemical and analytical methods</b> Gabriela N. CHIRITOIU, Cristian V.A. MUNTEANU, Marioara CHIRITOIU, Simona GHENEA, Andrei PETRESCU, Stefana PETRESCU Institute of Biochemistry of the Romanian Academy, Bucharest, Romania	<b>72</b>
<b>S3-O4</b>	<b>Lab-chip evaluation of bacterial cells' dynamics for rapid antimicrobial susceptibility testing</b> Raluca-Elena MUNTEANU (1,2), Daniela TUDOR (1,2), Sorin DAVID (1), Eugen GHEORGHIU (1,2), Mihaela GHEORGHIU (1,2) 1- International Centre of Biodynamics, Bucharest, Romania; 2 - Faculty of Biology, University Of Bucharest, Romania	<b>73</b>
<b>S3-O5</b>	<b>Novel reflected light microscopy assay for rapid, label free assessment of cellular processes and cellular status at single cell level</b> Daniela-Alexandra TUDOR (1,2), Raluca-Elena MUNTEANU (1,2), Cristina POLONSCHII (1), Eugen GHEORGHIU (1,2), Mihaela GHEORGHIU (1,2) 1- International Centre of Biodynamics, Bucharest, Romania; 2 - Faculty of Biology, University Of Bucharest, Romania	<b>74</b>
<b>S3-O6</b>	<b>ZrCu based thin films metallic glasses in medicine</b> A. VLADESCU (1), L.R. CONSTANTIN (1), G.A. JURAVLEA (1,2), A. PARAU (1) 1-National Institute for Optoelectronics INOE 2000, Department for Advanced Surface Processing and Analysis by Vacuum Technologies, Magurele, Romania 2-University Politehnica of Bucharest, Faculty of Material Science and Engineering, Bucharest, Romania	<b>75</b>
<b>S3-O7</b>	<b>MXenes-based bioanalytical sensors: design, characterization and applications</b> Reem KHAN, Silvana ANDREESCU Clarkson University Potsdam, USA	<b>76</b>
<b>S3-O8</b>	<b>Investigation of a truncated aptamer for ofloxacin detection</b> Gaëlle CATANANTE (1), Sondes Ben AISSA (1,2), Georges ISTAMBOULIE (1), Thierry NOGUER (1) 1- BAE-LBBM Laboratory, Université de Perpignan Via Domitia, Perpignan, France 2-Sensors and Biosensors Group, Laboratoire de Chimie Analytique et Electrochimie (LR99ES15), Faculté des Sciences de Tunis, Universitaire	<b>77</b>

	de Tunis El Manar, Tunis, Tunisia	
<b>S3-O9</b>	<b>Multifunctional nanomaterials an emerging area for biosensors</b> Akhtar HAYAT Interdisciplinary Research Centre in Biomedical Materials (IRCBM), COMSATS University, Islamabad, Lahore Campus, Pakistan	<b>78</b>
<b>S3-O10</b>	<b>Nano-engineered surface coatings on polymer substrates: the correlation between surface morphology, hydrophilic/hydrophobic properties and antibacterial activity</b> Sibu C PADMANABHAN (1), Sukhananazerin ABDULLA (1), Paula FERNANDEZ GARCIA (2), Malco-Cruz ROMERO (2), Joseph P KERRY (2), Michael A MORRIS (1) 1- Advanced Materials and BioEngineering Research (AMBER) Centre, & School of Chemistry, University of Dublin, Trinity College, Dublin 2, Ireland 2- Department of Food and Nutritional Sciences at University College Cork, Cork, Ireland	<b>79</b>
<b>S3-O11</b>	<b>Smart nanocoatings with thermo-switchable antibacterial activity</b> Joanna RACZKOWSKA (1), Yuriy STETSYSHYN (2), Kamil AWSIUK (1), Ostap LISHCHYNSKYI (2), Yana SHYMBORSKA (1), Svyatoslav NASTYSHYN (1), Andrzej BUDKOWSKI (1) 1- Smoluchowski Institute of Physics, Jagiellonian University, Kraków, Poland 2- Lviv Polytechnic National University, Lviv, Ukraine	<b>80</b>
<b>S3-P1</b>	<b>Comparative study on the antioxidant activity of extra virgin olive oil samples using a newly developed electrochemical method and DPPH spectrophotometric assay</b> Irina-Georgiana BULGARU (MUNTEANU), Constantin APETREI Department of Chemistry, Physics and Environment, Faculty of Sciences and Environment, “Dunărea de Jos” University of Galaţi, Galaţi, Romania	<b>81</b>
<b>S3-P2</b>	<b>IT technologies for medical data management</b> Liviu Doru DOGAR, Laura FLOROIAN Transilvania University of Brasov, Brasov, Romania	<b>82</b>
<b>S3-P3</b>	<b>Electrochemical and DFT analysis of quantum chemical reactivity parameters for electrochemical applications of an azulene-phenyloxazolone</b> Cornelia Elena MUSINA (BORSARU) (1), Alina-Giorgiana BROTEA (1), Ovidiu Teodor MATICA (1), Oana Iulia ENACHE (1), Amalia STEFANIU (2), Eleonora-Mihaela UNGUREANU (3) 1- Faculty of Chemical Engineering and Biotechnologies, University Politehnica of Bucharest, Romania 2- Institute of Organic Chemistry C. D. Nenitzescu of Romanian Academy, Bucharest, Romania 3- Doctoral School “Applied Chemistry and Materials Science”, University Politehnica of Bucharest, Romania	<b>83</b>
<b>S3-P4</b>	<b>Electrochemical properties of 2,6-bis((e)-2-(furan-2-yl) vinyl)-4-(azulen-1-yl) pyridine</b> Oana Iulia ENACHE (1), Ovidiu-Teodor MATICA (1), Magdalena-Rodica	<b>84</b>



	<p>BUJDUVEANU (1), Raluca ISOPESCU (1), Mihaela CRISTEA (2), Eleonora-Mihaela UNGUREANU (3)</p> <p>1- Faculty of Chemical Engineering and Biotechnologies, University POLITEHNICA of Bucharest, Romania</p> <p>2- Institute of Organic Chemistry C. D. Nenitzescu of Romanian Academy, 202B, Bucharest, Romania</p> <p>3- Doctoral School “Applied Chemistry and Materials Science”, University Politehnica of Bucharest, Romania</p>	
<b>S3-P5</b>	<p><b>Data management application in a smart hospital</b></p> <p>Liviu Doru DOGAR, Laura FLOROIAN</p> <p>Transilvania University of Brasov, Brasov, Romania</p>	<b>85</b>
<b>S3-P6</b>	<p><b>Biopolymeric compounds with medicinal applications</b></p> <p>Dana-Maria MIU (1,2), Ramona-Daniela PAVALOIU (1), Gabriela SAVOIU (1), Maria SPIRIDON (1), Mihaela-Carmen EREMIA (1)</p> <p>1- The National Institute for Chemical Pharmaceutical Research &amp; Development, Bucharest, Romania</p> <p>2- Faculty of Chemical Engineering and Biotechnologies, Bucharest, Romania</p>	<b>86</b>
<b>S3-P7</b>	<p><b>Folate-modified pullulan acetate nanoparticles loaded with curcumin for cancer therapy</b></p> <p>Dana-Maria MIU (1,2), Maria PETRESCU (1,2), Ramona-Daniela PAVALOIU (1), Fawzia SHA’AT (1), Mousa SHA’AT (3) , Gabriela SAVOIU (1), Maria SPIRIDON (1), Mihaela-Carmen EREMIA (1)</p> <p>1- The National Institute for Chemical Pharmaceutical Research &amp; Development, Bucharest, Romania</p> <p>2- Faculty of Chemical Engineering and Biotechnologies, University Politehnica Bucharest, Bucharest, Romania</p> <p>3- Faculty of Pharmacy, Department of Pharmaceutical Technology, Faculty of Pharmacy, “Grigore T. Popa” University of Medicine and Pharmacy, Iasi, Romania</p>	<b>87</b>
<b>S3-P8</b>	<p><b>From whole cyanobacterial cells of Synechococcus elongatus PCC 7942 to PSII: the effect of diuron on photosynthesis</b></p> <p>Robert RUGINESCU (1), Alina VASILESCU (2), Roberta Maria BANCUI (2), Szilveszter GASPAR (2), Cristina PURCAREA (1)</p> <p>1- Department of Microbiology, Institute of Biology Bucharest of the Romanian Academy, Bucharest, Romania</p> <p>2- International Centre of Biodynamics, Bucharest, Romania</p>	<b>88</b>
<b>S3-P9</b>	<p><b>ZrCu thin film metallic glasses as biomaterials for coating of metallic orthopedic implants</b></p> <p>Gabriela A. JURAVLEA (1,2), Alina VLADESCU (1), Anca C.PARAU (1)</p> <p>1- National Institute of Research and Development for Optoelectronics - INOE 2000, Magurele, Romania</p> <p>2- University Politehnica of Bucharest, Bucharest, Romania</p>	<b>90</b>
<b>S3-P10</b>	<p><b>In vitro evaluation of doped hydroxyapatite coatings electrochemically deposited on titanium nanostructured surface</b></p> <p>D.M. VRANCEANU (1), I.C. IONESCU (1), E. UNGUREANU (1), A.C. PARAU (2), R.I. ZAMFIR (1), I. TITORENCU (3), M. BADEA (4), C. ADOCHITE (4), S.</p>	<b>91</b>



	<p>COSTINAS (4), M. IDOMIR (4), A. VLADESCU (2), C.M. COTRUT (1)</p> <p>1 - University Politehnica of Bucharest, Bucharest, Romania</p> <p>2 - National Institute for Optoelectronics, Magurele, Bucharest, Romania</p> <p>3 - Institute of Cellular Biology and Pathology "Nicolae Simionescu, Bucharest, Romania</p> <p>4 - Faculty of Medicine, Transilvania University of Brasov, Brasov, Romania</p>	
<b>S3-P11</b>	<p><b>Influence of Mg and Zn content on the properties of hydroxyapatite based coatings</b></p> <p>D.M. VRANCEANU (1), I.C. IONESCU (1), E. UNGUREANU (1), A. KISS (2), A.C. PARAU (2), C.M. COTRUT (1)</p> <p>1 - University Politehnica of Bucharest, Bucharest, Romania</p> <p>2 - National Institute for Optoelectronics, Magurele, Bucharest, Romania</p>	<b>92</b>

## **PL.1. Optical biosensors for diagnostics of infectious viral disease: a recent update**

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The design and development of biosensors, analytical devices used to detect various analytes in different matrices, has emerged. Biosensors indicate a biorecognition element with a physicochemical analyzer or detector, i.e., a transducer. In the present scenario, various types of biosensors have been deployed in healthcare and clinical research, for instance, biosensors for blood glucose monitoring. Pathogenic microbes are contributing mediators of numerous infectious diseases that are becoming extremely serious worldwide. The recent outbreak of COVID-19 is one of the most recent examples of such communal and deadly diseases. In efforts to work towards the efficacious treatment of pathogenic viral contagions, a fast and precise detection method is of the utmost importance in biomedical and healthcare sectors for early diagnostics and timely countermeasures.

Among various available sensor systems, optical biosensors offer easy-to-use, fast, portable, handy, multiplexed, direct, real-time, and inexpensive diagnosis with the added advantages of specificity and sensitivity.

Many progressive concepts and extremely multidisciplinary approaches, including microelectronics, microelectromechanical systems (MEMSs), nanotechnologies, molecular biology, and biotechnology with chemistry, are used to operate optical biosensors.

A portable and handheld optical biosensing device would provide fast and reliable results for the identification and quantitation of pathogenic virus particles in each sample. In the modern day, the integration of intelligent nanomaterials in the developed devices provides much more sensitive and highly advanced sensors that may produce the results in no time and eventually help clinicians and doctors enormously.

This review accentuates the existing challenges engaged in converting laboratory research to real-world device applications and optical diagnostics methods for virus infections. The review's background and progress are expected to be insightful to the researchers in the sensor field and facilitate the design and fabrication of optical sensors for life-threatening viruses with broader applicability to any desired pathogens.

## **PL.2. Biosensing across continents: from advanced materials development to field deployable devices**

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The need for field portable devices that could respond to the today's requirements for low cost and rapid detection with on-site measurement capabilities continues to grow.

A key requirement in the development of these devices is to integrate materials with biological systems and create a suitable surface chemistry for the selective binding of target analytes.

This presentation will discuss development, manufacturing, analytical characterization and deployment of portable biosensors that incorporate functional redox active nanomaterials integrated with biological receptors. The modified surface integrates biorecognition, signal amplification and detection capabilities and can function as an all-in-one device that can be used for environmental, clinical and food monitoring.

The biosensors can be mass produced by printing which enables roll-to-roll manufacturing using a low cost, versatile and controllable process. Opportunities and challenges for translating this technology into practical applications will also be addressed.

## **S1. (Tele)diagnosis, (Tele)monitoring and (Tele)management in Medicine**

## S1-KN1. Towards point of use systems for fast and on-site detection

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Known as a sub-field of printed electronics, printed wearable electrochemical sensors are of special importance, due to their widespread applications in healthcare, food, agriculture and security. These analytical systems have opened new paths for body-integrated electronics that were once difficult to achieve. Much of the progress of printed wearable electrochemical systems relies on both innovations in materials engineering as well as novel combinations of conductive inks with diverse polymers or composites. Because of this advancement, new generations of printed electrochemical sensors include soft, light, flexible and anatomically-compliant electronics. The progress of wearable electrochemical sensors has relied on researchers' creativity in combining screen-printing techniques with unconventional platforms and substrates such as: gloves, medical bandages, mouthguards, and textiles, among others.

Several features need to be considered while developing wearable sensors: besides the good wearability and stretchability, wearable sensors should be highly sensitive, lightweight, low-cost, and low power consumption. To achieve such features, nanomaterials that possess a larger surface area are employed as building blocks in sensors. The incorporation of bioelements into wearable and single-use sensors is an important step in increasing their selectivity. A plethora of natural or biomimetic receptors was successfully tested, showing promising results in health and fitness monitoring as well as for other interesting applications.

The presentation will focus on the advancement of common electrochemical sensors toward development of point of use devices. Additionally, it will be discussed detailed requirements for the development of wearable electrochemical sensors. Taking into considerations these aspects, presenting in detail the performances of novel printed electrochemical sensors will be envisaged.

Several examples of wearable sensors applied in the quantification of different types of biomarkers will be presented with a focus on health and environmental analysis [1, 2]. Further growth of this field is foreseen due to the need for fast, sensitive, and easy-to-use sensing devices.

**Acknowledgments:** These studies have received funding from the European Union's Horizon 2020 Research and Innovation Programme under grant agreements no. 883484 (Pathocert) and no. 833787 (BorderSens).

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## S1-KN2. Long-term treatment with chloroquine for healthy aging in animal model

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Previous studies have shown that the polyamine spermidine increased the maximum life span in *C. elegans* and the median life span in mice. Since spermidine increases autophagy, we asked if treatment with chloroquine, an inhibitor of autophagy, would shorten the lifespan of mice. Recently, chloroquine has intensively been discussed as a treatment option for COVID-19 patients. To rule out unfavorable long-term effects on longevity, we examined the effect of chronic treatment with chloroquine given in the drinking water on the lifespan and organ pathology of male middle-aged NMRI mice. We report that, surprisingly, daily treatment with chloroquine extended the median life span by 11.4% and the maximum life span of the middle-aged male NMRI mice by 11.8%.

Subsequent experiments show that the chloroquine-induced lifespan elevation is associated with dose-dependent increase in LC3B-II, a marker of autophagosomes, in the liver and heart that was confirmed by transmission electron microscopy. Quite intriguingly, chloroquine treatment was also associated with a decrease in glycogenolysis in the liver suggesting a compensatory mechanism to provide energy to the cell. Accumulation of autophagosomes was paralleled by an inhibition of proteasome-dependent proteolysis in the liver and the heart as well as with decreased serum levels of insulin growth factor binding protein-3 (IGFBP3), a protein associated with longevity. We propose that inhibition of proteasome activity in conjunction with an increased number of autophagosomes and decreased levels of IGFBP3 might play a central role in lifespan extension by chloroquine in male NMRI mice.

## **S1-KN3. Portable Surface Plasmon Resonance system for blood marker analysis supporting rapid point-of-care diagnostics and treatment monitorin**

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Advances in rapid and point-of-care (POC) diagnosis are critical to gain ground against the high mortality associated with sepsis. Currently, blood biomarkers are analyzed in specialized laboratories by expensive and time-consuming procedures, however unsuitable for life threatening situations, where early detection is vital. To address this challenge, we developed a novel point-of-care system for fast and sensitive detection of biomarkers in blood, enabling early diagnostics (MarkerSense). By providing the means to detect rapidly (~10 min.), on-site and conveniently (using a single drop of blood) key parameters for assessing sepsis risk, MarkerSense will offer clinicians a valuable tool, allowing them to optimize and personalize the treatment and improve clinical outcome for the patients. Based on a proprietary Surface Plasmon Resonance technology developed by International Centre of Biodynamics<sup>1,2</sup> and a low-cost functionalization platform<sup>3</sup> optimized for high-sensitivity and reduced non-specific binding, MarkerSense simultaneously performs high signal-to-noise ratio reflectivity and phase angle resolved assays, in a portable format. The functionalization layer was tested for nonspecific binding and the limit of detection for a sepsis marker (i.e. soluble CD25) was ~2 ng/mL, as compared to relevant concentrations<sup>4</sup> for early diagnosis of sepsis (~3 ng/mL). Following successful clinical trials, MarkerSense is envisaged to be used for rapid on-site diagnosis of sepsis in hospitals (ICU, emergency departments).

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## S1-O1. Use of wearable devices for health and fatigue monitoring

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Modern wearable devices (smartwatches, wristbands, or rings) can incorporate several medical sensors. Therefore, those devices can be used to monitor the health condition of an individual. Machine learning algorithms can be used to predict fatigue or other health problems.

An Artificial Intelligence model for real-time prediction of fatigue using wearable devices will be developed. It will collect data using the sensors of the devices (Heart Rate, Blood Oxygen level saturation, Blood pressure and accelerometer) and it will be able to detect the incipient signs of fatigue.

For collecting the data the training data for the Machine Learning model, several volunteers with IRB agreements will wear the device for one week in their daily activities. The machine learning model will be trained locally, and then deployed in the Cloud. The medical data will be recorded by IoT device and then send to be processed in the Cloud. When fatigue is detected, an alert will be triggered and send to the wearable device.

Results and discussion: This Artificial Intelligence application can be used especially by drivers, to be warned in case of fatigue, but also by pilots, soldiers, or other workers. Therefore, it will be useful for keeping the physical and mental health, as well as for avoiding unwanted accidents.

The aim of this study is to prevent and detect the fatigue by using Artificial Intelligence methods. As it is crucial that the prediction is as accurate as possible, the ML model will be cross-validated using different dataset.



## S1-O2. Studies of determination of urinary bioactive metabolites

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The control of bacterial infections is starting to destabilize quickly, increasing the risk of sepsis even to immunocompetent patients. Frequently, pathogens involved in those infections are the multi-drug-resistance bacteria, which cause over 700,000 of deaths every year. 36 to 41 % of healthcare associated infections are the urinary tract infections (UTI), with 80% *Escherichia coli* strains as etiological pathogen.

It was demonstrated that some urinary metabolites are in a correlation with the bacterial growth in UTI and indicates the treatment direction. Therefore, there is a need to optimize the use of antibiotic therapy and propose rapid and specific techniques for the analysis of bioactive compounds of diagnostic importance in urine.

This study aims to identify and optimize an electrochemical method for the screening of uric acid, using a screen-printed carbon electrode (DRP 150) with a platinum auxiliary electrode, with a pH variation from 2.6 to 7, by differential pulse voltammetry and a limit of detection of 0.048 mmol/L. The results obtained sustains that this method is rapid and sensitive for testing uric acid and can be applied for testing urinary uric acid and as a model study for detection of other urinary metabolites.

**Key words:** multi-drug-resistance, uric acid, electrodes

### **S1-O3. Myo-inositol - never-ending resource in patients with polycystic ovary syndrome (PCOS)**

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Polycystic ovary syndrome (PCOS) is a disorder that affects women of childbearing age and it presents with any two of the following symptoms: hyperandrogenism, ovulation dysfunction and polycystic ovarian morphology. PCOS can be associated with obesity and/or insulin resistance that can worsen the symptoms of PCOS. Therefore, the management of obesity and insulin resistance in patients with PCOS is essential, being based on good nutrition, physical activity and medication, when needed.

Myo-inositol, a naturally occurring substance produced in the human body, plays an important role in insulin signaling by producing inositol phosphoglycans that act as signal messengers for insulin.

In recent years, supplementation with myo-inositol has shown promise in the treatment of symptoms in patients with PCOS. Some of the good effects that supplementation with myo-inositol has been shown to have are decreased insulin resistance, decreased hyperandrogenism and normalization of menstrual cycle. Moreover, there are some benefits to supplementation with myo-inositol in patients with PCOS undergoing *in vitro* fertilization, such as normalizing ovulation and improving oocyte and embryo quality.

Due to the fact that studies showing the benefits of supplementation of myo-inositol have been done on relative small groups of subjects with PCOS, the decision to administer myo-inositol to a patient with PCOS needs to be individualized to each patient's medical situation.

However, so far, the emerging evidence to support using myo-inositol in patients with PCOS is promising, especially regarding hormonal, ovulatory and metabolic benefits.

**Keywords:** myo-inositol, PCOS, ovulation, insulin resistance

## **S1-O4. Clinical and paraclinical evaluations of deficiency rickets**

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In the world Vitamin D deficiency is considered to be a major public health problem<sup>1</sup>, a high prevalence of vitamin D deficiency was reported. Vitamin D is important during childhood, for calcium absorption as well as for the accumulation and maintenance of calcium in the bones<sup>2</sup>.

Years ago, because there was no analytical method for dosing vitamin D, the disease was diagnosed using clinical and radiological parameters. Many laboratory techniques have been developed that allow dosing of vitamin D levels, in recent years. At present, 25(OH)D, serum-dosed is the most reliable marker of vitamin D status.

Plasma dosing of 25 (OH)-D is the most accurate marker for assessing the status of vitamin D in the body. This metabolite has a major role in calcium absorption but also in maintaining healthy bones.

The aim of this study was to assess the status of vitamin D among children between 1 month and 3 years in the central region of Romania.

A retrospective study was carried out by analyzing the values recorded in the hospital's electronic archive following the 25(OH)D dosing taken from 350 children aged < 3 years, admitted to the Children's Hospital of Sibiu, between January 2018 and January 2019. The 25(OH)-D dosing was performed from the serum, and for measuring concentration, an ELFA-type immunoassay test was used on a Vidas PC analyser, BioMerieux, France. The results were classified into 3 categories: patients with concentration 25(OH)D > 30 ng/mL (optimum), patients with concentration 25(OH)D levels ranging from 20 to 29 ng/mL (insufficiency) and patients with concentration 25(OH)D levels < 20 ng/mL (deficiency). The calcium, phosphorus and alkaline phosphatase concentrations were measured by spectrophotometric methods using the Architect c4000 analyzer.

25(OH)D deficiency it was registered in 38.2% of children and insufficient levels of vitamin 25(OH)D in 33.8% of children.

In the pediatric population from Romania vitamin D deficiency is increased. By studying the effect of this vitamin on bone health, it was possible to establish the optimal level of vitamin D in the body<sup>2</sup>.

In recent years, in addition to measuring the concentration of vitamin D, a method measuring the ultrasonic osteodensimeter (QUS) is used. This, measures mineral density and bone hardness based on the measurement of ultrasound attenuation and the speed of sound; the resulted data are used in calculating the calcaneal bone index.

The low serum levels of 25(OH)-D affect a large number of children, being necessary the implementation of vitamin D supplementation to prevent rickets in children.

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## **S1-05. Drug safety for elderly people – ethical, psychological and pragmatic aspects**

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The increasing number of elderly people represents both an individual benefit and a public health problem, as elderly patients need access to safe and effective medicines in the context of their polypharmacy and polypathology.

The purpose of the study is to present the main ethical aspects related to the introduction of a new drug in medical practice and its administration to elderly people, as well as those related to the therapeutic decision when the benefit-risk ratio must be evaluated.

A literature search was conducted using PubMed and Google Scholar for the time period between 1992 and 2022. Search terms included "Drug Safety", "Medical Ethics", "Patient Safety", "Drug Interaction" and "Adverse drug reactions". The articles for which at least the abstract was accessible were taken into account. Resources developed by the International Society of Pharmacovigilance and WHO were used in the analysis.

The introduction of drugs into medical practice is an old concern, since ancient times; there are a large number of examples where the introduction of a drug has had adverse effects. Also, the recent development of new drugs and the transition to personalized medicine, the need to minimize adverse reactions, as well as the tendency to regulate much more strictly the safety conditions of the use of a drug require a complex analysis of ethical aspects. The dynamics of the regulations show the fact that health professionals must cooperate not only at the level of those who apply a treatment but also those who can influence the attitude at the national and international level, requiring a complex analysis: marketing, psychology and a sociological approach to campaigns introduction of a new drug, along with knowledge of the results of medical researchers. The benefit-risk ratio must always be in favour of the benefit, and patient safety and drug safety must steer the therapeutic decision in the same direction.

The analysis and interpretations made will be a basis for both clinicians and people who manage quality in the health system, thus ensuring quality and safe care.

**Keywords:** drug safety, elderly people, ethics

## **S1-O6. Early puberty – clinical and paraclinical aspects**

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This study analyzes the cases of potential early puberty, evaluated in the INSMC Alessandrescu-Rusescu's Endocrinology Office, from 01.01.2021 to 30.04.2022. The aim is to assess the hormonal status for a group of 12 children, suspected of having early puberty, as well as to compare the mean values, medians, standard deviation and dispersion (using mean deviation) for FSH (Follicle-Stimulating Hormone), LH (Luteinizing Hormone), E2 (Estradiol), in the case of children from target group, compared to children from a group of control. Hormone dosages were multiple for the target group. Baseline values were followed for FSH, LH, E2, Testosterone and Cortisol tests, which were measured from blood collected at 4-6 months interval. Also, the children in the group target were stimulated with Diphereline, the dosing of the mentioned parameters being done before and after stimulation.

The determinations were performed on the Vidas PC analyzer. An enzyme-linked immunosorbent assay (ELFA) was used. On the inner surface of the solid phase (SRP) are bound specific anti-analyte (antigen) antibodies to be dosed. The specific antigen-antibody reaction takes place. In the final step, the conjugated enzyme hydrolyzes the substrate into a fluorescent product. The intensity of the fluorescent signal is directly proportional to the antigen concentration in the case of FSH and LH, and inversely proportional to the antigen concentration for E2, Testosterone and Cortisol. These weavings were performed for the target batch. For statistical purposes, in control group of children, from total dosages of FSH, LH and E2 were eliminated those belonging to boys (the target group consisted only of girls), those belonging to children born before 2013, as well as the dosages obtained as a result of stimulation processes. In the case of FSH out of a total of 476 determinations 113 were used, for LH out of a total of 488 determinations 108 were used, and for E2 out of a total of 458 determinations 106 dosages were used.

Not all children showed significantly increased values after stimulation, remaining under observation. Out of the total of 12 children in the target group, for 5 of them the stimulation test was clearly positive, one is at the border, and in the case of another 6 children the stimulation was negative. Regarding the statistical data, the indices obtained do not differ substantially between the 2 groups studied, which clearly shows that for the correct evaluation the cases must be treated in dynamics.

The complexity of the factors involved in the action and regulation of the phenomena related to puberty requires a coordinated evaluation from clinical, paraclinical and imagistic point of view. Early puberty is a potentially multifactorial pathology, requiring dynamic follow-up. Repeated and over a longer period of time hormonal investigations, in addition to the detailed clinical examination, allow the establishment of a correct diagnosis.

## **S1-O7. Towards an integrated automatic platform for rapid determination of antibiotic susceptibility of target bacteria**

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Rapid detection and assessment of antibiotic susceptibility of target pathogenic bacteria is demonstrated in a label-free format, further advancing our portable analytical platform [1, 2].

Immunomagnetic separation combined with periodic magnetic actuation and electrical impedance spectroscopy (EIS) monitoring are used in an innovative set-up allowing identification and determination of antibiotic susceptibility of target bacteria captured with specific magnetic nanoparticles (MNP).

Based on formation of Bacteria-MNP aggregates and their magneto-hydrodynamic behavior the EIS data provide rapid detection of intact target bacteria, and in conjunction with, microscopic modeling and complementary optical assays, to antibiotic susceptibility information.

Impedance data at two selected frequencies, highlight the magneto-hydrodynamic behavior contrast related to different antibiotic effects demonstrating method capability to both detect and assess antibiotic susceptibility of target bacteria.

We report on rapid pathogen detection and determination of antibiotic susceptibility directly from spiked samples.

All components of the assays are integrated into an automated system to enable rapid point of care diagnostic support.

### **Acknowledgements**

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## **S1-08. Evaluation of oxidative stress in the granulomatous disease**

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Oxidative stress caused by oxidative metabolism can cause disturbances in normal cellular signaling mechanisms, but on the other hand, neutrophils and other phagocytes manufacture O<sub>2</sub><sup>-</sup> (superoxide) for the purpose of killing invading microorganisms.

Chronic granulomatous disease (BGC) is a rare disease characterized by the inability of neutrophilic polymorphonuclear phagocytes to produce reactive oxygen species (ROS) is due to genetic mutations in the genes encoding the component subunits of NADPH oxidase.

To study the PMN response, various methods have been developed that are based on the evaluation of the oxidative explosion, by measuring the intracellular production of ROS or the formation of ROS in the extracellular space.

The following methods are widely used because of their convenience and accuracy:

- The method depends on the reduction of cytochrome c, which can be evaluated by photometry
- The second method is based on changes in the fluorescence properties of dihydrodhamine, which can be assessed by flow cytometry.
- NBT test - in detecting oxidative explosion in phagocytes Principle: NBT (nitrobluetetrazolium) is a yellow, water-soluble substance, phagocytosed by PMN and monocytes, then reduced intracellularly to formazane (dark blue to black) during oxidative explosion, under the action of reactive oxygen species (ROS) produced in phagocytes.

In the case of patients with a history of severe infections, after excluding more common diagnoses, it is necessary to check the immune status and in this context the existence of a BGC.

Although BGC is a rare disease, it may be underdiagnosed.

The NBT test is easy to perform and provides an indicative semi-quantitative assessment. The diagnosis of certainty requires confirmation by the Phagoburst test (quantitative assessment). Determination of genetic mutations in genes encoding NADPH oxidase subunits is the final diagnosis.



## **S1-O9. Lipid peroxidation and oxidative stress**

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Oxidative stress is the sum of all metabolites accumulated in tissues and biological fluids through radical oxygen species.

Lipid peroxidation is a major mechanism of cell damage and involves polyunsaturated fatty acids oxidation. As a result of this process, reactive species and toxic products are formed. Some of these species bind to functional groups of proteins, phospholipids, DNA and cause function`s changes.

One of the most important compounds obtained by lipid peroxidation is malonyl dialdehyde. When the plasma concentrations of reactive species reach critical values, the organism has not the ability to neutralize them. This is how it is explained the involvement in cellular physiology and pathology.

**Key words:** oxidative stress, lipid peroxidation, radical oxygen species

**S1-O10. Vitamin D status in the pediatric population in the pandemic context****M.A.VLAD (1,2), C.E.DELIA (1,2), M.D.HAIDAUTU (1), N.LACATUS (1), M.G.TOMA (1)**

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Vitamin D-Colecalciferol is a fat soluble vitamin. It is present in a few foods and is produced endogenously when ultraviolet rays from sunlight strike the skin and trigger vitamin D synthesis. Vitamin D promotes calcium absorption in the gut and maintain serum calcium and phosphate concentrations. Vitamin D sufficiency prevents rickets in children and osteomalacia in adults. It help protect older adults from osteoporosis. Vitamin D deficiency can be associated with secondary hyper-parathyroidism in adults, can increasing risk of diabetes, cardiovascular diseases, autoimmune diseases and some forms of cancer. Has a role in reduction of inflammation. Serum concentration of 25-hydroxyvitamin D [25(OH)D]=Calcidiol, is currently the main indicator of vitamin status. It reflects vitamin D produced endogenously and that obtain from food and supplements. Circulating 1,25-dihydroxyvitamin D [1,25(OH)2D]=Calcitriol is not a good indicator of vitamin D status because it has a short half-time, measured in hours.

The analyzed serum samples were obtained by collecting in a closed system, in coagulant free blood collection tubes. Samples were collected and analyzed from 3931 patients during 2020, and from 4389 patients during 2021. Data analysis was performed on several age groups: 0-3 years, 3-6 years, 6-10 years, 10-18 years and >19 years. The normal range given by our laboratory for vitamin D is: 75-175 nmol/L. The analysis of the samples was done with the Automatic Vidas PC, using appropriate kits for this analyzer. The assay principle combines an enzyme immunoassay competition method with a final fluorescent detection (ELFA) to 450 nm. The intensity of the fluorescence is inversely proportional to the concentration of vitamin D antigen present in the sample. At the end of the assay results are automatically calculated by the instrument in relation to the calibration curve stored in the analyzer memory.

In 2020 from a total of 3931 samples, in 0-3 years group were 821 patients; in 3-6 years group were 513 patients; in 6-10 years group were 635 patients; in 10-18 years group were 698 patients and in >19 years group were 1264 patients. The average concentration for vitamin D in 0-3 years group was 104,322 nmol/L with a standard deviation (SD) of 47,32; for 3-6 years group was 83,461 nmol/L with a SD of 31,17; for 6-10 years group was 72,131 nmol/L with a SD 27,16; for 10-18 years group was 64,526 nmol/L with SD of 29,38 and for >19 years group was 59,682 nmol/L with SD of 28.70. In 2021 from a total of 4389 samples, in 0-3 years group were 1176 patients; in 3-6 years group were 875 patients; in 6-10 years group were 937 patients; in 10-18 years group were 1042 patients and in >19 years group were 359 patients. The average concentrations for vitamin D in 0-3 years group was 109,837 nmol/L with a SD of 51,49; in 3-6 years group was 86,221 nmol/L with a SD of 30,66; in 6-10 years group was 78,052 nmol/L with a SD of 29,72; in 10-18 years group was 68,615 nmol/L with a SD of 27,93 and in >19 years group was 70,931 with a SD of 27,06.

It can be seen that the SD for the 0-3 years group is increased compared to the other age groups. This group has the highest average value of vitamin D but, also the highest SD. In our experience, vitamin D levels in newborns and in the first months of children s life are low. But, starting with vitamin D prophylaxis, these values are obviously improved and finally the average is much higher for that group compare with to the rest of the groups studied. This, once again highlights the importance of vitamin D prophylaxis. For both years (2020 and 2021) there is a decrease in the average values of vitamin D, with increasing age of patients. In the same time, there is an increase in the average values of vitamin D in 2021, compared to 2020 for all age group analyzed. This can be assumed to be due to the additional intake of vitamin D in the population in the context of the pandemic.

## **S1-O11. Telemedicine for older patients. Advantages and disadvantages**

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This presentation will underline a branch of medicine that it is considered very important, especially in the current pandemic. The purpose of this work is to raise awareness of the importance of telemedicine in medical practice. Being a vast subject, it was chosen telemedicine in the diagnosis and treatment of elderly patients.

The presentation will start with the beginnings and history of telemedicine. Therefore, we will return to the present – telemedicine during pandemic and then we will focus on elderly patients, as medical articles and books presented. Therefore, it will be presented the advantages and disadvantages of this type of medical practice.

**Keywords:** telemedicine, elderly, advantages, disadvantages

## S1-P1. Strategies to address the non-specific adsorption in electrochemical aptasensors for clinically relevant proteins

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The non-specific adsorption (NSA) is a major issue in biosensors, one that is often overlooked. It leads to lack of accuracy, selectivity, stability and reproducibility as well as to reduced sensitivity. The problem is particularly important in complex media such as serum or in samples containing oligomeric aggregates with exacerbated adsorption at interfaces compared to the monomeric forms. Modifying the surface with a self-assembled monolayer (SAM) of aptamer and filling the gaps in the surface coating with chemisorbed thiolated molecules such as mercaptohexanol or including polyethyleneglycol moieties is a very popular strategy for obtaining simple yet efficient electrochemical aptasensors for proteins. Nonetheless, low dilutions of serum samples or protein aggregates still pose significant practical challenges related to NSA. While the ionic strength, the composition of the interaction buffer help minimize the adsorption of unwanted compounds, they also affect the aptamer conformation and its interaction with the target protein.

We hereby present our investigation with respect to several thiolated compounds that were used for the prevention of NSA from serum samples and from lysozyme aggregation mixtures. We have found that a short methylated thiol, PEG4-SH was the most efficient among the tested compounds for preventing the adsorption of lysozyme amyloid fibrils to a gold surface. The evaluation of the adsorption effects was done based on cyclic voltammetry measurements, by comparing the intensities of anodic and cathodic peaks of the ferrocyanide/ferricyanide couple before and after the incubation with complex samples.

In a second strategy, the adsorption of lysozyme to gold surfaces was evaluated by coupled electrochemical and SPR measurements. Thiol-blocked, aptamer functionalized gold interfaces were compared with “blank” sensors, covered with thiol SAMs only. The specific and nonspecific binding of 200 µg/mL lysozyme was evaluated at three values of the applied potential, -0.65 V, 0 V and +0.2 V and it was found that applying a negative potential maximizes the specific over the non-specific binding. The results are discussed.

Combining a thiol coating that includes ethylene glycol groups with the application of an electrical field during the aptamer-protein interaction might prove a successful strategy for other aptasensors and other proteins' detection using a similar sensor design.

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## **S1-P2. The behavior of the medical staff and of the students from the Faculty of Medicine regarding the use of the mobile phone**

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The most microorganisms on the surface of mobile phones belong to the normal skin flora but have also been found bacteria that are involved in nosocomial infections. This is becoming a serious problem in the health system due to the increasing antibiotic resistance of the bacteria involved and the medical phones of the medical staff have become a source of rapid transmission of these types of bacteria.

The purpose of this study is to see if there is a connection between lifestyle of the medical staff and medical students and mobile phone contamination.

The group of participants, of which 89 are medical staff from hospitals in Brasov and 107 are students at the Faculty of Medicine. Both study groups (n=196) carry out activities in hospitals - work and semester internships, internships during the semesters, laboratories for certain disciplines. The questionnaire included a total of 50 questions, of which 43 questions include notions about mobile phone disinfection practices, mobile phone activities, measures taken to prevent mobile phone contamination.

In a majority of 93.88% (n=184) the study group uses a protective phone cover, and the most used material is silicone - 57.65% (n=113). In the case of phone disinfection frequency, the medical staff disinfects it at least once a day in a percentage of 50.56% (n=45), and the students chose in a majority percentage of 23.36% (n=16) the 'monthly' response option. Among the medical staff, only 1.12% (n=1) answered that they never disinfect their phone, while the students chose this option in a percentage of 14.95% (n=10). Both study groups believe that telephones play an important role in the spread of infections - 61.68% (n=66) of students and 70.79% (n=63) of medical staff.

Following the study, certain factors have been identified that may or may not favor the contamination of mobile phones. These factors are represented by the ~~gender~~, the time and place of use of the phone, the activities carried out using the mobile phone, the way of disinfecting the phone, the type of cover and protective foil.

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**Keywords:** mobile phone, screen protection, contamination, medical staff, medical student

### S1-P3. Vitamin D deficiency in diabetes

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Type 2 Diabetes Mellitus (T2DM) is the commonly endocrine disorder characterized by hyperglycemia. Vitamin D deficiency is associated with an increased risk of developing diabetes. Vitamin D has an important role in blood sugar control or in relieving diabetic complications. The probable mechanisms of glucose homeostasis in which vitamin D deficiency is involved are beta cells dysfunction and insulin resistance.

The obesity, aging and low physical activity are the risk factors that T2DM and vitamin D deficiency have in common. The best marker that highlights vitamin D status is 25-hydroxy vitamin D.

**Key words:** diabetes, vitamin D, insulin

## S1-P4. Vitamin D levels and pathology association in hospitalized patients

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Vitamin D deficiency is the most common medical condition, approximately one billion people have low levels of vitamin D. Vitamin D is associated with a pleiotropic effect (immune modulatory, anti-inflammatory and antiviral), which can be essential for a better immune response.

The aim of this study was to evaluate the prevalence of vitamin D deficiency considering sex and age distribution and to establish a correlation with different comorbidities.

In this cross-sectional study, 16572 patients (10647 women, 5925 men), mean age 54.8, who presented at the Central Military Hospital, Bucharest and had had the level of the vitamin D evaluated, were consecutively enrolled during almost 2 years- January 2020 to May 2022. The data was evaluated using SPSS.

27.61% (4575 patients) presented low levels of vitamin D; 7.68% (1274) had vitamin D insufficiency (mean 16.33, SD 2.26) and 19.91 (3301) had vitamin D deficiency (mean 8.58, SD 2.45). Low levels were more prevalent among women (59.61%), middle aged (32.39%) and elderly groups (39.93%). Moreover, it was a slightly predominance of the deficiency in spring-summer vs. autumn-winter (30.83% vs 24.84%). The prevalence of vitamin D deficiency was high at patients with COVID19 infection ( $p \leq 0.00001$ ), cardiac disease ( $p \leq 0.00001$ ), endocrine disorders ( $p \leq 0.00001$ ), neurological disorders ( $p \leq 0.045$ ), renal disease ( $p \leq 0.021$ ). It was not found a correlation with gastroenterological comorbidities ( $p \leq 0.6476$ ).

The prevalence of vitamin D deficiency was high and it was significantly associated with different comorbidities and an intense vitamin D supplementation should be taken into consideration.

**Keywords:** vitamin D deficiency, cross-sectional studies, COVID19

## **S2. New Trends in Nutritional Sciences and Food Control**



## **S2-KN1. Botanical extracts as new food additives: are they really new?**

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The consumer is increasingly aware and interested in his diet and the quality of the food he consumes. Food additives from plant sources have found interest from both food industries and consumers. In fact, different surveys have shown that consumers prefer additives of natural origin, in the common belief that what comes from nature is safe. Natural molecules have a long tradition as food preservatives, but other natural additives still have limitations and obstacles to overcome before they can be used in industry.

The presentation will try to give an overview of recent studies on additives of plant origin, their use, relative benefits, and risks.

## S2-KN2. Rapid and sensitive tools for enhanced chemical safety of food

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Recently, the European Parliament has called on Member States to strengthen their food safety control mechanisms, which has become a priority for the French government. Over the past few years, substantial progress has been made in microbiological safety, as advances in molecular biology have helped develop rapid, inexpensive, and sensitive methods that are now used by regulatory authorities to improve their control systems. However, on the chemical safety side, the technical and societal transition is lagging behind. The current system is mainly based on monitoring and inspection plans using high-performance reference methods able to detect toxic contaminants in traces level. Among these methods, chromatographic techniques as gas chromatography or HPLC have been shown to be suitable for the detection of toxins, but they need highly trained personnel and they require separation/preconcentration steps, thus limiting the frequency of analysis. Biosensors have been described for many years as good substitutive or complementary tools to conventional methods for the detection of food contaminants, due to their ability to provide real-time qualitative detection with minimum preparation.

In this work, we present two affinity-based sensors for the detection of meat contaminants:

Firstly, an electrochemical immunosensor for rapid and sensitive detection of fluoroquinolone (FQ) antibiotics is described based on the competitive binding of a synthesized electrochemical probe derivate of difloxacin and free quinolone present in sample on immobilized anti-quinolones antibodies. The proposed immunosensor allowed FQ detection from 0.005 µg/mL to 0.01 µg/mL with a detection limit of 0.003 µg/mL. The efficiency of the sensor and the adequacy of the extraction process were finally validated by analyzing different meat samples.

Secondly, we present the development of electrochemical aptasensors for polychlorinated biphenyls (PCBs) detection. This sensing platform is based on already described sequences, truncated using computer modeling in order to improve the sensitivities of aptamers. The truncated aptamers were validated using Aptamer-based Fluorescent Assay in view of their future integration in electrochemical sensors.

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## S2-O1. Electroanalytical method coupled with chemometry for detection of virgin olive oil adulteration

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The detection of olive oil adulteration with lower quality oils continues to be an important problem in the olive industry since fraud continues to be committed. The current methods used to detect these adulterations require complicated previous steps and are time and money consuming. Furthermore, these need high qualified persons. Therefore, there is a serious need to develop simpler, faster and cheaper methods for this purpose.

In the present research the potential of a novel electrochemical method based on screen printed electrodes modified with nanomaterials for the detection of extra virgin olive oil (EVOO) adulteration was developed.

The screen printed electrodes were modified with oils under study and the cyclic voltammograms were recorded in two different electrolyte solutions. The data were processed by means of multivariate data analysis and classification models were developed.

Extra virgin olive oil and samples containing different ratios of other oils such as pomace oil, sunflower, high oleic sunflower and corn were analysed. The electrochemical signals of the sensors immersed in the electrolyte solution are chemical fingerprints of the oils under study. These are mainly related with the presence of phenolic compounds and other compounds with antioxidant character from the samples. The PLS-DA and SIMCA models developed have shown that the samples could be correct classified in agreement with the EVOO contents, with an error below 2%. Probabilistic Neural Networks models success rate (correct predicted over total number of samples) was 99% for all adulterate oils analysed.

The electroanalytical method coupled with chemometrics could be used for the accurate detection of olive oil adulteration.

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**Keywords:** extra virgin olive oil, sensor, data analysis.

## **S2-O2. Develop a series of ultra-sensitive detection methods for mycotoxins**

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Mycotoxins are harmful to people's health because of their widespread existence and lethal toxicity. It is of great significance to realize the detection of mycotoxins at ultra-low concentration for both food safety and the development of high sensitivity detection method of biological small molecules. Here, we took mycotoxin as the target molecule, studied the structure of the aptamer, and enhanced the affinity between aptamer and mycotoxin by the evolution of the structure of the aptamer. Based on the new aptamers, we developed a series of aptasensors to achieve the ultrasensitive detection of mycotoxins.

Label-free fluorescence aptasensors were developed using crystal violet and SYBR Green I as probes. Simple label-free quencher fluorescence platforms were established by the quenching ability of guanine and G-quadruplex. And chemiluminescence aptasensor was developed based on split DNAzyme. We improved the limit of the detection from 20.0 nM to 0.10 nM.

## S2-O3. Sustainable approach for functional fermented product development

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In this study, sustainable development goal 12.3 regarding the reduction of food waste to achieve Sustainability by 2030 was applied for dairy industry. In this way, functional goat milk yoghurt with bioactive compounds, extracted from industrial tomato waste was developed and analysed. Selection of lactic acid bacteria determined the most promising combinations for use in the preparation of starter cultures for yoghurt development with high content of bioactive compounds from industrial tomato waste.

Goat milk samples were obtained from the individual farms and were investigated to chemical composition, rheology, physical, chemical and sensory properties. The strains of lactic bacteria for goat milk fermentation were developed. Several samples of tomato waste extracts were obtained in function of applied extraction parameters. Bioactive compounds and their antioxidant profile of were evaluated.

In goat milk was determined the mass fraction of proteins, which constituted 4.28 g. The fatty acid composition of goat milk was studied using GL-chromatography. Experimental data showed the content of 23 fatty acids. The strains of valuable native lactic bacteria of the species *Streptococcus thermophilus* and *Lactobacillus bulgaricus* were selected with stable technological characteristics. Research also included recipes and technological schemes development for functional yoghurt with increased biological value, due to the incorporation of industrial tomato waste extract, containing such antioxidants as beta-carotene, lycopene and tocopherol.

The obtained yoghurt samples are recommended for consumption as a valuable functional dairy product, designed in the context of sustainable and innovative food development.

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**Keywords:** sustainable tomato waste management, supercritical fluid extraction, lactic bacteria, fermented dairy products.

## **S2-O4. Harmonized in vitro digestion and Ussing chamber to investigate the effects of polyphenols on the intestinal physiology**

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The properties of several nutritional compounds depend on the digestion process. Plant polyphenols are an example, given that their physiological effects, bio-accessibility, and bioactivity can vary depending on the intestinal pH, interaction with other nutrients, gut microbiota composition, and several other factors.

Because of the dynamic processes involving complex enzymatic and physiological events that take place in different gastro-intestinal segments, the use of a single model to reproduce in vitro the digestion and the absorption of nutrients is difficult to simulate.

In-vitro digestion (IVD) protocols are widely used to address questions in the field of nutritional research. However, the biological relevance of the IVD protocols needs to be validated for each research question and several protocols exist, making a comparison between studies difficult.

The in-vitro simulation of the complex intestinal absorption is also challenging. Peristaltic movements are strictly associated with the gut functions by mixing and separating the lumen content. Additionally, the intestine hosts commensal microbial communities responsible for the intestinal homeostasis and immune system maturation.

The improvement of in-vitro models implemented to better represent the physiological conditions represents a big challenge for scientists. This intervention aims to describe the combination of a harmonized in vitro digestion method (INFOGEST), coupled with the Ussing perfusion chambers as a valuable model for the digestion and subsequent absorption of nutrients or bioactives in monogastrics and ruminants, with special emphasis on phenolic compounds

## S2-O5. Cider yeasts associated with Hardanger cider during fermentation process

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In the Hardanger area in Western Norway, the production of cider has a long tradition that goes back to the 12th century, when monks introduced apple growing in this area. Nowadays, this is also the main area of fruit production in Norway. Despite the strict regulation of the alcoholic beverage production in Norway, traditional cider is still produced on some farms in this area. Therefore, our aim was to study the ecology and biodiversity of the yeasts associated with the cider production in the Hardanger area during fermentation process; especially of traditional cider, which is produced by a spontaneous fermentation of apple juice, performed by naturally occurring indigenous yeasts that originate from the fruit or the surfaces of the processing equipment.

In our study, samples of fermenting juice/cider were taken during fermentation process from 12 producers, located in 12 different locations in Hardanger region. Classical cultivation methods using WL (Wallerstein Laboratories) agar medium with added chloramphenicol enable us to isolate a total of 530 yeast isolates that were stored in in-house yeast collection at the NIBIO and included also at the Wine Research Centre collection. Based on the sequencing of the D1/D2 domain of the 26S rDNA we managed to identify 357 isolates and distinguished 27 different yeast species as follows: *Aureobasidium pullulans*, *Candida californica*, *C. oleophila*, *C. sake*, *Hanseniaspora meyeri*, *H. uvarum*, *H. valbyensis*, *Kregervanrija fluxuum*, *Kregervanrija sp.*, *Metschnikowia andauensis*, *M. chrysoperlae*, *M. fructicola*, *M. pulcherrima*, *Metschnikowia sp.*, *Pichia fermentans*, *P. kluyveri*, *P. membranifaciens*, *P. nakasei*, *Piskurozyma capsuligena*, *Rhodotorula nothofagi*, *Saccharomyces bayanus*, *S. cerevisiae*, *S. paradoxus*, *S. pastorianus*, *Saccharomyces sp.*, *S. uvarum* and *Torulaspora delbrueckii*.

Even though we were not able to obtain samples in three different fermentation stages (beginning, middle and at the end of fermentation) from all producers, we could observe yeast succession during fermentation progress. Yeast diversity was higher at the beginning comparing to the middle of fermentation, when mostly different non-*Saccharomyces* yeast species prevailed, while in the middle of fermentation 11 species were detected (*Candida californica*, *H. uvarum*, *H. valbyensis*, *Kregervanrija sp.*, *K. fluxuum*, *Pichia membranifaciens*, *Metschnikowia pulcherrima*, *Saccharomyces sp.*, *S. bayanus*, *S. uvarum* and *S. cerevisiae*). On the other hand, at the end of fermentation mainly *Saccharomyces* species with high ethanol tolerance were present (*Saccharomyces sp.*, *S. cerevisiae*, *bayanus*, *S. uvarum* and *P. fermentans*).

In samples that were collected from three producers in all three fermentation stages also quality parameters were determined (ethanol, organic acids, sugars, biogenic amines) with in-house developed methods using HPLC-UV/RID. The most important sugars in ciders were fructose and glucose, as expected. Two producers added sugar to increase the level of ethanol in the middle of

fermentation, which is a common procedure in the Hardanger area. Ethanol and organic acid analysis indicated that fermentations went in the right direction, since all parameters were within normal limits. Including the acetic acid level, an indicator of low cider quality, was very low (average around 0,06 g/L). The alcohol incised from the beginning to end fermentation in all samples analysed and minimum concentration was 2,71 g/L. In ciders we detected four biogenic amines (putrescin, cadaverine, histamine and tyramine). The average amount was 32 mg/L and the most abundant was tyramine.

**Keywords:** indigenous yeasts; biodiversity; spontaneous fermentation; cider-making



## S2-O6. New educational trends in European teenagers obesity management

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Obesity is a chronic disease, considered a global epidemic by the WHO. According to international studies, the global rate of childhood obesity has increased tenfold in the last four decades, with the number of children diagnosed with obesity reaching about 124 million. For Romania, a 2015 study shows that in 7-9 age group studied the share of children with weight problems (overweight or obese) is over 25%.

The outcomes of this project are to provide to healthcare workers, nutrition experts and students, the knowledge and skills, methods and tools, to explore and address the management of obesity through transdisciplinary approaches (healthy lifestyle, stigma step-down, proper body perception, and education based on online training, videos, podcast, and PPT presentations).

Its an European project named C4H Project = “Connected4Health – A Medical and Humanities-based Approach to Navigating Obesity and Eating Disorders in Young People”, ERASMUS+ 2021 Call, KA220-HED – Cooperation partnerships in higher education, No 2021-1-RO01-KA220-HED-000032108, based on a partners network for 7 UE countries (Romania, Italy, Portugal, Serbia, Czech Republic, Lithuania and Spain), during 2022-2023.

This Erasmus+ project will develop and disseminate an interdisciplinary European guide, based on the following materials: (1) a Romanian and English Guide focused on cultural and historical perspectives on Obesity and Eating Disorders, (2) a Curriculum for various seminars/ conferences, which will target the multidisciplinary management of obesity in young people, and (3) a package of training Lectures for students about personalized management of young people with Obesity.

Nutritional community interventions are needed for the adoption of healthy eating behaviors, the stimulation of physical activity, and emotional management among European teenagers, tackling to reduce the frequency of risky behaviors and future chronic diseases.

**Keywords:** obesity, lifestyle, stigma, management, teenagers, nutrition guide

## S2-O7. Invasive knotweed species as a source of bioactivities

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Knotweed species, namely *Fallopia japonica* (FJ), *Fallopia. Sachalinensis* (FS) and their hybrid *Fallopia x bohemica* (FB) are considered among the most invasive plants in the world. They represent a big environmental problem, since they prevent growth of the native plants, which leads to decreased biodiversity. However, several recent studies showed that these plants are a very rich source of bioactive molecules, mainly from the group of polyphenols. Therefore, the aim of this study was to identify some of the most prevalent polyphenols, present in different tissues of all three taxons, as well as to study their biological activity, namely antioxidant, antidiabetic, antimicrobial and neuroprotective. Using LC/MS we identified a total of 13 polyphenols, in ethanol extracts prepared from rhizomes and young shoots, which all contributed to a substantial antioxidant activity with the highest values for FS and the minimum for FJ. Regarding antidiabetic activity, the highest ability to inhibit alpha-amylase activity was obtained for FS, with more than 80% inhibition already for the concentration of dry extract of 0.01 mg/mL. The antidiabetic activity of all tested extracts also showed a concentration-dependent pattern. Furthermore, all tested extracts showed antimicrobial activity against *Escherichia Coli*, *Listeria Monocytogenes* and *Candida Albicans* and were able to significantly prevent apoptotic and necrotic-like neuronal cell death caused by oxidative stress. In conclusion, our results show that it is possible to use all three invasive knotweed species as a source of powerful bioactive molecules to prevent or even treat several oxidative stress-related diseases.

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**Keywords:** knotweed species, antioxidants, antidiabetic activity, neuroprotection

## **S2-O8. Use trend and attitude of young people towards food supplements: comparison between surveys performed in 2011 and 2021**

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In the last decades, there has been an increasing interest in food supplements (FS) among consumers, including young people.

In 2011, a survey on plant food supplement consumption among six European countries was performed within the European project PlantLIBRA.

To investigate the trend evolution of FS use over the time, a similar survey was conducted ten years later (2021) among Italian and Romanian young people. An anonymous questionnaire, including questions about consumption and perception of FS, was submitted to students from both countries. Answers were compared to those selected from consumers of the same age range and countries of the PlantLIBRA survey.

Over the years, some differences have been observed in the attitude towards dietary supplements among both Italian and Romanian students.

One of them is related to the person/health professional who recommended the use of FS. In 2011, for both countries, most respondents indicated “my-self”; in 2021 medical/health professionals were generally selected as the main responsible of the advice. This difference could be due to a greater awareness in the use of supplements.

Although the perception of the health effects of food supplements was almost unchanged, interesting differences (among the same country and between Italy and Romania) in terms of use and habits were observed.

These considerations will be useful for better understanding the evolution of young consumers’ habits (e.g. frequency of use, cost range, etc.) together with efficacy and safety aspects of dietary supplements, that are increasingly available on the market.

**Keywords:** food supplements, PlantLIBRA, survey, Italy, Romania, students.

## S2-O9. Screening grapes for infection by *Botrytis cinerea*: detection of spores versus the evaluation of laccase activity

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*Botrytis cinerea* is a fungal pathogen responsible for high economic losses in vineyards worldwide. Various approaches have been considered for the detection of *Botrytis cinerea* [1], including gigahertz ultrasonic imaging of fungal spores [2], the fingerprint of the Raman spectrum of the fungal spores [3], detection from the mycelia using antibodies specific for *Botrytis* species [4], detection of the fungal DNA [5] or indirect detection through the measurement of indicators such as laccase activity, glycerol and gluconic acid [6]. While not all strains of *Botrytis cinerea* produce laccase, laccase activity evaluation is the most convenient and widely used test for screening for grape's infection by *Botrytis cinerea*. Enzymatic activity higher than 3U/mL indicates a high risk of oxidative degradation of wine [7].

In this work we present a paper based electrochemical assay for the evaluation of laccase activity which is simple, can detect activities in grape must down to 0.4U/mL in 5 minutes and is adequate for on-site analysis. Its application to monitoring the artificial infection of white, rosé and red grapes with two strains of *Botrytis cinerea* via the laccase activity evaluation is hereby described. In parallel we report studies towards the development of a molecularly imprinted polymer electrochemical sensor for *Botrytis cinerea* spores. The MIP was obtained through the electropolymerisation of the phenazine dye Toluidine Blue O. The initial studies were performed with commercially available lyophilized bacteria of *Micrococcus lysodeikticus* taken as a model. The spores of *Botrytis cinerea* and the bacteria used as model were characterized by Atomic force microscopy, Scanning electron microscopy and Raman spectroscopy. Our studies have shown that the non-specific adsorption is a major hurdle in obtaining an effective MIP and suggested that Raman spectroscopy can provide a more specific test for detecting the spores of *Botrytis cinerea* on grapes. When comparing the two approaches, i.e. the evaluation of laccase activity versus the detection of *Botrytis cinerea* spores, it becomes evident that each analytical tool is advantageous for a specific application. Thus, the evaluation of laccase activity is essential for screening grapes at processing points while a portable sensor for the (ideally) specific direct detection of spores would be preferable for screening grapes in the vineyard.

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## **S2-O10. Research on the use of *Saccharina latissima* seaweed as an innovative ingredient in the bakery industry**

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*Saccharina latissima* seaweed is attracting increasing interest as a sustainable food source in European countries because of their nutritional properties, especially due to high fibers and minerals content. The paper presents some research related to the use of sugar kelp (*Saccharina latissima* (class *Phaeophyceae*)) in the bakery industry, both for bread and snacks, researches develop in the frame of the project SUMAFOOD (Sustainable preservation of marine biomasses for an enhanced food value chain).

The seaweed obtained from a Spain ecological farm was dried in NTNU University at 5% humidity by using microwave under vacuum and milled under the size of 30 microns. Using ICP-OES method, the main mineral content were determined: K content: 62197 mg/kg; Ca content: 8273 mg/kg; Mg content: 6067 mg/kg; P content: 2290 mg/kg; I content: 16619 mg/kg.

After a preliminary microbiological test, four degrees of replacement of white wheat flour (type 480) from the bread recipe were used: 1.5%, 3%, 4.5%, and 6%. For the flour mixtures, the main rheological characteristics were analyzed, and baking tests were performed by direct method. Analysis started with: bread specific volume, porosity and crust/crumb colour.

The bread specific volume was determined according to SR 91:2007, AACC 2000. The ratio of the obtained data was the average of triplicate measurements of the fresh made bread loaf, expressing it in cm<sup>3</sup>/g. For porosity measurement, knowing the mass and density, the porosity was expressed % by measuring the total scale of holes, in a known crumb volume. The colour was evaluated using the Image-J software on the bread slices scanned with a HP2100 scanner.

Next, a sensory analyses was performed, as follows: a group of 10 specially trained panelist, with ages between 25 and 60, evaluated the bread samples, giving grades from 1 (lowest intensity) to 5 (highest intensity), for the following sensory attributions: crust color (degree of perceived brown color characterizing the crust), crumb color (degree of color darkness in the crumb ranging from white to dark brown), crumb pore uniformity (size of pores on the surface; (small/big), crumb softness (minimum force necessary to compress the sample), bitter taste (perceived by the back of the tongue and characterized by solutions of quinine, caffeine, and other alkaloids; usually caused by over-roasting), salty taste (fundamental taste sensation elicited by sodium chloride), sour taste (fundamental taste sensation evoked by acids, e.g., tartaric acid), specific aroma (aroma of fresh baked bread and odor associated with aromatic exchange from yeast fermentation), after-taste (flavor staying after tasting).

Also, a consumer overall acceptability in a 9-point hedonic scale (from 9 = i like it extremely to 1 = i dislike it extremely), was calculated, where 35 untrained panelists with ages between 21 and 60 (70% females and 30% males) have tasted the samples that were coded with 3 random letters in order to not influence their perception, and the results

were expressed as mean. All bread samples of flour mixtures P1-P4 were compared to the standards of 2 control samples P0 (wheat flour type 480 – white bread) and PN (wheat flour type 1250 – black bread).

The results show a good rheological behavior up to a degree of replacement of 4.5%, but due to the pronounced after-taste, consumers indicated a maximum acceptable level of 3% degree of replacement with algae. Other products developed and tested referred to snacks and biscuits, were after taste was reduced by using 2% of caraway seeds.

As conclusion, the Sugar Kelp seaweed flour could be use with good results as functional ingredient in bakery by addition in wheat flour up to a 3% replacement degree.

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**Key words:** *bakery, algae, Sugar kelp, rheological and sensorial analysis.*

## **S2-O11. Insect material as innovative feed ingredient in animal nutrition**

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The increasing global need to find alternative protein/energy sources has triggered research in the field of non-conventional feed ingredients, with insect proteins being one of the most promising. Insects contain high level of protein and fat; therefore, insect should be considered as promising alternative feed ingredients for livestock production. In addition to the nutritional value, they also represent a way by which food waste biomasses/streams can be upgraded to valuable feed ingredients.

This brief lecture outlines the main nutritional characteristics of insects, and also considers the legal framework involved.

The importance of the type of insect in term of species and development/metamorphosis and tailored substrates that could lead to the production of a premium feed is also described. For both ingredients a critical review of the safety issues is provided. Based on the current data available insects have an excellent potential use as alternative feed ingredients for livestock production. When produced in line with the criteria set by major feed/food authorities, they are characterized by high quality and safety standards. This makes them comparable to other feed materials and ingredients currently available on the market, although their full nutritional, functional, safety and sustainability evaluation cannot be considered complete



## **S2-O12. PlantMolecularTasteDB: a versatile tool in designing fortified food**

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PlantMolecularTasteDB is the first database dedicated to all types of orosensorially active phytochemicals (bitter, sweet, sour, umami, salty, pungent, astringent phytochemicals). It is freely available at <http://plantmoleculartastedb.org>, and includes over 1500 phytochemicals [1].

The phytochemicals can be searched by name, identifiers, molecular formula, chemical class, SMILES, taste or orosensation, associated taste receptors and other chemosensors, potential anti-inflammatory activity.

PlantMolecularTasteDB facilitates exploration of phytotastants for multiple purposes: finding phytocompounds matching a specific taste, finding the molecular taste of a plant extract/ingredient, studying the potential correlations between taste/orosensation and antiinflammatory activity. This may also contribute to design fortified food with anti-inflammatory potential and a certain organoleptic profile, in accordance with the consumers preferences.

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## S2-O13. Pigmented cereals and gluten related diseases

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The important role of oxidative stress and inflammation in celiac disease (CD) has been reported by several authors. The gluten free diet produces beneficial effects on the oxidative status of celiac subjects; however, it seems to improve only partially the functionality of the intestinal mucosa in celiac patients.

The aim of this study was to evaluate the possible antioxidant and anti-inflammatory properties of pigmented gluten free cereals in the framework of CD.

Extracts from pigmented cereals (rice and corn) were in vitro digested and characterized in term of phenolic compounds and antioxidant activity, using both spectrophotometric and chromatographic techniques. In parallel, for the evaluation of the anti-inflammatory activity of the extracts, Caco-2 cells were treated with digested gliadin in combination with other pro-inflammatory stimuli, to induce a celiac disease-related inflammatory status.

The in vitro digestion determined a significant reduction of the total anthocyanins content. However, no significant difference in term of phenolic compound content was observed after digestion and an increase in term of antioxidant activity was measured. These data agree with the bioactivity of the extracts; in fact, gliadin in combination with IFN- $\gamma$  and IL-1 $\beta$  enhances the release of CXCL10, that was inhibited by all the extracts tested (IC50s  $\approx$  200  $\mu$ g/mL), both before and after digestion.

This study underlines the potential antioxidant and anti-inflammatory activities of the extracts from pigmented cereals at the gut level. These results could be useful to partially clarify the role of dietary phenolic compounds in the protection of intestinal mucosa in CD.

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**Keywords:** Celiac disease, pigmented cereals, oxidative stress, anti-inflammatory activity

## S2-O14. Nutritional status in Crohn's disease

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In the last decades, the main entities of the inflammatory bowel disease spectrum, Crohn's disease and Ulcerative Colitis, have known an important hike in prevalence worldwide, especially in areas with hyperhygiene habits such as Western Europe and Scandinavian countries.

The aim of this presentation is to display the impact of malnutrition in Crohn's disease on the overall quality of life. In order to better understand the mechanisms behind the ailment of the nutritional status in Crohn's disease, the key notions of clinical presentation and physiopathology were listed below.

Crohn's disease is a chronic inflammatory disorder that may involve any part of the digestive tract from mouth to anus, but with a predilection for the distal small intestine and proximal large bowel. Inflammation in Crohn's disease often is discontinuous along the longitudinal axis of the intestine and can involve all layers from mucosa to serosa. Patients usually experience diarrhea, abdominal pain and weight loss. Common complications include strictures and fistulas, which often need surgery. A large variety of extra-intestinal manifestations also might be present.

Because most cases of Crohn's Disease affect the distal small intestine and the proximal large bowel, malabsorption of carbohydrates (disaccharides) and proteins (oligopeptides) occur. Also malabsorption of fats and fat soluble vitamins happen, due to the impairment of the enterohepatic recycling of bile salts. Even among patients whose disease has been in long standing remission, deficiencies such as of iron, folic acid, vitamin B12, calcium, magnesium and zinc were detected.

The catabolic state induced by intense inflammation can also increase energy and protein requirements. Unrecognized infection can additionally be a major contributing factor beyond the catabolism induced by the disease itself. A bypassing of small intestine by enteroenteric or enterocolonic fistulas can also contribute to undernutrition.

Unfortunately, up to the present, the etiology of Crohn's Disease is still incompletely understood and therapy, even though generally effective in alleviating symptoms, is not curative and cannot alleviate malnutrition.

## **S2-O15. Food - the history of a society**

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Ever since Homo sapiens discovered the fire, a greater interest occurred in the minds of the human beings of that time that transcended beyond the simple reason of eating because of hunger. Food rapidly became a way of showing power or seeking acceptance from a particular community.

Therefore, we can see different dietary patterns in today's nutrition as in the ancient one. Food is situated at the crossroads between nutrition and pharmacology and the old diets can confirm this. From the consumption of molded bread for healing wounds to the tapeworm diet or the use of radium in everything for everlasting health and youth, the history of food and society probably has not seen everything yet. The community has a major impact on the nutritional human behavior and I will outline its importance over ages in my presentation.

To find out more about the relationship between individuals and the medium in which they are raised, it was conducted a small qualitative and quantitative study, materialized in a questionnaire disseminated among different ethnical communities residing in Romania. The questionnaire consists of 17 items of different types (open-ended questions, close-ended questions and ratio-scaled questions), including 6 items regarding a series of data about the respondents (age, gender, ethnical community etc.).

## S2-P1. Phenolic profile and antioxidant activity of different corn and rice varieties

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Celiac disease (CD) is an autoimmune disease characterized by a permanent intolerance to gluten; to date, the only universally recognized treatment is the gluten-free diet (GFD). Despite patients maintain a GFD, a state of inflammation and oxidative stress could remain at intestinal level. Several components of the diet, such as phenolic compounds and anthocyanins with known antioxidant properties, could play a protective role in the inflammatory state of patients with CD. The objective of this study was the characterization of the phenolic profile and the antioxidant capacity of several pigmented cereals (rice and corn) from the Italian market and farms. The most interesting varieties were selected for subsequent studies.

Different *in vitro* methods were applied: 1) Folin-Ciocalteu assay 2) pH differential method 3) DPPH assay 4) TEAC assay 5) HPTLC technique (High Performance Thin Layer Chromatography).

All samples analyzed showed a good correlation between the soluble phenolic content (SPC) and the antioxidant capacity (AOA), measured by DPPH and ABTS tests ( $R^2 > 0.91$ ). Notably, SPC and AOA of pigmented varieties were significantly higher than those of non-pigmented varieties. Data obtained applying HPTLC technique and correlated with the spectrophotometric results were in agreement with data reported by the scientific literature.

According to our results, pigmented varieties are a valuable source of soluble polyphenols and anthocyanins with high antioxidant activity. They could be used as alternative ingredients for the formulation of gluten-free products.

**Acknowledgments:** This work was supported by Fondazione Celiachia Onlus (FC) Grant n°004\_FC\_2019.

**Keywords:** celiac disease, oxidative stress, pigmented cereals, phenolic compounds

## S2-P2. Electrochemical determination of hydroxytyrosol in extravirgin olive oils using screen-printed electrodes modified with single wall carbon nanotubes and tyrosinase

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*Olea europaea* L., commonly known as olive, is a small tree species distributed mainly in the Mediterranean countries, with the main derivative product being olive oil [1]. Olive oil is present in the Mediterranean diet, as it is considered a protective factor for the prevention of chronic diseases, due to its antioxidant, anti-inflammatory, lipid-lowering and even antimicrobial action [2].

The aim of this study is to develop a reliable, sensitive and selective tyrosinase-based biosensor (SWCNT-Ty-SPE) for the determination of hydroxytyrosol (HT) and its quantification from real samples of extra virgin olive oil (EVOO).

Tyrosinase (Ty) will be immobilized by casting and crosslinking, after prior modification of a screen-printed carbon electrode (C-SPE) with single-walled carbon nanotube (SWCNT-SPE). The characterization of the biosensor surface was performed both by electrochemical methods (CV, DPV, EIS) and by spectrometric methods (FTIR). The calibration curves of HT were performed by cyclic voltammetry (CV). Quantitative determinations were carried out from various samples of EVOO using CV.

The presence and the sensitive properties of SWCNT and Ty were highlighted by all applied electrochemical methods (CV, DPV, EIS). SWCNT-Ty-SPE have increased selectivity and sensitivity, confirming the biocatalytic activity of Ty immobilized on the biosensor surface. Following the application of several scanning rates (0.1-1.0 V/s), it can be stated that, in all cases, the reduction process is controlled by HT adsorption on the active surface, which is faster and more obvious in the case of the biosensor. Based on the superior sensing properties of the biosensor, the calibration curve was subsequently realized in a wide concentration range of 0.1  $\mu$ M - 300  $\mu$ M, with linearity in the range of 0.5-15  $\mu$ M and the limit of detection and quantification in the micromolar range. Subsequently, SWCNT-Ty-SPE was successfully used to quantify HT from real EVOO samples, using CV.

In conclusion, the new biosensor developed based on SWCNT and Ty has multiple advantages such as: sensitivity, selectivity, feasibility and low cost. SWCNT-Ty-SPE could also be used in routine analyzes for food quality control, such as vegetable oils containing HT.

**Acknowledgments.** This work was supported by a grant of the Romanian Ministry of Education and Research, CNCS—UEFISCDI, project number PN-III-P4-ID-PCE-2020-0923, within PNCDI III.

**Keywords:** hydroxytyrosol, tyrosinase, biosensor, olive oil

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## S2-P3. Consumer perceptions of functional foods with antioxidant and anticancer potential

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According to statistics, due to insufficient intake of essential nutrients, a person's condition, who was diagnosed with cancer, may worsen and the disease will progress.

The study involved the analysis of consumers' perception of functional foods with antioxidant and anti-carcinogenic potential and their development.

Main research methods: development of an online questionnaire, conducting a survey among respondents of the Republic of Moldova, analysis of the survey, creation of new food products with antioxidant and anti-carcinogenic potential, organoleptic analysis, development of technological maps and schemes, research of antioxidant activity of the plant raw materials used for the created products.

150 respondents took part in the survey. The survey revealed a lack of awareness among 62 (41%) respondents about functional products with antioxidant and anti-carcinogenic potential, this is one of the main factors hindering the observance of proper nutrition. Acknowledging the obtained personal data, oat bars with marmalade layers of sea buckthorn, grapes and black currant were developed. Among the bars, the most liked was the sea buckthorn marmalade layer sample, its appearance was rated at 4.8 points, taste – 4.9, aroma – 5, consistency – 4.9, color - 4.9. The highest values of the antioxidant activity of DPPH were obtained in samples of sea buckthorn and grapes, amounting to 92.52% and 90.45%, respectively.

Detailed information about nutrition for oncological diseases and functional products in general is important for the respondents, and they also consider it necessary to expand the range of this group of products on the territory of the Republic of Moldova.

**Acknowledgments.** The authors acknowledge the networking activities carried out within the Global Harmonization Initiative working group on “Consumer Perception” (<https://www.globalharmonization.net/wg-consumer-perception>) and especially “Innovative and Future Food Development” issue group.

**Keywords:** consumer perception, functional food, antioxidant and anticancer potential, technological process and food development.

## S2-P4. Facile specific detection of acetaldehyde in air samples using electrochemical enzyme biosensors

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Aldehydes are relevant markers in the assessment of human health, for the quality of food and of environmental air. Acetaldehyde is the major aldehyde in wine contributing to wine aroma but also influencing wine stability and its color, since it forms adducts with sulphur dioxide and serves as bridge between phenolic compounds leading to larger compounds with different chromatic characteristics. The group of Mitsubayashi developed “biosniffer” sensors based on fluorescence or electrochemical detection and membranes impregnated with dehydrogenase enzymes for the assessment of several compounds including acetaldehyde in gas phase [1-3].

Inspired by the advantages and simplicity of these methods, we developed simple electrochemical tests for the detection of acetaldehyde in aerosols and vapor samples. The chronoamperometric detection is sensitive and requires low-cost equipment. For a specific detection of aldehydes, we employed the two recombinant dehydrogenase enzymes from the Antarctic *Flavobacterium* PL002 strain, ALDS2 and F-ALDH characterized by different Michaelis-Menten constants and substrate specificity profiles [3]. These enzymes catalyze the conversion of aldehydes to the respective carboxylic acids with the simultaneous reduction of the enzymatic cofactor nicotinamide adenine dinucleotide. The electrochemical detection of NADH is achieved either via direct oxidation at +0.5 V vs Ag/AgCl on carbon nanotube electrodes or via mediated oxidation at +0.1 V using Meldola Blue-modified carbon electrodes.

Several approaches were compared: placing the enzymes together with the enzymatic cofactor  $\text{NAD}^+$  in solution on the electrodes, using a single-use, pre-impregnated membrane or modifying the electrodes with an ink containing the enzymes immobilized on magnetic nanoparticles. The experimental setup was examined as well: we tested aerosol samples obtained by spray-ing known amounts of aldehyde solution in plastic tubes. In another approach we took advantage of the vapor pressure to sample aldehyde vapor from the headspace of tubes containing known amounts of aldehyde solution. A custom-made pump-based gas delivery system was used to bring the vapor samples in test tubes containing the electrodes modified with enzymes. The characteristics of each system are discussed for the development of an optimized application for the detection of acetaldehyde in wines.

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## **S2-P5. Consumer perceptions of functional foods for type 2 diabetes in Romania and Republic of Moldova**

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Type 2 diabetes is increasingly prevalent and accounts for about 90 to 95 percent of all diagnosed cases of diabetes in adults. Functional foods can alleviate the problem due to their ability to fight chronic diseases, diseases of the cardiovascular system, digestion, etc.

The analysis of consumer perception of functional foods, level of awareness of this type of food and attitudes towards the development and introduction to the market of this type of functional foods.

The study included elaboration of online questionnaires, analyzes of obtained survey data, creating of functional foods and analyzing the quality properties of developed foods.

Results and discussion: As a result of survey, it was revealed that the market of Romania and Moldova lacks functional foods for type 2 diabetes, and there is an interest and demand for such foods among population: 77.8% and 94.4% of the respondents in Romania and Moldova agree with this. There is a great demand among consumers for such functional products as confectionery and sweets. Marmalade-like product and mousse using stevia and inulin were developed. Research included recipes and technological schemes development. The organoleptic, physical and chemical properties were subject to careful analysis and compared with reference sample.

The experimental data regarding perception of functional foods for type 2 diabetes by consumers, as well as the development of new functional foods, can make a positive contribution to the coverage of the problem, as well as improve the quality of life of people with type 2 diabetes.

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**Keywords:** geographical regions and consumer perceptions, functional food for diabetes, new food development and evaluation.

### **S3. New Trends in (Bio)engineering Sciences Applied in Life Sciences**

### **S3-KN1. In situ remote monitoring of novel biodegradable cardiovascular stents - design & validation**

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A biodegradable metal stent implantation restores flow in a clogged or damaged artery back to a healthy artery and then completely disappears in 18-24 months. Magnesium-based stents have advantages due to their radial strength and biocompatibility.

**Aim:** We aimed to study novel magnesium-based biodegradable tubes and propose a wireless scheme to monitor on-site the stent's degradability.

To fully explore the potential benefits of this alternative approach, research and development are needed on biodegradable stent materials, stent designs and delivery systems, deployment techniques, adjunctive antithrombotic therapies, and sensing technologies for stent degradation monitoring.

In the proof-of-concept study, funded by Sidra Medicine:

We investigated the significant progress made over the last two decades in the development of Mg-based alloys and in their performance characterization for both in vitro and in vivo deployments.

We developed design and manufacturing guidelines and evaluated the performance bounds of biodegradable stenting and monitoring solutions. The mechanical and structural design of the stent was optimized using multiphysics simulation models. The key design elements considered include the selection of length and diameter, deciding on open vs. closed-cells, surface area, number, and thickness of struts connecting the support segments.

We presented a non-invasive technique to progressively monitor the degradation of a magnesium-based biodegradable cardiovascular stent using mm-waves.

We completed the tasks of manufacturing Mg-alloy tubes and develop microstructure-property relationships and studied the stent's geometry design parameters for optimum performance. The proposed frequency range can provide sufficient spatial resolution and penetration without radiation damage to the tissue.

**Acknowledgments:** This study was made possible by Sidra Internal Research Fund grant number SIRF 200041 from Sidra Medicine (a member of the Qatar Foundation).

**Keywords:** magnesium-based bioresorbable stent, corrosion, degradation monitoring

### **S3-KN2. Synthesis and characterization of nanostructured silver based antibacterial coatings on protective polymer foils of touch screen devices**

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The use of electronic devices that are frequently touched can contribute to the spread of various pathogens, leading to bacterial and fungal infections. In this study we investigate the use of very thin layers of silver nanoparticles embedded into a dielectric layer, as antimicrobial thin films on the surface of self-adhesive polymer covers for touch screen devices. Physical vapor deposition technique was used, combining High-Power Impulse Magnetron Sputtering (HiPIMS) for the deposition of Ag nanoparticles with Radio Frequency (RF) sputtering for the deposition of oxides. The characterization of the coatings includes: elemental and phase composition, texture, structure, topography of the coatings, evaluation of optical properties. The antimicrobial activity of the foils was assessed, using E-coli bacteria at different concentrations. Thin films in the range of ~30 nm thickness, including Ag as active antimicrobial agent and SiO<sub>2</sub> and TiO<sub>2</sub> as dielectric matrices, were obtained and characterized. Good transparency in the visible spectral range was proven along with good responsiveness to touch. The presence of nanostructured Ag was demonstrated also by spectrophotometric measurements and XRD measurements, indicating the presence of ~10nm size nano-grains. Comparisons between different film compositions and antibacterial testing methods is also provided.

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**Keywords:** antimicrobial, nanostructured silver, magnetron sputtering

### S3-O1. Development of nanotechnologies and their impact on public health

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The nanotechnology industry has become more and more developed with applicability in many fields, including the medical one. A series of tests showed the toxic effects of these nanoparticles (NPs) on human health (immunotoxicity, hepatotoxicity, genotoxicity, etc.).

The aim of this study is to analyze the impact of the development of nanotechnologies on human health.

For this review study, articles from the last 15 years were selected, and as inclusion criteria, the selected articles were intended to be in English.

The presence of NPs is becoming more common in the environment, but also in certain products for daily use. NPs production is expected to reach 58,000 tonnes by 2020. With this increase in the production of NPs-containing materials, together with the constant discovery of new applications of NPs, it is surprising that knowledge about the health effects of exposure to NPs is still limited. However, the number of efforts to determine the health risks associated with exposure to NPs continues to increase. Exposure, absorption, distribution and degradation of NPs in the environment are very important notions that influence the degree of toxicity on public health. The main determinant of the efficacy or toxicity of NPs is their ability to interact with the cell. As in the case of target organ toxicity, the size of nanomaterials plays a major role in their cellular interaction and also determines the half-life of the systemic circulation and their biodistribution. Cellular absorption differs from that of small molecules and understanding these differences is essential for understanding and limiting the cellular toxicity of NPs. Factors influencing NPs and cell interactions and absorption are related to physicochemical and biophysical properties of cell membranes.

With the application of nanotechnologies in many fields, the impact of nanoparticles on cell and animal models needs to be tested and considered for possible toxic effects. The development of complete toxicological profiles of nanoparticles and the development of relationships between physico-chemical structure and their activity will help identify key physical or chemical properties that cause their toxicity and help design safer strategies to minimize toxicity while maximizing efficacy.

**Keywords:** nanoparticles, nanoparticle toxicity, public health, health program

### S3-O2. Functional Cell Sorting and Cellular Spectroscopy: New technologies for the study of cellular heterogeneity

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Cellular heterogeneity is an important characteristic of biological systems that allows them to function. For analysing cellular heterogeneity, in medicine and biology, novel technologies that characterise cells at the structural and functional level are needed. Here we will present two new technologies that improve our capabilities for characterising the structure and the function of the cell. First, the functional phenotype flow cytometer (FPFC), can analyse and sort cells based on their intracellular function. In this purpose the FPFC performs a cascade of steps on a microfluidic platform: first the cells change their intracellular environment; second the cells are incubated in the new extracellular environment and thirds the cellular florescence is monitored all through this process. Finally, cells are sorted based on the magnitude of their fluorescence changes - linked to intracellular responses to stimuli. The validation of this device is performed with a human lymphoma cell line Ramos by separating cells based on their intracellular calcium changes upon an exposure to an antiBCR antibody.

The second technology presented here is a cavity enhanced absorption spectrometer that promises to deliver single cell absorption spectroscopy. In this purpose a novel concept that circulates the light multiple times through the environment of interest is proposed. The circulated light interacting with matter generates an enhanced spectral fingerprint characteristic for the analysed sample. Here we are proposing the use of this spectrometer for characterising the internalisation of drugs in living cells.

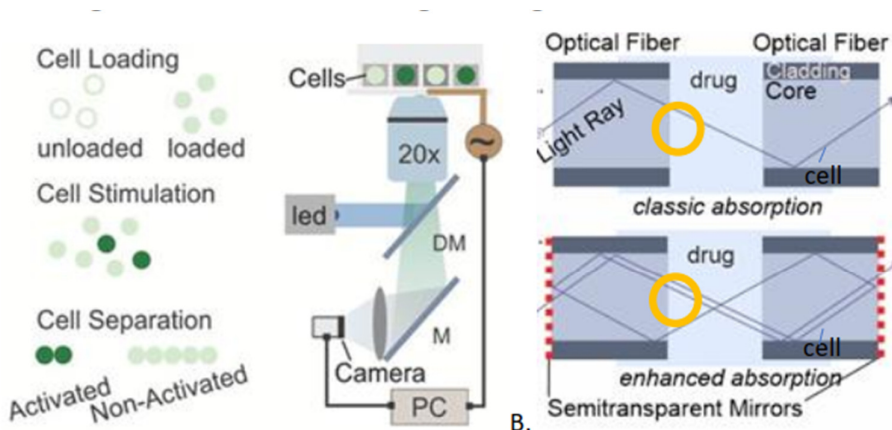


Figure 1. A. Functional Cell Sorting (1). B. Cellular Spectroscopy

Reference:

P. Nikiforov et al., Functional Phenotype Flow Cytometry: On Chip Sorting of Individual Cells According to Responses to Stimuli. *Advanced Biology* 2021, 5, 2100220.

### **S3-03. Defining protein function and activity associated to different pathologies by combining molecular, cellular, biochemical and analytical methods**

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Although recently, new strategies such as balanced diets, reduction of alcohol consumption and tobacco, exercise, limited UV exposure are proposed as prevention measures for different maladies such as cancer, neurological, cardiovascular diseases etc., their incidence is still growing worldwide. Thus, the molecular and biochemical investigation of different proteins dysregulated in such pathologies is of high interest for the prevention, stadialization and for the treatment of different disorders.

Taking advantage of different available tools in gene regulation such as knock-down, stable-overexpression or knock-out, we modulate expression of different proteins in order to study their function or control their activity in cell lines. We have achieved these goals by coupling cellular tools with biochemical methods such as: SDS-PAGE, Western Blot, Immunofluorescence, gradient ultracentrifugation and analytical ones such as Mass Spectrometry (MS) and Liquid Chromatography (LC).

More precisely, we showed that tyrosinase N-glycans, a tumor antigen overexpressed in melanoma cells, are important for the immune response activation by using single or triple tyrosinase glycosylation mutants overexpressed in A375 amelanotic melanoma cells.

Additionally, since perturbation of Endoplasmic Reticulum (ER) homeostasis it is associated with various pathologies, we focused our attention on modulating the expression of Endoplasmic Reticulum Associated Degradation (ERAD) components able to restore ER homeostasis. Our results demonstrated that downregulating or stable overexpression of ER degradation-enhancing  $\alpha$ -mannosidase-like proteins (EDEM1 and EDEM2), highlighted new proteins from the degradation complex involved in cellular homeostasis restauration, such as SEL1L and DERL1, components of retrotranslocation complex. Moreover, by identifying new ERAD substrates our studies contributed to the enrichment of the immune repertoire associated to melanoma, able to elicit an immunological response.

All these data suggest that combining genetical tools with biochemical and analytical methods can result in the identification of new components potentially involved in prognostic and treatment of different pathologies.

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### S3-O4. Lab-chip evaluation of bacterial cells' dynamics for rapid antimicrobial susceptibility testing

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Availability of methods for the Rapid identification of microorganisms and for Rapid testing of antimicrobial susceptibility (AST) suitable for Point of need (PoN) analyses is an yet unmet need towards accelerated clinical diagnosis and treatment of infectious diseases, food and water safety control.

The aim of the study was to develop multiplexed, high resolution (micro-spot) analyses and bacterial micro-chemostats with chromogenic media as a lab-chip approach for rapid, sensitive and specific quantitative assessment of microorganisms' dynamics, translatable in bench top and portable formats.

Implementation of Optical-based electrical impedance spectroscopy assessment was coupled with Differential Interference Contrast (DIC) and Reflected Light Microscopy assays for extensive characterization of the phenotypic features of single cells, including the cellular size, motion, morphology, membrane integrity and electrical parameters and enable discrimination of "persisters" from "resistant" cells upon antibiotic exposure.

Bacterial cells (gram negative *Escherichia coli*) are spotted on custom developed ITO chips, layered with agar strips and investigated in peptone medium with an inverted microscope in transmitted light (DIC) and in reflected light (red light - LED). The coupling with electrical actuation [1] is tested in conjunction with various electromagnetic field geometries for rapid, quantitative electro-optical, label-free multiparametric assessment.

This novel bioanalytical platform allows real-time tracking of events taking place at the cellular level and evaluation of the division of individual bacterial cells as a reliable phenotypic feature for rapid AST and cell identification.

The multiplexed, high resolution (micro-spot) analyses allows establishing the set of representative cellular parameters that quickly (in couple of hours) reflect bactericidal, bacteriostatic effects towards real-time tracking of cellular effects of (novel) antimicrobial compounds.

**Acknowledgments** Support of PNIII projects P2-2.1-PED-2019-5155, -5185, -4932 is gratefully acknowledged.

**Keywords:** bacteria, bioanalytical platform, electro-optic analyses

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### **S3-O5. Novel reflected light microscopy assay for rapid, label free assessment of cellular processes and cellular status at single cell level**

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Whole cell-based biosensing systems are uniquely capable of providing functional information on the impact of a sample on cell physiology including related toxicity or pharmacology effects. As a result cell-based sensing grounded on dynamic, real-time, label-free, and non-invasive analysis of cellular events, (including cell adhesion, cell viability, cell morphology, and cell motility) is aimed for as a significant enabling technology for biological research, environment assessment and pharmaceutical industry.

The aim of the study was to develop a rapid, label free and sensitive assay to investigate cell viability and cytotoxicity at single cell level compatible with cellular platforms format and useful in: cell-cell, pathogen-host interactions evaluation.

Endothelial bEnd-3 cells under normal and (chemical, microbiological) stress conditions, were monitored, in real time, using reflected light dark field microscopy configuration. The MTT (methyl-thiazolyl-tetrazolium) assay, a gold standard in viability evaluation, is deployed in single cell analysis format to enable method calibration: high resolution quantitation of cytological mechanisms of MTT uptake, reduction to formazan nanocrystals, formazan compartmentalization and its extrusion is quantitatively assessed in conjunction with the type and extent of stressors. The (scattered) reflected light intensity and cell localization changes reveal dynamic differences between cells cultured in normal and stress conditions enabling label free assessment of cellular processes and cellular status (viability, membrane integrity, organelle/vesicles dynamics, cell surface contacts) at single cell level. The progress of individual steps in the known MTT cytological mechanisms of uptake, reduction to formazan nanocrystals, formazan compartmentalization and its extrusion is innovatively used to demonstrate the dynamic, quantitative access to phenotypic and functional cell analysis.

This method assures results in a shorter time, compared to gold standard MTT and allows real-time single-cell analysis and an accurate quantification of formazan crystal products, in a dynamic manner.

**Acknowledgments:** Support of PNIII projects P2-2.1-PED-2019-5155, -5185, -4932; PN-III-P4-ID-PCE-2020-2432 ZEISS Romania support in providing reflected light dark field microscopy set-up.

**Keywords:** single-cell, nanocrystals, MTT, reflected light dark field microscopy

### S3-O6. ZrCu based thin films metallic glasses in medicine

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Biomaterials represent a widely used category of materials for different types of medical related issues, such as physical replacement of a diseased biological part, as bones, teethes, types of joints and so on, or just for repairing. Usually, these materials need to develop a strong bond between them and the tissue that comes in direct contact with it and should be able to coexist without having unwanted and inappropriate effects. Also, restoring the integrity of the damaged bone leads to the fact that the patient life should be prolonged and the quality improved.

Several studies reinforce the idea that, because of their technological promise for practical applications and scientific significance in the biomedical realm, bioactive glasses have sparked extensive study attention. Recently, they seem to improve the biocompatibility and bioactivity and have the tendency to integrate well with the tissue, in comparison with the metal implants and also, including the facilitation of the tissue regeneration as studies have shown.

The goal of the present study is to investigate the novel ZrCu-based ternary Thin Film Metallic Glasses (TFMGs) as a solution for orthopedic implants. The coatings consisted in ternary coatings based on ZrCu with small addition of Si, Mg, Ca, Sr, Mo. The coatings were performed by cathodic arc technique using a system with high deposition rate. The used substrates were 316L steel and Ti6Al4V alloy, commonly substrates used in medicine. The elemental and phase compositions of the coatings were determined. Hardness, adhesion, roughness and corrosion performance at 37°C in SBF, in comparison with the characteristics of uncoated alloys, were investigated. All of the coatings were more proper for coatings of Ti6Al4V alloy, than of the 316L steel.

We acknowledge the support of the Romanian Ministry of Education and Research, CNCS - UEFISCDI, project PN-III-P4-ID-PCE-2020-1264 (PCE95/2021), within PNCDI III, and Romanian National Core Program no.PN18N-01-02/2019.

### **S3-O7. MXenes-based bioanalytical sensors: design, characterization and applications**

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MXenes are an emerging class of 2D layered nanomaterials that provide large surface area, hydrophilicity, high ion transport properties, low diffusion barrier, biocompatibility and ease of surface functionalization. Due to their unique features, MXenes have gained substantial attention in fields such as batteries and supercapacitors and their application in chemical and biological sensors is growing. Their composition and layered structure makes MXenes particularly attractive for biosensing applications.

This presentation will discuss the use and application of MXenes in the development of electrochemical biosensors as an immobilization matrix, signal transducer and amplifier of biomolecular recognition. An example of a portable MXenes-based biosensing platform fabricated in our lab for detection of glucose will be illustrated in which  $\text{Ti}_3\text{C}_2$  MXenes was used as transducer surface and supporting material for the immobilization of the glucose oxidase enzyme.

Opportunities for developing wearable sensors and systems with integrated biomolecule recognition will be highlighted.

### S3-08. Investigation of a truncated aptamer for ofloxacin detection

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The revolutionizing medicinal properties of antibiotics as Fluoroquinolones (FQs) have increased their overuse, mainly in veterinary medicine for promoting animal growth and optimizing large-scale breeding programs. Among FQs, ofloxacin is primarily used in treating infections of the respiratory, digestive, and urinary systems in both humans and animals. Despite the remarkable features of this quinolone, its contribution to the expansion of bacterial resistance remains inevitable.

The consumption of animal-derived foodstuffs with high residual ofloxacin concentrations can cause adverse reactions in the human body. Therefore, it is crucial to control the fluoroquinolones residues level in animal-derived foodstuffs before consumption. So, we suggest the development of a rapid aptamer based bioassay for ofloxacin detection. Our approach harnesses the fluorescence quenching of the fluorescein-tagged aptamer (FAM-APT) induced by its partial hybridization to a tetramethyl rhodamine-labelled complementary ssDNA (TAMRA-cDNA). In such structure, dye labels brought into proximity act as a FRET pair. Upon ofloxacin addition, an affinity competition occurs to form a more stable FAM-APT/OFL complex, thus unquenching the FAM-APT signal. The recovered fluorescence intensity correlate with the antibiotic's concentrations at 0.2 – 200  $\mu$ M in HEPES buffer, with a linear response ranged between 0.2 – 20  $\mu$ M. The rapid apta-assay achieved limits of detection and quantification of 0.12 and 0.40  $\mu$ M, respectively.

The truncated aptamer shown an improved specificity toward OFL than other quinolones, compared to the original full-length aptamer described in previous works. Finally, the practical application was confirmed to detect OFL quinolone in spiked milk samples with satisfactory recoveries ranging between 97.4% and 111.4%.

**Keywords:** aptamers, antibiotics, bioassay

### **S3-O9. Multifunctional nanomaterials an emerging area for biosensors**

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The recent years have witnessed a major influence of nanotechnology in the field of biosensors. The integration of nanomaterials in the construction of biosensors is aimed to achieve better analytical figures of merit in terms of limit of detection, linear range, assays stability, low production cost, etc. Nanomaterials can play a variety of roles in the development of responsive, easy-to-use, and field portable biosensors. In this line, significant research efforts have been made towards exploration and synthesis of various types of nanomaterials for subsequent use in the fabrication of biosensors.

Nanomaterials can act as an immobilization support, signal generating probe, signal amplifier, signal quencher, mediator and artificial enzyme label etc.

The present work will define the key roles of nanomaterials and relate the nano-based features to the analytical performance of the biosensor design.

### **S3-O10. Nano-engineered surface coatings on polymer substrates: the correlation between surface morphology, hydrophilic/hydrophobic properties and antibacterial activity**

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The demand for surface engineered coatings with controlled hydrophilic/hydrophobic properties are ever-increasing owing to their diverse applications in fields such as biomedical technologies, engineering, sensors, automotive, defense, space, aerospace, apparels and so on. Previous research has shown that these properties can be tuned by both chemical and surface texture engineering. In this contribution, we will discuss in detail the tunability of titania (TiO<sub>2</sub>) coatings applied on polymer (polyethylene terephthalate (PET)) substrates and the role of chemical and surface engineering on their hydrophilic to hydrophobic transition.

We will show that the inherent hydrophilicity of titania coatings can be altered to impart hydrophobicity by the applied chemical and surface engineering via doping or compositing and controlling coating procedure. We will further discuss the effect of these functionality modifications on the antibacterial activities against Gram-positive (*Staphylococcus aureus*) and Gram-negative (*Pseudomonas fluorescens*) microbial cultures, and will discuss the effect of nanocoating size, distribution, morphology, topography and doping/compositing on the activity. The correlation between surface hydrophilicity/hydrophobicity and antibacterial properties will be discussed.

### S3-O11. Smart nanocoatings with thermo-switchable antibacterial activity

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The diminished potency of current biocides and antibiotics, leading to increased mortality and higher medical costs has motivated the intensive search for alternative methods of antibacterial defence. One of the most perspective approaches includes the application of silver nanoparticles (AgNPs), as most of the antibiotic resistance mechanisms of bacteria are irrelevant for NPs, which bind directly to bacterial cell walls causing membrane disruption and elimination of bacteria.

The main goal of the project was to combine the unique properties of thermo-responsive polymer brushes and AgNPs in smart nanocoatings with controlled, thermo-switchable antibacterial activity. Such coatings should be safe for human cells and environment and may be applied for various applications, including anti-bacterial cell culture dishes and scaffolds for tissue engineering.

Antibacterial activity of the coatings was tested against model bacterial strains, *E. coli* and *S. aureus* at 4 and 37 °C using serial dilution method.

At 4 °C no significant difference between the amounts of bacteria accounted on the coatings and the control sample was observed. In contrast, at 37 °C almost no bacteria were observed for coating with AgNPs, whereas the growth of bacteria remained not disturbed for the “pure” coating, indicating the strong temperature-dependent antibacterial properties of AgNPs integrated into brushes

The fabricated novel thermo-switchable nanocomposites are promising materials for applications in food packaging, medical laboratories as well as in air-conditioning systems where the enhanced proliferation of bacteria is a common and unwanted tendency.

**Keywords:** smart nanocoatings, antibacterial properties, silver nanoparticles, thermo-switching



### **S3-P1. Comparative study on the antioxidant activity of extra virgin olive oil samples using a newly developed electrochemical method and DPPH spectrophotometric assay**

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Olive oil, which is the main source of dietary fat, is a complex mixture of natural compounds having multiple beneficial effects on the body. The phenolic content of olive oil confers to this product antioxidant, free radical scavenging, cardioprotective, and anticancer properties.

The present study describes the development of an electrochemical method based on a screen-printed carbon electrode modified with carbonaceous nanomaterials used for the determination of antioxidant activity of several extra virgin olive oil samples. At the same time a correlation between the data obtained by the proposed electrochemical method and those obtained by applying DPPH (2,2'-diphenyl-1-picrylhydrazyl) spectrophotometric method has been performed.

The optimised method based on cyclic voltammetry was used to determine the antioxidant properties of extra virgin olive oil samples and the results were compared and correlated with the results obtained by DPPH spectrophotometric assay.

The cyclic voltammetry (CV) measurements were carried out in the hydroalcoholic extract of the extra virgin olive oil samples and in solutions of catechin of different concentrations. The analysis of the radical scavenging properties of the real samples and catechin solutions by DPPH method were correlated with the electrochemical data expressed as the area under the anodic branch of the cyclic voltammogram. The CV method results were in good agreement with the results obtained by the spectrophotometric method.

The results obtained from this study indicate that the extra virgin olive oil extracts possess antioxidant properties and could serve as free radical inhibitors or scavenger or, acting possibly as primary antioxidants. Moreover, the developed method could represent a valuable alternative for the analysis of antioxidants in complex samples than the classical spectrophotometric method that is prone to interference from colour and turbidity of the samples and also usually requires pre-treatment of the complex samples.

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**Keywords:** olive oil, antioxidant activity, cyclic voltammetry, DPPH spectrophotometric assay

### **S3-P2. IT technologies for medical data management**

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This paper presents the main approaches of new IT technologies used in healthcare.

These technologies refer to Big Data, Big Data Analytics (BDA) and Internet of Things (IoT). The new Big Data technologies can help physicians choose the correct and faster treatment, based on information collected by another healthcare professional.

Patients can benefit from more appropriate and on time treatment to be better informed about health care providers. The BDA addresses the challenges generated by: the increasing share of unstructured and multi-structured data generated by highly prolific and widespread data sources (such as social networks, sensor networks, the Internet of Things) and the growing gap in the amount of important data and the ability to process them in time for decision support.

### S3-P3. Electrochemical and DFT analysis of quantum chemical reactivity parameters for electrochemical applications of an azulene-phenyloxazolone

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The azulene derivative 4-((5-isopropyl-3,8-dimethylazulen-1-yl) methylene)-2-phenyl oxazole-5(4H)-one (L) was investigated by electrochemical methods. L was previously synthesized and characterized by physical methods. Previous computational approaches were done for designing novel ligands with enhanced electrochemical properties.

The aim of this work was the establishment of L ability to recognize heavy metals (HMs) for analytical purposes, building CMEs for sensing; obtaining complete structural insights for further electrochemical properties estimation by DFT.

There were used different methods: differential-pulse voltammetry, cyclic voltammetry (CV), rotating disk electrode, DFT theoretical approach using B3LYP/6-31 G\* density functional.

L was electrochemically studied on glassy carbon in 0.1M TBAP/ CH<sub>3</sub>CN. L's films were obtained by successive cycling at different potentials or by controlled potential electrolysis. The formation of the films was proved by recording the CV curves of the chemically modified electrodes (CMEs) in ferrocene solutions. CMEs prepared for various charges or potentials were used to detect heavy metal (HM) ions. Synthetic samples of HMs solutions (Cd (II), Pb (II), Cu (II), Hg (II)) were analysed using these CMEs. The DFT studies based on the correlation of electrochemical oxidation and reduction potentials with energy levels corresponding to the HOMO or LUMO, respectively, resulted from quantum computations were done. The complexing capacity of L was discussed also based on electrostatic potential map (Figure 1) and charge distribution.

Good results for Pb(II) analysis were obtained on L-CMEs, complete structural insights for further electrochemical properties. DFT methods afford access to new hybrid azulene ligands with promising characteristics for organic sensors materials in view of HMs analysis.

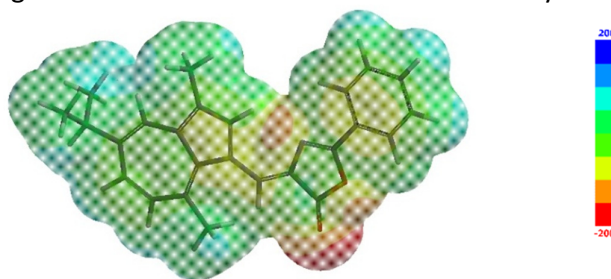


Figure 1: Predicted molecular electrostatic potential map for L.

**Keywords:** DFT quantum chemical reactivity parameters, redox potentials, electrochemical applications.

### S3-P4. Electrochemical properties of 2,6-bis((E)-2-(furan-2-yl) vinyl)-4-(azulen-1-yl) pyridine

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Azulene derivatives were investigated by electrochemical methods to establish their ability to recognize heavy metal (HM) ions for analytical purposes, such as sensors. 4-(azulen-1-yl)-2,6-bis((E)-2-(furan-2-yl)vinyl)pyridine (L) ligand was previously synthesized and characterized by physical chemical methods.

Starting from this premise, the present study focused on the investigation of L, to evaluate its sensory response for HM ions.

The electrochemical study of L was performed on glassy carbon working electrode by using cyclic voltammetry (CV), differential pulse voltammetry (DPV), and rotating disk electrode voltammetry (RDE). Polymeric films were formed by electrochemical oxidation either by cycling the potential in the anodic scans or by controlled potential electrolysis at different anodic potentials and charges. The obtained modified electrodes were tested as sensors for heavy metal ions.

The formation of thin films is confirmed by the comparative recorded cathodic CV curves in ferrocene (Fc) solution of the obtained L-CMEs and of the bare electrode. Stripping peaks for Cd, Pb, Cu and Hg ions (each having a  $10^{-5}$  M concentration) were noticed at different potentials. For each investigated HM ion, the DPV curves in the tested accumulation solutions showed the corresponding stripping peak currents which are higher than for bare electrode indicating that these ions have been retained in the electrogenerated films, ensuring the immobilization into L complexing units in different percentages.

One of the results shows that the estimated detection limit of Pb species is less than  $10^{-8}$  M.

**Acknowledgments:** This research was funded by Romanian National Authority for Scientific Research, UEFISCDI, under grant PN-III-P2-2.1-PED-2019-0730, contract no. 293PED/2020.

**Keywords:** 2,6-bis((E)-2-(furan-2-yl) vinyl)-4-(azulen-1-yl) pyridine, voltametric methods, chemically modified electrodes, heavy metal ions detection

### S3-P5. Data management application in a smart hospital

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This paper proposes a system based on cloud technology, which can provide a transmission of inter and intra hospital information in real time, so as to reduce the time, stress and possible delays caused by human errors.

At the same time, it can restrict access to certain confidential data, but also make it easier to search the medical history, by reducing the time spent searching in archives or avoiding unforeseen situations, such as losing medical records or destroying them in a fire.

By creating a web and mobile application specific to a smart hospital, capable of facilitating the management of patient data, each doctor will receive all the medical tests and other data needed to decide on the actions to be taken to treat patients.

**Keywords:** management, hospital, medical history

### S3-P6. Biopolymeric compounds with medicinal applications

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Biopolymers derived from nature sources or microbial have grown in importance in the medical field. They are used alone or in polymeric complexes, in tissue engineering for tissue reconstruction, or as drug delivery systems in combination with drugs to accelerate regeneration. Among the various biomaterials available for therapeutic applications, polyhydroxyalkanoates (PHAs) offer new properties as biomaterials of interest due to their high biocompatibility, as well as their biodegradability and various thermo-mechanical properties.

This paper takes into account the leading medical representatives of biopolymers with medical applications (PHAs and polysaccharides), with a particular focus on obtaining a scaffold hybrid biomaterial composed of biopolymers (e.g., polyhydroxybutirate and chitosan) and bioceramic material.

To create the hybrid biomaterial, each biopolymer was dissolved in a trifluoroacetic solution at a temperature of 35-40°C. After thoroughly mixing the two polymeric components, the bioceramic was added and stirred for approximately one hour at 45°C. The mixture was poured into a Petri dish and left to evaporate overnight.

Following the trials, distinct flexible and translucent films, easily processed for medical purposes, were obtained depending on the ratio of the component polymers.

Although only PHB has been approved by the FDA for medical applications, its demonstrated favorable properties (immunologically inert, biocompatible, rapid tissue growth, bioresorbable, slow biodegradable tissue scaffolds), as well as a large number of promising studies with other PHAs, indicate an optimistic outlook for the development of those biopolymers alongside natural biopolymers.

**Acknowledgments:** This work was supported by a grant of the Ministry of Research and Innovation, project number PN-19-410401 (Contract 34 N/2019).

**Keywords:** polyhydroxyalkanoates, polysaccharides, bioceramics, scaffolds

### **S3-P7. Folate-modified pullulan acetate nanoparticles loaded with curcumin for cancer therapy**

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Folate-modified nanosystems have the potential to improve several drawbacks of conventional chemotherapy including nonspecific biodistribution and targeting, poor water solubility and poor bioavailability.

The aim of study was to obtain and characterise folate-modified pullulan acetate nanoparticles for cancer therapy.

Folate-modified pullulan acetate nanoparticles were prepared by a solvent diffusion method using folate-modified pullulan acetate, an amphiphilic block copolymer stabilizer (Pluronic F127) and curcumin as anticancer agent. The nanoparticles were characterised based on entrapment efficiency, size, polydispersity index, stability and release profile. In vitro curcumin release from was performed in phosphate-buffered saline, pH 7.4 and pH 5 by using dialysis bag method under sink conditions.

Folate-modified pullulan acetate nanoparticles displayed nanometric size, narrow distribution, satisfactory entrapment efficiency, and good storage stability in distilled water at 4°C for three months with no significant curcumin loss. Curcumin was released from the nanoparticles in a biphasic profile with a fast release rate in the first 6h followed by a slow release. The maximum curcumin release was reached after 72 hours.

The results demonstrated that folate-modified pullulan nanosystems could serve as a potential nano-carrier for cancer therapy. Further studies are needed to demonstrate anti-cancer activity and targeting ability of the systems.

**Acknowledgments:** This work was supported by a grant of the Ministry of Research and Innovation, project number PN-19-410401 (Contract 34 N/2019).

**Keywords:** cancer therapy, folic acid, nanoparticles, nanotechnology, drug delivery

### S3-P8. From whole cyanobacterial cells of *Synechococcus elongatus* PCC 7942 to PSII: the effect of diuron on photosynthesis

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The cyanobacterial strain *Synechococcus elongatus* PCC 7942 was longtime used as a model in biosensors for the detection and study of photosynthesis-inhibiting pesticides [1]. Choosing the optimal bioreceptor providing the best combination of stability, high photocurrents and good sensitivity to herbicide is a key point for developing efficient analytical devices. Whole cells and subcellular preparations of thylakoids and photosystem II (PSII) particles present each particular advantages. In this context, the current investigation compared the photosynthetic activity and sensitivity to diuron of whole cells, thylakoid membranes and PSII particles from *S. elongatus* PCC 7942. *Synechococcus elongatus* PCC 7942 was cultivated in BG-11 medium up to an OD (730 nm) of 0.9, then lysed to obtain thylakoid membranes [2]. PSII particles were obtained from thylakoids by extraction with detergent, ultracentrifugation and separation in sucrose gradient [3].

The stability of the investigated photosynthetic preparations at -20° C in the presence of DMSO, glycerol and PEG-8000 as stabilizers was also investigated.

The photosynthetic activity was measured via amperometry using screen-printed 3-electrode cells that include a working carbon nanotube electrode, a carbon counter electrode and an Ag reference electrode (DRP-110 D CNT, from Metrohm Dropsens, Spain). The measurements were conducted in 20 mM phosphate buffer pH 7, with 0.15 mM NaCl and 1mM MgCl<sub>2</sub>, in the presence of 0.5 mM 2,6-dichlorobenzoquinone (DCBQ) as electrochemical mediator. The cell was illuminated with white light for 1 min light/15 min dark cycles. All samples were diluted to have 7.5 µg/mL chlorophyll in the cell. The intensity of the photocurrent after 1 min of illumination was compared to that measured in similar conditions after the incubation with 60 ppb diuron. It was confirmed that the all preparations tested were suitable for the electrochemical detection of diuron.

Thylakoids, showing good stability and sensitivity were studied in more detail. The morphology of thylakoid membrane from *S. elongatus* PCC 7942 was studied by atomic force microscopy (AFM). Moreover, thylakoids were immobilized on CNT-electrodes with redox polymer G43 using PEDGDE cross-linker [4]. In this case, no additional mediator was needed so the measurements were performed in buffer without DCBQ, but the photocurrents were reduced. It was confirmed that sensors based on thylakoids immobilized on top of electrodes are adequate to study photosynthesis-inhibiting herbicides. In perspective, we aim to address the selectivity of the sensors and test real water samples.

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### **S3-P9. ZrCu thin film metallic glasses as biomaterials for coating of metallic orthopedic implants**

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In the current study, ZrCu-based thin film metallic glasses were developed as biomaterials especially for coatings of metallic orthopedic implants. The following coatings were proposed: ZrCu containing small amounts of Si, Ca, Sr, Mo and Mg. The films were deposited with a high deposition rate by a cathodic arc technique. Mostly utilized for orthopedic implants, Ti6Al4V alloy was used as substrates.

EDS and XRD techniques were used to analyze the elemental and phase compositions of the films. Scratch tests and surface profilometry were used to examine hardness, adhesion, and roughness. The investigation of the film corrosion resistance played an important role. The corrosion rate was calculated after these experiments, which were carried out in SBF at 37°C. For all of the tests, uncoated alloys were used as a reference sample. After electrochemical tests, the morphology of the generated films was examined.

All of the films enhanced the qualities of the uncoated substrates, according to the tests. The electrochemical behavior revealed that by adding Si, Mo, and Sr to the ZrCu structure, it improves the corrosion rate of uncoated and coated substrates significantly.

**Acknowledgements:** We acknowledge the support of the Romanian Ministry of Education and Research, CNCS - UEFISCDI, project PN-III-P4-ID-PCE-2020-1264 (PCE95/2021), within PNCDI III, and Romanian National Core Program no.PN18N-01-02/2019.

**Keywords:** metallic glasses, thin films, corrosion

### **S3-P10. In vitro evaluation of doped hydroxyapatite coatings electrochemically deposited on titanium nanostructured surface**

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In medicine, the coatings field is rapidly diversifying, aiming to improve the osseointegration of the metallic implant and to tackle the bioinert character of the metallic materials.

The aim of this research was to evaluate the *in vitro* behaviour of Mg or Zn doped HAp coatings obtained by electrochemical technique in pulsed galvanostatic mode on titania nanotubes.

The surfaces were analysed in terms of wettability, bioactivity, electrochemical behaviour, cell interaction and antibacterial efficiency.

Addition of Zn or Mg into HAp has enhanced the wettability and *in vitro* cell behaviour, while on the antimicrobial efficiency the best results were attributed to the Zn doped HAp.

Overall, it can be said that addition of the Mg or Zn can enhance the *in vitro* behaviour of the HAp based coatings in accordance with to the clinical needs.

**Acknowledgments:** This research has been funded by the Romanian Ministry of Education and Research, CNCS - UEFISCDI, project number PN-III-P1-1.1-TE-2019-1331, within PNCDI III (project no. TE 172/2020; 3B-CoatED).

**Keywords:** hydroxyapatite; bioactivity; MTT assay; antibacterial efficiency.

### S3-P11. Influence of Mg and Zn content on the properties of hydroxyapatite based coatings

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Biomaterials are an integrated part of modern medicine, and their development and improvement are essential.

Thus, the aim of this research was to obtain hydroxyapatite (HAp) based coatings doped with different concentration of Mg ( $H/MgX$ , where  $X$  is 1 and 1.5 mM of  $Mg(NO_3)_2 \cdot 6H_2O$  in the electrolyte) and Zn ( $H/ZnY$ , where  $Y$  is 0.015 and 0.020 mM of  $Zn(NO_3)_2 \cdot 6H_2O$  in the electrolyte) on Ti nanostructured surface through electrochemical techniques, and to evaluate the influence of the doping elements content on the physico-chemical properties of HAp. All electrolytes were prepared with respect to  $(Ca+Mg)/P$  ratio of 1.67, where  $M$  is Mg or Zn concentration.

The HAp based coatings were electrochemically deposited in galvanostatic pulsed mode on titania nanotubes. The materials were analysed by SEM-EDS, XRD and FTIR. The coatings adhesion on the substrate was also investigated.

All coatings present a morphology that consists in ribbon like crystals. XRD and FTIR have confirmed the formation of HAp phase in all coatings. Additions of the doping elements in smaller amounts don't alter the coatings adhesion.

The results have indicated that addition of Zn or Mg in smaller amounts enhances the properties of HAp coatings, compared to higher ones.

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