Book of Abstracts

Nano-Biotechnologia PL

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Bionanomaterials Centrum opening and Laboratory of Nanostructures for Photonic and Nanomedicine CEPT presentation

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17 September 2012

NanoForCE InfoDay and Mat&Nano Brokerage Day

18 September 2012

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Programme

Monday, 17 September

Nano-Biotechnology Poster session

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8:50	Poster	B1

Preparation and properties of the nanocrystalline Ti-6Zr-4Nb alloy with Ca-P coating as a biomaterial for hard tissue implant applications

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Formation of the Ti-6Zr-4Nb porous nanocrystalline alloy with Ca-P coating was described. The alloy was prepared by mechanical alloying followed by pressing and sintering. The porous bioactive surface was produced in two steps electrochemical treatment: anodic oxidation in 1M H_3PO_4 + 2% HF electrolyte at 10V for 30min and subsequent cathodic Ca-P layer deposition at -5V for 1h, using a solution mixture of $Ca(NO_3)_2$ + $(NH_4)_2HPO_4$ + HCl. The microstructure and morphology of the biofunctionalized surface was studied by TEM, XRD, SEM, and optical profiler. The corrosion potentiodynamic tests were performed in Ringer's solution. The corrosion current density does not significantly change after surface modification, indicating good corrosion resistance of the biofunctionalized surfaces. Biocompatibility was investigated by in vitro tests using normal human osteoblasts (NHOst) which showed good cells behavior. The nanocrystalline Ti-6Zr-4Nb alloy with modified surface is a possible candidate for hard tissue implant application.

8:50	Poster	B2

Production Of Carbon Nanotubes CNTS Using Alcohol Catalytic Chemical Vapour Deposition (ACCVD) Technique: A Review

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Abstract. The mass production of high purity Carbon Nanotubes CNTs at cheaper rate is becoming the most important factor in the application point of view and most industries today are opting for the Chemical Vapour Deposition CVD technique. The CVD method is the most suitable synthesis method in terms of product quality and quantity. This is due to its flexibility, inexpensive,

energy efficient, simple and low cost of fabrication, and ability to produce CNTs in large quantity. This paper reviewed the production of CNTs using the Alcohol Catalytic Chemical Vapour Deposition (ACCVD) technique with emphasis on the influence of hydrocarbon, reaction temperatures, catalyst and substrate materials on the growth of CNTs;

BRIEF INTRODUCTION OF THE PROJECT.

Carbon in its various forms can be referred to as soot, diamond, graphite, coke, Buckey Balls, carbon nanotubes and Fullerenes. Carbon nanotubes (CNTs) are allotropes of carbon with a cylindrical nanostructure, also referred to as sheets of graphite rolled into tubes and possess excellent properties due to their symmetric structure (Barros et al., 2006). They are broadly classified into single-walled carbon nanotubes (SWNTs) and multi-walled carbon nanotubes (MWNTs). CNTs possess exceptional properties and wide range applications as a result of their high strength, stiffness, thermal and electrical conductivity (Wang et al., 2000), CNTs are designated as attractive material in applications range from nanoelectronics (Tsukagoshi et al., 2002), sensors (Rivas et al., 2007), ultracapacitor (Adewuyi et al., 2007), and field emitters to composites (Ajayan et al., 2000). Reliable growth techniques capable of yielding high-purity material in desirable quantities are critical to realize CNTs potential applications. A number of technological advancements ranging from agriculture, electronics, medicine, aerospace, power, to automobiles can be traced to the applications of CNTs. CNTs can be produced by electric arc discharge method (Iijima 1991), chemical vapour deposition method (Jose- Yacaman et al., 1993), pulsed laser vaporization technique (Guo et al., 1995), and high-pressure carbon monoxide conversion (HiPCO) process (Nikolaev et al., 1999).

In pulse laser vaporization and arc discharge methods, although high quality materials can be produced, the high temperature employed for evaporating the carbon atoms from solid carbon sources (over 2700°C) make them difficult to scale up the process in a cost-effective way. Also, arc discharge and pulse laser vaporization methods produce CNTs as powder samples in bundles while CVD offers synthesis of CNTs on substrates (Seidel et al., 2003) as well as in powder form (Nikolaev et al., 1999). Additionally, using CVD the diameter (Li et al., 2001) length and orientation (Huang et al., 2003) of CNTs can be effectively controlled.

Hence, chemical vapour decomposition (CVD) method is the most suitable synthesis method in terms of product quality and quantity (Zheng et al., 2002). This is due to its flexibility, inexpensive, energy efficient, simple and low cost of fabrication, to uniformly deposit thin films of materials, even onto nonuniform shapes and ability to produce CNTs in large quantity. Since, there is a huge demand for CNTs production and application for various industrial uses, it is necessary to maximize the yield and minimize the production cost. The major applications of CVD technique take advantage of the unique characteristics of the process, such as the capability of producing materials of exceptionally high purity. The CVD method is currently the best hope for large-scale production of SWNTs. CVD processes can be used to deposit a wide range of conducting, semiconducting, and insulating materials. The aim of this project therefore is to initially developed a model for the synthesis and growth of carbon nanotubes and later proceed to the actual production process.

8:50	Poster	B3

Albumin-HMchitosanGTMAC-based nanoparticles as potential controlled release delivery systems of curcumin

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Curcumin is a natural polyphenol derived from a traditional herbal remedy. It shows the promising anti-inflammatory, anti-cancer and anti-oxidant activities. It is, however, only sparingly soluble in water and therefore its bioavailability is very low.As a solution to that problem we propose a new type of hybrid nanocarrier for effective curcumin. The carrier is based on bovine serum albumin (BSA) and N-dodecyl-N-glycidyltrimethylammonium-chitosan (HMchitosanGTMAC). Albumin is known for its extraordinary ligand binding capacity and non-covalent reversible binding characteristics. Chitosan is a cationic, biocompatible polymer. Stable aqueous dispersion of bovine serum albumin (BSA) nanoparticles NPs containing curcumin was obtained by a coacervation process followed by electrostaticly driven adsorption of cationic N-dodecyl-N-glycidyltrimethylammonium-chitosan (HMchitosanGTMAC) on the NP surfaces. Variuos concentrations of HMchitGTMAC were used for coating of BSA NPs to stabilize the colloidal system and to control the release of curcumin. The physicochemical properties of the resulting objects were then studied. The size and surface potential of the nanoparticles were evaluated using dynamic light scattering (DLS) measurements. The morphology of the unloaded and curcumin-loaded nanocarriers was evaluated by Atomic Force Microscopy (AFM) and by Scanning Electron Microscopy (SEM). The binding constant (K_a) of curcumin to bovine serum albumin (BSA) and curcumin release profiles were determined. The high K_a value and plausible release profiles suggest that our novel delivery system, based on albumin, may act as an effective carrier for curcumin.

Acknowledgements: Project operated within the Foundation for Polish Science Team Programme co-financed by the EU European Regional Development Fund, PolyMed, TEAM/2008-2/6.

8:50	Poster	B4

Adsorption study of pharmaceutical substances on the silica particles surface

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Among different materials investigated as drug carriers (liposomes, micelles, polymeric nanoparticles and metal oxides) interesting are silica particles. The properties of silica particles (chemical and mechanical stability, hydrophilicity, biocompatybility) give an opportunity to use this material as a new universal drug delivery system [1]. Silica has an interesting capability due to possibility for tailoring their surface reactivity and realising desirable electrical surface potential (zeta potential) that can be obtained by the surface modification. The separated silica particles possess a negative charge what is demanded. Negatively charged particles are repelled by the negatively charged cell membrane so these particles should be slowly detected and rejected. This effect helps the particles avoid elimination from the body. SiO_2 degrades in the body by hydrolysis of siloxane bonds into $Si(OH)_4$, which is eliminated through the kidneys [2].

As a model drugs we used substances such as methylene blue, diclofenac sodium and famotidine. Drugs were loaded onto silica spherical particles (diameter size 150 nm) by soaking and filtration, and after centrifugation solution was analyzed by a spectrophotometer. In order to analyze adsorption data in aqueous media, Langmiur and Freundlich models were used.

We have found that the chemical structure of the drugs influence the drug-loading amount. Simple structure of diclofenac sodium causes low level of the adsorption (0.42%). The best adsorption results we have obtain for the methylene blue (36%) which have more difficult structure and better possibility to connect with silica.

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fects after implantation. Biomaterials 2000, 21, 193-198.

8:50	Poster	B5

Antibiotics adsorption and desorption processes from the silica particles surface

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Recently nanotechnology has been rapidly coming out in biomedical applications, such as drug or gene delivery carriers, cancer therapy and diagnosis of diseases. A significant advances in drug-delivery systems have enabled more effective drug administration. A range of organic systems, such as: micelles, liposomes, dendrimers and polymeric nanoparticles have been designed to deliver drugs to specific organs. All mentioned organic systems suffer from limitations, including poor thermal and chemical stability and rapid elimination by the immune system [1]. Inorganic materials, especially silica has a fundamental advantages over the organic materials. Amorphous SiO_2, in contrast to crystalline silica, offers a biocompatible, stable and promising alternative. The advantages of silica lie also in well-developed silane chemistry and easy preparation. Bioactive molecules (drugs) can be adsorbed on the silica surface and encapsulated within silica materials by the sol-gel method or emulsion chemistry. Sol-gel method enable the production of various materials with homogeneous drug distribution and permit ambient temperature processing, necessary for handling active molecules and biological. The silica materials employed as a matrix are chemically inert towards the encapsulated substances [2].

In this work we are presented the study of the adsorption and desorption of some antibiotics (ceftazidime, chloramphenicol and gentamicin) on/from silica particles surface. Silica particles have diameter size about 150 nm and were obtained by the sol-gel method. Loading of the particles was performed by immersion method, in which the particles were diped into a drug solution with definite concentration. After the centrifugation the supernatant was analyzed using a spectrophotometer. Langmuir and Freundlich isotherms were applied to the experimental data obtained for researched antibiotics.

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8:50	Poster	B6

PPUH MEDGAL Józef Borowski - Implanty antyalergiczne z powłoką węglową o budowie nanokrystalicznej

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Firma MEDGAL jako pierwsza na świecie wprowadziła na rynek medyczny stalowe implanty do osteosyntezy, dodatkowo pokryte antyalergiczną warstwą węglową o budowie nanokrystalicznej. Proces pokrywania implantów odbywa się przy wykorzystaniu metody opatentowanej przez firmę MEDGAL wraz z Politechniką Łódzką.

Dzięki zastosowaniu powłoki węglowej implanty zyskują następujące cechy:

- bardzo wysoka biotolerancja i biozgodność

- bardzo wysoka odporność na korozję w środowisku tkanek i płynów ustrojowych

- wyeliminowanie metalozy i powikłań odczynowych w obszarze zespolenia kości

- wyeliminowanie niekorzystnych dla organizmu reakcji toksycznych i alergicznych, a przez to znaczące zmniejszenie powikłań pooperacyjnych.

8:50	Poster	B7

Novel polymeric inhibitors of human coronavirus NL63

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Human coronavirus NL63 (HCoV-NL63) is one of the most important pathogens which may cause a common cold and severe upper and lower respiratory tract diseases. It is estimated that HCoV-NL63 causes 1-10% of all respiratory infections per year, especially in winter and early spring. Thus, there is a need to obtain more effective methods of preventing and treating these infections.

The aim of this study was to develop and explore anticoronaviral activity of polymer-based compounds. Four polycations - a cationically modified chitosan derivative (HTCC), hydrophobically-modified HTCC, poly(allylamine hydrochloride), and cationically modified polyvinyl alcohol were investigated. In order to appropriately evaluate the inhibitory activity of the tested polymers there was performed in vitro study (LLC-MK2 cells). The cytopathic effect (CPE) was correlated with a quantitative RT-PCR based assay. The cytotoxicity was examined by an XTT assay and a Neutral Red assay.

The results show that cationically modified polymers are effective inhibitors of HCoV-NL63 rep-

lication. A cationically modified chitosan derivative HTCC seems to be the best inhibitor of replication of HCoV-NL63 at non-toxic concentrations. Non modified polymers (chitosan, polyvinyl alcohol) did not show any antiviral properties.

Acknowledgements

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8:50	Poster	B8

Bioactive glass-modified composites for bone tissue engineering

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Artificial bone substitutes include metals, polymers and ceramics that are selected for clinics by their ability to support bone cell growth and osteogenic differentiation of progenitor cells, i.e. they display osteoconductive properties. Yet, one-phase materials are not able to fulfil the requirements for the ideal bone substitute and very few biomaterials are osteoinductive, i.e. they induce osteogenesis on their own. We have tested the hypothesis the incorporation of gel-derived bioactive glasses (SBG) into polymer (i.e. poly(L-lactide-co-glycolide; PLGA) or titanium dioxide (TiO_2) will modify the nanostructure of resulting composite materials, improve their mechanical properties, surface activity (i.e. bioactivity) and result in composite osteoinductivity in human mesenchymal stem cell (hMSC) cultures. Composites made of either TiO_2 or PLGA were combined with either A2 or S2 SBG of respective CaO/SiO_2 ratios: 54/40mol% (A2) or 16/80mol% (S2). PLGA was combined with 21%vol of A2 or S2 S-BG, whereas TiO_2 with 25, 50 or 75 wt% of A2 or S2 SBG. PLGA-SBG composites were next fabricated into 3D scaffolds, whereas TiO_2-SBG composites into cell growth surfaces. Human MSC were harvested from bone marrow of adult patients, seeded onto scaffolds and surfaces and stimulated in culture with either dexamethasone (Dex) or recombinant human BMP-2 up to 20 days. Bone-like hydroxyapatite structures formed mainly inside the PLGA-SBG scaffolds. Human MSC cultures on bioactive PLGA-A2 scaffolds increased ALP activity, osteopontin and BMP-2 mRNAs without Dex or BMP-2 treatments. When hMSC on PLGA-A2 scaffolds were treated with BMP-2, cells elevated ALP activity, RANK-L, osteoprotegrin and osteocalcin mRNAs compared to plain PLGA.
In addition, BMP-2 treated hMSC cultured on PLGA-SBG scaffolds elevated several osteogenic

genes compared to Dex-treated hMSCs. Either Dex or BMP-treated hMSC cultured on PLGA-SBG scaffolds elevated collagen type I and matrix mineralization compared to plain PLGA. TiO_2-SBG surfaces developed hydroxyapatite-like forms mostly on the surface of composites containing 50 or 75wt% of either SBG. Higher content of A2 or S2 SBG in TiO_2 decreased the viability of hMSC and thiswas observed regardless of the time and culture conditions. Incorporation of 25wt% S2 SBG increased almost twice ALP activity in Dex-treated hMSC compared to Dex-treated cells on plain TiO_2.
In contrast, increased A2 SBG contents in the TiO_2 composites significantly enhanced both matrix mineralization and cell-mediated collagen production. Our data indicate the human MSC osteogenic response can be enhanced by bioactive composites made of either PLGA or TiO_2 enriched with gel-derived bioactive glasses A2 or S2. We believe the osteoinductive properties of the reported PLGA- or TiO_2-based SBG composite scaffolds in human MSC cultures may prove useful in several bone tissue engineering strategies and therapies where cell and/or growth factor delivery are not sufficient to induce proper bone formation.

8:50	Poster	B9

The influence of platinum nanoparticles on human epidermal keratinocytes.

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Nanoparticles (NPs) have diverse applications in industry, medical devices, therapeutic agents and cosmetics. Nanoparticles are defined as a structures that have at least one dimension of nanometer scale. The unique physicochemical properties of engineered nanoparticles are attributable to their small size, large surface area, chemical composition, surface reactivity, charge, shape and media interactions. Although impressive from the perspective of material science, the novel properties of NPs could lead to adverse biological effects with the potential to create toxicity. Because of their strong catalytic activity, platinum nanoparticles are commonly used in converters of automobiles causing emission of PtNPs to the environment. In recent years PtNPs have attracted even more attention due to the fact that they have been shown to quench reactive oxygen species. They are added to supplements or cosmetics due to their anti-oxidative and anti-aging properties. While the commercialization of platinum nanoparticles expands rapidly, their impact on health and environment is still not well understood. In this study, the cellular influences induced by 4-5 nm polyvinylpyrrolidone-coated PtNPs on human primary keratinocytes was investigated. Cytotoxicity, genotoxicity, morphology, metabolic activity and changes in the expression of proteins were studied in PtNPs- treated cells. Also the antimicrobial properties of PtNPs were verified. My studies show, that despite the positive effects, nanoparticles may present possible danger, concerning activation of genotoxic processes, although no cytotoxic effects were observed. It was also found out that PtNPs are effective against Gram- negative bacteria. This data might contribute to better understanding of effects triggered by PtNPs on cells and may provide valuable information to develop safety regulations for the use of nanoparticles in various applications.

8:50	Poster	B10

Diamond nanoparticles - antiangiogenic factor in brain cancer therapy?

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Gliomas are the most common primary brain tumors, glioblastoma multiforme (GBM) being the most aggressive subtype. One of the most effective forms of antiglioma therapy is antiangiogenic therapy. Inhibition of tumor angiogenesis suppresses both tumor growth and metastasis. A large number of pro- and anti-angiogenic cellular factors regulate angiogenesis in glioblastoma. Among them, vascular endothelial growth factor (VEGF) has been implicated as a major mediator in the pathogenesis of glioblastoma. The increased protein level of VEGF is linked to the higher permeability of the blood vessels and their untypical structure, which is characteristic for tumors. Lowering of the VEGF level contributes to the normalization of blood vessels, facilitating the infiltration of other factors into the tumor, while inhibiting VEGF expression limits the transport of nutrients and oxygen into cancer cells. Recently, the new biologically active substances have appeared that can be useful in antiangiogenic therapy: nanoparticles of carbon allotropes [1] (C60 fullerenes, graphite nanoparticles, multi-walled carbon nanotubes, single-walled carbon nanotubes and diamond nanoparticles). Moreover, carbon nanoparticles are bioactive and biocompatible [2].

We evaluated interaction between diamond nanoparticles and angiogenesis in inducted tumor. The study was conducted on multiforme glioblastoma cells (U87MG) cultured on the chicken embryo chorio-allantoic membrane (CAM). The analysis were made on stereoscope microscope, transmission electron microscope (TEM) and scanning electron microscope (SEM). Moreover the expression of proangiogenic genes: FGF-2 (basic fibroblast growth factors), VEGF (vascular endothelial growth factor) and VEGFR (vascular endothelial growth factor receptor) was evaluated. Determination of vascular permeability was performed with using dextran-FITC particles (70 000 kDa).

Nanoparticles of diamond significantly decreased tumor mass and volume, and vessels' area. We also found effect on blood vessels structure. Under the influence of diamond nanoparticles was observed the decrease blood vessels permeability and different structure of blood capillaries in glioblastoma tumors. qPCR analysis showed down regulated gene expression of VEGF and FGF-2 on mRNA level [3].

The present results demonstrate antiangiogenic activity of carbon nanoparticles, making them the potential factors for anticancer therapy.

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8:50	Poster	B11

Polisacharydowo-białkowe membrany do rekonstrukcji rogówki

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Rogówka jako wypukła zewnętrzna warstwa gałki ocznej. Uważana jest za pierwszy i najsilniej załamujący światło element układu optycznego oka. Histologicznie składa się z pięciu warstw z czego najbardziej zewnętrzną jest nabłonek złożony z 5-7 regularnie ułożonych warstw.

Choroby, infekcje i uszkodzenia mechaniczne mogą spowodować uszkodzenia nabłonka rogówki, co może doprowadzić nawet do ślepoty. By temu zapobiec zaleca się całkowity przeszczep rogówki. Jednakże z powodu braku odpowiedniej ilości dawców nauka poszukuje pewnych alternatyw. Jedną z nich jest inżynieria tkankowa pozwalająca na wykorzystanie komórek samego pacjenta, które namnaża się stosując rusztowania komórkowe.

Celem badań było otrzymanie biodegradowalnych układów polimerowych w postaci membran jako potencjalnego materiału do uzyskania podłoży do hodowli komórek nabłonka rogówki gałki ocznej. Głównym składnikiem układu jest chitozan czyli wysoce biozgodny, biodegradowalny, łatwodostępny, oraz stosunkowo tani polisacharyd pochodzenia naturalnego, który został usieciowany za pomocą genipiny - naturalnego, pozyskiwanego z owoców gardenii jaśminowej związku sieciującego. Membrany winny spełniać szereg stawianych im wymagań m. in. biokompatybilność, biodegradacja, wysoka wytrzymałość mechaniczna, elastyczność oraz wysoka przepuszczalność dla wielu substancji biologicznie czynnych i tlenu. W tym celu do układu wprowadzono dodatkowy białkowy składnik
w postaci keratyny.

Wykorzystując mikroskop optyczny oraz mikroskop sił atomowych (AFM) przeprowadzono analizę morfologiczną membran oraz zależność topografii ich powierzchni od składu. Wyznaczono stopień spęcznienia, kąt zwilżania układów oraz zbadano możliwość wykorzystania membran jako podłoży do hodowli komórkowych stosując komórki nabłonka.

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8:50	Poster	B12

Microbial and cell growth on nanoporous anodic titanium oxide (ATO) layers

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The most commonly used materials for bone-implants are titanium and its alloys. It is due to their good biocompatibility, high strength to weight ratio and excellent corrosion resistance [1]. Such materials are therefore widely used in orthopedic, dental and other implants, as well as in medical devices (e.g. screws and plates). Unfortunately, there are some drawbacks connected with titanium implants, e.g. their inertness and long-term osseointegration via the natural oxide (TiO_2) existing on surface. Therefore, nanoporous materials on Ti surfaces become a novel solution for bone implants [2].

Nanoporous anodic titanium oxide (ATO) layers on Ti formed by electrochemical anodization have been proposed as a potential nanostructured material for bone implants [3-4]. The main advantage of such materials is direct growth of TiO_2 on Ti surface, using simple and cost-effective method such as anodic oxidation. Anodization allows to precisely control nanopore size and structure porosity [5]. The presence of nanoporous layer guarantees an excellent inertness of its surface, allowing it to readily heel into the bone tissue.

There are two main aspects connected with bone-implants, osseointegration and risk of post-operative infections. The osseointegration, especially its speed, is a major factor for implant success. The surface topography and surface chemistry are crucial for the short- and long-term success of the osseointegration process. In terms of biomaterial development and implant technology, the cellular response can be affected by topographical structure of the surface. It has been proven that cells sense and react to nanotopography in vitro by exhibiting changes in cell morphology, orientation, cytoskeletal organization, proliferation, signaling and gene expression [6].

One of the most serious side effects connected with implant surgeries is a very high risk of post-operative infection. It is well-known that formation of biofilms by human pathogenic bacteria on medical titanium-based implants can be dramatic, leading to failure of the devices and resulting in the necessity of implant removal [7]. Strains of *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonasaeruginosa* are reported to be significant contributors to infections associated with orthopedic implants. Hence, finding materials that are less adherent to bacteria, yet good for cell growth are of great importance.

ATO films on Ti foil were prepared via a three-step anodization in glycol ethylene and glycerine based solutions containing NH_4F (0.38 wt.%) and H_2O (1.79 wt.%) under a constant voltage of 40 V. The duration of first and second anodizing step was 3h. The third step lasted 10 min for glycol ethylene and 1 h for glicerine based electrolytes. The titanium foil (99.5 % purity) was used as both working and counter electrodes. The anodization was performed in two-electrode cells at constant temperature of 20 °C. Some of the samples were then annealed in the furnace in two dif-

ferent temperatures (400 and 1000 °C) for achieving different polymorphic structures of TiO_2 (anatase and rutile). One part of the samples were then used for the cell growth, whereas others for microbial examinations. Adipocyte derived stem cells obtained from abdominal liposuction were seeded onto TiO_2 surfaces. Cell viability, proliferation and phenotype were assessed by the measurement of redox reactions in the cells, cellular DNA, tritiated thymidine ([3H]-TdR) incorporation and alkaline phosphatase (ALP) production. For the ATP evaluation, the ATPLite™ kit was used. For the cytotoxicity determination, the CytoTox96® Radioactive Cytotoxicity Assay was used. *Staphylococcus aureus* bacteria were used for examining an antimicrobial character of nanoporous TiO_2 surfaces on Ti. The evaluation of the inhibition of microorganisms growth and possibility of a biofilm formation on various nanoporous TiO_2 surfaces were examined. Nanoporous TiO_2 surfaces were placed either on the surface of Mueller-Hinton agar which has been inoculated with microorganism before or stroked perpendicularly into the inoculated selected broth. In both techniques the area of growth inhibition on broths was measured. Prior to each experiment, the plates with TiO_2 surfaces were sterilized in 170 °C for 2 hours in a Sanyo Sterilizer. The cultivation of microorganisms on broths was carried out in incubators Mermmet at 37 ºC for 24 h. For biofilm formation tests, nanoporous TiO_2 surfaces were placed in liquid broths with suspension of microorganism. The cultivation at 37 ºC were continued for 5 days. After that time the occurrence of biofilm on plates was investigated with a microscopic and UV radiation methods.

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Acknowledgements

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This research was partially supported by the Polish Ministry of Science and High Education (Grant No N N204 213340).

8:50	Poster	B13

Up-conversion and down-conversion processes observed in Er^{3+}, Yb^{3+} and Mn^{2+} doped $ZnAl_2O_4$ nanoparticles

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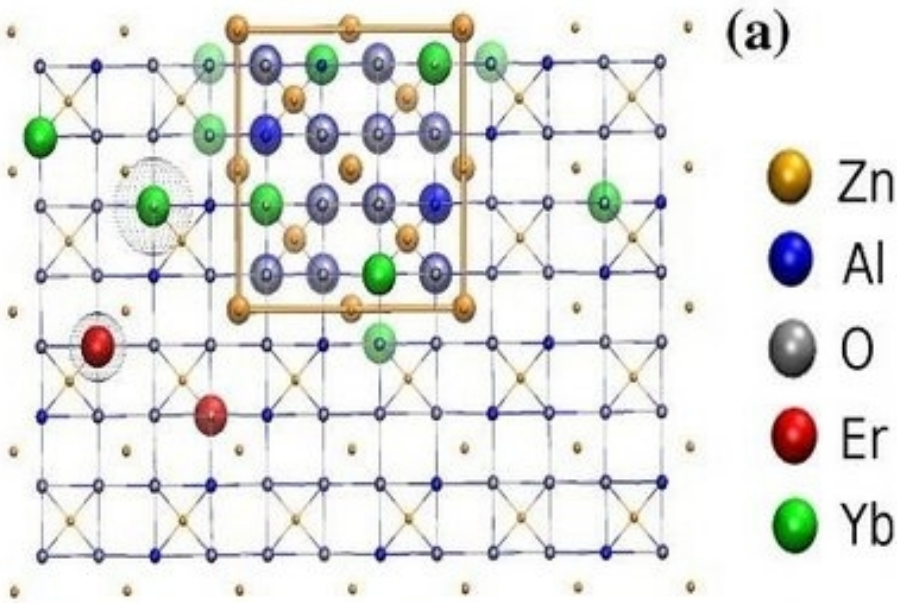
Our objective was to synthesize hydrophilic zinc-aluminum spinel nanostructures doped with rare earth ions such as Er^{3+} and Yb^{3+} (fig. 1a) and ions from transition metal group such as Mn^{2+}. Spinels are isomorphic metal oxides AB_2O_4, where A and B are divalent and trivalent ions, respectively. The material shows a close-packed face centered cubic structure with *Fd*3*m* space group symmetry. They are applicable as useful luminescent bio-marker due to a significant improvement in the signal to background ratio and high resilience to photo-bleaching.


(a)
Zn
Al
O
Er
Yb

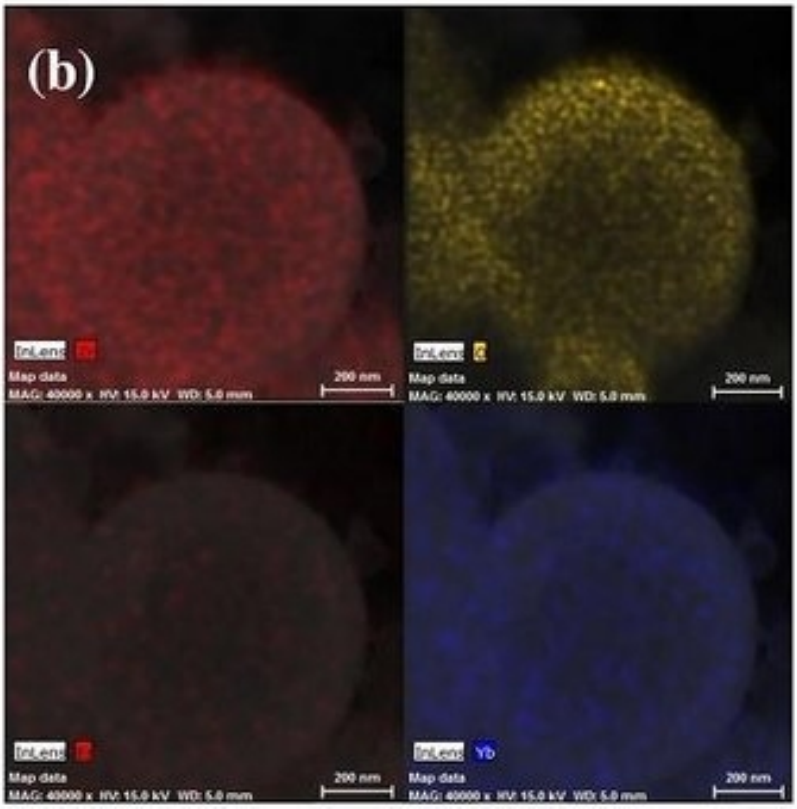

(b)
InLens
Map data
MAG: 40000 x HV: 15.0 kV WD: 5.0 mm
200 nm
Yb

Fig. 1. (a) Spinel $ZnAl_2O_4$: Er^{3+}, Yb^{3+} unit cell geometry (upconversion process) [2], (b) Chemical distribution maps of elements: Zn, O, Er, Yb.

A wide variety of emission spectra could be obtained by changing the concentrations and proportions of the rare earth ions in the crystal host. Although upconversion can be expected, in principle, from most lanthanide-doped crystalline host materials, efficient UC occurs only by using a limited number of well selected host-dopant combinations [1].

The nanoparticles were synthesized in aerosol solutions consisting of droplets (4μm in diameter) by injecting a solution containing the reductors and oxidants to a furnace at the temperature of $1000^{o}C$. The structures of the materials were characterized by transmission electron microscopy and X-ray diffraction.
We obtained regular spherical polycrystalline nanoparticles with a broad size distribution from 20 to 800 nm. To evaluate morphological changes of nanoparticles surface and to visualize the surface topography we used scanning electron microscope. In addition, we performed the EDX material analysis in order to determine distribution of elements in the samples. The results confirmed the presence of Zn, Al, O and Er and Yb (fig. 1b). To check the toxicity of the $ZnAl_2O_4$: Er^{3+}, Yb^{3+} nanoparticles, we introduced them to the body of the living nematode (*Caenorhabditiselegans*). *C. elegans* is a frequently used model organism in biological research, such as process development, embryogenesis, morphogenesis and aging. Confocal microscope images confirm the presence of the nanoparticles inside the gastrointestinal tract of *C. elegans*.

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8:50	Poster	B14

Ultradźwiękowa technika pokrywania polimerowych rusztowań dla zastosowań biomedycznych

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Rusztowania przeznaczone do regeneracji kości umożliwiają wzrost kości w przypadku, gdy ubytek jest zbyt duży, by mógł być zaleczony w sposób naturalny. Takie sytuacje są często spotykane w przypadku złamań, po resekcji nowotworu czy też chirurgii kości twarzoczaszki lub szczęki. Istniejące obecnie na rynku materiały nie spełniają wymagań stawianych przez lekarzy.

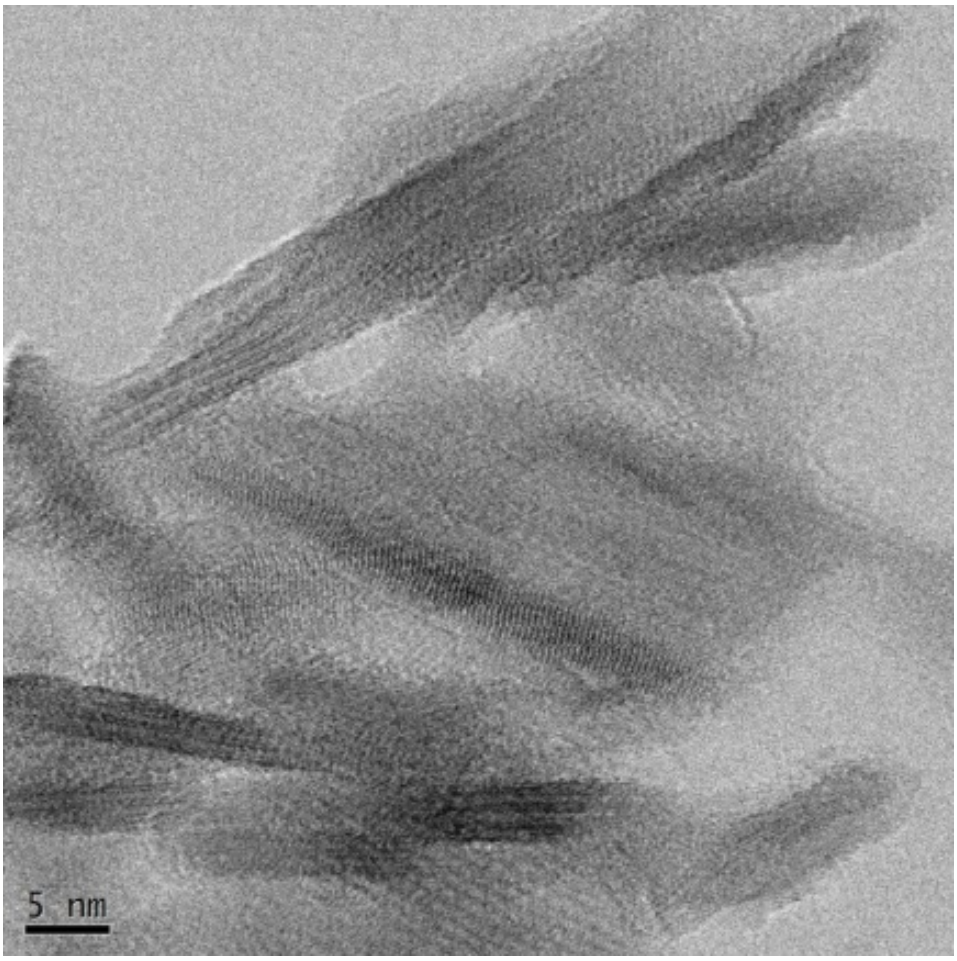
Proces regeneracji zajmuje zbyt dużo czasu, przestrzeń ubytku wypełniana jest nie dość dokładnie, a ich mechaniczne właściwości są zbyt słabe. W praktyce często pojawiają się też procesy zapalne w miejscu ubytku, co utrudnia lub uniemożliwia proces gojenia.

Ludzka kość jest zbudowana w części przez kryształy hydroksyapatytu. Jednak hydroksyapatyt syntetyczny, dostępny na rynku, jest materiałem uznanym za nieresorbowalny przez organizm ludzki. W Laboratorium Nanostruktur dla fotoniki i nanomedycyny IWC PAN opracowano proces mikrofalowej syntezy, który umożliwia otrzymanie w pełni resorbowalnego nano-hydroksyapatytu „*GoHAp”*.

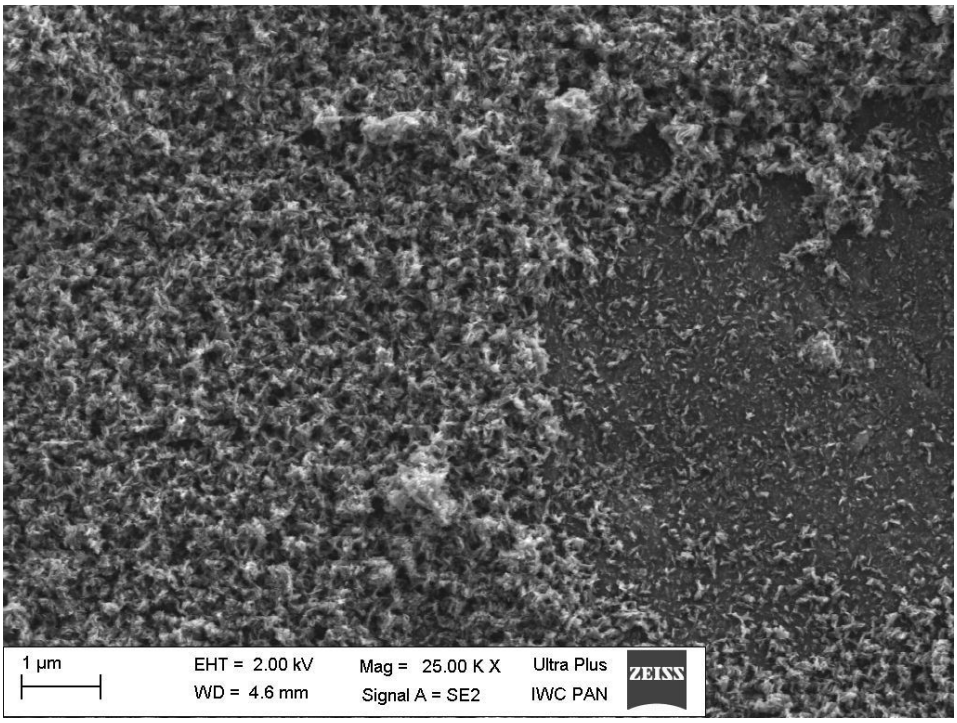
Testy rozpuszczalności proszku *GoHAp*, przeprowadzone zgodnie z Europejskimi Standardami ISO 10993-6: 2007 wykazały, że ma on dużo wyższą rozpuszczalność w porównaniu do komercjalnie dostępnego proszku hydroksyapatytu (NanoXIM). Struktura materiału analizowana była za pomocą transmisyjnej mikroskopii elektronowej (TEM) (Rys.1), a także z pomocą metod dyfrakcji rentgenowskiej. Otrzymano kryształy proszku o morfologii płytkowej, wielkości krystalitów od 5 do 30 nm, oraz powierzchni właściwej do 240 m^2/g.

W ramach przeprowadzonych badań polimerowe rusztowanie pokryto nanoproszkiem hydroksyapatytu *GoHAp z wykorzystaniem* techniki ultradźwiękowej. Warstwa taka umożliwi narastanie komórek kostnych na jego powierzchni z powodu wysokiej biozgodności proszku.

Uzyskaną warstwę analizowano za pomocą skaningowej mikroskopii elektronowej (SEM) (Rys.2.), mikroskopii sił atomowych (AFM) oraz mikroanalizy rentgenowskiej (EDX). Powyższymi metodami określono teksturę, skład oraz wytrzymałość połączenia ceramika/polimer. Wyznaczono także wartość kąta zwilżania powierzchni dla wody. Na podstawie pomiaru metodą atomowej spektrometrii emisyjnej (ICP) oznaczono całkowitą zawartość naniesionego nanoproszku.



Rys. 1. Zdjęcie TEM nanoproszku hydroksyapatytu (*GoHAp*), otrzymanego w Instytucie Wysokich Ciśnień PAN.



Rys. 2. Zdjęcie SEM polimerowego rusztowania pokrytego nanoproszkiem hydroksyapatytu.

8:50	Poster	B15

Core-shell P(LLA-CL) based electrospun nanofibers for peripheral nerve tissue engineering

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Unhealed peripheral nerve defects caused by inflammation or trauma are one of the major problems in today's regenerative medicine. Existing methods of fracture treatment of longer gaps (more than 20 mm) requiring use of sensor autografts do not give a guarantee of full recovery and functionality of the nerve tissue. Schwann cells, which support neurons in peripheral nervous systems and are involved in conduction of nervous impulses along axons play a significant role in nerve tissue engineering. Laminin is an extracellular matrix (ECM) component synthesized after nerve injury and it plays a crucial role in cell migration and axonal growth. A chemical and biological composition of the engineered scaffolds along with its mechanical stiffness influence proliferation and spreading of the cells. Electrospinning is a versatile and simple method, which allows to produce nanofibers with high surface area to volume ratio. Advance techniques such as coaxial

electrospinning enables fabrication of nanofibrous scaffolds with bioactive agents encapsulated within the core of the fiber allowing its controlled delivery.

The aim of this study was to fabricate P(LLA-CL), blended P(LLA-CL)/Laminin and core-shell Laminin/P(LLA-CL) nanofibers and characterize their structure, morphology along with mechanical properties and biocompatibility towards Schwann cells. We obtained P(LLA-CL), blended P(LLA-CL)/Laminin and core-shell Laminin/P(LLA-CL) nanofibers with diameter of 302±101 nm, 350±112 nm and 316±110 nm respectively. The biological study confirmed that use of Laminin/P(LLA-CL) core-shell nanofibers increases proliferation and spreading of the Schwann cells compared to pure polymeric and blended nanofibers. Finally, it could be concluded that the obtained core-shell nanofibers show high potential to be a suitable substrate for peripheral nerve tissue engineering.

8:50	Poster	B16

Silver nanowire sensor for sensitive and quick detection of H_2O_2 in the company of various interfering substances

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In the recent times, considerable interest in a precise determination of hydrogen peroxide (H_2O_2) traces has been observed. This compound is an essential intermediary in food, pharmaceutical, clinical and environmental analyses. Moreover, hydrogen peroxide is a final product of many enzymatic reactions, so its concentration may be used as a direct indicator of the reaction progress. Consequently, the detection of hydrogen peroxide has been recognized as a very important task in many fields of research [1]. For example, it has been found that reactive oxygen species (ROS) – also H_2O_2 – can be considered as the mediators of the biochemistry of cellular pathology and may be involved in etiology of several neurodegenerative diseases, such as Parkinson's disease [2]. Therefore, sensitive and selective determination of H_2O_2 is of great practical importance and many scientific groups have been working on development of new methods for hydrogen peroxide determination. Conventional methods of detection of H_2O_2 are not able to meet many requirements posed to them, including speed, reliability and ease of implementation. The best alternatives seem to be the electrochemical methods which are characterized by good performance, high sensitivity and relatively low production costs [3]. In real samples, such as biological fluids, there are electroactive interfering substances such as ascorbic acid (AA), uric acid (UA) and acetaminophen (AP) that may generate electricity and thus interfere with the detection of hydrogen peroxide.

A nanostructured electrochemical H_2O_2 sensor was prepared by cathodic electrodeposition of silver inside nanoporous anodic aluminum oxide (AAO) templates. AAOs were synthesized via a simple and cost-effective two-step anodization of aluminum. After a suitable treatment, free standing Ag nanowire arrays were obtained and investigated as amperometric sensors for the detection and determination of the hydrogen peroxide in the presence of interfering substances such as glucose and ascorbic acid. Typical electrochemical techniques, including cyclic voltammetry (CV) and chronoamperometry were applied for examination of fabricated sensors. CV tests were used to select an appropriate potential for the reduction of hydrogen peroxide. The chronoamperometric measurements, carried out in a phosphate buffer solution (pH 7.4), were used to study the response

of electrochemical sensors to different concentrations of the analyte.

The results showed that interfering substances do not affect the sensor response (Fig. 1).

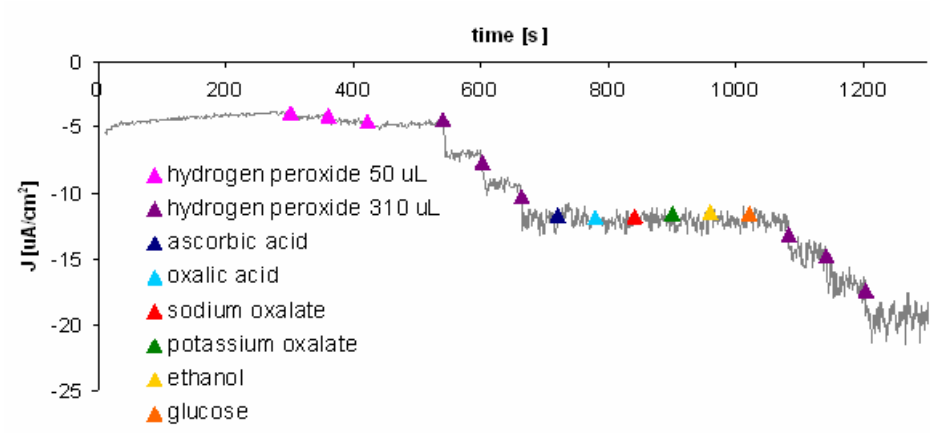

Figure 1. Amperometric response of the Ag nanowire-sensor to the addition of hydrogen peroxide and various interfering substances.

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8:50	Poster	B17

Optyczne urządzenie do leczenia niewielkich zranień

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Zastosowanie światła widzialnego na pograniczu fali niebiesko-fioletowej (405-460 nm) do leczenia niewielkich zranień ma duże znacznie dla gojenia się ran zwłaszcza u osób chorych na hemofilię czy zażywających leki antykoagulacyjne, a także u dzieci czy podczas interwencji chirurgicznych.

Podstawą leczenia niewielkich zranień jest fakt, że w tym zakresie długości fali światła widzialnego hemoglobina, która jest naturalną nanoczasteczką, absorbuje światło w większym stopniu, niż inne składniki obecne w tkance skóry jak np. melanina, co pozwala na selektywne dostarczanie energii optycznej wprost do krwi. Zgromadzona w ten sposób energia wywołuje wzrost temperatury krwi a przy przekroczeniu pewnego poziomu dostarczonej energii występuje proces

koagulacji.

Prace opublikowane w literaturze światowej pozwalają przypuszczać, że efektem leczenia światłem niebiesko-fioletowym jest przyspieszenie gojenia się ran w wyniku szybkiego zamknięcia naczyń krwionośnych. Cały proces leczenia jest bezpieczny dla zdrowia pacjenta [1,2], a eksperymentalnie wykazano przydatność takiego postępowania np. przy ekstrakcji zębów [3].

Pozytywne efekty wywołane w tkance biologicznej przez promieniowanie niebiesko - fioletowe umożliwiło podjęcie prac nad tanimi urządzeniami, które wykorzystują źródło światła diody elektroluminescencyjnej [4-6].

Firma SKA-Polska zaprojektowała i dostarczyła urządzenie optyczne do powszechnego użytku, a także do precyzyjnego naświetlania trudno dostępnych miejsc. Obie wersje urządzenia zostały zbudowane w ramach projektu Light+ter (nr projektu FP7-SME-2008-1-232397).

Firma SKA-Polska była odpowiedzialna w tym projekcie za układy zasilania diody oraz sterowania urządzeniem. Podjęła także prace badawcze nad samym urządzeniem i wywoływanymi przez nie efektami.

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8:50	Poster	B18

Importance and trends in nanomedicine according European Nanomedicine Platform survey

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The definition of nanomedicine will be given and its strategic research agenda according to the European Nanomedicine Platform. Some data on the nanomedcine market shares and dynamic advance will be given. Chances and challenges will be shortly discussed.

8:50	Poster	B19

Layersome systems, used in control delivery, stabilized by polyelectrolytes

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This paper is a presentation of our results from studies based on a development of novel liposomal nanocarriers which can be applied in delivery of many biologically substances such as proteins or antiarrhythmic drugs like phenytoin. There is a appreciable interest in liposomal carriers which could be used for medical and pharmacological applications. Because of low stability of liposomes these applications are challenging. Some external factors such as pH, temperature or other substances present in an environment may have damaging effect on the liposomes.

Proteins are known to play very important role in functioning of living organisms. They have mechanical-structural functions but also they can act as biocatalysts (enzymes), neurotransmitters or can be involved in enabling transport of small molecules and ions in tissue. Alkaline phosphatase [ALP], belonging to the class of hydrolases, was choosen as model protein. ALP is responsible for the dephosphorylation processes of various compounds (including proteins). The highest activity of ALP is strictly related to high pH of solutions. The main sources of alkaline phosphatase are: liver, bone tissues, kidneys or placenta. Due to this fact, ALP can be used as a biomarker. That's why it can be used in diagnostic for some diseases. Moreover, alkaline phosphatase is related to biomineralization process.

Other kind of encapsulated drug was phenytoin. It was used as a model drug which can be used for reducting the electrical conductivity of the brain cells. But one of the most important application is being an antiarrhythmic agent. Due to low solubility in agueous media and high toxicity, phenytoin is delivered into liposomal bilayer.

Because of low stability of liposomes, they were coated by derivatives of poli(vinyl alcohol) [PVA]. It was choosen due to good solubility in water, high resistance to organic solvents and wide use in cosmetic or pharmaceutical applications. We obtained the hydrophobic and ionic derivatives of PVA to use them later to coat the outer surface of vesicles.

Acknowledges:

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8:50	Poster	B20

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- facilitate development of innovative technologies,
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8:50 Poster B21

Selective adsorbents of adenosine-5'-triphosphate

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The purpose of this studies was the synthesis and characterization of hybrid polymeric-inorganic materials which can be used for selective adsorption and sensing of adenine-containing compounds, in particular adenosine triphosphate (ATP), which is a biomarker for neutrophilic airway inflammation and ischemia and may have considerable potential in the management of ischemic heart disease and stroke.

Silica gel particles were coated with a polyanion bearing thymine chromophores. The polymer coated particles were found to selectively adsorb adenine and ATP, as compared to other nucleobases and nucleotides, respectively. The adsorption was enhanced by the irradiation of the particles in the presence of adenine which resulted in the molecular imprinting of adenine and improved selectivity vs. other nucleobases and nucleotides. It was found that ATP adsorption was strongly pH-dependent.

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8:50 Poster B22

Surface modification of PCL scaffold changes protein adsorption

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Biomaterial surface plays a key role in the cell-material interaction and adsorption of proteins. However, the processes occurring at the implant interface are still not fully understood. The role of biomolecules and their influence on initial adhesion has received little attention. There are several stages occurring on the surface after implantation into biological environment: adsorption of proteins, cell adhesion and proliferation and differentiation of cells. Knowledge of the parameters characterizing the surface of material (surface topography, chemical composition, surface energy)

can be used for the selective adsorption of proteins (and cell) on the surface of the biomaterial. Surface chemistry and topography are known as two of the most important factors affecting biological reactions. Ideal biomaterials should encourage good cell-substrate interaction and stimulate the cells growth. The surface characteristics, such as chemical composition, wettability, ionic charge, texture and topography can be modified using different surface modification methods (chemical, physical and radiation). Modifications are intended to improve wettability, charge, ions and reactive groups (such as –COOH and -NH2) distribution. It has been reported that cellular adhesion and proliferation are closely dependent on the topographical nature of the biomaterial surface.

In this study chemical modification of the surface was conducted by NaOH treatment under varying time intervals. Results from our in vitro study NaOH treatment causes increase hydrophilicity and roughness of topography which can have desirable effect on protein adsorption. The proteins adhesions (albumin and fibronectin) were measured using AFM technique.

This work demonstrates simply method of surface modification enables changes in surface properties which have influence on protein adsorption.

8:50	Poster	B23

Use of aliphatic polyesters and poly(ester-carbonate)s for the controlled release of paclitaxel

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Paclitaxel (PACL) is one of the most widely used anti-cancer chemotherapeutic drugs. It`s usually applied to treat patients with lung, ovarian, breast, head and neck cancers. However, therapeutics efficacy and safety of drugs administered by conventional methods is limited. In the recent years, considerable attention has been focused on the research of the polymeric drug delivery systems. Polymeric conjugates have been extensively studied and applied as promising delivery systems to augment therapeutic efficacy of chemotherapeutic agents in the treatment of cancer. Some problems, however, such as allergic reactions, heart and blood vessel effects and infections have limited their use due to a low white blood cell count, hair loss, joint and muscle pain, irritation at the injection site, a low red blood cell count, mouth or lip sores, numbness, tingling or burning in the hands and/or feet, stomach upset and diarrhea as well as decrease in urine output and/or swelling of the hands, face or feet [1-3].

The aim of the present study is to synthesize polyester-PACL and poly(ester-carbonate)-PACL conjugateswith different molecular weights and characterization of their physicochemical properties. The degree of degradation of the macromolecular conjugates was tested *in vitro* under different conditions.

We believe that the obtained polyester and poly(ester-carbonate) conjugates of PACL can find practical applications as effective implantation drug delivery systems.

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The work was supported by the research program (ProjectMNiSzW-0451/B/H03/2010/39, N N209 045139) oftheMinistry of Science and Higher Educationin Poland.

8:50	Poster	B24

Atomic force microscopy characterization of corneocytes

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Atomic force microscopy (AFM) is a novel technique for *ex vivo* analysis of epidermal stratum corneum cells (corneocytes). In this study, AFM was used to evaluate topography, elasticity and adhesion of corneocytes before and after treatment with moisturizer. Cream was applied daily on the forearm for a period of 5 days. The material for test was collected from the skin surface of volunteers using Cuderm tape strips before and after the treatment. First, changes in the morphology of the cells were evaluated. It was found that corneocyte surface was smoother after the application of the cosmetic product. The analysis also included Young's modulus measurements. Young's modulus is a measure of elasticity (flexibility) of the tested material. The study results prove the existence of statistically significant differences in the mechanical properties of corneocytes taken after cosmetic treatment compared to the control sample. Control corneocytes were more rigid than corneocytes taken after the 5-day cosmetic treatment. The measurements of adhesive forces was based on the estimation of the force needed to detach the AFM tip from the corneocyte surface. The results revealed a relatively wide distribution of adhesive forces in similar way to Young's modulus values. A bigger fraction characterised by stronger tip-surface interactions was recorded for samples taken after the cosmetic treatment. The AFM analysis of corneocytes taken by *tape-stripping* proved that they became smoother, less rigid, with stronger adhesive bonds between them after the cosmetic treatment. In conclusion, AFM can be used as a very sensitive tool for early detection of changes in corneocytes after moisturizer usage.

8:50	Poster	B25

Zastosowanie układów mikroprzepływowych w izolacji nasienia

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Preparatyka nasienia jest ważnym elementem technik wspomaganego rozrodu (ang. assisted reproductive technology, ART). Izolacja plemników z nasienia ma na celu oddzielenie plemników od plazmy nasienia, usunięcie substancji hamujących zdolność do zapłodnienia, zapoczątkowanie procesów kapacytacji ułatwiających zapłodnienie komórki jajowej oraz selekcję najbardziej prawidłowych plemników na podstawie uznanych parametrów – na przykład prawidłowej morfologii i ruchliwości.

Pomimo znacznego wzrostu zainteresowania tymi technikami, obecnie stosowana metodologia tego procesu pozostaje niezmienna. Obecnie do izolacji plemników najczęściej stosuje się separację na nieciągłym gradiencie podłoża bądź migrację wstępującą (ang. swim up). Metody te jednak nie mogą być zastosowane w przypadkach próbek o bardzo małej koncentracji plemników . Niektóre badania sugerują również, że techniki te mogą uszkodzić DNA także poprzez produkcję wolnych rodników tlenowych. Ponadto są one praco i/lub czasochłonne. Idealna technika izolacja plemników powinna być zatem szybka, prosta i atraumatyczna, dając jednocześnie dobre wyniki bez względu na ilość i jakość pierwotnej próbki nasienia. Poprawa jakości technik separacyjnych i izolacyjnych powinna zwiększyć skuteczność ART.

Coraz wyraźniej widoczne są zatem tendencje zmierzające do poprawy jakości technik separacyjnych np. poprzez zastosowanie układów mikroprzepływowych. Układy tego typu testowane w naszych badaniach stworzone zostały z zastosowaniem metody fotolitografii. Składają się one z płaskiego szklanego podłoża, które za pomocą plazmy tlenowej połączone zostało z płytką poli(dimetylosiloksanową) (PDMS) zawierającą system rozgałęzionych mikrokanałów. W mikrokanałach przepływ cieczy jest laminarny skutkując mieszaniem się płynów ze strumieni tylko na zasadzie dyfuzji. Dodatkową zaletą tego układu jest to, że przepływ spowodowany jest siłą grawitacji, co eliminuje konieczność stosowania różnego rodzaju pomp i tłoków.

Prowadzone są badania nad skutecznością systemu umożliwiającego separację ruchliwych i morfologicznie prawidłowych plemników z nasienia nawet w próbkach o bardzo słabych parametrach jakościowych. W przypadku pozytywnych wyników badań testowych, prezentowana metoda separacji może być doskonałą alternatywą dla tradycyjnie stosowanych metod separacji nasienia.

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8:50	Poster	B26

Antybakteryjna folia wykorzystujaca nano-srebro z certyfikartami europejskimi dla zastosowań medycznych i w przemyśle sporzywczym

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Do uzupełnienia

8:50	Poster	B27

Emulsion-core and (bio)polyelectrolyte shell nanocapsules for neuroprotective drug delivery

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Nanocapsules are colloidal systems (r < 100nm), composed of either solid or liquid core surrounded by a polymer membrane (shell). Emulsion-core nanocapsules provide good environment for encapsulation, protection and delivery of poorly water soluble drugs improving their in vivo bioavailability and efficiency while reducing undesirable side effects. Therefore, encapsulation of therapeutic agents inside biocompatible nanocarriers is among the major research topics in nanopharmacology and nanomedicine.

Biocompatibility of nanoproducts is the principial prerequisite for nanomedicine applications, therefore, the aim of this work was to synthesize polyelectrolyte-coated nanocapsules with emulsion cores by the layer-by-layer (LbL) technique, as described by Szczepanowicz et al [1],[2], and to examine the biocompatibility of synthesized nanocapsules with human neuroblastoma SH-SY5Y cells. Nanoemulsions were stabilized by polyelectrolyte-surfactant complexes withsuitable surface properties. Chemicals used in nanocapsules preparation were docusate sodium salt (AOT), isopropyl myristate (IPM), chloroform (HCCl3) and various polyelectrolytes: PLL – poly(L-lysine hydrobromide), PAH – poly(allylamine hydrochloride), PDADMAC – poly(diallyl dimethyl ammonium chloride)], PGA – poly(L-glutamic acid) sodium salt, ALG – alginic acid sodium salt, CHI – chitosan, PSS – poly(sodium 4-styrenesulfonate). Prepared nanocapsules were characterized for particle size and electrical charge of successive layers adsorbed on the emulsion core by measuring their hydrodynamic diameter (DLS technique) and zeta potential (LDE technique), respectively. The biocompatibility of synthesized nanocapsules on human neuroblastoma SH-SY5Y cells using biochemical assays (MTT, LDH) was examined. Nanocapsules were tested in various concentrations and for different times of incubation.

The results obtained show that desired concentrations of several nanocapsules are non-toxic to SH-SY5Y cells and they can be used as drug-loaded nanocarriers. As the next step, the neuroprotective potential of nanoencapsulated model drugs will be evaluated against oxidative stress cytotoxity (e.g. H_2O_2 induced) in the same cell culture.

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8:50	Poster	B28

Polysiloxanes - based multilayer nanocoatings

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Polysiloxanes are biocompatible polymers with very high flexibility of the chains that may be very important for the formation of the functional coatings. Especially modified polysiloxanes, which are soluble in water and have low glass transition temperatures may be of high interests. They may be used for the formation of flexible coatings of biological relevance as well as for the fabrication of nano(micro)capsules that may serve as containers for drug delivery or photoreactors for conversion of sunlight energy.

This work presents synthesis and characterization of novel charged polysiloxanes and their usage for the formation of ultrathin multilayer films on model surfaces using “layer-by-layer” approach. The obtained films were characterized using Atomic Force Microscopy and Spectroscopic Ellipsometry. The films will be further used as functional coatings for biomedical and photochemical applications.

The authors would like to thank Polish Ministry of Science and Higher Education for the financial support (“Ideas Plus” grant no. IdP2011 000561).

8:50	Poster	B29

Wytwarzanie preparatu enzymatycznego przydatnego do otrzymywania nanocelulozy.

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Nanostruktury uzyskiwane z materiałów ligninocelulozowych stały się jednym z bardziej pożądanych komponentów biokompozytów. Bardzo duże zainteresowanie nanocelulozą, zwłaszcza przemysłu rolno-spożywczego i papierniczego, wymusza opracowanie szybkich i wydajnych metod rafinacji biomasy roślinnej. Obecnie nanowłókna celulozy otrzymywane są głównie metodami fizyko-chemicznymi, które generują stosunkowo dużą ilość uciążliwych dla środowiska odpadów. Wymusza to konieczność opracowania nowych, ekologicznych metod konwersji biomasy, którymi mogą być metody enzymatyczne [1].

Biorafinacja materiałów ligninocelulozowych wymaga zastosowania kilku enzymów o różnej specyficzności substratowej, wśród których jedne są odpowiedzialne za oczyszczenie struktury celulozy (ligninazy, pektynazy) a inne za jej kontrolowany rozkład do nanowłókien (ksylanazy i celulazy) [2]. Szczególnie ważną rolę w finalnym procesie otrzymy-

wania nanocelulozy odgrywa endo-1,4-β-glukanaza hydrolizująca celulozę w jej amorficznych obszarach. Niestety biosynteza tego enzymu w warunkach hodowli mikroorganizmów jest zazwyczaj ściśle powiązana z biosyntezą β-glukozydazy, której aktywność skutkuje rozkładem nanostruktur celulozy
do pojedynczych cząsteczek glukozy.

Celem projektu jest wytworzenie preparatu celulaz przydatnego
do ukierunkowanej na otrzymywanie nanocząstek celulozy, biorafinacji biomasy (słomy lnianej i konopnej), w oparciu o hodowle solid-state pleśni *Aspergillus niger* [3,4]. W ramach badań zoptymalizowano warunki hodowli (w tym sposób wyjaławiania pożywki), dobrano metodę ekstrakcji białek enzymatycznych
i częściowego ich oczyszczenia. Stwierdzono pozytywny wpływ ozonowania podłoża na stosunek aktywności endo-1,4-β-glukanazy i β-glukozydazy w uzyskiwanym preparacie enzymatycznym.

Badania są finansowane z projektu 01.01.02-10-123/09 współfinansowanego
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8:50	Poster	B30

Chitosan-Silver Nanocomposites: synthesis, characterization and antibacterial activity against Staphylococcus aureus

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Nowadays, there is a great interest in development of bionanomaterials exhibiting antibacterial activity against multi-drug resistant bacterial strains. Appearance of antibiotic resistant ones has moved the researchers attention to searching for new alternative bactericidal agents, which could possibly replace so popular antibiotics. Despite the discovery of a various active agents, very few exhibit desirable properties as biocompatibility, biodegradability and non-toxicity. Among a number of nanotechnological strategies, fabrication of polymer-metal nanoparticles bionanocomposites draws a lot of attention[1, 2].

The present investigation concerns chitosan-silver nanocomposites fabrication, physicochemical

characterisation and their antibacterial activity evaluation against two Gram-positive, *biofilm forming* bacterial strains *Staphylococcus aureus.*

Chitosan, biocompatible and biodegradable polymer was used as a reducing and stabilizing agent in the optimized silver nanoparticles synthesis. Nanoparticles and nanocomposites were characterized by UV-vis, IR, TGA, SEM, TEM, *etc*. techniques. Obtained nanoparticles, trapped in the polymeric network showed high antibacterial activity against biofilm forming *S. aureus* and caused significant morphological changes in the bacterial cell walls (Fig.1).

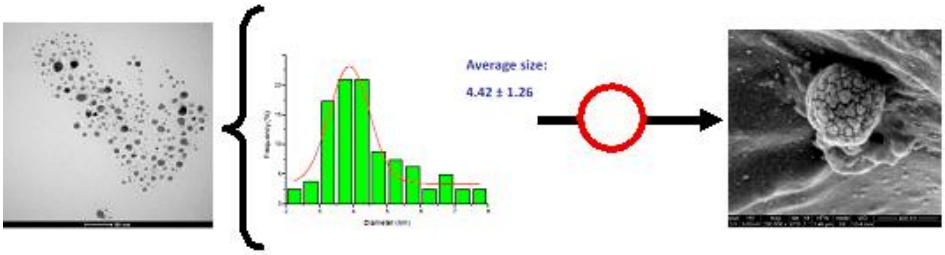

Fig.1. Chitosan based silver nanoparticles (<10 nm), revealing high bactericidal activity against *S. aureus*.

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8:50	Poster	B31

TiO_2 nanotubes based composite layers for biomedical applications

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Titanium when used in orthopaedic devices and oral implants was considered to be inert. However, recently, it has been shown that it can be toxic or induce allergic reactions. It is possible for the biological properties of titanium to be improved by the application of appropriate surface treatments. The aim of this was to develop methods to modify the surface of titanium, which would allow precise control of its morphology and chemical composition, and achieve beneficial biomedical properties. To achieve this objective a two-step procedure for surface modification was introduced. In the first step the Ti substrates were electrochemically pretreated in solutions containing fluoride. These processes produced specific surface morphology, namely nanotubular structure. In the second stage, biomimetic calcium phosphate (Ca-P) coatings were formed on the

nanoporous titanium oxide layer by immersion of the pretreated Ti in simulated body fluid (Hanks' solution). The results revealed TiO_2 nanotubes provided beneficial substrates for the deposition of a uniform Ca-P coating. The main component of coating was octacalcium phosphate, which is considered to be a precursor for the in vivo formation of hydroxyapatite. The preliminary results of the U2OS cells response to the modified surfaces indicate that the number of cells on the TiO_2 nanotubes possessing the deposited Ca-P coatings is distinctly higher than for an unmodified Ti surface. In addition, the albumin preadsorption on the Ca-P coating was shown to give further improvement of the adhesion of the osteoblast-like cells. In order to minimize the likelihood of biomaterial induced infections the TiO_2 nanotubes layers were loaded with silver nanoparticles using a sputter deposition technique. The presence of Ag nanoparticles on the TiO_2 nanotube layers led to a significant reduction in the adhesion of S. epidermidis cells and biofilm formation on the surfaces. Additionally, the Ag/TiO_2 nanotubes composite layers were shown to have a positive effect on the adhesion and differentiation of the osteoblast-like cells. The proposed titanium surface modification procedure may be a promising method for fabricating novel materials for internal applications so that components to meet the physical functional requirements maintain biocompatibility and, very importantly, prevent infection.

8:50	Poster	B32

Protoporphyrine IX and its PEI derivatives as possible photosensitizer in antibacterial photodynamic therapy

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The progressive increase in bacterial resistance to antibiotics makes it conducted a search for new treatments for bacterial infections. One of the new method is antibacterial photodynamic therapy. The possibility of its application depends on the availability of appropriate photosensitizers (PS), which will allow for efficient cell destruction of pathogens. Photosensitizers currently used in cancer therapy are generally not suitable for the treatment of bacterial infections. There is a particularly their ineffectiveness against infections caused by Gram (-). The main reason for this is specific to the construction of these pathogens. In contrast to Gram (+), they have an outer membrane composed primarily of lipopolysaccharides. It functions as an impermeable barrier (negatively charged), protecting against the effects of bacterial cells with a photosensitiser. Therefore, for effective treatment, it becomes necessary to use the PS are able to overcome this barrier, or early treatment of bacterial substances that destroy the structure of their outer membrane, such as EDTA or cationic polypeptides. An example of effective photosensitizers against both Gram (-) and Gram (+) proved to be conjugate of polyethyleneimine chlorin or chlorin with poly-L-lysine.

The present work is focused on attaching protoporphyrin (Pp) to the polymer chain (PEI).[1]
Polyethyleneimine (PEI) is a positively charged polymer that has gained recent attention as a transfection agent. PEI can easily interact with negative charged molecular bilayer.
Physicochemical and photophysical properties of this systems were determined.
The interaction of the polymeric photosensitizers with lipid membranes were next studied. Using fluorescence properties of the analyzed compounds, the kinetics of penetration to liposomal system, the distribution constant between aqueous and lipid phase of liposomes and fluorescence quenching of the porphyrin units by copper sulfate were determined. The influence of addition of

the PEI photosensitizers on stability of the liposomes was analyzed using turbidmetric titration with solution of Triton x-100. Using the measurement of dynamic light scattering, the size and zeta potencial of obtained systems were evaluated.

Acknowledges:
The study was conducted according to the TEAM Foundation for Polish Science, co-funded by European Regional Development Fund: PolyMed, TEAM/2008-2/6.

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8:50	Poster	B33

Synthesis and properties of up-converted $NaYF_4$: Er, Yb nanoparticles for biomedical applications

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We synthesized potential biosensors, based on Er, Yb doped $NaYF_4$ nanoparticles, with up-conversion of infrared light capable to emit in the visible and ultraviolet range. They could be potentially applied for targeted therapy in cancer, through up-conversion to the ultraviolet radiation. Biological systems excited by the near infrared energy exhibit less autofluorescence than if excited by the visible or the ultraviolet light. Visible emission permits to observe physiologically relevant processes within cells, and the emission of the ultraviolet light allows for the selective elimination of cancer cells by generating free radicals.

We were able to synthesize $NaYF_4$ nanoparticles doped with different amounts of rare earth ions, providing an efficient energy up-conversion. After functionalization by PVP or oleic acid, we were able to introduce these particles into HeLa cancer cells, where we examined their localization in the function of both, incubation time and particles concentration. We performed preliminary attempts to bio-functionalize nanoparticles for specific locations in the cancer cells.

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8:50	Poster	B34

Kontrola procesu wytwarzania nanocząstek srebra w NMMO

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N-tlenek N-metylomorfoliny (NMMO) jako bezpośredni rozpuszczalnik celulozy wykorzystywany jest do otrzymywania zarówno włókien jak i nanowłókien celulozowych [1]. Związek ten stosowany jest na skalę przemysłową do otrzymywania włókien celulozowych znanych pod nazwą Lyocell. Metoda ta charakteryzuje się prostym procesem technologicznym jest ekologiczna, a dodatkowym jej atutem jest możliwość otrzymywania modyfikowanych włókien o ściśle zaprogramowanych właściwościach poprzez wprowadzanie zarówno dodatków organicznych jak i nieorganicznych. Jednym ze stosowanych modyfikatorów jest nanosrebro, które pozwala na otrzymywanie włókien mających szerokie spektrum działania bioaktywnego. Proces generowania nanocząstek srebra może być kontrolowany poprzez wiele czynników, mających wpływ na ich podstawowe parametry (np. temperatura i czas syntezy, obecność czynników redukujących takich jak NMMO, czy celuloza) [2]. Do określenia wpływu zastosowanych warunków syntezy na wielkość i kształt nanocząstek srebra zamkniętych w materiale włókna posłużyła metoda, Dynamicznego Rozpraszania Światła (DLS) oraz metoda Transmisyjnej Mikroskopii Elektronowej (TEM). W prezentowanych wynikach badań skupiono się syntezie nanocząstek srebra bezpośrednio w NMMO. Oszacowano wpływ temperatury i czasu procesu na m.in. na wielkość i kształt znajdujących się w tworzywie włókna nanocząstek srebra. W prezentowanych badaniach skupiono się na włóknach bioaktywnych, w których zawartość srebra w matrycy polimerowej jest na poziomie 500ppm (0,05%). Na podstawie wcześniejszych badań stwierdzono bowiem, że jest to optymalne stężenie srebra we włóknach, które zapewnia dobre działanie antybakteryjne wobec bakterii *Escherichia coli* oraz *Staphylococcus aureus* [3].

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8:50	Poster	B35

Wpływ warunków generowania nanocząstek srebra na barwę włókien

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Od niepamiętnych czasów człowiek podejmował próby manipulowania materią. Próby te przeprowadzane najczęściej nieświadomie lub w oparciu o dość przypadkowe obserwacje czasami kończyły się sukcesem. Ciekawym przykładem może być puchar Likurga, w którym obecność nanocząstek wpływa na jego zaskakujące efekty barwne. Wraz z postępem nauki i technologii zaczęto badać, analizować i wyjaśniać pewne niewytłumaczalne wcześniej zjawiska jednocześnie

dążąc do kontrolowania wszelkich procesów. Szczegółowa wiedza dotycząca procesów i reakcji chemicznych, w coraz większym stopniu pozwala osiągnąć z góry oczekiwany rezultat, a potem powtórzyć go tyle razy ile tylko zapragniemy.

W prezentowanych wynikach badań pokazano możliwości kontrolowania barwy włókien celulozowych modyfikowanych nanocząstkami srebra generowanych w trakcie procesu wytwarzania roztworu przędzalniczego. Jedną z wielu zalet tej metody jest to, iż pozwala ona uzyskać włókna o szerokiej gamie kolorów, wykorzystując do tego celu tylko jeden rodzaj prekursora o danym stężeniu, a zmieniając jedynie parametry prowadzenia procesu syntezy nanocząstek srebra. Interesujące, głębokie i połyskujące w świetle efekty barwne otrzymanych włókien uzyskano dzięki połączeniu nanotechnologii z technologią otrzymywania włókien celulozowych typu Lyocell. W omawianych badaniach nanocząstki srebra generowano w różnych warunkach (np. w świetle słonecznym, w ciemni, bezpośrednio w NMMO lub bezpośrednio na celulozie) i na różnych etapach otrzymywania roztworu przędzalniczego. Do oceny barwy otrzymanych włókien wykorzystano Spectroreflectometr Mackbeth Color Eye 200. Na podstawie uzyskanych wyników stwierdzono, że barwa włókien jest w ściśle uzależniona od zastosowanych warunków reakcji syntezy nanocząstek srebra i w efekcie od wielkości i kształtu ziaren srebra.

Głównym celem badań w tym obszarze jest możliwość przyszłego programowania i kontroli barwy włókien. Otrzymane włókna celulozowe, ze względu na ich właściwości bakteriobójcze będą mogły znaleźć zastosowanie głównie w medycynie jako materiały opatrunkowe i odzież ochronna. Unikatowe walory estetyczne włókien, których nie można uzyskać na drodze tradycyjnych metod barwienia pozwolą również na ich wykorzystanie między innymi do celów dekoracyjnych i odzieżowych.

8:50	Poster	B36

Synthesis of highly biocompatible hydroxyapatite nanopowders

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Today, the most common approach for regrowth of bone in case of large bone defects is to use autografts. It is providing the primary factors for effective bone regeneration but this technique requires two or more operations which is increasing risk for patient and is impossible to use in case of large, complicated bone gaps. There is also an emerging market for bioactive ceramics like hydroxyapatite (HAp), which is used in form of paste and granules to fill small bone defects. For large bone gaps regrowing there are still unsolved barriers like: low regrowth rate, poor mechanical properties, high risk of inflammantory and low resorption rate. Therfore the main objectives of the current regeneration medicine projects is to developed the technology for bioactive scaffold with improved, comparing to the state of the art, control of shape, mechanical properties, bioactivity and resorbability. One of the ways to achieve these goal is to produce nonstoichiometric nano-

particles of hydroxyapatite with grain size lower than 10nm and shape close to the natural HAp which will be used as a material for bioactive, mechanically strong scaffolds. Such nanoparticles due to their calcium deficiency and high surface to volume ratio may achieved necessary solubility level and increased osteoblasts adhesion.

The Institute of High Pressure Physics of the Polish Academy of Science (IHPP) is an expert in synthesis of doped nanoparticles with narrow size distribution, at relatively low temperatures by using Microwave Solvothermal Synthesis (MSS) technology. The MSS technology permits synthesis of nanoparticles with precise control of reaction time, temperature and pressure. IHPP and the Faculty of Materials Engineering, Wasrsaw University of Technology, created a joint Center for Biomaterials and started cooperation to developed materials for bone resorable scaffolds.

Thanks to unique MSS technology for nanoparticles synthesis, IHPP is able to synthesize unque HAp nanoparticles using the standard reaction between calcium hydroxide and phophoric acid. The reaction is carried out in water solution in time lower than 5 minutes. The specific surface area is almost $240m^2/g$ the average grain size 6nm (basing on TEM investigations) with shape in the form of plates mimicking natural bone particles. 28 days of degradation test conducted according to norm ISO 10993-14 indicated material solubility equal $20mg/dm^3$.
Research subject was carried out with the use of CePT infrastructure financed by the European Union – the European Regional development Fund within the Operational Programme “Innovative Economy” for 2007-2013

8:50	Poster	B37

Transmission Electron Microscopy characterization of gold nanoparticles

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The interest in noble metals particles like gold nanoparticles is caused by wide possibilities of applications in areas such as nanomedicine, nanophotonics or the protection of the natural environment. Nanoparticles of noble metals show the phenomenon of Localized Surface Plasmon Resonance (LSPR). This phenomenon is related to the oscillation of free electrons in the metal and therefore depends on nanoparticles size [1]. The LSPR has been used for the ablation of cancer cells - Plasmonic Photothermal Therapy (PPTT) [2]. The electromagnetic radiation of frequency corresponding to the resonance, excited localized plasmons which leads to a local overheating and damage cancer cells. This technique is minimally invasive method for healthy cells, but very effective in the destruction of cancer cells.

The most spectacular application of gold particles is to use them to generate the bioluminescence of plants, so far from the family *Bacopa caroliniana* [3]. Leaves of plants in which gold particles (~ 10 nm) were placed can light in the dark. The bioluminescence, induced by the presence of gold particles, can be further used for the production of energy efficient lighting such as tree in city streets.

Transmission Electron Microscopy (TEM) gives valuable information for nanotechnology. The paper presents results of investigations of gold colloids containing particles of different sizes (2 nm - 50 nm). The knowledge of the size and shape of gold nanoparticles is very important because have a significant influence on the optical properties. In the paper we determined the crystal struc-

ture and analyzed the size of nanoparticles. TEM methods and especially one of them: Energy Electon Loss Spectroscopy (EELS) allows to collect spectra from individual particle, which can not be done by optical methods.

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8:50	Poster	B38

Polylactic acid - calcium phosphates composites

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Bioactive ceramics such as hydroxyapatite (HAp) and tricalcium phosphate (TCP) are one of the most popular implant materials in the reconstructive surgery to repair damaged hard tissues. Calcium phosphates (CaP) have excellent biomaterial properties due to their similarity to the inorganic component of the bone matrix. However, CaP clinical application is restricted due to their inherent brittleness and poor shape ability. To capitalize the advantage of CaP and overcome some drawbacks, CaP is combined with polylactic acid to generate highly porous biocomposite materials. Polylactic acid (PLA) has good mechanical properties and low toxicity.

CaP – PLA composites have received a great deal of interest in orthopedic and dental applications, which is attributed to their good osteoconductivity, biodegradability and high mechanical strength.

In this study CaP powder was prepared by wet chemical precipitation method from calcium hydroxide suspension and ortophosphoric acid solution. Multiple factors like suspension temperature, pH, acid addition rate and mixing determine phase composition of the product. Lower temperatures and acidic pH values promote formation of β-TCP phase, but slightly alkaline pH and higher temperatures promote formation of HAp phase. It is possible to obtain calcium phosphate product with diverse phase composition, by varying these parameters

There is a problem with homogenization of CaP with the PLA due to different chemical characteristics of hydrophilic CaP and hydrophobic PLA. Organic solvents normally used for dissolving PLA could not be used to effectively disperse the CaP powder. The aim of this study was to resolve homogenization and intermixing problem raised on the ceramic – polymer biomedical composite system.

Fourier-transform infrared spectroscopy (FT-IR) is used to determine the characteristic functional groups in the sintered calcium phosphate samples. X−ray diffractometry (XRD) is used to analyze

phase composition of obtained bioceramics samples. Scanning electron microscopy (SEM) is used to characterize the morphology of the obtained samples.

8:50	Poster	B39

Polymeric Nanocapsules for Controlled Delivery of Lipophilic Drugs

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Development of the techniques of designing and production of controlled drug delivery systems represents one of the most important area of contemporary science. Traditional drug carriers are ineffective because of partial degradation and toxic as a consequence of lack of control over concentration. It makes they are expensive, often uncomfortable for patients and cause side effects thus need to be replaced by more effective, cheaper and better defined systems.

The ideal drug carrier should be inert, biocompatible, biodegradable, non-toxic, mechanically strong, easy to fabricate, efficient in loading and release of drugs and simple to administer. Polymers satisfy this requirements and are believed to be sufficient materials for biomedical applications. There are many different types of polymeric materials suitable for fabrication of drug delivery systems, such as nanoparticles, nanocapsules, brushes, graft and block polymers.

Amphiphilic polyelectrolytes attract attention due to their wide range of practical applications. The presence of hydrophilic groups makes them soluble in water while presence of hydrophobic groups leads to spontaneous aggregation of polymer in aqueous solution which causes formation of hydrophobic domains in which poorly soluble organic compounds can be solubilized. It makes such systems can be used as nanoreactors or nanocontainers, e.g. drug carriers.

The aim of my research is to synthesize and characterize amphiphilic polyelectrolytes which enable encapsulation of liphophilic drugs and their controlled delivery. Nanocapsules are created via “layer by layer” technique which relies on consecutively alternating adsorption of oppositely charged polyelectrolytes and leads to formation of multilayer walls. By using controlled radical polymerization techniques it is possible to control molecular weight and architecture of synthesized polymers. Stability of capsules depends on both of that features hence possibility of control of them is crucial for biomedical applications where it is important to have stable drug carriers.

Presented results include both, experimental and theoretical characterization of the model system. Experimental studies include spectroscopic methods (UV/VIS, IR), measurements of dynamic light scattering and optical microscopy Theoretical calculations include quantum-chemical Born-Oppenheimer molecular dynamics on the semiempirical level of theory (as implemented in MSINDO software package).

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8:50	Poster	B40

1-methylnicotinamide (MNA) reduces inflammatory responses in human endothelial cells as revealed by nanoindentation spectroscopy using an AFM tip

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1-Methylnicotinamide (MNA), previously considered to be a biologically inactive metabolite of nicotinamide (NA), has been recently presented as a potential anti-inflammatory agent. It has been suggested that MNA may regulate the human endothelium cell inflammatory response processes caused by external agents, such as tumor necrosis factor alpha (TNF-α). For example, the incubation of endothelial cells with TNF-α (10 ng/ml) for 1 h causes a significant increase in cell stiffness and a reduction in nitric oxide NO production. In the presented work, the anti-inflammatory potential of MNA (100 nM) was investigated using an in vitro model of endothelial cell dysfunction induced by incubation with TNF-α (10 ng/ml). Nanoindentation spectroscopy with an AFM tip, F-actin fluorescent staining and the measurement of NO production and PGI2 release were used to characterize the changes in the endothelial cell phenotype influenced by TNF-α and MNA. Therapeutic and protective effects of MNA against inflammation of the endothelium in vitro were observed by the return of measured parameters (elasticity, NO production) to the reference level. The measurements confirmed that MNA acts as an anti-inflammatory agent for endothelial cells, and this result was consistent across all 4 complementary methods used. The results using our in vitro model demonstrate that MNA can serve both as a preventive and therapeutic compound. Acknowledgements MNA has been kindly provided by Prof. Jerzy Gebicki, Technical University of Lodz. Numerous discussions with Prof. Stefan Chlopicki are gratefully acknowledged. This work was supported by the European Union from the resources of the European Regional Development Fund under the Innovative Economy Program (grant coordinated by JCET-UJ, No POIG.01.01.02-00-069/09)

8:50	Poster	B41

Effect of silver nanoparticles on human epidermal keratinocytes

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Nanoparticles are gaining much importance for biological application in biomedicine, biotechnology and other life sciences. For their unique physicochemical properties they are used in molecular diagnostics, cancer therapy, as well as drug and gene delivery systems. Among all nanoparticles, silver ones (AgNPs) have the highest degree of commercialization. For their strong antimicrobial, antiviral and antifungal activity, they have been used extensively in a range of medical settings especially in wound dressings. Nanoparticles are defined as structures that have at least one dimension in 1-100 nm range. Their ultra-small size in comparison to enormous surface area,

makes AgNPs very reactive forms. Small size also affirms grate particles mobility and has impact on their cellular distribution. Depending on the size, shape and the type of a carrier, AgNPs demonstrate different physicochemical properties. This study was undertaken to examine the potential toxicity effects of 15 nm PVP-coated AgNPs on Normal Human Primary Keratinocytes (NHEK). Cells were treated with different concentrations of AgNPs and then cell viability, morphology and metabolic activity were studied. To determined a role of AgNPs in signal transduction pathway, we examined an activation of several proteins involved in cell response to extracellular factors. We observed that 15 nm PVP-coated AgNPs decreased cell viability, metabolic activity, as well as they caused inhibition of cells proliferation and migration. Moreover, they induced DNA damage and increased caspase 3/7 activity. AgNPs significantly activated p38 MAPK, as well as caused weak activation of Erk 1/2 kinase and p53 protein. AgNPs increased level of mRNA coding for pro-apoptotic proteins from Bcl-2 family, as Bax and Puma. Used nanoparticles have a toxic impact on NHEK cells and they are acting in time- and/or dose-dependent manner. Our studies show, that despite the positive effects, AgNPs may present possible danger, concerning activation of genotoxic and cytotoxic processes.

8:50	Poster	B42

Superparamagnetyczne nanocząstki tlenku żelaza opłaszczane pochodnymi chitozanu – nowe kontrasty do MRI

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W ostatnich latach obserwuje się ogromny wzrost zainteresowania tematyką superparamagnetycznych nanocząstek tlenku żelaza (SPION). Wynika to głównie z faktu bardzo szerokiego potencjalnego ich zastosowania, w szczególności w zaawansowanej diagnostyce i terapii przeciwnowotworowej. SPION mogą być wykorzystane jako nowoczesne kontrasty w technice rezonansu magnetycznego, w leczeniu hipertermią, w celowym dostarczaniu leków czy detoksykacji organizmu [1,2,3].

Naszym głównym celem było otrzymanie i charakterystyka biokompatybilnych SPION o określonym rozmiarze oraz właściwościach magnetycznych odpowiednich do zastosowania nanocząstek w technice obrazowania MR. Dodatkowo przeanalizowaliśmy wpływ różnych warunków syntezy (temperatura, zastosowanie ultradźwięków, ilość wykorzystanego polimeru itp.) na właściwości otrzymanych SPION. Na podstawie przeprowadzonych dotychczas pomiarów zaobserwowano, iż jednoetapowa synteza z równoczesnym pokryciem nanocząstek biokompatybilną, kationową pochodną chitozanu pozwala na otrzymanie nanocząstek o najlepszych parametrach.

Głównymi technikami pomiarowymi wykorzystanymi w trakcie badań była ocena wielkości, ładunku oraz stabilności nanocząstek za pomocą dynamicznego rozpraszania światła, a także transmisyjna mikroskopia elektronowa potwierdzająca wielkość pojedynczych cząstek oraz ich strukturę krystalograficzną. Magnetyczne właściwości zbadano przy wykorzystaniu magnetometru z wibrującą próbką natomiast zawartość polimeru otaczającego nanocząstki poprzez analizę termograwimetryczną.

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8:50	Poster	B43

Stiffness changes of larynx carcinoma HEp2 cells correlates with the inhibition and release of TRAIL-induced apoptosis pathways sensitized by Actinomycin-D

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In this work, atomic force microscopy (AFM) based techniques (imaging and measurements of cell stiffness) and fluorescence staining were used to monitor apoptosis in larynx carcinoma HEp2 cells. HEp2 were exposed to TRAIL (TNF-related apoptosis inducing ligand), Actinomycin-D (a chemotherapeutic drug) or to a simultaneous action of these agents. For incubation with TRAIL, an increase in the stiffness of the cells was observed and correlated with the rearrangement of cell cytoskeleton, particularly actin fibers. However, the detection of apoptotic cells using fluorescence markers revealed the resistance of HEp2 cells to TRAIL-mediated apoptosis. Most probably, TRAIL initiates apoptosis in HEp2 cells, but the apoptosis process was terminated by anti-apoptotic agents. Unblocking of apoptosis by sensitization of HEp2 cells with a chemoterapeutic drug Actinomycin-D is related to the depolymerization of F-actin with the decrease in the cell stiffness. Most probably, the depolymerization of actin results from downregulation of Rho protein, which in turn is accompanied by a lower activity of Bcl-2 and in consequence releases the intrinsic apoptotic channel. The presented results reveal a promising application of nanoindentaion spectroscopy with AFM tip as a novel tool for monitoring the apoptosis inhibition processes.

8:50	Poster	B44

Novel nanostructural photosensitizers for photodynamic therapy

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Photodynamic therapy is currently used as a popular form of photochemotherapy for a wide

variety of clinical applications. This treatment requires the presence of a drug, called photosensitizer, activating light of a specific wavelength and molecular oxygen to produce a localized damage in tumor tissues and/or their vasculature. Many efforts have been undertaken to develop new agents with a high efficacy for photodynamic treatment of cancer and some other diseases. Porphyrins and their derivatives are studied as potential sensitizers because of their high anti-cancer efficiency. The factors limiting the use of porphyrins in PDT are their poor solubility in water, low chemical purity and high dark toxicity [1].

This paper presents the results of our studies on the development of the efficient nanoformulations of porphyrin for PDT. In the first step three PEG-functionalized porphyrins (p-THPP-PEG_{350}, p-THPP-PEG_{2000} and p-THPP-PEG_{5000}) were synthesized by covalent attachment of PEG chains of various molecular weights (350, 2000 and 5000 Da) to commercially available 5,10,15,20-tetrakis(4-hydroxyphenyl)porphyrin (p-THPP). To improve their ability to penetrate the hydrophobic cell membrane, the conjugates were solubilized in liposome vesicles.

The usefulness of novel hybrid systems as photosensitizers in photodynamic therapy was tested in vitro. Dark and photo cytotoxicity of these photosensitizers delivered in solution or embedded in liposomes were evaluated on two cell lines: HCT 116 and DU 145 and compared with these treated with free p-THPP. The kinetics of cellular uptake and the intracellular co-localization of p-THPP-PEG_{2000} formulations (which exhibit the most significant photodynamic activity) were studied by means of flow cytometric analysis and confocal laser scanning microscopy with the application of properly chosen fluorescence markers. Additionally, the time course and the mechanism of cell death after PDT were determined [2].

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8:50	Poster	B45

Chitosan in a new and innovative approach of metal nanoparticles synthesis for biomedical applications

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There is a need for the development of procedures for the atmospheric synthesis of metal nanoparticles (NPs) that remain stable for sufficiently long time, especially in biomedical field i.e. antitumor, antimicrobial and antifungal applications. In this regard, substantial attention is paid to creating conditions, where metal nanoparticles (Ag, Au, Cu, Sn etc.) could be formed with desired and controlled size, shape and structure. Silver has been regarded as a versatile healing tool and been used since ancient times to control infections. In recent times, colloidal Ag has been proposed for antimicrobial [1] and anticancer therapy [2]. However, the use of Ag-based nanomaterials receives currently a lot of attention since of their widespread and diverse applications. Particularly, the potential adverse effects of Ag NPs on humans and environment are under a lot of scrutiny. Copper could be an excellent replacement of silver in biomedical applications. Importantly, Cu is known to have significant antibacterial and antifungal properties [3, 4]. Furthermore, there are described sequestering systems for Cu releasing out of the body [4, 5]. However, commercial use of Cu nanomaterials is still a huge challenge for researchers, mainly because of their ease of

oxidation in aqueous systems (i.e. living systems) and in open atmosphere. In this regard, new synthetic strategies promising methods for well-defined Cu nanomaterials as well as for the scale-up of their production are highly required. In this work we present development of a new method of synthesis of stable and well-defined silver and copper nanoparticles by chemical reduction with application of natural polymer – chitosan. Physicochemical characterization of the prepared nanomaterials by such techniques as: (i) UV-vis and IR spectroscopy, (ii) structure imaging by scanning (SEM) and transmission (TEM) electron microscopy, (iii) chemical analysis by energy-dispersive X-ray spectroscopy (EDS) and (iv) structural analysis by X-ray powder diffraction (XRD) were executed. The best nanomaterials were selected for in vitro tests and preliminary evaluation of their biological activity against human cancer cells and fungi were performed. [1] Kalishwaralal K, Kanth SBM, Pandian SRK, Deepak V, Gurunathan S. Silver nanoparticles impede the biofilm formation by Pseudomonas aeruginosa and Staphylococcus epidermidis. Colloids and Surfaces B: Biointerfaces. 2010;79:340-4. [2] Sanpui P, Chattopadhyay A, Ghosh SS. Induction of Apoptosis in Cancer Cells at Low Silver Nanoparticle Concentrations using Chitosan Nanocarrier. ACS Appl Mater Interfaces. 2011;3:218-28. [3] Theivasanthi T, Alagar M. Studies of copper nanoparticles effects on microorganisms. Annals of Biological Research. 2011;2:368-73. [4] Iakovidis I, Delimaris I, Piperakis SM. Copper and its complexes on medicine: a biochemical approach. Molecular Biology International. 2011. [5] Mallick S, Sharma S, Benerjee M, Ghosh SS, Chattopadhyay A, Paul A. Iodine-stabilized Cu nanoparticle chitosan composite for antibacterial application. ACS Appl Mater Interfaces. 2012;4:1313-23.

8:50	Poster	B46

Biodegradable nanofibrous coatings electrospun from polymers derived from renewable resources.

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Research goal:

The research goal of this work is to elaborate production methods of fibrous materials from nano- and micro-fibres by electrospinning technique from solutions of biodegradable polymers (including medical purity) for the production of medical products.

Technology and polymer feedstocks:

Nowadays electrospinning is the most effective and most often used technology of the production of nano-fibres from polymer solutions. Institute of Biopolymers and Chemical Fibres for a few years has been conducting research concerning the usage of this technology to obtain various forms of materials from nano- and micro-fibres, mainly from chitosan (also alginate, starch, collagen) and from synthetic polymers of medical purity as PLA and co-PLA (Böringer's Resomers). Apparatus that is used for electrospinning is the laboratory equipment which is the Institute's own technical solution that enables the production or modification of materials in form of sheets sized 20cm x 20cm and 20cm x 50cm. Production of these materials requires preparation of applicable spinning solutions with polymer concentration typically in the range 2 - 10 wt%. Solvents such as water, acids, chloroform, DMSO and others are usually used for this. Due to special requirements which are the effect of medical applications, produced materials are often subjected to additional

processes in order to give them their final form. They are also subjected to mechanical examination as well as to various biological and chemical examinations which define level of their purity. In the final stage these materials have to be subjected to sterilization, mostly sterilization by radiation.

Produced fibrous materials – application directions:

implants' modification (surgical meshes, vascular prosthesis, polyurethane wound dressing sponges, substructures of modified bacterial cellulose) – one-sided covering of permanent implants' surface with layer of biodegradable nano-fibres in order to obtain biocompatible and biostimulative character for wound healing acceleration. In case of some implantable materials the other side is left in "slipping" form, which protects the organism from fusing an implant and the treated organ.

wound dressings in form of self-supporting fibrous coatings

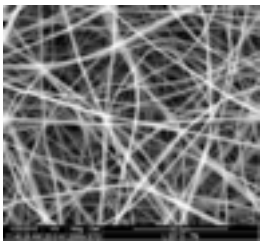
culture substrates for cell cultures

We are extending given directions to new 2D forms with aligned fibres, and also we are trying to produce nano-fibrous 3D objects. We would like to add collagen, alginate or starch to biodegradable polymers that we already use: chitosan, PLA and co-PLA.

Why nano-fibres?

The results of numerous world news report that living cells proliferate much better in contact with nano-fibres than in contact with the surface of much thicker textile fibres. As the porosity of nano-fibrous wound dressing materials is small, they create a barrier for pathogensenabling at the same time an air access to the surface of the wound. Such materials in form of coating covering implants of thicker and mechanically stronger structure, can also biologically enrich the surface of the implants or they can seal the implants to prevent outflow of blood components from vascular prosthesis. We have here ability to influence the structural features of nanofibers and a wide range of polymer materials and the technique of manufacture from polymer solutions enables on the introduction of nano-sized functional additives, which preferably modify the fiber material or as a result of release them to organism, creates the environment with favorable conditions to assist the healing process. Some fibrous structures can be obtained in forms similar to natural tissues occurring in living organisms (biomimetics).

Figures:

		
Nanofibrous scaffold from chitosan / PEO composite in a state of high flexibility in the wet environment.	Zdjęcie SEM powłoki okrywającej implant z nano i mikrowłókien PLA	Laboratory installation for electrospinning nanofibers from polymer solutions.

Acknowledgement:

This work was carried out as a part of the research projects: PBZ-MNiSW-01/II/2007; DWM/233/MATERA/2006 and the statutory project (P22) IBWCh/2012, all supported by the Ministry of Science and Higher Education, Poland.

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8:50	Poster	B47

Biomimetic medical coatings with improved hemocompatibility

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Introduction

We developed a simple and effective method of coating polymer surfaces with highly biocompatible, non-thrombogenic coating that mimics the outer membrane of cells. The coating is based on phospatydylcholine (PC), the main component of cell membrane, which creates biopasive, non-adhesive layer onto polymer substrate.

The obtained coatings were characterized in terms of chemical composition, morphology and platelet adhesion. The experiments proved that PC was successfully incorporated onto polymer surface and the deposition was homogenously through the surface. Experiments with human blood and platelet rich plasma in both static and dynamic condition showed significantly reduced surface platelet adhesion.

Materials and methods

As a polymer substrate was further modification medical grade polyurethane was used (ChronoFlex, AdvanSource Biomaterials). The ChronoFlex pellets were cleaned with alcohol, dried and dissolved in dimethyacrylamide (20% w/v) and subsequently used to prepare polyurethane films: the solution was poured onto glass and dried until the constant mass. The modifying solution was prepared as follows: the soybean – derived PC (POCh, Poland) in concentration of 0, 0.5, 1, 1.5 or 2% (w/v) was added to polyurethane/dimethylacrylamide (1% w/v) solution. 10-mm-diameter discs were cut from polyurethane film and dipped in the modifying solution for 15 seconds, dried under the cover and washed.

Results

The modified surfaces was stained with rhodamine 6G, which due to its highly lipophilic properties is a suitable dye to visualize PC-modified surfaces. The results of fluorescence microcopy

imaging revealed that PC was successfully and evenly incorporated through the polymer surface. The platelet adsorption studies reveled a significant reduction in number of platelets adhered to the surface compared to unmodified polymer (Fig. 1). Moreover, the platelets morphology was normal, without any changes characteristic for activated platelets. The dynamic experiments with whole blood, performed under arterial flow simulated conditions, confirmed high hemocompatibility of prepared surfaces. The value of platelet consumption was much more lower for PC-modified surfaces compared to the pristine polyurethane. Additionally, the number of aggregates created onto PC-modified surfaces was very low; the majority of aggregates were small aggregates (2-3 platelets).

8:50	Poster	B48

Nanowłókna PLA modyfikowane nanocząstkami metali

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Cel:

Badania miały na celu wytworzenie specjalnych filtrów, zbudowanych z nanowłókien, posiadających zdolność zatrzymania niebezpiecznych związków chemicznych, zanieczyszczeń oraz mikroorganizmów.

Materiały oraz metodyka:

Nanowłókna wytwarzano z naturalnego, biodegradowalnego polimeru PLA. Pozyskiwano je na drodze elektroprzędzenia z roztworu, nanosząc na powierzchnie włókniny lnianej, stosowanej jako ich nośnika. Odbiór odbywał się za pomocą aparatury zaopatrzonej w obrotową elektrodę odbiorczą. Stosowano dwa rodzaje roztworu przędzalniczego. Pierwszy stanowił PLA rozpuszczony w chloroformie, z którego otrzymano czyste nanowłókna. Drugi roztwór przędzalniczy zawierał dodatkowo nanocząstki srebra i cynku. Czyste nanowłókna PLA zmodyfikowano poprzez napylanie nanocząstek srebra oraz cynku, w postaci mgiełki, na ich powierzchnię, których zawiesinę otrzymano poprzez homogenizację metali za pomocą ultradźwięków w alkoholu etylowym. Nanowłókna następnie klimatyzowano przez okres 5 dni w temperaturze 30 stopni.

Zmodyfikowane oraz niezmodyfikowane nanowłókna PLA poddano badaniom aktywności przeciwdrobnoustrojowej wobec 3 gatunków bakterii:

Escherichia coli – bakterie gram-ujemne, wskaźnik sanitarno-higieniczny

Staphylococcus aureus – bakterie gram-dodatnie, patogenne dla człowieka

Bacillus subtilis – bakterie gram-dodatnie, przetrwalnikujące

The study has been carried out within the Key Project – POIG.01.03.01-00-004/08 Functional nano- and micro textile materials - NANOMITEX co-financed by the European Union with the

financial resources of the European Regional Development Fund and the Ministry of Science and Higher Education within the framework of the Innovative Economy Operational Programme, 2007-2013.

8:50	Poster	B49

Nadanie funkcyjności bakteriobójczej nanowłóknom PLA/PHB poprzez wprowadzenie chlorheksydyny do ich struktury

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Cel:

Celem badania było wytworzenie zmodyfikowanych za pomocą chlorheksydyny nanowłókien posiadających zdolność zahamowania rozwoju drobnoustrojów.

Materiał oraz metodyka:

Nanowłókna uzyskano w procesie elektroprzędzenia ze zhomogenizowanej mieszaniny naturalnych i biodegradowalnych polimerów PLA i PHB oraz 0,1 % chlorheksydyny. Otrzymane nanowłókna poddano badaniom mikrobiologicznym w celu oznaczenia ich właściwości przeciwdrobnoustrojowych wobec 3 gatunków bakterii:

Escherichia coli – bakterie gram-ujemne, wskaźnik sanitarno-higieniczny

Staphylococcus aureus – bakterie gram-dodatnie, patogenne dla człowieka

Bacillus subtilis – bakterie gram-dodatnie, przetrwalnikujące

Poprzez odpowiednią modyfikację struktury nanowłókien PLA/PHB chlorheksydyną można nadać im właściwości bakteriobójcze i bakteriostatyczne.

The study has been carried out within the Key Project – POIG.01.03.01-00-004/08 Functional nano- and micro textile materials - NANOMITEX co-financed by the European Union with the financial resources of the European Regional Development Fund and the Ministry of Science and Higher Education within the framework of the Innovative Economy Operational Programme, 2007-2013.

8:50	Poster	B50

Otrzymywanie nanostruktur celulozy metodami rafinacji

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Polimer złożony z nanowłókien celulozowych, którego właściwości funkcjonalne determinowane są przez strukturę nanofibryli, nazywany jest nanocelulozą. Włókna naturalne, których przynajmniej jeden wymiar zamyka się w 100 nm łączą w sobie cechy substratów makroskopowych ze specyficznymi właściwościami nanomateriałów. Większa powierzchnia nanocelulozy w stosunku do jej formy natywnej skutkuje wzrostem interakcji z otaczającymi substancjami, np. wiązania różnorodnych nanocząsteczek. Ponadto, w stanie stałym nanoceluloza charakteryzuje się występowaniem w jej strukturze nanoporów, podczas gdy w zawiesinie lub też w kompozycie jest ona przeźroczysta [1].

Założeniem badań realizowanych w ramach projektu jest opracowanie metody wytwarzania nanocelulozy z biomasy roślinnej, która następnie zostanie wykorzystana do produkcji biodegradowalnych tworzyw sztucznych oraz jako komponent
w dziedzinie włókiennictwa.

Założono otrzymanie nanostruktur celulozowych z biomasy roślinnej metodą top-down, wykorzystując do tego celu metody rafinacji o charakterze: fizycznym, chemicznym oraz enzymatycznym (celulazy, pektynazy, ksylanaz i ligninazy). Metody te polegają na rozdrobnieniu materiału wyjściowego i usunięciu z komórek roślinnych składników innych niż celuloza, takich jak: pektyny, hemicelulozy, ligniny i minerały. Następnie, poprzez odpowiednią obróbkę włókien celulozowych można zwiększyć dostępność grup hydroksylowych, zmienić stopień krystaliczności, rozwinąć powierzchnię wewnętrzną, zerwać wiązania wodorowe, co spowoduje wzrost reaktywności celulozy. Prowadzony w odpowiednich warunkach proces może doprowadzić do rozdzielenia włókien celulozowych na mikro i nanofibryle.

W ramach obecnie prowadzonych badań opracowywane są warunki wstępnej obróbki mechanicznej oraz parametry procesu enzymatycznej hydrolizy lignin. Kontrolę wielkości włókien prowadzono z wykorzystaniem mikroskopu optycznego (Olympus BX-51) i oprogramowania CellSens Dimension. Natomiast zawartość lignin po procesie sprawdzano poprzez oznaczanie liczby Kappa.

Badania są finansowane z projektu 01.01.02-10-123/09 współfinansowanego ze środków Europejskiego Funduszu Rozwoju Regionalnego w ramach Programu Operacyjnego Innowacyjna Gospodarka 2007-2013.

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8:50	Poster	B51

Preparation of CAB/organoclay nanocomposites by solution casting

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Celullose acetate butyrate/organo-montmorillonite (CAB/OMMT) nanocomposites were prepared using solution casting method with acetone as a solvent. All samples obtained were characterized using Fourier transform infrared spectroscopy (FT-IR) and X-ray diffraction (XRD). Various

amounts of organoclay were used for nanocomposites preparation in order to investigate the effect of nanofiller amount on the mechanical properties of the samples. The results revealed that CAB/OMMT nanocomposites showed the intercalation of polymer inside the layered silicate structure. The addition of organoclay improved mechanical properties of CAB/OMMT nanocomposites in comparison with unfilled polymer.

8:50	Poster	B52

Synteza i charakterystyka fizykochemiczna nanokapsułek polipirolowych wypełnionych związkami neurochemicznymi

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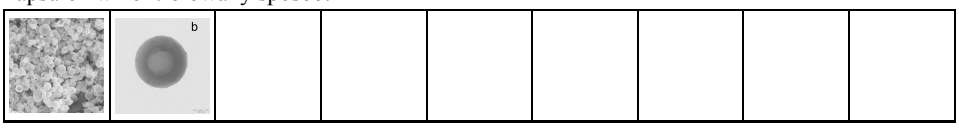
W ostatnich latach obserwuje się intensywny rozwój badań nad nanokapsułkami polimerowymi, gdyż mogą one przynieść wiele rewolucyjnych zastosowań w chemii, medycynie i farmacji. Nanokapsułki polipirolowe są wielką nadzieją badaczy, gdyż mają one kształt sferyczny, co pozwala na zamykanie w środku praktycznie dowolnych substancji chemicznych. Stwarza to możliwości ich wykorzystania np. jako nośników leków, gdzie substancja czynna uwalniana jest z kapsułek w kontrolowany sposób.



Rys. 1. a) Zdjęcie ze skaningowego mikroskopu elektronowego (10 μm x 10 μm) kapsułek polimerowych otrzymanych metodą fotopolimeryzacji pirolu. b) Zdjęcie z transmisyjnego mikroskopu elektronowego pojedynczej kapsułki.

Na konferencji przedstawione zostaną sposoby syntezy oraz charakterystyka fizykochemiczna nanokapsułek polipirolowych wypełnionych kwasem γ-aminomasłowym (GABA). GABA jest endogenną substancją neuroaktywną o działaniu hamującym w układzie nerwowym. Neuroprzekaźnik ten zamknięty w kapsułkach polimerowych stanowi doskonały układ modelowy do badań uwalniania leków z nanonośników w warunkach *in vivo,* gdyż po zadziałaniu bodźca uwalniającego neuroprzekaźnik, efekt fizjologiczny może być w łatwy sposób zarejestrowany z wykorzystaniem standardowych metod medycznych (np. pomiar ciśnienia tętniczego krwi i parametrów oddechowych).

Do badań właściwości kapsułek wypełnionych GABA wykorzystano metody mikroskopowe (SEM, TEM), spektroskopię w podczerwieni oraz pomiary termograwimetryczne.

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8:50	Poster	B53

Surface modifications of ZnO-based quantum dots

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Quantum dots are among the most promising tools in the nanomedicine area. The potential uses of these colloidal semiconductor nanocrystals include bioengineering, bioanalytical detection, bioimaging, as well as targeted drug delivery, *in vivo* monitoring, and photodynamic therapy. The possibility of such wide applications is attributed to their unique size-dependent photophysical properties, like narrow and symmetrical emission spectra, broad absorption spectra, high quantum yield, and simultaneous excitation by a single light source. However, toxicity of the QDs (especially those based on Cd) is a serious limitation in biomedical applications. Synthesizing biocompatible nanocrystals, composed of weakly toxic ZnO became a leading route in quantum dots nanofabrication.

However, uncapped ZnO QDs tend to aggregate and are not stable due to high surface reactivity. In this work, we present various approaches which can be useful to eliminate these drawbacks. Introducing of inorganic shell made of higher band-gap semiconductor as well as stabilizing with oleic acid were the routes to obtain better stability and protection of the ZnO core. Oleate-capped ZnO QDs as well as ZnO/MgO and ZnO/ZrO_2 core-shell QDs synthesized in a sol-gel process were characterized by UV-vis and fluorescence spectroscopies, transmission electron microscopy (TEM), and powder X-ray diffraction (XRD), showing good stability and preserved luminescent properties.

As water-stabilityis the fundamental criterion to apply these nanoparticles in biological applications, several strategies were evaluated to disperse primarily hydrophobic ZnO QDs in aqueous media. One of the methods used by us was silanization with 3-aminopropyltrimethoxysilane and 3-[2-(2-aminoethylamino)ethylamino]propyl-trimethoxysilane, which resulted in exchange of hydrophobic ligands into hydrophilic ones. In terms of bioimaging such modified QDs were further conjugated with small selected biomolecules (folic acid, L-glutathione or gluconic acid) used as targeting ligands. The effects of structural properties of biomolecules on the formation, stabilization, photoluminescence of ZnO-core QDs have been evaluated. The obtained results demonstrated that aminosiloxane-capped ZnO QDs coupled either with folic acid, glutathione or gluconic acid show good optical properties.

Obtained results encourage our team to work on cytotoxicity of modified-ZnO QDs. First tests have demonstrated that synthesized materials are very promising and should be more developed to be successfully used in medical applications, such as e.g. cancer therapy.

8:50	Poster	B54

Nanotechnologies for load-bearing implants

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In load-bearing implants the nanotechnologies are applied for different purposes, generally to increase the mechanical strength and structure-enhanced bioactivity. Such solutions may involve development of both microroughness and nanoroughness for titanium dental implants, which makes adhesion of enamel ceramic and/or polymer coatings to the metallic implant better. Another application includes nanometric reinforcement of dental resins to increase their mechanical properties. In orthopaedics, the conventional PMMA-based cements can be successfully substituted by materials containing hydroxyapatite nanoparticles.

The nanotechnologies are applied at Gdansk University of Technology to improve the biocompatibility and bioactivity of load-bearing implants based on a new concept, i.e. titanium scaffold filled in with a biodegradable and mechanically resistant polymer – ceramic core material. In order to improve the bioactivity, the nanoxide layers are created from the base rutile oxide structure by preferential dissolution in presence of fluorides. Such oxide layers demonstrate the increased bioactivity and because of their specific structure may attract and release such important constituents of an implant as antibiotics, nanosilver and growth factors. Another possible solution is a deposition of nanometric hydroxyapatite coating by sol-gel or electrochemical technique, which adhesion and elasticity are substantially better than that of conventional plasma sprayed coatings. Even for thicker ceramic coatings, their nanoroughness is important for better adhesion of core material. An application of silver in form of single atoms or isolated clusters of nanometric size into bulk of nanotubes of the oxide layer, into hydroxyapatite coating or core material, or as an element of hybride material, can be crucial for achievement of better bioactivity, i.e. preventing the adhesion of bacteria on the surface of metallic implant and then inflammation processes.

Nanomedicine session 1

Monday morning, 17 September, 9:00

Chair: Urszula Narkiewicz

9:00	Oral

Welcome address

Witold Łojkowski[1], Malgorzata Lewandowska[2], Urszula Narkiewicz[3]

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Acknowledgments for support if the conference to:

Warsaw University of Technology
Faculty of Materials Science, WUT
Institute of High Pressure Physics, PAS
Centre for Preclinical Research and Technology

9:10	Invited oral

Carbon nanotubes applications for drug and gene delivery

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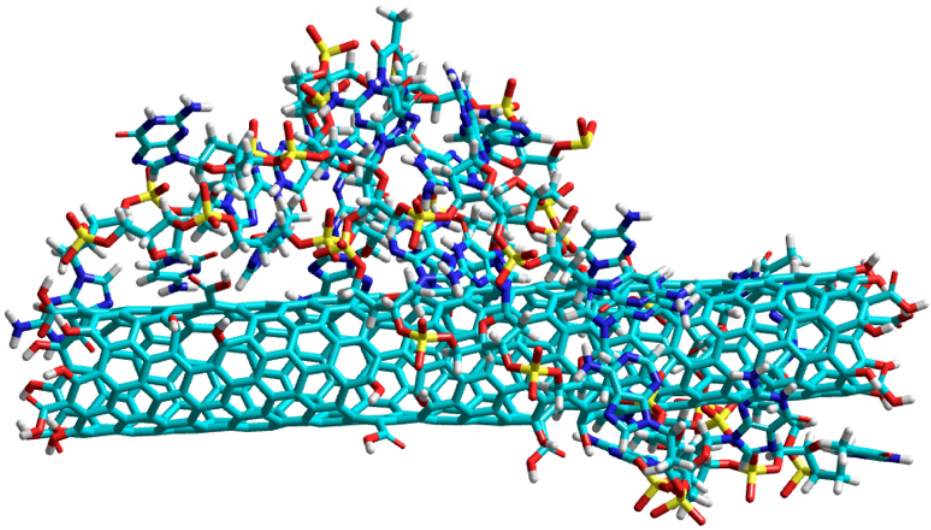

Single DNA strand immobilized on SWCNT-COOH surface

The world production of carbon nanotubes reaches recently 2000 Mg/year. However, the world demand for CNTs material nowadays reaches hardly 300 Mg/year resulting in a considerable over-saturation of the market. Consequently, challenge is generated for scientists and manufacturers to find new, efficient applications for this fascinating material. Medicine is one of potential application area, where CNTs can be applied in, for instance, drug delivery systems, gene transfection and scaffolds for tissue regeneration. However, thus far poorly understood effects of CNTs on human organism requires detailed risk analysis and balancing potential risks and benefits from such applications.

In the case of industrial applications of CNTs it is necessary to examine thoroughly in this respect every step of such involvement of CNTs starting from their manufacturing through their functionalization, incorporation into a matrix (e.g. polymer), checking relevant properties of final products, then methods of their administration and, finally, after the product death also their

degradation. Despite a technical feasibility study of the scale up-grading a careful analysis of possible risks for human health and environment at each stage from manufacturing to the final product, recycling and/or destruction is indispensable.

9:30	Oral

Biological properties of graphene

Ewa Sawosz Chwalibóg, Sławomir Jaworski, Marta Grodzik, Mateusz Wierzbicki, Marta Prasek

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Graphene - one of the allotropic forms of carbon is regarded as a structure that will revolutionize the electronics. This allotropic form of carbon consists of a single layer of sp^2 bonded atoms and is considered as the finest and most durable layer capable of free existence. Because of its unique structure, graphene exhibits chemical properties, which can have vital biological applications. Graphene is vulnerable for the self-organization processes with organic molecules, it is nearly transparent and impermeable to the other atoms, It has also gas sorption properties and it is resistant to water and body fluids. Only few studies investigated the effect of graphene on selected biological molecules and organisms such as bacteria and cell lines cultured in vitro but the results are conflicting and not conclusive.

The objective of the present studies was to determine the potential aplicabilty of graphene as a modifier of the biological functions of living organisms.

The non-functionalized graphene (SkySpring Nanomaterials Inc., Houston, USA) was applied and its effects on the morphology, toxicity, expression of selected genes and proteins in experimental *in vitro* models with proteins and nucleic acids, bacteria Salmonella Enteritidis and glioblastoma cell lines, and in *in vivo* animal models with chicken embryos and mice.

The results showed relatively low toxicity of graphene, both to cell lines and animal models. Furthermore, there was a strong tendency of single cell organisms to settle on specific areas of graphene flakes, indicating possibilities of constructing special bacterial “rafts”, which can be used for diagnosis or immuno-modulations. Our preliminary studies also indicated the possibility of using graphene as a platform for delivering DNA or signal proteins. Furthermore, we observed that a super thin graphene flake might act as a "cell blocker" of abnormally developing cells.

These preliminary studies indicate a wide range of applications of graphene in biology and medicine. However, the future research has to be carried out with graphene with precisely known physical characteristics, and this requires a close interdisciplinary cooperation between physicists and biologists.

9:50 Invited oral

"Superstructures of protein-based amyloid nanofibrils as a scaffold material for SERS platforms"

Wojciech Dzwolak

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Nanostructuring of noble metal (such as Au, or Ag) surfaces with biomorphic templates facilitates variety of applications of surface enhanced Raman scattering (SERS). On the other hand, there is a growing interest in possible applications of amyloid fibrils – linear, beta-sheet-rich aggregates of misfolded protein molecules, studied so-far mostly in the context of degenerative disorders such as Alzheimer's disease. Here we show that the newly reported insulin amyloid superstructures [1] may be employed as stable nanoscaffolds for metallic Au films providing an effective substrate for surface enhanced Raman scattering on covalently bound aromatic molecules [2]. The self-assembled superstructures of insulin fibrils are capable of templating nanopatterns in sputtered Au layers without overlapping the SERS spectra of the Raman scatterer with vibrational bands stemming from the protein.

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[2] Wojcik S, Babenko V, Dzwolak W, **Langmuir** 26 (2010) 18303–18307.

10:10 Oral

Studies towards stereoselective bionanocatalysis on gold nanoparticles

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Among different nanomaterials, gold nanoparticles are especially interesting for biocatalysis due to great affinity of gold to thiols and amino groups, which are present in enzymes. Such interactions may cause changes in a structure of an enzyme and modify its substrate specificity. In literature, there is no information about influence of gold nanoparticles on enzyme stereoselectivity. Therefore systematic studies were performed to verify this phenomenon. Selected enzymes were immobilized on gold nanoparticles using different protocols. Obtained bionanocatalysts were used in a model reaction. Reaction progress was monitored using standard procedure and enantioselectivity was determined. The influence of bionanocatalysts structure on the stereochemical course of the reaction studied will be discussed.

Acknowledgments: This work was spported by project "Biotransformations for pharmaceutical and cosmetics industry" No. POIG.01.03.01-00-158/09-01 part-financed by the European Union within the European Regional Development Fund.

10:25	Invited oral

Applications of selected nano-particles in nano-medicine

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Some nano-particles have great potential for applications in nano-medicine. These include especially nano-silver, nano-magnetite and carbon nano-tubes (CNT´s).

Nano-silver has an antimicrobial effect. Nano-magnetite is a ferrimagnetic substance which can be excited in an alternating magnetic field. Injected into tumors, cancer cells die via hyperthermia. Modified carbon nano-tubes can be used as drug carriers with defined release characteristics.

In the first step, the nano-particles are to be synthesized either by a top-down or bottom-up process. For best efficiency - in case of nano-silver and nano-magnetite - particle size distribution should have a maximum at 10 nm.

For thermodynamic reasons, nano-particles tend to re-agglomerate in order to lower surface-energy. This weakens the intended effects and must be prevented in any case and under all circumstances. Therefore a nano-stabilization by chemical additives is required, imprinting resulting surface charges. Poisson-Boltzmann differential equation is suitable to calculate surface potential and enable a targeted selection of additives.

The characterization of the nano-particles with selected analytical methods, e.g. scanning electron microscopy, x-ray fluorescence, x-ray diffraction, photon correlation spectroskopy and atomic emission spectroscopy ist the next step. Here the trace analysis with AAS/AES is an essential element to proof nano-safety.

Most important step to enable application is system-integration, which means the combination of nano-technology with polymer-technology.

The paper explains the above steps in detail with examples and international references.

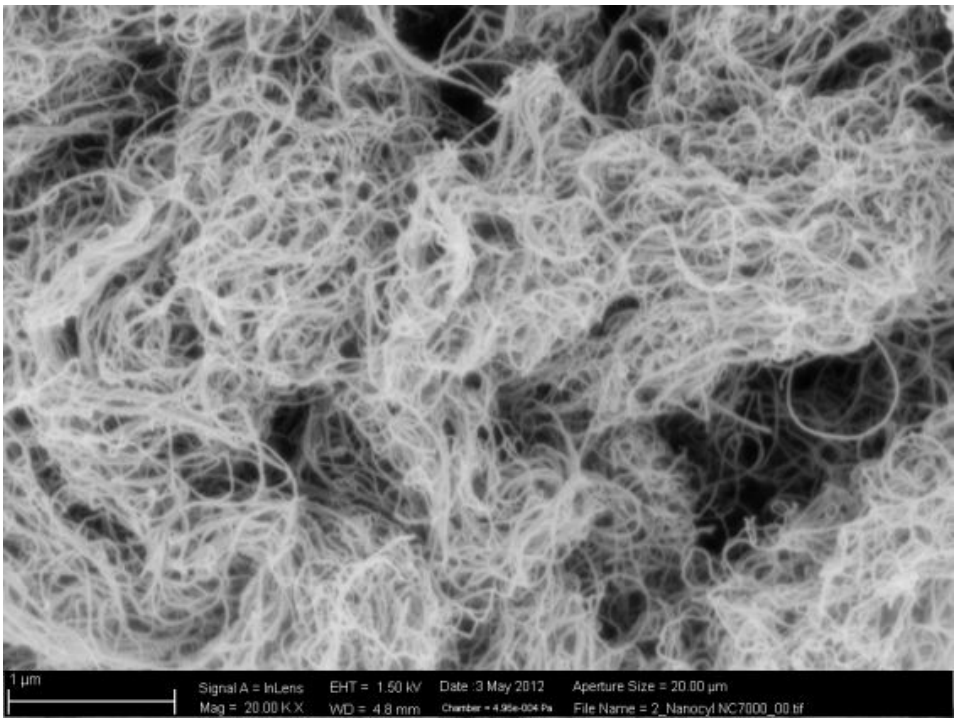

Figure 1: SEM-record of CNT´s suggested for contolled drug-delivery.

COFFEE

Monday morning, 17 September, 10:50

Nanomedicine session 2

Monday morning, 17 September, 11:10

Chair: Witold Łojkowski

11:10	Invited oral

Nanomedicine in combating cancer - How multidisciplinary research enhances innovation and accelerates path to the clinic

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Nanotechnology will provide novel, paradigm shifting solutions to medical problems. In oncology, nanomaterials are enabling targeted delivery of imaging agents and therapeutics to cancerous tissue; nanoscale devices are providing for multiplexed sensing in early disease detection and therapeutic monitoring.

Nanotherapeutics are capable of increasing treatment effectiveness while limiting side effects.

Next generation of nanotherapies for cancer is expected to use active targeting of tumor-specific cell markers to deliver entirely new modalities of cancer treatment, including triggered release of cytotoxic molecules, genetic material, heat, or cellular disruption. Given that positive cancer outcomes are associated so closely with early detection, another important goal of cancer nanotechnology efforts is to improve diagnostic capabilities, through *in vivo* imaging contrast enhancement and *in vitro* device development. Magnetic resonance imaging, ultrasound, positron emission tomography (PET) will all benefit from the development of these new contrast agents. Furthermore, those constructs can be made to operate in multi-functional manner; whether it is ability to probe and monitor tumor microenvironment in addition to imaging tumor mass itself, capability of multi-modality imaging, or performing theranostic functions of diagnosis and subsequent treatment. Advances in microfluidics and nanodevices will greatly accelerate the genetic and proteomic analysis of cancer subtypes *in vitro* and the monitoring of markers of early or premalignant stage cancer and premetastatic disease.
In order to further these research goals, National Cancer Institute (NCI) formed a program called Alliance for Nanotechnology in Cancer which was initiated in 2004. This presentation will describe the current advances of cancer nanotechnology, future strategies and prospects of the field, and details behind the organization of the Alliance.

11:45	Invited oral

Polysaccharide nanoparticles for anticancer drug delivery

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Introduction

Growing level of hygiene and medical care in western civilization is resulting in the change of disease types we are facing. Currently, the most dangerous, as expressed in the number of lethal cases, are cardiovascular disease (CVD) and cancer, and the last one is slowly taking the first position in the infamous statistics of death causes. Among contemporary citizens of Europe probability of developing cancer during the life time is about 42%; male 45%, female 39%. The most frequent are breast, prostate, colon and lungs. Carcinogenesis arises from a single transformed cell but it is a multistep process resulting from a combination of environmental and congenital factors. Thus, the appearance of cancer in our body is an effect of several subsequent events triggered by various factors mostly genetic predispositions, viruses, radiation or certain chemicals. They elicit many genetic and cytoplasm events including fragmentation of chromosomes, deactivation of tumor suppressor genes (protein 53, retinoblastoma protein…), functional retardation of mitochondria, what finally leads to cancer.

Cancer, as a disease was for the first time described by ancient doctors almost 30 centuries ago but the word “carcinoma” and first effective surgical procedures were introduced by Avicenna (Ibn Sina, *Al-Quanunfi al-tibb*, Canon Medicae, 1020). At the beginning cancer grows without any symptoms, when it is big enough to be noticed by a patient in most of the cases it already started to spread forming new colonies in our body. First help of a doctor is a surgical removal of primary cancer. When it is already in the metastasis special anticancer drugs are administered. Contemporary cancer pharmacotherapy began in 1940s, firs by application of nitrogen mustard and folic acid

antagonist (aminopterin and methotrexate), early examples of rational drug design. With the development of chemical and medical sciences new anticancer drugs appeared: taxanes, vinca alkaloids, anthracyclines, fluorouracil... Beginning of chemotherapy brought a new problem - side effects, which are frequently devastating for the patient's organism, and drug resistance. To achieve efficient elimination of cancer cells we started using poisons at almost lethal doses, what heavily destroys also healthy cells and organs in our body. In 1965 combinatorial - multidrug cancer treatment was introduced. This new approach was based on the hypothesis that cancer chemotherapy should follow the strategy of antibiotic therapy for tuberculosis with combinations of drugs, each with a different mechanism of action. Cancer cells could conceivably mutate to become resistant to a single agent, but by using different drugs concurrently it is more difficult for the tumor to develop resistance. Application of few drugs in specially designed "cocktails" proved to be much more efficient. With the successes of combinatorial chemotherapy and the discovery of many new agents, there was a feeling, at this time, that all cancers could be treated, if one could properly deliver to the tumor the proper combination of drugs, but it was just a feeling... Nonetheless, cancer remains a major cause of illness and death, and conventional cytotoxic chemotherapy has proved unable to cure most cancers after they have metastasized (spread). Recently a new hope on efficient and safe delivery of anticancer agents is associated with nanoparticles (NP), objects of diameter below a fraction of micrometers.

Nanotechnology

Nanoparticles show a few very important properties, which can help to fight the beast. First, they can encapsulate cytostatic drugs which are frequently strong poisons and causes local necrosis at the place of delivery as well as systemic side effects. Nanoparticles have already been introduced in drug delivery and even in cancer therapy (liposomes with doxorubicine, taxanes-albumin coniugates). Liposomal drug carriers are not stable enough in our circulatory system. Blood vessels in tumor are found to be leaky with pores of 20-150 nm, what leads to the increased resident time of NPs in this area and are called enhanced permeability and retention (EPR) effect. NP can be equipped in surface antibodies or other special recognition moieties which increase their affinity to cancerous cells. To achieve passive (EPR) or active targeting NPs should be able to stay in our circulatory system long enough. Unfortunately our immunological system is efficiently removing any foreign objects from our bloodstream so the NP to stay there should be carefully designed and made. To escape recognition by the complement immune system and prevent coating by small plasma proteins, special coatings of NP have been developed; those are polyethylene glycol and polysaccharides. They form a sort of hydrogel which has very low surface energy and binds water around. NP can serve as a carrier for multiple drugs with various mechanisms of action; such "one package delivery" will prevent development of drug resistance of cancer cells. Additionally NP can carry molecules which are normally unable to get into the cells, like siRNA, which can trigger apoptosis. Polysaccharide NPs are especially promising due to their biocompatibility since polysaccharides are widely present in our organism. Polysaccharides are also very easy for chemical modification so various targeting molecules and multiple drugs can be covalently bound in their structure and at the surface. There is also another advantage of having glucose residues at the NP surface. Malignant rapidly-growing tumor cells typically have glycolytic rates that are up to 200 times higher than those of their normal tissues of origin. This phenomenon was first described in 1930 by Otto Warburg and is referred to as the Warburg effect. This phenomenon may simply be a consequence of the mitochondria damage in cancer, or an adaptation to low-oxygen environments within tumors, or a result of cancer genes shutting down the mitochondria because they are involved in the cell's apoptosis program which would otherwise kill cancerous cells. Despite the unclear reasons Warburg phenomenon has important medical applications; it is utilized clinically to

diagnose and monitor treatment responses of cancers by imaging uptake of 2-18F-2-deoxyglucose (FDG) (a radioactive modified glucose) with positron emission tomography (PET). We plan to utilize the same mechanism by building outer surface of NP from polysaccharides. Presence of abundant glucose chains at the surface, which will be "swallowed" by glucose transporters on cancer cell wall, will cause attachment of NP to the cell surface followed by internalization. Inside, in the endosome, due to low pH and enzymatic (lizozyme) activity NP will break apart releasing drugs.

Presented work describes technology which allows direct formation of polysaccharide NPs in water solution at room temperature without the use of aggressive chemicals. This is very important since cleaning technologies of NPs suspensions are sometimes complex and expensive. First polysaccharides are partially oxidized to form reactive aldehyde groups along the chain. Specific oxidation agents are applied, those are periodates and hydrogen peroxides with catalyst. Then hydrophilic polysaccharide backbone is modified with hydrophobic moieties, like lipophylic amino acids or aliphatic or aromatic amines. Polysaccharides, when dissolved in water are in the form of more or less loosely coiled chain, sometimes with some branching, which are closely surrounded by water molecules. In the case of ionized polysaccharide derivatives, like hyaluronan or chitosan, since interactions with water molecules are stronger and also electrical mutual repulsion appears, the chain is less coiled – more straight. When some of the sugar rings are substituted with hydrophobic molecules side groups (hydrocarbons, fatty acids, hydrophobic amino acids), hydrophobic moieties are "trying" to be together, to minimize the internal energy of molecule. This process forces the formation of NP in water environment due to self assembly. Obtained NPs are stable and can be lyophilized and stored as dry powder, after immersion in water hydrophobic – hydrophilic interactions forms NPs again. Preliminary experiments conducted on cancer cell lines and mice are very promising. Polysaccharide NPs proved to be able to efficiently enter cancerous cells by endocytosis and to release their load in lysosomes, when pH decreases. This is a very important phenomenon, in normal body pH nanoparticles are stable for many days, in the low pH after endocytosis NP breaks down and release the load.

Various drugs have already been covalently linked inside polysaccharide NP: Daunorubicine, Doxorubicine, Taxol, 5-F Uracile, Cytarabin, Gemcitabin… Obtained NP showed to efficiently eliminate various types of human cancer cell lines, during in vitro experiments, while polysaccharide NPs without the drug load are nontoxic for human cells and mice. Currently a series of animal experiments are in the preparation stage.

12:15	Invited oral

Bio-based nanofibres - manufacture, processing and practical application

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In recent years at Institute of Biopolymers and Chemical Fibres (IBWCh) an extensive research has been carried out on development of methods for producing nanofibers and other nanostructures (e.g. nanoparticles, nanofibrids) from various polymers using different techniques. These techniques include biotechnological (bio-catalysis, biosynthesis), chemical and physico-mech-

anical (electrospinning, mechanical grinding, ultrasonication) methods. Functional nano-fibrous forms have a wide range of applications including medicine, filtration materials, electronics and the manufacture of bio-composites.

Electrospinning is the most widely used and most effective technique for production of micro-and nano-fibrous forms of biodegradable polymers from renewable sources, including medical grade chitosan, cellulose, PLA and co-PLA [1,2]. Nano-fibrous coatings produced by this method can be applied in dressing materials as a barrier against pathogens or in surface modification of implants (surgical mesh, vascular grafts), giving them biostimulative and biocompatible character.

The Institute has also developed the production technology of chitosan and chitosan-alginate micro- and nano-fibrids [3]. The manufacturing process is carried out under dynamic conditions using a flow reactor. Developed polymeric micro- and nano-forms can be applied in haemostatic wound dressings produced in the form of non-wovens and sponges which are characterized by high fluid absorption [4] and also in modification of surgical implants.

In IBWCh research is conducted into obtaining cellulose nanofibers from waste plant biomass, which is a rich source of the polymer. Processes of cellulose extraction from vegetal raw materials and of production of micro- and nanofibrous forms involve thermo-mechanical, chemical, biotechnological as well as physical-mechanical methods. Cellulose nano-fibers get more and more attention because of their excellent mechanical properties which allow their application in production of various types of composites.

Another source of cellulose nanofibers is bacterial cellulose produced by certain strains of bacteria, especially of the genus *Acetobacter*. Bacterial cellulose is synthesized in the form of microfibrils creating highly entangled network of micro-and nanofibers. The resulting bio-nano-cellulose is characterized by high chemical purity, lack of lignin and hemicellulose, high crystallinity and degree of polymerization, which distinguishes it from other types of cellulose [5,6]. An important feature of bacterial cellulose is that it can be modified during its biosynthesis. Research on the production of bacterial cellulose-modified chitosan has been conducted at IBWCh for many years [5-7]. Modified nano-bio-cellulose may have numerous medical applications e.g. as an occlusive hydrogel wound dressing [8], a material of artificial blood vessels [9], or as a component of multilayer surgical mesh for hernia treatment [10]. Research was also conducted at IBWCh on the use of modified bacterial cellulose for manufacture of high-grade loudspeaker membranes [11].

Acknowledgment

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12:45	Oral

Optyczna charakteryzacja powierzchni tkaniny poddanej działaniu promienia laserowego

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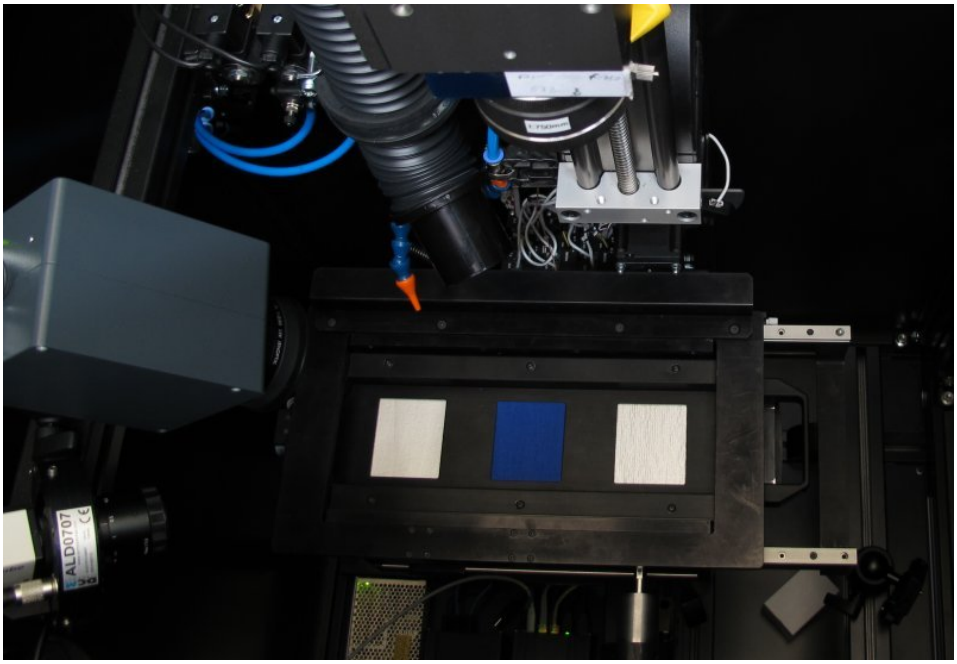
Nowe rozwiązania laserów przemysłowych – tzw. lasery ręczne – emitują w obszarze roboczym dużą gęstość mocy (do 6 KW mocy ciągłej na powierzchnię rzędu kilku mm^2) w zakresie bliskiej podczerwieni (λ = 800-1100 nm). Są wykorzystywane głównie jako narzędzia spawalnicze i czyszczące. Pomimo wysokich kwalifikacji operatorów tych narzędzi, odnotowywane są przypadki porażeń ciała przypadkowo ukierunkowanym promieniem lasera bądź jego odbiciem. Zniszczenie tkanki jest zwykle głębokie i związane z jej absorpcję wysoką absorpcyjnością promieniowania przy typowych dla laserów długościach fali. Opracowanie metod ochrony skóry przed takimi porażeniami stanowi poważne wyzwanie technologiczne.

Dotychczas wykorzystywane metody oceny jakości tkaniny jako środka ochrony bazują na rozwiązaniach stosowanych przy zabezpieczaniu przed ogniem czyli tzw. ciepłem strukturalnym. Metody te są tylko częściowo stosowalne do ochrony przez promieniowaniem laserowym z uwagi na inną wrażliwość typów tkanki na ciepło i na promieniowanie NIR. Również gęstość mocy promienia laserowego wielokrotnie wyższa od gęstości mocy ognia strukturalnego.

Kierunki projektowania tkanin chroniących skórę przed promieniem lasera, to zmniejszenie gęstości mocy poprzez odbicie części promieniowania od powierzchni oraz rozproszenie i absorpcja (transformacja w ciepło) wewnątrz tkaniny. Wynikowy strumień energii na skórze powinien

być na tyle mały by w określonym nie spowodować oparzeń i na tyle duży by wywołać odruchowe wycofanie części ciała z zagrożonej strefy. Kształtowanie odbicia odbywa się poprzez dobór odpowiedniego pokrycia tkaniny cienką warstwą zawierającą nanocząstki rozpraszające kierunkowo odbite promieniowanie. Natomiast na wewnętrzne rozpraszanie i absorpcję wpływa się poprzez dobór mikrowłókien, z których wykonana jest tworząca tkaninę przędza, oraz odpowiedni jej splot. Alternatywne rozwiązania obejmują wyposażenie tkaniny w układ pozwalający na wyłączenie lasera.
Nieodłącznym zagadnieniem badawczym jest opracowanie metody charakteryzacji tkaniny pozwalającej na ocenę jej właściwości ochronnych. Do tego celu firma CIM-mes Projekt wykorzystała pośrednią metodę wyznaczania parametrów optycznych (refleksyjności, transmisyjności i absorpcyjności) poprzez pomiar kątowej charakterystyki odbicia i transmisji (tzw. funkcji BRDF i BTDF), który dodatkowo pozwala na wyznaczenie stopnia rozproszenia odbicia.. Metoda została wdrożona poprzez zaprojektowanie stanowiska badawczego, gdzie w miejsce zaawansowanej sfery Ulbrichta został wykorzystany mechanizm goniometru wraz z ruchomym stolikiem pomiarowym pozwalającym na jednoczesne badanie 3 próbek tkaniny. Urządzenie jest wyposażone również w kalorymetr do przeprowadzania testu Stoll-Chianty. Stanowisko pracuje w cyklu automatycznym bezpiecznym dla operatora. Wyniki pomiaru są przetwarzane przy wykorzystaniu dedykowanego oprogramowania w środowisku LabVIEW. Pomiary na zbudowanym urządzeniu stanowią część planowanej procedury kwalifikacyjnej projektowanej euronormy dotyczącej ochrony skóry przed promieniowaniem laserowym. Stanowisko zostało zainstalowane w dwóch jednostkach badawczych. Prace badawcze i projektowe zostały sfinansowane przez Komisję Europejską w ramach projektu PR7 nr NMP2-SE-2009-229165 o akronimie PROSYS LASER.



LUNCH

Monday afternoon, 17 September, 13:00

Nanomedicine session 3

Monday afternoon, 17 September, 13:40

Chair: Danuta Ciechańska

13:40	Invited oral

Application of micro- and nanostructured biomaterials in regenerative medicine

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The number of critical tissues defects caused by injury, cancer or aging of the world population, is increasing. Novel biodegradable biomaterial natural or synthetic might have broad applications in tissues regeneration. Scaffolds made of them could support regeneration of the damaged site and then, undergo complete degradation. The degradation rate of those biomaterials must be carefully adjusted to ensure mechanical stability of the scaffold when the new tissue is being formed. Moreover, the biomaterials should give proper micro- and nano-environment for stimulation of a new tissue formation.

The aim of the study was to show a high potential of using novel biomaterials for regeneration of different tissues. Biomaterials such as biodegradable polyesters, and their copolymers as well as composites comprised of synthetic polymers and bioceramics and/or natural polymers were analyzed for different applications. Several methods of processing of biodegradable materials such as blending, solid freeform fabrication, electrospinning have been investigated. These methods allowed fabrication of 3D porous micro and nano-structures, which could form scaffolds for new tissues formation. The structure, chemical and mechanical properties, and also biocompatibility and bioactivity of these 3D structures were analyzed.

In summary, the study shows that biodegradable materials have high potential to be utilized in tissue engineering of different tissues such as bones or nerves. However, designing and fabrication of the bioactive biomaterials with required properties over the time of exposure to implant conditions is still a big challenge.

ACKNOWLEDGMENTS: This work was partially financed by the European Regional Development Fund within the Innovative Economy Operational Programme in the frame of BIO-IMPLANT project - „Bioimplants for the treatment of bone tissue lesions in oncological patients" (*POIG.01.01.02-00-022/09*).

14:10 Oral

Synthesis and optical properties of ultrasmall inorganic optical markers based on lanthanides emission for bio-medical applications

Artur Podhorodecki, Mateusz Banski, Agnieszka Noculak, Bartlomiej Sojka, J. Misiewicz

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Introducing to medicine and biology concept of optical markers in tremendous way has changed the recent status of these two important disciplines. This was mainly due to strong development in imaging techniques which recently allow us to investigate both static as well dynamic properties of living cells, their components and their interactions with external factors.

Recently used molecular markers including organic dyes, fluorescent proteins or chelates containing lanthanide ions have several significant limitations. One of the alternatives for molecular markers are inorganic quantum dots (ie. CdSe, CdS) which are recently commonly used in many academic works. However, even if they are much better from physico-chemical point of view, from the application point of view at this moment they are rather useless mainly because of their high risk of toxicity. One of the solution combining advantages of both concepts is to make non-toxic inorganic nanocrystals doped by lanthanide ions.

In this work, we will present optical results obtained for $NaYF_4$, $NaGdF_4$ and GdF nanocrystals doped by different lanthanide ions (Eu, Tb, Nd, Tm, Yb). The aim of this work was to design and to synthesize these markers and to understand physical processes responsible for their emission/ excitation and to optimize these processes to the physical limits. Additionally, potential use of such nanocrystals in nano-bio medicine will be discussed.

14:25 Oral

Advanced microtechnologies for cytogenetic analysis

Dorota Kwasny, Indumathi Vedarethinam, Pranjul Shah, Maria Dimaki, Asli Silahtaroglu, Zeynep Tumer, Winnie E. Svendsen

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Cytogenetic and molecular cytogenetic analyses, which aim to detect chromosome abnormalities, are routinely performed in cytogenetic laboratories all over the world. Traditional cytogenetic studies are performed by analyzing the banding pattern of chromosomes, and are complemented by molecular cytogenetic techniques such as fluorescent *in situ* hybridization (FISH). To improve FISH application in cytogenetic analysis the issues with long experimental time, high volumes of expensive reagents and requirement for trained technicians need to be addressed. The protocol has recently evolved towards on chip detection of chromosome abnormalities with the development of microsystems for FISH analysis. The challenges addressed by the developed microsystems are mainly the automation of the assay performance, reduction in probe volume, as well as reduction of assay time.

We present here our efforts to introduce automation in the cytogenetic laboratories at a microscale. We have developed membrane based micro perfusion systems capable of expansion of lymphocytes in a shorter time and at a smaller scale. The simulated and experimental results show very efficient exchange of the growth medium to the hypotonic solution and fixative. These are commonly used solutions required for proper preparation of a metaphase chromosomes analysis. Further we developed a microfluidic chip for preparation of metaphase chromosome spreads and their analysis by metaphase FISH on chip. All developed devices are capable of performing the entire metaphase FISH protocol in a shorter time and at the same quality as standard methods.

14:40	Invited oral

ZnO nanoparticles effect on integrity of human endothelial cells

Ewa Stępień[1,2], Dominik Jakubiak, Jacek Wojnarowicz[3], Agnieszka Opalińska[3,4], Witold Łojkowski[3,5]

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Disruption of the intercellular interactions between endothelial cells leads to endothelial dysfunction. The role of nanoparticles in plasma membrane stability, actin cytoskeleton organization and intercellular junctions is unclear. Human umbilical vein endothelial cells were treated with zinc oxide NPs *in vitro*. Cell shape, adhesiveness and plasma membrane integrity were analyzed by means of optical fluorescence microscopy, scanning electron microscopy and flow cytometry methods. Additionally, lactate dehydrogenase activity assay and annexin V staining were performed. The scanning electron microscopy analysis showed changes in morphology and surface topography. The F-actin organization was typical for migrating cells. Cell membrane damage (significant increase in lactate dehydrogenase release and annexin V staining) was observed in the concentration of ZnO nanoparticles above 30µg/ml. The relationship between ZnO nanoparticles and endothelial dysfunction was clearly established and the importance of cytoskeleton reorganization and loosening of the continuous endothelial monolayer after nanoparticles exposure has been documented. Furthermore, loosing of cell contact diminishes contact-inhibition and activates endothelial cells. Contact-inhibition universally regulates migratory cell proliferation in non-transformed normal cells when neighboring cells are in contact. This phenomenon explains poor regenerative capacity of in vivo human endothelial cells during aging, injury, and surgery. Disrupting of contact inhibition regulated by Ca2+-related adhesion molecules (cadherins), actually activates the canonical Wnt signaling and induces canonicl activation of Wnt pathways. This may also explain why NPs which varied in shape and size and are made from the same substance may have divergent effect on NF-kB-dependent genes or inflammatory response of endothelial cells.

COFFEE

Monday afternoon, 17 September, 14:55

AFM in Nanomedicine

Monday afternoon, 17 September, 15:15

Chair: Ewa Stępień

15:15 Oral

Novel strategies for nanoindentation spectroscopy of cells

Grzegorz D. Brzezinka[1], Jacek Szczerbinski[1], Agnieszka M. Szczygiel[1], Marta Targosz-Korecka[1], Marc Richter[2], Marek Szymonski[1]

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Various cell signalling pathways (i.e. mechanotransduction processes) and cytoskeletal rearrangements are triggered by changes in the physical stress and forces applied to the cells. In recent years, in addition to biochemical and physiological studies, research on mechanical properties of cells has been performed. Among those, a great potential of atomic force microscopy (AFM) and nanoindentation spectroscopy techniques was demonstrated for determination of cell shape, membrane structure its stiffness and adhesive properties. Recent achievements in the processing of large amounts of data and the optimization of AFM for biological applications, such as Quantitative Imaging mode provided by JPK Nanowizard 3, have offered methods for deeper insight of cell membrane structure. Microtubules are composed of actin fibers, which create a net below the cellular membrane. Thus, this structure directly responds to local cell membrane contraction. Both fluorescent staining and AFM contact mode topography allows for the visualization of the stress fibers structure. However, these techniques do not provide information about the mechanical properties of the microtubules. We demonstrate that the elasticity of the cell may be mapped with resolution sufficient to distinguish details of membrane structure. Furthermore, the features in topography and elasticity maps correlate spatially, which confirms the observation.

15:30 Oral

Atomic Force Microscopy (AFM) of polyelectrolyte multilayer systems.

Marcin Strawski[1], Marek Szklarczyk[1], Ludomira Granicka[2], Magdalena N. Borkowska[2]

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In this presentation we are going to show the results on polyelectrolyte multilayers systems (PEM, polyeletrolyte membranes) studies. Obtained in the layer-by-layer self-organization process (LbL) membranes can be proposed for several applications. One of the most spectacular is the use of PEM in cell transplantation. Encapsulated cells are protected against the immune attack by recipient organism. The respective polyelectrolyte properties which forms the multilayer system has a large impact on the PEM character, while the used membrane will influence the cell, affecting its shape, or changing the form and strength of interaction with the environment. Atomic Force Microscopy (AFM) can help us to characterize several properties of the produced multilayers. The

morphology and thickness determination for multilayers based on poly-L-lysine, polyethylenimine and polystyrene sulfonate will be discussed.

15:45	Oral

Narzędzia dla Nanotechnologii

Ignacy Mościcki, Grzegorz Kaszyński

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Firma IGHT jest młodą firmą założoną przez ludzi zafascynowanych nowoczesnymi rozwiązaniami sprzętu laboratoryjnego, a w szczególności służącego nanotechnologii. Ta fascynacja uwidacznia się w doborze sprzętu, jaki znajduje się w naszej ofercie. Dokładamy wszelkich starań, aby rozwiązania, które sprowadzamy do Polski były jedyne w swoim rodzaju, najnowocześniejsze i najbardziej innowacyjne w swojej klasie. Doskonałym przykładem są dwaj nasi najwięksi partnerzy: NT-MDT oraz HYSITRON.

NT-MDT jest producentem systemów SPM (Scanning Probe Microscopy), który wyróżnia się unikalnym podejściem do swoich urządzeń. Są to systemy modułowe: można rozpocząć od prostej głowicy SPM i rozwinąć system do ponad czterdziestu technik SPM, mikroskopii optycznej, konfokalnej laserowej, a nawet badań TERS (Tip-Enhanced Raman Spectroscopy). Pozwala to na tworzenie doskonałych, uniwersalnych laboratoriów, które można przekonfigurować z badań np. materiałowych na chemiczne lub biologiczne w ciągu paru minut.

Hysitron jest liderem w badaniach właściwości mechanicznych w skali mikro i nano (zarówno nm jak i nN), a ich urządzenia wykraczają daleko poza standardowe pojęcie nanoindentacji. Systemy TriboIndenter pozwalają na prawdziwe ilościowe pomiary nanomechaniczne, a nie wyłącznie badania jakościowe. Z kolei PicoIndenter przenosi pomiary właściwości mechanicznych w świat mikroskopii elektronowej.

W naszej prezentacji skupimy się na zaprezentowaniu praktycznych zastosowań oferowanych przez nas urządzeń. Pokażemy najnowsze publikacje oraz wyniki badań i skupimy się na aplikacjach biologicznych, farmaceutycznych oraz medycznych.

NANOFORCE Presentations

Monday afternoon, 17 September, 16:00

16:00	Invited oral

Nanocząstki srebra, potencjał i bezpieczeństwo.

Tomasz Niemiec[1], Marta Grodzik[1], Ewa Sawosz Chwalibóg[1], Agnieszka Muszyńska[1], Andrzej Chwalibóg[2]

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Nanocząstki srebra są jednym z najbardziej skomercjalizowanych produktów nanobiotechnologii.

Świętują swoje triumfy na rynku chemicznym, spożywczym, kosmetycznym, odzieżowym, a także wśród asortymentu artykułów gospodarstwa domowego. Przemysł medyczny stosunkowo opornie korzysta z potencjału nanocząstek srebra, co w dużym stopniu wynika z udokumentowanej badaniami wiedzy na temat ich toksyczności. Należy jednak pamiętać, że w wielu obszarach medycyny nigdy nie zrezygnowano ze stosowania związków srebra, a nowe nano-odpowiedniki mając większą powierzchnię oddziaływania i tym samym mniejszą koncentrację efektywną oraz mniejszą toksyczność stają się bezkonkurencyjne (Sintubin i in., 2012).

Pomimo dużych ograniczeń do dzisiaj azotan srebra wykorzystywany jest do kauteryzacji naczyń krwionośnych lub ziarniaków (*granuloma*), obliteracji jamy opłucnej czy dezynfekcji oczu w zabiegu Credego. Z kolei sulfadaizyna srebra od lat sprawdza się w powszechnym leczeniu oparzeń. Obecnie metaliczne srebro o rozmiarach nano podbija potencjał i zakres aplikacyjny tego pierwiastka w medycynie i biologii. Już od dziesięciu lat komercyjnie sprzedawane są opatrunki z powłoką nanokrystaliczną, znajdując szerokie zastosowanie w leczeniu ran pooparzeniowych, martwicy naskórka, przewlekłych owrzodzeń i pęcherzyc. Działanie antybakteryjne wykazuje obiecujące efekty w walce z niebezpiecznymi i opornymi na antybiotyki szczepami szpitalnymi. Nanocząstki srebra wchodzące w skład cewników, kateterów, implantów czy cementu stosowanego w chirurgii kości zwiększają istotnie antymikrobiologiczne bezpieczeństwo ich użytkowania (Chaloupka i in., 2010). Szczególne zainteresowanie nanocząstki srebra wzbudziły w branży chemii gospodarczej, której asortyment nie tylko stosowany jest do odkażania pomieszczeń szpitalnych ale także na dużą skalę stosowany jest w utrzymaniu prawidłowych warunków zoochigienicznych w budynkach inwentarskich. Na etapie eksperymentów znajduje się obecnie ocena interakcji nanocząstek srebra z mikroflorą przewodu pokarmowego zwierząt gospodarskich (Sawosz i in., 2007; Chwalibog in., 2010; Sawosz i in., 2011). Konieczność poszukiwania bezpiecznej alternatywy dla wycofanych po 2006 roku antybiotykowych stymulatorów wzrostu była okazją do prac nad nanokoloidami srebra. Także szerszy aspekt intensyfikacji produkcji zwierzęcej wydaje się być wysoce kompatybilny z biopotencjałem nanotechnologii (Sawosz i in., 2010; Sikorska i in., 2010).

Nie powinien więc dziwić fakt, że skomercjalizowane nanosrebro stało się szerokodostępne, a ekspozycja na nie ludzi i zwierząt jest jeszcze bardziej powszechna i permanentna. Konsekwencją tego jest większy dostęp srebra do tkanek i komórek żywych organizmów (Lem i in. 2012).

Pomimo ponad dziesięcioletnich intensywnych badań, naukowcom nie udało się wypracować wspólnego konsensusu w sprawie bezpieczeństwa stosowania nanocząstek srebra. Fakt ten nie tylko wynika z niedopracowania standardów metod produkcji i wytwarzanego asortymentu ale także z rezultatów wielu badań stwierdzających między innymi neurotoksyczność nanocząstek srebra zarówno *in vitro* jak i *in vivo* (Chen i in., 2008; Lem i in., 2012). Zatem konieczny jest monitoring producentów i produktów zawierających srebro oraz jak najszybsze wypracowanie standardów i metod ich restrykcji. Z drugiej strony, winno wymagać się od biotechnologów "konstruowania" bardziej bezpiecznych form nanocząstek, nie tylko dla ludzi i zwierząt ale także dla środowiska, aby wynikające z tego korzyści ustanowiły XXI wiek, wiekiem nanobiotechnologii.

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16:20	Oral

Leveraging potential - open access to nano technologies for bio applications

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The trend towards personalised health care and the demands of an aging society provide a growing market for innovative solutions with nano and/or micro enabled components. EUMINAfab offers technologies for prototype development of individual components or to R&D investigations related to Point of Care Systems and lab-on-a-chip solutions.

This includes fabrication of components and moulds with the possibility of short series replication, e.g. components for medical applications from metals, ceramics and polymers, the coating of tools and components also fabrication of micro fluidic channels and through holes for disposable devices.

Surface functionalisation of polymers, metals ceramics and bio-patterning of substrates (e.g. scaffolds), is also possible.

In the area of biosensors applications are invited for functional patterning of substrates combining different (bio-compatible) materials such as polymers and metals. Additionally, nano patch clamping and imaging of components is possible.

This paper presents an overview of the types of application found for the life science area and the maturity of the types of tasks requested .

Aim/Introduction

Nano and Microsystems technologies are widely accepted to be cross cutting technologies with a relevance for application in a broad range of areas. Innovative ideas based on solutions using micro and nano fabrication technologies require access not only to high-end equipment but also the essential highly skilled personnel. It is not possible for SMEs or even most research departments to justify investment in a comprehensive range of technologies and trained personnel, especially when the need is to try out the feasibility of a new idea or develop a one off tool.

European Research Infrastructures (ERIs) aim to overcome these barriers by enabling open access to such high-end technologies. However, newly founded ERIs face the challenge of finding users for their equipment. Personal contacts between technologists resident at the infrastructure and their colleagues from outside, as well as contacts with relevant networks, technology platforms and associations give an important starting point for advertising the services. However questions are still to be raised as to how best to market the availability of technologies and expertise to a wider but still relevant community; a community which in fact may not be familiar with the capabilities of the technologies offered to find solutions to their challenging problems, or indeed generate new ideas.

EUMINAfab (www.euminafab.eu) is a European Research Infrastructure funded under the EU FP7 Capacities Specific Programme. The focus of this ERI is on multimaterial micro and nano fabrication and characterization technologies. An important project aim is to increase the realization in the wider MNT community of the possibilities created by the use of high-end micro and nano equipment. The environment of open innovation allows not only the no-fee access to the technologies but also the necessary technology expertise and advice. The FP7 funding enables this infrastructure to offer users access to technologies and the related expertise in micro and nano patterning, thin film deposition, replication and characterization. The aim is to facilitate the use of high end nano and micro technologies to enable next generation products.

EUMINAfab began services as a Technology platform – that is a group of partners which marketed the technologies in the areas of micro nano structuring, thin films, replication and characterisation. Since the launch of user access to EUMINAfab in September 2009 more than 100 proposals have been received. The technologies in the portfolio are, especially when considered with the skills of the operators, unique capabilities within Europe.

As a Consortium it was necessary to market the facilities and indeed to show the impact and innovation relevance of the technologies. Therefor it was necessary to evaluate the User projects and map the maturity of the technologies employed.

Even though users are required to publish the results of their work it was not possible to use normal impact factors such as numbers of publications to determine the success of EUMINAfab as a ERI, since it takes many months or even longer to reach a publication. Additionally the work typically carried out in EUMINAfab is usually part of a larger project – a part solution of a future publication or patent, this increases the time need to reach publication. The aim was therefor to investigate the application relevance of the proposals received and to in turn use this to reach new user communities, and consequent to this assemble the complete value chain from material , technology task made, sub application are and main application area.

Presentation of the Laboratory of Nanostructures for Photonics and Nanomedicine, Center of Bio-Nanomaterials, CePT

Monday afternoon, 17 September, 16:40

16:40	Invited oral

Presentation of the Laboratory of Nanostructures for Photonics and Nanomedicine, Center of Bio-Nanomaterials, CePT

Witold Łojkowski[1,2], Tadeusz Chudoba[1], Aleksandra Kędzierska[1], Sylwia Kuśnieruk[1], Iwona E. Malka[1,3], Jan Mizeracki[1], Agnieszka Opalińska[1], Adam Presz[1], Dariusz Smoleń[1], Joanna Sobczyk[1], Anna Swiderska - Sroda[1], Jacek Wojnarowicz[1]

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The research infrastructure, research team and the research programs of the Laboratory of Photonics and Nanomedicine, Center for bio-nanomnaterials, CePT will be presented.

POSTERS, FOOD and DISCUSSIONS

Presentation of the Laboratory of Nanostructures for Photonics and Nanomedicine, Center of Bio-Nanomaterials, CePT

Monday afternoon, 17 September, 17:00

17:00	Poster	C1

Badania rozkładu wielkości cząstek i potencjału zeta w Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny CePT

Agnieszka Opalińska[1,2], Jacek Wojnarowicz[1], Aleksandra E. Kędzierska[1], Witold Łojkowski[1]

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Kierunki badawcze Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny IWC PAN skoncentrowane są na syntezie nanoproszków, ich charakterystyce oraz możliwości zastosowania ich w medycynie, optyce, optoelektronice, farmacji i kosmetyce.

Poprawna charakterystyka nanometrycznych proszków jest kluczowym etapem w technologii otrzymywania nanomateriałów. Ze względu na rozmiar ziaren nanometrycznych (średnia wielkość ziarna do 100nm) tradycyjne metody charakteryzacji mogą dawać nieprawidłowe wyniki.

Specjalizujemy się między innymi w badaniach stabilności roztworów koloidalnych oraz w pomiarach wielkości cząstek/ aglomeratów w tych roztworach. Pomiary te wykonujemy na poniższych

urządzeniach:

Analizator rozkładu wielkości oraz potencjału zeta z automatycznym systemem titracji MPT-2– Zetasizer NanoSeries, MALVERN.

Zasada działania urządzenia oparta jest na technologii dynamicznego rozproszenia światła (Dynamic Light Scattering - DLS) i może mierzyć trzy wielkości charakteryzujące cząsteczki czy molekuły w dyspersjach cieczowych: wielkość cząsteczek, potencjał zeta, masę cząsteczkową.

Analizator rozkładu wielkości cząstek– NS500, NANOSIGHT.

Zasada działania urządzenia opiera się na bezpośredniej technice NTA „Nanoparticle Tracking Analysis" detekcji i wizualizacji indywidualnych nanocząstek w zawiesinie, która dostarcza informacji o rozmiarze, dystrybucji rozkładu i koncentracji cząstek.

W Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny IWC PAN zajmujemy się charakterystyką nanoproszków otrzymanych przez nasz zespół, jak również świadczymy usługi badawcze.

17:00	Poster	C2

Wpływ parametrów materiałowo-technicznych na właściwości optycznego czujnika tlenu na bazie nano-ZrO_2

Anna Swiderska - Sroda, Mateusz Szymański, Witold Łojkowski, Agnieszka Opalińska

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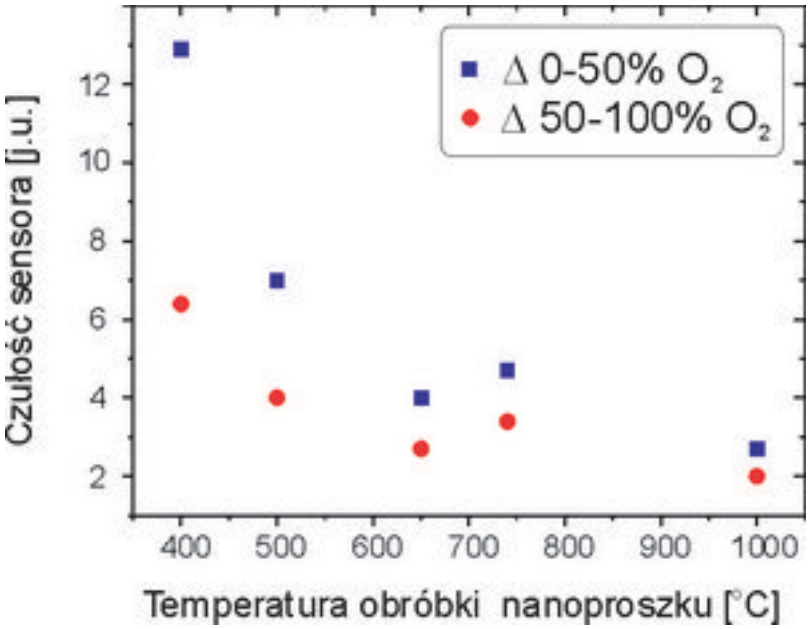
Kontrola zawartości tlenu w atmosferze jest niezbędna w różnych dziedzinach życia ludzkiego, na przykład w anestezjologii, zabezpieczeniach pracowników pracujących w pomieszczeniach/ przestrzeniach do których dostęp powietrza jest ograniczony (górnicy), czy narażonych na działanie gazów trujących (praca w kanałach). Czujniki tlenu są również konieczne dla prawidłowej pracy różnego typu urządzeń np. silników spalinowych, pieców, hutniczych.

W Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny IWC PAN podjęto badania nad nowym sensorem tlenu, pracującym w zakresie od temperatury pokojowej do 400°C. Bazą dla planowanego rozwiązania było zarejestrowane w trakcie wcześniejszych eksperymentów, zjawisko czułości luminescencji nanokrystalicznego ZrO_2 na udział tlenu w otaczającej atmosferze. Przeprowadzono cykl komplementarnych badań, które pozwoliły na ustalenie optymalnych parametrów materiałowych oraz konstrukcyjnych urządzenia. Analizowano wpływ składu chemicznego nanoproszku sensora, w zakresie rodzaju i ilości stosowanej domieszki, na przykład Tb, Gd, Eu w przedziale 1-10 mol%. Badano warunki obróbki proszku stosując wygrzewanie w zakresie temperatur 400-1000°C. Na rysunku poniżej pokazano wpływ temperatury obróbki nanoproszku ZrO_2: 8 mol%Eu na czułość sensora. W konsekwencji, obecnie jako materiał na sensor stosowany jest dwutlenek cyrkonu domieszkowany 8 mol% Europu, wygrzewany w temperaturze 400°C. Skonstruowano również prototyp urządzenia czujnika. Zbudowany na bazie komercyjnych, tanich podzespołów, autorski sensor ciśnienia parcjalnego tlenu wykazuje dużą czułość już od temperatury pokojowej. Niska temperatura pracy jest istotną zaletą tego urządzenia. W prezentowanej pracy przedstawione będą wybrane wyniki badań, uzyskane w trakcie tworzenia nowego czujnika tlenu.



17:00	Poster	C3

Characteristic of hydroxyapatite dense nanoceramic produced by The High Pressure Consolidation

Elżbieta Pietrzykowska, Aleksandra E. Kędzierska, Dariusz Smolen, Tadeusz Chudoba, Witold Łojkowski

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Regenerative medicine is an emerging multidisciplinary field. The aim is to restore, maintain or enhance tissues and hence organ functions. Nowadays bioactive scaffold is developing. Their's role is to mimic biological system and initiate the regeneration of tissue in the body. However, currently available materials do not satisfy every clinical application, so t**here is still a need for new materials which can stimulate the body's own regenerative mechanism and heal tissue.**

As it is known bones tissue is consisted of biological systems, which are built up from nanoscale biomolecules. Therefore, nanomaterials can be a key for radical improvement in medicine. The Institute of High Pressure Physics of the Polish Academy of Sciences (IHPP) has successfully developed the High Pressure Consolidation Technology (HPCT), which gives nanograin size ceramic. The hydroxyapatite nanoceramics produced in IHPP offers great oportunity for obtaining bioactive and resorbable scaffolds.

The HPCT method allows to produce dense ceramic by using extreme high pressure and low temperature. The process conditions are high pressure in range from 4 to 8GPa and temperature from 24 to 500ºC. This conditions cause the acceleration of the driving force for sintering and reduce pores in the material. Due to this method the nanopowder structure is maintain. The nanopowder

structure is significance for material's bioactivity and resorbability.

The mechanical properties of nanoceramic was investigated. The obtained material is characterized by 8.3 GPa nanohardness, 124 GPa Young's Modulus. During the process, the grain size is maintained in the range from 20−40 nm.

These materials are attracting interest in regenerative medicine because of theirs nanostructure which gives good bioactivity, resorbability and mechanical prosperities.

Acknowledgements: The authors wish to acknowledge the financial support from the project SONOSCA.

17:00	Poster	C4

Badania nanoproszków przy pomocy technik DSC-TG/ QMS-FTIR w Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny CePT

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Specjalizacją Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny Instytutu Wysokich Ciśnień (PAN) są synteza i kompleksowe badania nanomateriałów. Prace w tym obszarze wymagają zastosowania nowoczesnych, zaawansowanych aparatów pomiarowych. Jednym z nich jest system do badań stabilności temperaturowej, zakupiony w roku 2010 dzięki współfinansowaniu z europejskiego projektu CePT (Centre for Preclinical Research and Technology). Jest to układ złożony z trzech komplementarnych urządzeń. Główną częścią układu jest aparat do jednoczesnej analizy termicznej (Simultaneous Thermoanalysis-STA) firmy Netzsch, łączący dwie techniki badawcze: termograwimetrię (Thermogravimetry-TG) i skaningową kalorymetrię różnicową (Differential Scanning Calorymetry-DSC). Urządzenie to połączone jest z dwoma spektrometrami: masowym i podczerwieni (Quadrupole Mass Spectrometer- QMS i Fourier Transform Infrared Spectroscopy-FTIR), umożliwiającymi analizę składu chemicznego gazów wydzielających się z badanej próbki w trakcie jej ogrzewania. Tak zaprojektowane stanowisko badawcze daje możliwość realizacji specjalistycznych badań, niezbędnych w charakteryzacji nanomateriałów. Na przykład, dla nano-hydroksyapatytu ogrzewanego do 1300ºC rejestrowano zarówno desorpcję wody jak i przemianę fazową, co jest niezwykle trudne do zaobserwowania innymi technikami badawczymi. Badania DSC-TG-QMS wykorzystano do ustalenia warunków obróbki termicznej nano–ZrO_2, stosowanego jako czujnik tlenu. W przypadku materiału zdolnego do magazynowania wodoru w fazie stałej (nanokrystaliczny MgH_2 z dodatkami katalitycznymi) stwierdzono, że wodór nie jest jedynym gazem, który się uwalnia podczas dekompozycji próbki. Zaprezentowane przykłady, które stanowią jedynie część badań prowadzonych w Laboratorium Nanostruktur, wskazują na istotność, a zarazem uniwersalność metody DSC-TG-QMS-FTIR w badaniach nanomateriałów.

17:00	Poster	C5

Solwotermalna synteza domieszkowanego nanotlenku cynku dla projektu NanoFATE. Otrzymywanie wodnych nanozawiesin domieszkowanego tlenku cynku.

Jacek Wojnarowicz, Agnieszka Opalińska, Dariusz Smoleń, Sylwia Kuśnieruk, Tadeusz Chudoba, Witold Łojkowski

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W dzisiejszych czasach nanotlenek cynku cieszy się coraz większą popularnością. Kwestią wciąż nierozstrzygniętą jest obieg nanokrystalicznego tlenku cynku w przyrodzie po zakończeniu czasu użytkowania kosmetyków czy wyrobów wzbogaconych nanoproszkiem ZnO. Zdefiniowanie „cyklu życia" tlenku cynku w przyrodzie, w szczególności wpływ formy nanokrystalicznej na życie organizmów żywych jest przedmiotem projektu NanoFATE. Celem tego projektu jest określenie wpływu toksyczności nanocząstek pochodzących z powszechnie stosowanych produktów na środowisko naturalne.

Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny IWC PAN jest ekspertem w syntezach domieszkowanych nanocząstek o małym rozkładzie wielkości ziaren przy użyciu technologii solwotermalnej syntezy mikrofalowej (MSS). Technologia MSS pozwala na dokładne kontrolowania parametrów syntezy nanocząstek takich jak czas reakcji, temperatura i ciśnienia. W ramach projektu NanoFATE laboratorium odpowiedzialne jest za dostarczenie domieszkowanego tlenku cynku. Tlenek cynku domieszkowany jonami kobaltu o kontrolowanej zawartości domieszki i rozkładzie wielkości cząstek jest otrzymywany za pomocą reaktora mikrofalowego MSS w procesie opisanym poniższym równaniem:

$$z(CH_3CO_2)_2Zn + y(CH_3CO_2)_2Co \rightarrow Zn_{(1-x)}Co_xO$$

Synteza nanotlenku cynku domieszkowanego kobaltem przebiega w roztworze glikolu etylenowego w czasie 25 minut. Powierzchnia właściwa domieszkowanego ZnO wynosi 30-50 m2/g, a wielkość krystalitów mieści się w zakresie od 20 do 50 nm.

Opracowano technologię otrzymywania wodnych zawiesin nanotlenku cynku. Wielkość aglomeratów ZnO:Co wynosi od 60 do 150 nm, wartość potencjału zeta zawiesin mieści się w przedziale od 24 do 34 mV. Zawiesiny charakteryzują się wysoką czystością. Otrzymuje się je bez użycia środków powierzchniowo czynnych oraz stabilizatorów.

17:00	Poster	C6

Badanie stabilności i rozpuszczalności zawiesin z nano ZnO i nano ZnO:Co otrzymanych w Laboratorium Nanostrutur dla Fotoniki i Nanomedycyny CePT

Jacek Wojnarowicz, Agnieszka Opalińska, Dariusz Smoleń, Sylwia Kuśnieruk, Tadeusz Chudoba, Witold Łojkowski

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Kwestią wciąż nierozstrzygniętą/ niezdefiniowaną jest obieg nanokrystalicznego tlenku cynku w przyrodzie po zakończeniu czasu użytkowania kosmetyków czy wyrobów wzbogaconych nanoproszkiem ZnO. Zdefiniowanie „cyklu życia" tlenku cynku w przyrodzie, w szczególności wpływ formy nanokrystalicznej na życie organizmów żywych jest przedmiotem projektu NanoFATE. NanoFATE jest projektem europejskim realizowanym przez 12 partnerów z 9 krajów europejskich. Celem projektu NanoFATE jest określenie wpływu toksyczności nanocząstek pochodzących z powszechnie stosowanych produktów na środowisko naturalne. Od wprowadzenia ich do produktu, poprzez przetwarzanie odpadów do ich ostatecznego miejsca składowania i utylizacji. Jednym ze sposobów śledzenia nanocząstek w środowisku naturalnym jest ich znakowanie poprzez domieszkowanie wybranymi jonami.

Od ponad 10 lat Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny rozwija solwotermalną technologie wytwarzania nanocząstek. Proces opiera się na technologii MSS Mikrofalowej Solwotermalnej Syntezy. Prekursory reakcji (roztwory, zawiesiny) są zamknięte w zbiorniku ciśnieniowym i w wyniku ogrzewania energią mikrofalową temperatura rośnie powyżej punktu wrzenia. Proces MSS pozwala zapobiegać zanieczyszczeniu syntezy, poprzez szczelne zamknięcie naczynia reakcyjnego wykonanego z chemicznie obojętnego materiału, dzięki czemu otrzymujemy ultra-czysty produkt. W reaktorze mikrofalowym dodatkowo występuję efekt mieszania, dzięki czemu otrzymany produkt jest jednorodny. Innowacją technologii MSS jest możliwość kontrolowania wielkości krystalitów nanocząsteczek w wąskim rozkładzie wielkości. ZnO-NPS, ZnO:Co-NPS są syntetyzowane w reakcjach, które można wyrazić następująco:

$$(CH3COO)_2Zn \rightarrow ZnO$$

$$z(CH3CO2)2Zn + y(CH3CO2)2Co \rightarrow Zn(1-x)CoxO$$

Dzięki unikalnym na skale światową reaktorą mikrofalowych do solwotermalnych syntez, jesteśmy w stanie syntetyzować nanocząstki ZnO o kontrolowanych właściwościach. Powierzchnia właściwa otrzymanych nanoproszków ZnO naszych reaktorach mieści się w zakresie od 9 do 53 m2/g a wielkość ziarna wynosi od 20 do 150 nm. Na potrzeby projektu NanoFATE opracowano technologię otrzymywania wodnych zawiesin nanotlenku cynku. Wielkość aglomeratów ZnO, ZnO:Co wynosi od 60 do 150 nm, wartość potencjału zeta zawiesin mieści się w przedziale od 24 do 34 mV. Zawiesiny koloidalne charakteryzują się wysoką czystością. Otrzymuje się je bez użycia środków powierzchniowo czynnych oraz stabilizatorów.

Otrzymane zawiesiny scharakteryzowano pod wzgledem stabilności, rozpuszczalności, zmian wielkości aglomeratów, zmian właściwości fizykochemicznych. Badania rozpuszczalności nanoczastek przeprowadzane przy użyciu analizy kolorymetrycznej. Metoda kolorymetryczna polega na oznaczeniu zawartości danej substancji na podstawie intensywności zabarwienia jej roztworu i porównaniu z barwą roztworów wzorcowych tej substancji. Przy badaniach stabilności i zmian wielkości aglomeratów wykorzystywano analizatory DLS (Dynamic Light Scattering - DLS) oraz NTA (Nanoparticle Tracking Analysis).

17:00	Poster	C7

Nanoparticle Tracking Analysis (NTA): Applications in biotechnology

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NanoSight's Nanopartilce Tracking Analysis (NTA) is a technique for characterising nanoparticles in its native suspension.

This technique provides real time and multi-parameter characterisation of nanoparticles (down to 10-20nm diameter) achieved by video analysis of a laser illuminated sample through conventional microscopy. Analysis of the Brownian motion of individual particles in the defined field of view allows their size to be determined as well as their concentration.

Each particle is individually but simultaneously analysed for i) size; ii) high resolution particle size distribution profile; iii) light scattering properties (refractive index); iv) fluorescence allowing phenotyping or speciation; v) zeta potential through the application of electric fields and, vi) concentration (particles per millilitre) of any given size class of nanomaterial. This has a broad range of applications in biotechnology.

Exosomes and microvesicles: Fluorescently labelled exosomes can be successfully tracked and analysed allowing phenotyping of subpopulations in complex sample types. NTA is capable of selectively analysing such populations through the use of antibody-mediated fluorescent labelling. This approach allows the user to detect, analyse and count only the specific nanoparticles to which the fluorescently-labelled antibody binds, with background non-specific particulates being excluded.

Drug delivery: The ability to characterise polymer nanoparticles used for systematic drug delivery is an important tool in biomedicine. For example, NTA has recently been applied for monitoring drug renal retention, the specific targeting properties of drugs and the performance of newly developed medicines.

Nanotoxicology: NTA is widely used as a preliminary characterisation technique prior to exposure. This has been applied to a range of materials including metals, metal oxides and polymers.
The ability to characterise and count individual nanoparticles ensures NTA will continue to be a valuable tool in a range of biotechnological applications and holds an important role in the advancements of this field.

17:00	Poster	C8

Synteza nano-ZnO przy pomocy reaktora Mettler Toledo w Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny CePT.

Sylwia Kuśnieruk[1], Jacek Wojnarowicz[2], Witold Łojkowski[1,2], Tadeusz Chudoba[2], Agnieszka Opalińska[2,3], Dariusz Smoleń[4]

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Tlenek cynku jest stosowany zarówno w przemyśle jak i w medycynie.

Jedną z głównych zalet nano-ZnO jest możliwość kontrolowania rozmiaru cząsteczek ZnO, ponieważ jak wskazują badania, właściwości cytotoksyczne cząsteczek ZnO zależą od ich rozmiaru. Mniejsze cząsteczki wykazują większą toksyczność [1].

Nano materiały ZnO o rozmiarze większym niż 100 nm uważane są za relatywnie biokompatybilne, oraz za bezpieczne, co sprawia, że mogą być stosowane jako nośniki leków. Jedną z głównych przewag w rozważaniu nano-ZnO do użycia przeciw nowotworom jest ich immanentna preferencyjna cytotoksyczność przeciwko komórkom raka in vitro [2,3]. Właściwości elektrostatyczne nano-ZnO są kolejną użyteczna cechą do zastosowań biomedycznych.

Reaktor Mettler Toledo służy do opracowania procesów i reakcji chemicznych. Za pomocą tego urządzenia można optymalizować planowany proces oraz charakteryzować jego przebieg. Kontrola czasu syntezy oczekiwanego produktu daje możliwość decydowania o każdym etapie prowadzonego procesu.

Za pomocą reaktora Mettler Toledo otrzymano nano-ZnO. Synteza w Reaktorze Mettler Toledo pozwala na obserwowanie poszczególnych etapów prowadzonej syntezy wraz ze zmianą temperatury. Zaobserwowano, że produktem pośrednim procesu otrzymania tlenku cynku jest wodorotlenek cynku. Nano-ZnO otrzymano w temperaturze około 160°C.

Uzyskany produkt scharakteryzowano różnymi metodami fizykochemicznymi takimi jak: proszkowy dyfraktometr rentgenowski, analizator DLS, SEM (Skaningowa Mikroskopia Elektronowa). Za pomocą dyfrakcji rentgenowskiej zbadano skład fazowy otrzymanego proszku. Analizator DLS pozwolił zmierzyć wielkość cząsteczek oraz potencjał zeta. Rozkład wielkości aglomeratów dla uzyskanego nano-ZnO to około 80 nm. Do wizualizacji otrzymanego proszku wykorzystano Skaningową Mikroskopię Elektronową. Tak otrzymany nano-ZnO może mieć zastosowanie w medycynie jako nośnik leków.

1.Hanley C, Thurber A, Hanna C, et al. The influences of cell type and ZnO nanoparticle size and immune cell cytotoxicity and cytokine induction. Nanoscale Res Lett 2009;4:1409–20.

2.Hanley C, Layne J, Punnoose A, et al. Preferential killing of cancer cells and activated human T cells using zinc oxide nanoparticles. Nanotechnology 2008;19:295103–13.

3. Wang H, Wingett D, Engelhard MH, et al. Fluorescent dye encapsulated ZnO particles with cellspecific toxicity for potential use in biomedical applications. J Mater Sci Mater Med 2009;20:11–22.

17:00	Poster	C9

Application of the Zeiss Ultra Plus scanning microscopy in bio-nanotechnology

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Selected applications of the Zeiss Ultra Plus scanning electron microscope to study bio-materials in the Laboratory of Nanostructures for Photonics and Nanomedicine will be presented.

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END of the DAY
Monday evening, 17 September, 19:00

Tuesday, 18 September

REGISTRATION NanoInfoDay
Tuesday morning, 18 September, 8:30

NanoForCE InfoDay Poster session
Tuesday morning, 18 September, 9:00
NanoForCE InfoDay

9:00	Oral

Otwarcie NanoInfoDay, Powitanie

Witold Łojkowski

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9:00	Poster	N1

Nanotechnologia bezpieczna dla ludzi i przyjazna środowisku

Anna Swiderska - Sroda[1], Joanna Sobczyk[1], Witold Łojkowski[1,2]

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W ciągu ostatnich dwóch dekad nastąpił ogromny rozwój nanotechnologii. W tym samym czasie podstawowe pytania na temat wpływu nanomateriałów na środowisko i ludzi pozostają bez odpowiedzi. Dlatego władze Unii Europejskiej podjęły wielorakie działania, skierowane na bezpieczeństwo nanotechnologii. Rezultatem tych działań jest międzynarodowa / inter-dyscyplinarna współpraca w tematyce nano-bezpieczeństwa, podejmowana w ramach projektów europejskich.

Jednym z nich jest projekt Nanoforce, który w maju 2012 roku wszedł w drugi rok realizacji. Obecny etap prac poświęcony jest znalezieniu metod charakteryzacji nanoproszków, które z jednej strony wykażą unikalne właściwości nanostruktur, z drugiej zaś zweryfikują obawy w kwestii zagrożeń, jakie mogą powodować nanoproszki dla ludzkiego zdrowia oraz dla środowiska.

W ramach projektu Nanoforce podejmowane są prace nad: określeniem procedur oceny ryzyka,

określeniem standardów charakteryzacji nanoproszków, testami toksykologicznymi, oraz nad stworzeniem propozycji scenariusza toksykologicznego dla trzech wybranych nanomateriałów.

Partnerzy projektu wyselekcjonowali trzy nanoproszki, spośród tych, które są najbardziej popularne i najczęściej używane w nanoproduktach komercyjnych. Nanoproszki ZnO, nano-Ag, TiO_2 będą testowane w czterech laboratoriach partnerów projektu Nanoforce, między innymi w Laboratorium Nanostruktur IWC PAN. Badania obejmują także testy toksykologiczne, które pozwolą stwierdzić, jaki jest wpływ tych nanoproszków na środowisko i ludzkie zdrowie.

W Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny w Instytucie Wysokich Ciśnień Polskiej Akademii Nauk będą prowadzone prace dotyczące charakterystyki mikrostrukturalnej nanoproszków. Laboratorium dysponuje nowoczesną aparaturą, która gwarantuje realizację tego zadania według najlepszych standardów w tej dziedzinie.

W niniejszej pracy zaprezentowane będą techniki badawcze dostępne i stosowane w Laboratorium Nanostruktur do oceny nanomateriałów, jak również obszary specjalizacji laboratorium w dziedzinie nanotechnologii.

9:00	Poster	N2

Infrastruktura i specjalizacje nanotechnologii w Polsce na podstawie wyników ankiety NANOFORCE

Anna Swiderska - Sroda, Joanna Sobczyk, Iwona E. Malka, Witold Łojkowski

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Rynek nanoproduktów osiągnął wysoki stopień zaawansowania i spodziewany jest dalszy jego wzrost, zarówno pod względem różnorodności asortymentu, jak i oczekiwanych dochodów. W tym świetle, nanotechnologia jest szansą rozwoju ekonomicznego Europy, szczególnie krajów Europy Centralnej. Dlatego, podejmowanych jest wiele inicjatyw, które mają zabezpieczyć ważną pozycję Europy na globalnym rynku nanotechnologii, oraz przyspieszyć rozwój europejskiego nano-biznesu. Przykładem takich działań jest projekt NANOFORCE, który ma wspomóc rozwój nanotechnologii w poszczególnych regionach Europy Centralnej poprzez aktywację współpracy pomiędzy przemysłem a jednostkami naukowymi, jak również pomiędzy instytucjami państwowymi i prywatnymi.

Jednym z zadań NANOFORCE jest rozpoznanie infrastruktury oraz specjalizacji nanotechnologicznych w krajach partnerów projektu, a także analiza stanu wiedzy i uregulowań prawnych w dziedzinie nano-bezpieczeństwa. W tym celu została przeprowadzona akcja ankietowa, którą objęto zarówno ośrodki nanukowo-badawcze, jak i producentów zainteresowanych nanotechnologią.

W Polsce badanie spotkało się z dużym zainteresowaniem wśród instytucji naukowych, oraz bardzo nikłym w sektorze przemysłowym. Potwierdza to opinię o pilnej potrzebie popularyzacji nanotechnologii wśród krajowych producentów, w tym ukazanie im potencjału dochodowego tej grupy wyrobów. Pozytywnym aspektem stanu nanotechnologii w Polsce jest silne zaplecze naukowo-badawcze, na bazie którego możliwy jest rozwój rodzimego nano-biznesu.

W oparciu o wyniki ankiety stwierdzono, że polską specjalnością w dziedzinie nanotechnologii są nano-metale i materiały kompozytowe. Zauważono przy tym, że znaczna część obszarów zaint-

eresowania naukowców nie pokrywa się z obszarami ważnymi dla reprezentantów przemysłu. Wskazuje to na konieczność ściślejszej współpracy pomiędzy tymi dwoma środowiskami, jak również na potrzebę zmiany systemu wyboru priorytetowych tematów badawczych tak, aby w większym stopniu były one powiązane z oczekiwaniami przemysłu.

Ważną częścią ankiety były zagadnienia dotyczące bezpieczeństwa w nanotechnologii. Okazało się, że według jednej trzeciej respondentów nanomateriały mogą stanowić zagrożenie dla zdrowia ludzi i środowiska, przy czym świadomość ta była wyższa w sektorze państwowym niż w prywatnym.

Pomimo, że wśród respondentów dominowała opinia, iż nanomateriały są bezpieczne, to większość z nich postulowała konieczność wprowadzenia uregulowań prawnych, dedykowanych tej grupie substancji. Oczekiwana jest w tym względzie ścisła współpraca pomiędzy środowiskami związanymi z nanotechnologią a jednostkami rządowymi, odpowiedzialnymi za tego typu działania.

W niniejszej pracy przedstawiony będzie stan nanotechnologii w Polsce, opisany na podstawie wyników wspomnianych wyżej badań ankietowych.

9:05	Invited oral

Nanoforce - wielki mały projekt

Krzysztof Łokaj

Polska Izba Pzemysłu Chemicznego (PIPC), Śniadeckich, Warszawa 00-654, Poland

e-mail: krzysztof.lokaj@pipc.org.pl

Myślenie w skali nano jest zupełnie nowym sposobem postrzegania świata. Tu obowiązują inne reguły gry i to zarówno w warstwie fizykochemicznych właściwości substancji, podejściu opinii publicznej do nanomatriałów, a także stanowionego prawa. Projekt Nanoforce ma przed sobą szereg ambitnych celów i zadań, których realizacja może mieć istotny wpływ na rozwój nanotechnologii w Europie Centralnej i całej Unii Europejskiej. Badania, wdrożenia, fundusz inwestycyjny i rekomendacje dla Komisji Europejskiej – tak wiele robimy w tak „malutkiej" sprawie.

9:25	Invited oral

Kryteria przystąpienia i zasady uczestnictwa w Nanoforce

Dariusz Bugajski

Polska Izba Pzemysłu Chemicznego (PIPC), Śniadeckich, Warszawa 00-654, Poland

e-mail: dariusz.bugajski@poczta.neostrada.pl

W trakcie wystąpienia przedstawione zostaną podstawowe kryteria, jakie muszą spełnić przedsiębiorstwa, ośrodki naukowe oraz inwestorzy, aby wziąć udział w Projekcie Nanoforce i móc skorzystać z jego bazy kontaktów oraz możliwości uczestniczenia w tworzonym Międzyregionalnym Nanotechnologicznym Funduszu Inwestycyjnym. Przybliżony zostanie także proponowany sposób współpracy pomiędzy prowadzącymi Projekt Partnerami a jego Uczestnikami.

9:45	Invited oral

Regulacje prawne w nanotechnologii

Andrzej Krześlak

Polska Izba Pzemysłu Chemicznego (PIPC), Śniadeckich, Warszawa 00-654, Poland

e-mail: andrzej.krzeslak@ichp.pl

Polska i Unia Europejska nie posiadają zbioru regulacji dedykowanych nanomateriałom i nanotechnologii w ogólności. Jeśli w ustawodawstwie pojawia się odniesienie do nanomateriałów, to niemal każdy sektor posiada odmienne normy i definicje. W tracie prezentacji przedstawione zostaną najważniejsze z obowiązujących zapisów oraz wstępne wnioski co do przyszłych rozwiązań.

NanoForCE InfoDay

Regulation in Nanotechnology
Tuesday morning, 18 September, 9:00
NanoForCE InfoDay
Chair: Anna Świderska-Środa

9:00	Oral

Otwarcie NanoInfoDay, Powitanie

Witold Łojkowski

Polish Academy of Sciences, Institute of High Pressure Physics (UNIPRESS), Sokolowska 29/37, Warszawa 01-142, Poland Instytut Wysokich Ciśnień PAN (IWC), Sokołowska 29/37, Warszawa 01-142, Poland

e-mail: wl@unipress.waw.pl

9:00	Poster	N1

Nanotechnologia bezpieczna dla ludzi i przyjazna środowisku

Anna Swiderska - Sroda[1], Joanna Sobczyk[1], Witold Łojkowski[1,2]

1. *Polish Academy of Sciences, Institute of High Pressure Physics (UNIPRESS), Sokolowska 29/37, Warszawa 01-142, Poland* **2.** *Instytut Wysokich Ciśnień PAN (IWC), Sokołowska 29/37, Warszawa 01-142, Poland*

e-mail: annas@unipress.waw.pl

W ciągu ostatnich dwóch dekad nastąpił ogromny rozwój nanotechnologii. W tym samym czasie podstawowe pytania na temat wpływu nanomateriałów na środowisko i ludzi pozostają bez odpowiedzi. Dlatego władze Unii Europejskiej podjęły wielorakie działania, skierowane na bezpieczeństwo nanotechnologii. Rezultatem tych działań jest międzynarodowa / inter-dyscyplinarna współpraca w tematyce nano-bezpieczeństwa, podejmowana w ramach projektów europejskich.

Jednym z nich jest projekt Nanoforce, który w maju 2012 roku wszedł w drugi rok realizacji. Obecny etap prac poświęcony jest znalezieniu metod charakteryzacji nanoproszków, które z jednej strony wykażą unikalne właściwości nanostruktur, z drugiej zaś zweryfikują obawy w kwestii zagrożeń, jakie mogą powodować nanoproszki dla ludzkiego zdrowia oraz dla środowiska.

W ramach projektu Nanoforce podejmowane są prace nad: określeniem procedur oceny ryzyka, określeniem standardów charakteryzacji nanoproszków, testami toksykologicznymi, oraz nad stworzeniem propozycji scenariusza toksykologicznego dla trzech wybranych nanomateriałów.

Partnerzy projektu wyselekcjonowali trzy nanoproszki, spośród tych, które są najbardziej popularne i najczęściej używane w nanoproduktach komercyjnych. Nanoproszki ZnO, nano-Ag, TiO_2 będą testowane w czterech laboratoriach partnerów projektu Nanoforce, między innymi w Laboratorium Nanostruktur IWC PAN. Badania obejmują także testy toksykologiczne, które pozwolą stwierdzić, jaki jest wpływ tych nanoproszków na środowisko i ludzkie zdrowie.

W Laboratorium Nanostruktur dla Fotoniki i Nanomedycyny w Instytucie Wysokich Ciśnień Polskiej Akademii Nauk będą prowadzone prace dotyczące charakterystyki mikrostrukturalnej nanoproszków. Laboratorium dysponuje nowoczesną aparaturą, która gwarantuje realizację tego zadania według najlepszych standardów w tej dziedzinie.

W niniejszej pracy zaprezentowane będą techniki badawcze dostępne i stosowane w Laboratorium Nanostruktur do oceny nanomateriałów, jak również obszary specjalizacji laboratorium w dziedzinie nanotechnologii.

9:00	Poster	N2

Infrastruktura i specjalizacje nanotechnologii w Polsce na podstawie wyników ankiety NANOFORCE

Anna Swiderska - Sroda, Joanna Sobczyk, Iwona E. Malka, Witold Łojkowski

Instytut Wysokich Ciśnień PAN (IWC), Sokołowska 29/37, Warszawa 01-142, Poland

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Rynek nanoproduktów osiągnął wysoki stopień zaawansowania i spodziewany jest dalszy jego wzrost, zarówno pod względem różnorodności asortymentu, jak i oczekiwanych dochodów. W tym świetle, nanotechnologia jest szansą rozwoju ekonomicznego Europy, szczególnie krajów Europy Centralnej. Dlatego, podejmowanych jest wiele inicjatyw, które mają zabezpieczyć ważną pozycję Europy na globalnym rynku nanotechnologii, oraz przyspieszyć rozwój europejskiego nano-biznesu. Przykładem takich działań jest projekt NANOFORCE, który ma wspomóc rozwój nanotechnologii w poszczególnych regionach Europy Centralnej poprzez aktywację współpracy pomiędzy przemysłem a jednostkami naukowymi, jak również pomiędzy instytucjami państwowymi i prywatnymi.

Jednym z zadań NANOFORCE jest rozpoznanie infrastruktury oraz specjalizacji nanotechnologicznych w krajach partnerów projektu, a także analiza stanu wiedzy i uregulowań prawnych w dziedzinie nano-bezpieczeństwa. W tym celu została przeprowadzona akcja ankietowa, którą objęto zarówno ośrodki nanukowo-badawcze, jak i producentów zainteresowanych nanotechnologią.

W Polsce badanie spotkało się z dużym zainteresowaniem wśród instytucji naukowych, oraz bardzo nikłym w sektorze przemysłowym. Potwierdza to opinię o pilnej potrzebie popularyzacji

nanotechnologii wśród krajowych producentów, w tym ukazanie im potencjału dochodowego tej grupy wyrobów. Pozytywnym aspektem stanu nanotechnologii w Polsce jest silne zaplecze naukowo-badawcze, na bazie którego możliwy jest rozwój rodzimego nano-biznesu.

W oparciu o wyniki ankiety stwierdzono, że polską specjalnością w dziedzinie nanotechnologii są nano-metale i materiały kompozytowe. Zauważono przy tym, że znaczna część obszarów zainteresowania naukowców nie pokrywa się z obszarami ważnymi dla reprezentantów przemysłu. Wskazuje to na konieczność ściślejszej współpracy pomiędzy tymi dwoma środowiskami, jak również na potrzebę zmiany systemu wyboru priorytetowych tematów badawczych tak, aby w większym stopniu były one powiązane z oczekiwaniami przemysłu.

Ważną częścią ankiety były zagadnienia dotyczące bezpieczeństwa w nanotechnologii. Okazało się, że według jednej trzeciej respondentów nanomateriały mogą stanowić zagrożenie dla zdrowia ludzi i środowiska, przy czym świadomość ta była wyższa w sektorze państwowym niż w prywatnym.

Pomimo, że wśród respondentów dominowała opinia, iż nanomateriały są bezpieczne, to większość z nich postulowała konieczność wprowadzenia uregulowań prawnych, dedykowanych tej grupie substancji. Oczekiwana jest w tym względzie ścisła współpraca pomiędzy środowiskami związanymi z nanotechnologią a jednostkami rządowymi, odpowiedzialnymi za tego typu działania.

W niniejszej pracy przedstawiony będzie stan nanotechnologii w Polsce, opisany na podstawie wyników wspomnianych wyżej badań ankietowych.

9:05	Invited oral

Nanoforce - wielki mały projekt

Krzysztof Łokaj

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9:25	Invited oral

Kryteria przystąpienia i zasady uczestnictwa w Nanoforce

Dariusz Bugajski

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e-mail: dariusz.bugajski@poczta.neostrada.pl

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Międzyregionalnym Nanotechnologicznym Funduszu Inwestycyjnym. Przybliżony zostanie także proponowany sposób współpracy pomiędzy prowadzącymi Projekt Partnerami a jego Uczestnikami.

9:45	Invited oral

Regulacje prawne w nanotechnologii

Andrzej Krześlak

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e-mail: andrzej.krzeslak@ichp.pl

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NanoForCE InfoDay

NANOFORCE- Nanotechnology for Chemical Enterprises
Tuesday morning, 18 September, 10:05
NanoForCE InfoDay
Chair: Joanna Sobczyk

10:05	Invited oral

Nanotechnologies – standardization issues

Maria Zielecka

Industrial Chemistry Research Institute (ICRI), Rydygiera 8, Warszawa 01-793, Poland

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As many authorities predict the applications of nanotechnologies will change all areas of life and will enable dramatic advances to be realized in all areas of communication, health, manufacturing, materials and knowledge-based technologies. Due to these facts there is an obvious need to provide industry and research the suitable tools to aid the development and application of those technologies. Moreover, the reliable measurement systems and evaluation protocols supported by well-founded and robust standards are needed for regulator and health and environmental protection agencies.

The answer on the development of nanotechnology was the establishment of the International Organization for StandardizationISO Technical Committee ISO / TC 229; Nanotechnology, IEC / TC 113 [3] Standards of nanotechnology devices and systems for electrical and electronic Electrotechnical International Commission and CEN / TC 352 Nanotechnology in the European Committee for Standardization. Committees ISO / TC 229 and CEN / TC 352 work closely together - according to the Vienna Agreement.

In Poland the normalization in the area of nanotechnology until 30 December 2011, dealt with two

technical committees TC 207 - Incremental machining and KT 60 - Power electronics and semiconductor devices. Increasing need of standardization in the field of nanotechnology in Technical Committee 207 justified the launch of the establishment of the technical committee especially focused on nanotechnologies. As a result of these actions, on 30 December 2011, Technical Committee 314 Nanotechnology was appointed, which took over responsibility for the standardization work carried out in the Technical Committee 207 in the field of nanotechnology.

Technical Committee 314 scope includes issues relating to the matter and processes at the nanoscale, typically, but not exclusively, of dimension less than 100 nm in one or more directions, where the properties depend on the dimensions enable new applications, use the properties of materials at the nanoscale to produce improved materials, equipment and systems using these new features, terminology and nomenclature, metrology and instrumentation, including specifications for reference materials, research methodologies, modeling and simulation, with based on scientific aspects of health, safety and environmental protection.

10:25 Oral

Toxicity of paints containing TiO_2 and ZnO nanoparticles after environmental exposure scenarios

Nina Bednarsek, Elsa Fabbretti, Urska L. Stangar

University of Nova Gorica, Nova Gorica 5000, Slovenia

e-mail: nina.bednarsek@gmail.com

Although the increasing use of the nanoparticles in many consumer products, there are distinct knowledge gaps regarding the potential release and exposure of manufactured nanomaterials. There is also an urgent need for more detailed studies on the environmental behaviour and fate of nano-based products. We tried to address these gap through our current research acitivites within the Central European project NANOFORCE. The objective of this study was the evaluation of the behaviour of different paints containing nano-sized TiO_2 and ZnO after distinct exposure scenarios: leachability release and UV exposure. The testing systems for toxicity were based on cytotoxicity (using MTT activity) and ecotoxicity (inhibition of luminescence in *Vibrio fisheri*). Characterization of leachates was performed by using CLS and PIXE in order to determine NPs size distribution and released amounts in the leachates. Some of the leachates, although containing NPs in small amounts, were cytotoxic and ecotoxic. In the case of ZnO-based NP paints, it seems that the toxicity of leachates was due to the presence of the dissolved Zn2+ ions and not of NPs.

10:40 Invited oral

NANOFORCE- Nanotechnology for Chemical Enterprises – how to link scientific knowledge to the business in the Central Europe

Sonja Hartl

BioNanoNet Research Association, Elisabethstraße 9-11, Graz 8010, Austria

e-mail: sonja.hartl@bionanonet.at

Introduction: The 30 months long NANOFORCE project, which is performed in 8 regions of Central Europe belonging to 7 Countries, is developed by national & regional chemistry associations

and R&D Centers of the Central Europe area. As result of the gradually increasing nanotechnology sector there is the necessity of a larger international cooperation and research's coordination to overcome disciplinary boundaries, to fill the gap between more and less experienced regions and to turn investments in R&D in industrial innovations.

One of the main objectives of NANOFORCE project is to improve the environmental, health and safety knowledge and develop a state-of-the-art report on regulations in the field of nanotechnology to identify needs and gaps and give possible recommendations for the European Commission. Therefore the first actions of the NANOFORCE project was the development of questionnaires for producers, downstream users, researchers and financiers with the goal to collect information about the nanotechnology sector in the Central Europe region concerning development, financial status, international cooperation within joint ventures, safety and nanotoxicology. The results achieved are presented in regional related reports on nanotechnology business as well as a state of the art report on existing safety procedures and nanotech related legislation.

The BioNanoNet Forschungsgesellschaft mbH is an Austrian network which combines a wide range of expertise in numerous disciplines of medical and pharmaceutical research in nanomedicine and nanotoxicology and acts as work package leader in the NANOFORCE project. Additionally the BioNanoNet functions as coordinator of the European Center for Nanotoxicology (EURO-NanoTox), an Austrian example of a European hub in nanotoxicology. Within the aim of EURO-NanoTox, the BioNanoNet offers regulatory expertise, dissemination and communication service, scientific reviewing of testing methods and a panel of standardized in vitro and in vivo toxicity tests, with the overall goal of driving innovative interdisciplinary research.

Focus: The general objective of NANOFORCE is to foster the innovative nanotechnology-sector networks across Central Europe regions by bringing together public and private organizations (enterprises, research centers, venture capitalists and public institutions) to carry out collaborative and interdisciplinary researches on nanomaterials (in the frame of REACH Regulation) and to turn the most promising laboratory results into innovative industrial applications.

Within this contribution the main objectives of the NANOFORCE project will be presented as well as first results. Furthermore the BioNanoNet will present its work and the EURO-NanoTox as well as the ONLINE-Journal "EURO-NanoTox-Letters".

DISCUSSION

Tuesday morning, 18 September, 10:45
NanoForCE InfoDay
Chair: Anna Świderska-Środa/Joanna Sobczyk

COFFEE

Tuesday morning, 18 September, 11:10

Mat&Nano Brokerage Day

Tuesday morning, 18 September, 11:40

Abstracts

in author alphabetical order

Oral

Wrapping up

Witold Łojkowski[1], Urszula Narkiewicz[2], Malgorzata Lewandowska[3]

1. *Polish Academy of Sciences, Institute of High Pressure Physics (UNIPRESS), Sokolowska 29/37, Warszawa 01-142, Poland* **2.** *West Pomeranian University of Technology, Szczecin (ZUT), Szczecin 70-310, Poland* **3.** *Warsaw University of Technology, Faculty of Materials Science and Engineering (InMat), Wołoska 141, Warszawa 02-507, Poland*

e-mail: wl@unipress.waw.pl

Welcome. Acknowledgements. Background and aims of the conference. Technical details.

Poster

Biodegradable nanofibrous coatings electrospun from polymers derived from renewable resources.

Wacław Tomaszewski, Danuta Ciechańska, Marek Szadkowski, Michał Kudra

Instytut Biopolimerów i Włókien Chemicznych w Łodzi, ul. M. Skłodowskiej-Curie 19/27, Łódź 90-570, Poland

e-mail: nanotech@ibwch.lodz.pl

Research goal:

The research goal of this work is to elaborate production methods of fibrous materials from nano- and micro-fibres by electrospinning technique from solutions of biodegradable polymers (including medical purity) for the production of medical products.

Technology and polymer feedstocks:

Nowadays electrospinning is the most effective and most often used technology of the production of nano-fibres from polymer solutions. Institute of Biopolymers and Chemical Fibres for a few years has been conducting research concerning the usage of this technology to obtain various forms of materials from nano- and micro-fibres, mainly from chitosan (also alginate, starch, collagen) and from synthetic polymers of medical purity as PLA and co-PLA (Boehringer's Resomers). Apparatus that is used for electrospinning is the laboratory equipment which is the Institute's own technical solution that enables the production or modification of materials in form of sheets sized 20cm x 20cm and 20cm x 50cm. Production of these materials requires preparation of applicable spinning solutions with polymer concentration typically in the range 2 - 10 wt%. Solvents such as water, acids, chloroform, DMSO and others are usually used for this. Due to special requirements which are the effect of medical applications, produced materials are often subjected to additional processes in order to give them their final form. They are also subjected to mechanical examination as well as to various biological and chemical examinations which define level of their purity. In the final stage these materials have to be subjected to sterilization, mostly sterilization by radiation.

Laboratory installation for electrospinning nanofibers from polymer solutions.

Produced fibrous materials – application directions:

- implants' modification (surgical meshes, vascular prosthesis, polyurethane wound dressing sponges, substructures of modified bacterial cellulose). – one-sided covering of permanent implants' surface with layer of biodegradable nano-fibres in order to obtain biocompatible and biostimulative character for wound healing acceleration. In case of some implantable materials the other side is left in "slipping" form which protects the organism from fusing an implant and the treated organ.
- wound dressings in form of self-supporting fibrous coatings
- culture substrates for cell cultures

We are extending given directions to new 2D forms with aligned fibres, and also we are trying to produce nano-fibrous 3D objects. We would like to add collagen, alginate or starch to biodegradable polymers that we already use: chitosan, PLA and co-PLA.

Nanofibrous scaffold from chitosan / PEO composite in a state of high flexibility in the wet environment.

Technology and polymer feedstocks:

Nowadays electrospinning is the most effective and most often used technology of the production of nano-fibres from polymer solutions. Institute of Biopolymers and Chemical Fibres for a few years has been conducting research concerning the usage of this technology to obtain various forms of materials from nano- and micro-fibres, mainly from chitosan (also alginate, starch, collagen) and from synthetic polymers of medical purity, as PLA and co-PLA (Böringer's Resomers). Apparatus that is used for electrospinning is the laboratory equipment which is the Institute's own technical solution that enables the production or modification of materials in form of sheets sized 20cm x 20cm and 20cm x 50cm. Production of these materials requires preparation of applicable spinning solutions with polymer concentration typically in the range 2 - 10 wt%. Solvents such as water, acids, chloroform, DMSO and others are usually used for this. Due to special requirements which are the effect of medical applications, produced materials are often subjected to additional processes in order to give them their final form. They are also subjected to mechanical examination as well as to various biological and chemical examinations which define level of their purity. In the final stage these materials have to be subjected to sterilization, mostly by radiation.

Zdjęcie SEM powłoki okrywającej implant z nano i mikrowłókien PLA

Why nano-fibres?

The results of numerous world news report that living cells proliferate much better in contact with nano-fibres than in contact with the surface of much thicker textile fibres. As the porosity of nano-fibrous wound dressing materials is small, they create a barrier for pathogens enabling at the same time an air access to the surface of the wound. Such materials in form of coating covering implants of thicker and mechanically stronger structure, can also biologically enrich the surface of the implants or they can seal the implants to prevent outflow of blood components from vascular prosthesis. We have here ability to influence the structural features of nanofibers and a wide range of polymer materials and the technique of manufacture from polymer solutions enables on the introduction of nano-sized functional additives, which preferably modify the fiber material or as a result of release them to organism, creates the environment with favorable conditions to assist the healing process. Some fibrous structures can be obtained in forms similar to natural tissues occurring in living organisms (biomimetics).

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