XXVIII Conference
Polish Chitin Society
Gdańsk
September, 25-27th, 2023

NEW ASPECTS ON CHEMISTRY AND APPLICATION OF CHITIN AND ITS DERIVATIVES

Photo: Mateusz Ptak

XXVIII Conference of Polish Chitin Society

„NEW ASPECTS ON CHEMISTRY AND APPLICATION OF CHITIN AND ITS DERIVATIVES”

Gdańsk, September 25-27th 2023

Polish Chitin Society

XXVIII Conference

„New aspects on chemistry and application of chitin and its derivatives"

Łukasiewicz
Lodz
Institute of
Technology

SCHEDULE OF CONFERENCE

September 25th 2023 – Monday

16^{00}-18^{30}	**Registration**
18^{00}-20^{00}	**Buffet dinner**

September 26th 2023 – Tuesday

9^{00}-9^{30}	**Opening ceremony** **Katarzyna Struszczyk-Świta, Ph.D., President of PTChit**
	Why it is worth to publish in Progress on Chemistry and Application of Chitin and its Derivatives (PCACD) Journal **Magdalena Gierszewska, Ph.D., Editor-in-Chief**
9^{30}-9^{55}	**Prof. Henryk Struszczyk prize - giving ceremony**

Session A		
Chairman		**Prof. Małgorzata Jaworska, Ph.D., D.Sc.**
9^{55} - 10^{15}		**<u>Anke Wunder</u>, Katja Heppe, Andreas Heppe** **BioLog Heppe GmbH – CHITOSAN PRODUCTS, TECHNOLOGIES AND APPLICATIONS FOR 30 YEARS**
A1	**10^{15}-10^{35}**	**<u>Justyna Kozłowska</u>, Klaudia Brzezińska, Julia Rypińska, Klaudia Istal, Emilia Golińka** **CHITOSAN AND CHITOSAN-BASED MATERIALS IN COSMETIC APPLICATIONS**
A2	**10^{35}-10^{55}**	**<u>Anna Rył</u>, Piotr Owczarz, Johanna Hafner, Claude Oelschlaeger, Norbert Willenbacher** **CHARACTERISTICS OF LOCAL MECHANICAL PROPERTIES OF CHITOSAN HYDROGELS USING PASSIVE MICROREOLOGY TECHNIQUES: PRELIMINARY RESEARCH**
A3	**10^{55}-11^{15}**	**Małgorzata Gnus** **EXTERNAL MAGNETIC FIELD – AFFECT ON SEPARATION PROPERTIES OF MAGNETITE CONTAINING CHITOSAN MEMBRANES IN ETHANOL DEHYDRATION PROCESS**
11^{15}-11^{35}		**Coffee/tea break**

Session B		
Chairman		Justyna Kozłowska, Ph.D., D.Sc.
B1	11^{35}-11^{55}	Magdalena Paczkowska-Walendowska, Judyta Cielecka-Piontek CHITOSAN AS A PLATFORM FOR THE LOCAL DELIVERY ANTI-INFLAMMATORY SYSTEM PROVIDING NATURAL COMPOUNDS
B2	11^{55}-12^{15}	Aleksander Smolarkiewicz-Wyczachowski, Halina Kaczmarek, Jarosław Piskorz, Paweł Nowak, Marta Ziegler-Borowska CHITOSAN COMPOSITES CONTAINING BODIP-Y COMPOUNDS AS MATERIALS FOR BIOMEDICAL APPLICATIONS
B3	12^{15}-12^{35}	Dominik Sikorski, Zbigniew Draczyński PRELIMINARY STUDIES ON THE PREPARATION AND PROPERTIES OF CHITOSAN FILMS MODIFIED WITH ACID VAPORS WITH CIPROFLOXACIN
B4	12^{35}-12^{55}	Mateusz Data, Danuta Ciechańska, Łukasz Horajski, Bogumił Brycki, Maria Wiśniewska-Wrona, Justyna Wietecha, Dorota Kaźmierczak, Jagoda Jóźwik-Pruska HYGIENIC PAPERS MODIFIED WITH THE ADDITION OF FUNCTIONAL BIOPOLYMER-BIOCIDE COMPOSITIONS PRODUCED ON A PILOT RESEARCH INSTALLATION
13^{00}-14^{15}		Lunch
14^{30}-19^{00}		Visiting the monuments of Gdańsk with a guide
20^{00}-24^{00}		Gala dinner

September 27th 2023 – Wednesday

9^{00}-10^{00}	General Assemble of the Polish Chitin Society *(only for PTChit members)*

10^{00}-11^{00}	Session C1: poster session
P1	Karol K. Kłosiński, Radosław A. Wach, Bożena Rokita, Renata Czechowska-Biskup, Małgorzata K. Girek-Bąk, Damian Kołat, Żaneta Kałuzińska-Kołat, Barbara Kłosińska, Łukasz Duda, Zbigniew W. Pasieka CARBOXYMETHYL CHITOSAN HYDROGELS - THEIR MECHANICAL PROPERTIES AND BIOCOMPATIBILITY
P2	Michał Arabski, Arkadiusz Kuś, Monika Sikora, Maria Wiśniewska-Wrona, Katarzyna Gałczyńska, Oleg Łyżwiński, Małgorzata Kujawińska LASER INTERFEROMETRIC DETERMINATION OF AMPICILIN DIFFUSION THROUGH CHITOSAN-BASED MEMBRANE DEDICATED FOR BACTERIOPHAGES APPLICATION
P3	Justyna Kozłowska, Julia Rypińska, Klaudia Brzezińska CHITOSAN-BASED HYDROGEL WITH ACTIVE SUBSTANCES FOR DERMATOLOGICAL APPLICATION

P4	**Emilia Szymańska, Katarzyna Winnicka** **EFFECT OF TOPICAL CHITOSAN EMULGEL COMPOSITION ON HYDROCORTISONE PERMEABILITY ACROSS BIOMIMETIC STRAT-M MEMBRANE**
P5	**Dorota Chełminiak-Dudkiewicz, Miloslav Macháček, Aleksander Smolarkiewicz-Wyczachowski, Kinga Mylkie, Sebastian Drużyński, Rafał Krygier, Marta Ziegler-Borowska** **NATURAL PLANT EXTRACTS AS ACTIVE COMPONENTS IN CHITOSAN-BASED MATERIALS FOR BIOMEDICAL APPLICATION**
P6	**Karolina Rolińska, Ewelina Jakubowska, Małgorzata Żmieńko, Katarzyna Łęczycka-Wilk** **DEEP EUTECTIC SOLVENTS AS PLASTICIZER AND ACTIVE AGENT IN CHITOSAN-BASED FILMS**
P7	**Janek Weißpflog, Christine Steinbach, Rahma Boughanmi, Anke Wunder, Katja Heppe, Simona Schwarz** **ADSORPTION OF DIFFERENT HEAVY METAL IONS AND OXYANIONS ONTO CHITOSAN**
P8	**Tomasz Jóźwiak, Urszula Filipkowska** **THE USE OF CHITIN FOR THE REMOVAL OF NITRATES AND PHOSPHATES FROM GREENHOUSE WASTEWATER**
P9	**Paulina Król, Klaudia Piekarska, Cesar Hernandez, Marcin Kudzin, Katarzyna Śledzińska, Maciej Boguń, Piotr Kaczmarek, Aleksandra Dyrała, Marzena Dymel, Krystyna Guzińska, Zdzisława Mrozińska, Gabriela Pałucka** **WHAT ABOUT THESE WOUNDS - WHAT DRESSING TO CHOOSE AND HOW THEY DIFFER?**
P10	**Monika Sikora, Michał Arabski, Maria Wiśniewska- Wrona** **CHITOSAN-BASED MATRIX AS A CARRIER FOR BACTERIOPHAGES**

Session D		
Chairman		**Katarzyna Małolepsza-Jarmołowska, Ph.D., D.Sc.**
D1	**11^{00}-11^{20}**	**Izabela Dziedzic, Hermann Ehrlich** **PATENTOLOGY OF CHITINOUS BIOMATERIALS**
D2	**11^{20}-11^{40}**	**Paweł Poznański, Wacław Orczyk** **CHITOSAN AND SYNTHETIC FUNGICIDES: SYNERGY OF ANTIFUNGAL ACTIVITY AGAINST FUSARIUM GRAMINEARUM**
D3	**11^{40}-12^{00}**	**Eryk Jędrzejczak, Patrycja Frąckowiak, Marcin Wysokowski** **MULBERRY SILKWORMY (*BOMBYX MORI*) AS AN ALTERNATIVE SOURCE OF CHITIN – ISOLATION AND ANALYSIS OF THE RESULTING POLYSACCHARIDE**
12^{00}-12^{15}		**Coffee/tea break** *The coffee break also lasts during the poster session*

12^{15}-13^{00}	**Session C2: poster session**
P11	**Łukasz Pawłowski, Szymon Mania, Adrianna Banach-Kopeć, Michał Bartmański, Anna Ronowska, Kacper Jurak, Aleksandra Mielewczyk-Gryń, Natalia Karska, Sylwia Rodziewicz-Motowidło, Andrzej Zieliński** **ELECTROPHORETIC DEPOSITION AND CHARACTERIZATION OF RGD PEPTIDE-FUNCTIONALIZED CHITOSAN COATINGS ON TI13NB13ZR ALLOY**
P12	**Maria Wiśniewska-Wrona, Klaudia Piekarska, Anna Wojtala, Marek Warzala, Anna Pietruszka, Longina Madej-Kielbik, Monika Sikora, Karolina Gzyra-Jagiela, Konrad Sulak, Wieslaw Adamiec, Piotr Cichacz, Przemysław Wiecek** **DEVELOPMENT OF THERMOPLASTIC CHITOSAN ENRICHED WITH UNIQUE BIOMODIFIERS INTENDED FOR PROCESSING BY EXTRUSION**
P13	**Magdalena Gierszewska, Ewa Olewnik-Kruszkowska, Mohamed Bouaziz** **RELATION BETWEEN CHITOSAN FILM COMPOSITION, OVERALL MIGRATION, AND SWELLING IN VARIOUS FOOD SIMULANTS**
P14	**Katarzyna Małolepsza-Jarmołowska, Hanna Bazan** **PROTECTION OF THE ESOPHAGEAL MUCOSA WITH CHITOSAN GELS**
P15	**Katarzyna Małolepsza-Jarmołowska, Hanna Bazan** **STUDIES ON CHITOSAN GELS WITH POLYVINYL ALCOHOL PROTECTING THE ESOPHAGEAL MUCOSA**
P16	**Bożena Grimling, Bożena Karolewicz** **RESEARCH ON THE PHYSICOCHEMICAL PROPERTIES OF DERMATOLOGICAL APPLICATIONS IN THE PRESENCE OF CHITOSAN CONTAINING AN EXTRACT FROM *HUMULUS LUPULUS L*.**
P17	**Katarzyna Struszczyk-Świta** **MICROWAVE-ASSISTED EXTRACTION OF CHITOSAN FROM FILAMENTOUS FUNGAL BIOMASS**

13^{00}-13^{15}	**Closing of the conference**
13^{15}-14^{15}	**Lunch**

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SMART CHITOSAN-BASED COMPOSITE COATINGS ELECTROPHORETICALLY DEPOSITED ON TITANIUM OR ITS ALLOYS FOR IMPLANT APPLICATIONS

Post-operative bacterial infections are one of the main causes of implant failure. To prevent bacteria from settling on the surface of the implant, coatings with antibacterial properties are now intensively produced. However, the problem is the controlled release of the therapeutic substance from the coating over a long time of implant use while maintaining adequate osteointegration [1,2].

The base of the proposed coatings was chitosan, a natural biopolymer that exhibits biocompatibility and antimicrobial activity. In addition, chitosan belongs to the group of so-called smart biopolymers that respond to external environmental effects, such as temperature change, pH change, UV-Vis radiation, or electric and magnetic fields. The sensitivity of chitosan can be exploited in controlled drug delivery systems for peri-implant tissues, as inflamed tissues show a local reduction in the pH of their environment.

The ongoing research aimed to produce smart coatings on the surface of titanium or its alloys by electrophoretic method, based on chitosan with the addition of other polymers such as poly(4-vinylpyridine) or Eudragit E 100 and metallic nanoparticles - silver or copper - with bactericidal activity, which will release the active substance (metallic nanoparticles) in a controlled manner - only when inflammation occurs. The microstructure, surface roughness, coating thickness, chemical composition, mechanical and electrochemical properties, silver release rate at different pH of simulated body fluid solution (SBF), wettability, antimicrobial and cytotoxic properties were investigated.

The coatings, as a biopolymer blend, were successfully deposited on metallic substrates by one-step electrophoretic deposition. Deposited coatings affected the surface roughness, wettability, and corrosion resistance of the titanium substrate. An increase in antimicrobial activity compared to the bare substrate was observed. More consistent release of silver from the fabricated systems was noted in the acidified SBF solution compared to the neutral pH solution. The produced coatings enable the adhesion and proliferation of osteoblast cells, but further research work is needed to select the optimal chemical composition of these systems. Such biopolymer coatings deposited on titanium or its alloy surface may contribute to reducing the risk of implant-related infections and the necessity of antibiotic therapy and re-implantation.

References:
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[2] Ł. Pawłowski, J. Wawrzyniak, A. Banach-Kopeć, B. M. Cieślik, K. Jurak, J. Karczewski, R. Tylingo, K. Siuzdak, A. Zieliński, Antibacterial properties of laser-encapsulated titanium oxide nanotubes decorated with nanosilver and covered with chitosan/Eudragit polymers, Biomater. Adv. 138 (2022) 212950.

Session A

Justyna Kozłowska, Klaudia Brzezińska, Julia Rypińska, Klaudia Istal, Emilia Golińka

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CHITOSAN AND CHITOSAN-BASED MATERIALS IN COSMETIC APPLICATIONS

The Cosmetics Regulation applies to cosmetic products defined as: "any substance or mixture intended to be placed in contact with the external parts of the human body (epidermis, hair system, nails, lips, and external genital organs) or with the teeth and the mucous membranes of the oral cavity with a view exclusively or mainly to cleaning them, perfuming them, changing their appearance, protecting them, keeping them in good condition or correcting body odours" [1].

Chitosan is commonly used in cosmetic and skin care applications. According to the CosIng database, the functions assigned to chitosan are chitosan are film forming and hair fixing [2]. However, we know that not all activities of chitosan are included in the CosIng database. Other interesting biological properties of interest for top skin applications can be found in chitosan, such as antifungal and wound healing activities, bio-adhesivity, non-toxicity, and biodegradability [3]. Chitosan can act as a cosmetic ingredient due to its specific properties or as a carrier of other active ingredients due to its technological properties.

The presented research aimed to use chitosan in various new cosmetic applications, e.g., chitosan-based capsules or hydrogel matrices.

Acknowledgements:

This work was supported by the Center of Excellence „Towards Personalized Medicine" operating under Excellence Initiative – Research University.

References

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[2] https://ec.europa.eu/growth/tools-databases/cosing/details/75065

[3] Aranaz, I.; Mengíbar, M.; Harris, R. et al. Functional characterization of chitin and chitosan. Curr. Chem. Biol. 2009, 3.

Anna Rył[1], **Piotr Owczarz**[1], **Johanna Hafner**[2], **Claude Oelschlaeger**[2], **Norbert Willenbacher**[2]
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CHARACTERISTICS OF LOCAL MECHANICAL PROPERTIES OF CHITOSAN HYDROGELS USING PASSIVE MICROREOLOGY TECHNIQUES: PRELIMINARY RESEARCH

The growing interest in research conducted to characterize polymer hydrogels results from their potential application in tissue engineering and controlled drug release systems. In both cases, the architecture and mechanical properties of the obtained scaffolds are extremely important because they will directly affect the release profile of the active substance as well as support the tissue regeneration process. Classically, the above properties are determined on a macroscopic scale using mechanical tests (determination of the Young's modulus value or the use of the texture profile analysis test [1]), rheological measurements and microscopic tests. It should be emphasized that the obtained results do not necessarily reflect the local state of the structure on the living cells scale length; especially in the case of highly heterogeneous lattices.

The aim of the work is to discuss the theoretical foundations of selected techniques of passive optical microrheology and the possibility of their application to characterize local mechanical properties of injectable and implamantable chitosan hydrogels.

Based on the classical mechanical studies of implamantable chitosan hydrogels cross-linked with EGDE (ethylene glycol diglycidyl ether), it was impossible to formulate unambiguous conclusions about the macroscopic properties of the structure due to the significant uncertainty of the obtained results - strong phase separation. Contrary, the use of the video multiple particle tracking microrheology (V-MPT) allowed for undisputed confirmation of the strong heterogeneity of the tested samples, but also enabled the quantitative assessment of the local state of the structure. It was shown that the increase in the concentration of both chitosan and the cross-linking substance causes an increase in the value of the storage modulus G'; this effect is more intense in the case of a higher concentration of EDGE.

In the case of studies on injectable chitosan scaffolds undergoing a thermoinduced sol-gel phase transition, the use of microrheological measurements based on the dynamic light scattering (DLS) technique allowed to track the progressive formation of a spatial polymer network under the influence of temperature increase. The courses of dynamic modules as a function of oscillation frequency at low temperatures are consistent with the spectra obtained during bulk measurements using a rotational rheometer [2]. The greatest discrepancies are observed for measurements carried out at 40°C; in contrast to rheological measurements, no occurrence of a constant value of dynamic modules independent of the applied deformation was observed for microrheological measurements. This may be due to prolonged formation of the hydrogel in the perikinetic regime induced solely by stochastic Brownian motions [3].

Finally, the advantages and limitations of the passive optical microrheology methods used will be discussed, and further research directions and perspectives will be pointed out.

Acknowledgements:

The research was partly funded by the FU²N project "Application of passive optical microrheology techniques to determine the effect of the injection application on the local mechanical properties of the polymer network".

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[2] Owczarz, P., Ryl, A., Modrzejewska, Z., & Dziubiński, M. The influence of the addition of collagen on the rheological properties of chitosan chloride solutions. Progress on Chemistry and Application of Chitin and its Derivatives, (22), 176-189, (2017).

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EXTERNAL MAGNETIC FIELD – AFFECT ON SEPARATION PROPERTIES OF MAGNETITE CONTAINING CHITOSAN MEMBRANES IN ETHANOL DEHYDRATION PROCESS

A hybrid materials composed of polymer and magnetic particles are of great interest due to their potential use as anticancer materials, magnetic resonance imaging, magnetic recoverable, catalysts, hyperthermia treatment, bio-separation and drug release agent. These materials can be applicated also in water treatment or membrane separation processes.

In last work, the magnetic particles were arranged in the membrane matrix by an external magnetic field which allowed to distribute particles perpendicular as well as parallel in membrane matrix. Differences in orientation of magnetic particles affected membrane's separation properties.

In present work, influence of external magnetic field during pervaporation process on hybrid chitosan-magnetite membrane properties was examined. For this purpose, series of chitosan membranes with different amount of oriented (perpendicular or parallel) or not magnetite particles were tested in ethanol-water pervaporation process during which membrane chamber was additionally placed in external magnetic field.

This studies showed, that additional magnetic field can either improve or deteriorate the properties of the material depending on its initial preparation.

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Session B

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CHITOSAN AS A PLATFORM FOR THE LOCAL DELIVERY ANTI-INFLAMMATORY SYSTEM PROVIDING NATURAL COMPOUNDS

Periodontal disease is a significant global oral health burden, and severe perio-dontitis accounts for multiple tooth losses in the adult population worldwide. One of the key plant materials used in oral infections is Scutellariae baicalensis radix (Baikal Skullcap Root) showing anti-inflammatory, antioxidant, and antibacterial properties [1].

To increase the pro-health benefits of the plant materials, their extracts should be combined with functional polymers with additional health-promoting properties. One such substance is chitosan.

The aim of the studies was to establish the influence of chitosan on the preparation of systems containing Scutellariae baicalensis radix extract. The Design of Experiment (DoE) approach was used to choose the best chitosan system parameters.. The optymalization experiment design was developed, using Statistica 13.3 software (TIBCO Software Inc., Palo Alto, CA, USA), for two independent variables that were assigned three levels of values (32 full factor design). The deacetylation degree of chitosan and its ratio in regards to lyophilized extract were selected as independent factors.

An increase in the deacetylation degree of chitosan used in the system improved the potential for reducing free radicals and inhibiting the hyaluronidase enzyme. Also, increasing the degree of chitosan deacetylation results in increased resistance of the carrier to biodegradation and an extended baicalin release profile, which is also associated with an increase in the viscosity of the chitosan-based system. In total, the system of a freeze-dried extract with chitosan 90/500 in the ratio of 2:1 turns out to be the one with the best physicochemical (high percentage of baicalin release and the highest viscosity conditioning the prolonged stay at the site of administration) and biological properties (the highest antioxidant and anti-inflammatory activities), resulting in the highest potential for use in the treatment of oral inflammatory diseases.

As a result of the application of the design of experiment (DoE) approach, it was possible to optimize the obtaining of chitosan systems with S. baicalensis radix extracts. The rheological properties of the prepared chitosan-based systems and the profile of their biological activity confirm the appropriate approach to the search for therapeutic solutions in the treatment of periodontal diseases based on the use of the anti-inflammatory potential of plant materials.

Acknowledgements:
This research was funded by National Science Center (Poland), under Sonata grant (number 2020/39/D/NZ7/01824).

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Aleksander Smolarkiewicz-Wyczachowski[1,2], **Halina Kaczmarek**[1], **Jarosław Piskorz**[3], **Paweł Nowak**[1], **Marta Ziegler-Borowska**[1]

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CHITOSAN COMPOSITES CONTAINING BODIP-Y COMPOUNDS AS MATERIALS FOR BIOMEDICAL APPLICATIONS

Photodynamic therapy (PDT) is one of the known methods of fighting cancer. This method uses the action of light, photosensitizer, and oxygen present within cancer tissues, which leads to the formation of reactive oxygen species capable of damaging pathological cells. However, PDT has several limitations, including those related to using photosensitizers, e.g., excessive drug aggregation in the body or unspecified photosensitizer targets. Therefore, it is crucial to look for new materials that could be carriers for photosensitizers and would minimize the above limitations. As a biopolymer, Chitosan is a promising material that can be used to design of new drug carriers. Combined with active substances, it can constitute systems capable of gradually releasing the drug at the target in the body, and it can also act as a protective coating for the drug - preventing its degradation.

BODIP-Y compounds, due to their properties(i.e., the ability to strongly absorb and emit light, chemical stability in various environments, easy modification of the structure, and the ability to generate singlet oxygen), are ideal photosensitive drugs that can be used in Photodynamic Therapy.[1].

In this work, we obtained chitosan composites doped with BODIP-Y type compounds differing in substitution in the main core in positions 2 and 6. The spectral properties of those materials (both in solid and in solution) were characterized using UV-ViS, infrared spectroscopy, and spectrofluorimetry. The surface morphology was described using Scanning Electron Microscopy (SEM), Atomic Force Microscopy (AFM), and also by measuring the contact angle. In the context of using the tested material in biomedical applications was essential to check the degree of release of the dye from the chitosan matrix and the interaction with proteins (i.e., human serum albumin and alpha-acid glycoprotein).

References:

[1] A.Smolarkiewicz-Wyczachowski, H.Kaczmarek, J.Piskorz, P.Nowak, M. Ziegler-Borowska, Chitosan Composites Containing Boron-Dipyrromethene Derivatives for Biomedical Applications, International Journal of Molecular Sciences,(2023), 24.

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PRELIMINARY STUDIES ON THE PREPARATION AND PROPERTIES OF CHITOSAN FILMS MODIFIED WITH ACID VAPORS WITH CIPROFLOXACIN

Chitin is a polysaccharide commonly found in the structure of sponges, corals, shells of marine invertebrates, insects and cell walls of fungi [1-4].

The mechanisms of action of chitosan against bacteria have been studied and described in many articles. The antimicrobial properties of chitosan can be improved by chemically modifying its structure. The two reactive sites -NH2 and -OH present in chitosan open great possibilities for its chemical modification. These groups allow for sulfonation, amination and carboxymethylation reactions [5-6].

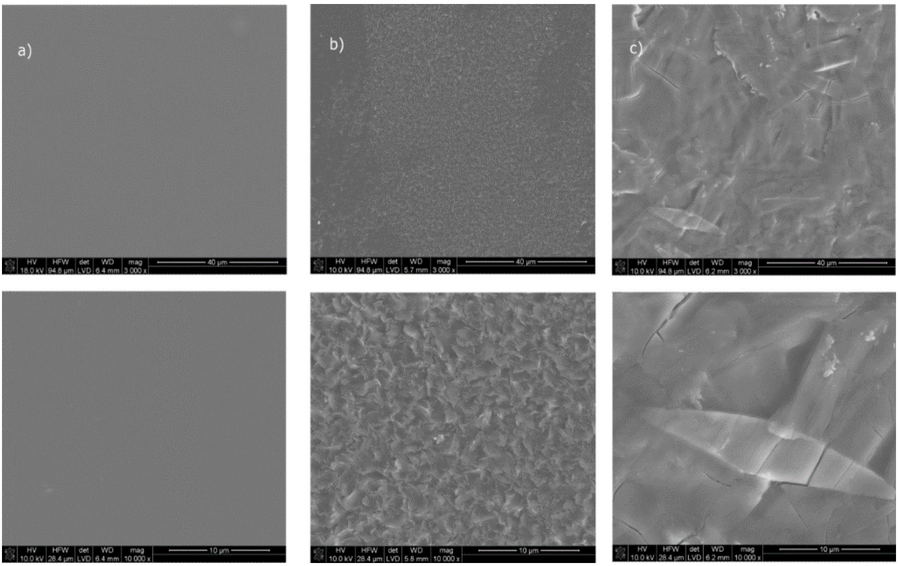

Figure 1. Microstructure images of CS film treated with CIP sodium salt (b) and subsequently treated with acetic acid vapours (c).

The purpose of the research was to develop methods for modifying nonwoven fabrics and chitosan films in the gas phase (using acid vapors). The research was planned to use various acids: acetic, propionic, butyric and valeric acids, as well as hydrochloric acid. In addition, membranes containing ciprofloxacin on their surface for use as a model drug were produced. A unique structure was obtained that not only imparts new properties to the films, but also changes the coating and surface structure. The form in which the drug binds to the surface was confirmed. The determination of antimicrobial activity against E. coli and S. aureus was planned.

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Mateusz Data[1], <u>Danuta Ciechańska</u>[1], Łukasz Horajski[1], Bogumił Brycki[1], Maria Wiśniewska-Wrona[2], Justyna Wietecha[2], Dorota Kaźmierczaka[2], Jagoda Jóźwik-Pruska[2]

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HYGIENIC PAPERS MODIFIED WITH THE ADDITION OF FUNCTIONAL BIOPOLYMER-BIOCIDE COMPOSITIONS PRODUCED ON A PILOT RESEARCH INSTALLATION

The aim of the work was to verify and optimize the process of manufacturing and applying functionalizing additives using a pilot research installation. Physical, mechanical and microbiological properties as well as susceptibility to biodegradation of the produced prototype hygienic papers were tested. Compositions prepared in EPICOM and Łukasiewicz- Łódzki Instytut Technologiczny were used in the studies. In particular, the issues related to: assessment of the uniformity of paper surface coverage, determination of the minimum amount of biopolymer composition that will ensure sufficient coverage of the paper surface, determination of the minimum amount of active bioactive agent ensuring the assumed requirements for the biological activity of the finished product against S.aureus, B.subtilis and E.coli bacteria, being more than 90% biodegradable in a compost environment. Based on the studies carried out, it was found that the uniformity of the coverage of the paper web by the biopolymer composition is influenced by all three parameters, i.e. the rotational speed of the nozzles, the linear speed of the paper web and the total output. During the tests, the maximum value of the total expenditure was verified, which does not cause problems with movement on the machine, i.e. web breaks caused by weakening of the paper due to excessive web moisture. The power of the dryer needed to dry the web to the assumed level was verified in relation to the total spraying rate. As part of the optimization studies, a series of papers functionalized with biopolymer-biocide compositions based on chitosan, starch, alginate and geminisurfactants was prepared: Parameters for the production of papers functionalized with biopolymer compositions with assumed functional properties were developed.

Acknowledgements:

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Session C

POSTER SESSION

P1

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CARBOXYMETHYL CHITOSAN HYDROGELS - THEIR MECHANICAL PROPERTIES AND BIOCOMPATIBILITY

Hydrogels hold significant amount of water, but they have properties of solid substances, thus are useful for medical applications, e.g., in systems for controlled release of drugs or as wound dressings [1]. Ionizing radiation was proven to be a suitable tool for carboxymethyl chitosan (CMCS) hydrogel synthesis and could be of use for synthesis of biomaterials, as it may simultaneously sterilize the product [2,3].

The aim of this study was to evaluate the properties and action of biopolymer CMCS hydrogels to select the best compositions for future research towards wound-dressing applications. The hydrogels were formed from aqueous solutions of (CMCS) of high concentration via electron beam radiation-initiated crosslinking, with the presence of a crosslinking agent: poly(ethylene glycol) diacrylate. The elasticity of the gel depends on the concentrations of the components and the irradiation dose employed to synthesize the hydrogel, therefore the resulting hydrogels might be fragile, easy to break or elastic, but durable [4]. Initial biocompatibility screening of hydrogels done with the XTT and Live/Dead tests using human fibroblasts, confirmed that the material is not cytotoxic. The biocompatibility studies of implantation, carried out on laboratory rats, showed no adverse effect of hydrogels towards animal tissue. Those suggest that CMCS in the form of hydrogels could be considered for further evaluation towards wound-healing dressings.

Acknowledgements:

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P2

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LASER INTERFEROMETRIC DETERMINATION OF AMPICILIN DIFFUSION THROUGH CHITOSAN-BASED MEMBRANE DEDICATED FOR BACTERIOPHAGES APPLICATION

Chitosan has a positive effect on the healing process of skin wounds. In our previous study, we prepared chitosan-based dressing material in the form of a film dedicated to adding active phages (ChM). This material had antibacterial activity against *Pseudomonas aeruginosa* PAO1 under wound healing conditions. It seems to be important to consider the possibility to use this ChM in combination with another bactericidal agent in would treatment such as antibiotics. This summary effect could be crucial from a clinical point of view for the effective eradication of bacteria from the wound care environment. So, the transport properties of antibiotics through ChM should be characterized. In this study, we used single-shot polarization-based Mach-Zehnder interferometer for a phase-retrieval-based approach to quantifying the diffusion of ampicillin through ChM in function of time. The aim of the study was to check the transport properties of ampicillin (as a model of antibiotic) through ChM, as a carrier dedicated for bacteriophages. Our results show that the presented laser interferometer setup could be used for quantitative, spatio-temporal investigations of the diffusion properties of antibacterial agents through new materials based on chitosan as drug carriers.

Acknowledgements:

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P3

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CHITOSAN-BASED HYDROGEL WITH ACTIVE SUBSTANCES FOR DERMATOLOGICAL APPLICATION

The skin is exposed to abrasions every day. The most common cause is rubbing by clothing or new uncomfortable shoes. Chafing is also caused by rubbing one area of skin against another, such as on the thighs, when wearing shorts, skirts, or dresses. Despite such a common problem, few preparations on the market can remedy it.

The study aimed to develop a biodegradable hydrogel mask to address the problems caused by skin abrasions. We created a material that immediately soothes skin problems caused by abrasion damage.

The first step was to optimize the preparation method of the hydrogel matrix based on chitosan. Subsequently, appropriate active ingredients were selected to provide a soothing effect on skin irritation and redness and moisturize the skin -Centella asiatica extract, panthenol, and royal jelly.

The obtained hydrogel with active ingredients fulfills its purpose, i.e., non-irritating, intensely moisturizing, and reducing transepidermal water loss (TEWL), confirmed by skin tests. The addition of active substances also has a significant impact not only on the biophysical parameters of the skin but also on the hydrogel's physical properties. Our hydrogel could find potential applications in the field of biomedical and cosmetic sciences, especially in dermatology.

Acknowledgements:

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P4

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EFFECT OF TOPICAL CHITOSAN EMULGEL COMPOSITION ON HYDROCORTISONE PERMEABILITY ACROSS BIOMIMETIC STRAT-M MEMBRANE

Low molecular weight chitosan, a biodegradable multifunctional polycation with ability to enhance penetration and promote skin healing is extensively studied in pharmaceutical technology of topical formulations (1-2). Emulgel, a semi-solid formulation being a combination of emulsion and gel, is characterized by favorable application properties including emollient and soothing effect when compared to conventional creams or ointments.

The aim of the study was to investigate the effect of novel chitosan emulgel composition on the penetration and accumulation profile of hydrocortisone as model corticosteroid commonly used to treat skin inflammation, e.g. upon eczema or dermatitis. Pomegrate oil with anti-inflammatory properties and skin repairing potential was employed as lipophilic phase for emulgel preparation (3).

Methodology: Two emulgels comprising of 3% (w/w) chitosan (Heppe GmbH, Mw 232kDa) in 1% (w/w) lactic acid mixed with pomegrate oil in a weight ratio 4:1 (E o/w) or 1:4 (E w/o) were prepared by homogenization technique (15000 rpm, 60 min) with using lecitihin as emulsifier. Hydrocortisone was then uniformly suspended in emulgel bases in a final concentration of 1% (w/w). Permeability studies were performed in in-line cell system equipped with thermostated diffusion chambers (SES GmbH Analysesysteme) across Strat-M membrane mimicking the human skin barrier properties (4). The applied model examined solely the passive diffusion of the drug. Hydrocortisone analysis was achieved at 254 nm by isocratic elution in RP-HPLC Agilent Technologies 1200.

Results: Hydrocortisone slowly permeated across biomimetic membrane and profound differences in diffusion pattern among tested emulgels were noticed. Higher content of pomegrate oil in emulgel composition sped up the passive diffusion and the drug appeared in the acceptor medium at earlier time points. In addition, E w/o exhibited 3-fold greater permability across strat-M membrane when compared to E o/w. An observed enhanced penetration behavior most probably resulted from lower values of viscosity and consistency observed for E w/o. In turn, greater amount of chitoson in emulgel composition E o/w was found to increase hydrocortisone accumulation in biomimetic barrier. Drug penetration and retention was not affected by the amount of the soluble hydrocortisone fraction in emulgel sample as solubility studies displayed comparable concentrations of drug in dissolved state in both tested emulgels E w/o and E o/w.

Conclusions: The obtained results demonstrated a potential of emulgels composed of chitosan in combination with pomegrate oil in enhancing permeability and accumulation of hydrocortisone but further studies are needed to examine whether designed formulations are able to reduce inflammatory response and impact hydrocortisone activity.

Acknowledgements:

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P5

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NATURAL PLANT EXTRACTS AS ACTIVE COMPONENTS IN CHITOSAN-BASED MATERIALS FOR BIOMEDICAL APPLICATION

Chitosan is a natural, biodegradable polymer with a high molecular weight and a wide range of applications, including medicine, cosmetology, and biotechnology. This natural compound is widely utilized for its film-forming ability and antimicrobial properties. In addition, chitosan supports tissue regeneration and acts actively as a hemostatic agent [1-3].

The introduction of natural plant extracts enhances the properties of chitosan. Historically, plants and plant-based materials have been extensively used to treat wounds. Medicinal plants promote disinfection and debridement and provide a moist atmosphere, thus facilitating a natural healing environment. Today, some folk cultures use many other plants to treat cuts, wounds, and burns, albeit without knowing their activity and healing mechanisms.

This study aimed to obtain, test, and mutually compare three dried hemp extracts derived from *Cannabis sativa* (prepared in different solvents) as active ingredients in chitosan-based films. Therefore, three different extract-containing chitosan-based films were prepared and further evaluated regarding the effect of extracts incorporation on their structural, physicochemical, and biological properties. The obtained materials exhibited antioxidant and anti-inflammatory properties so that they could find potential applications in the field of biomedical sciences, including wound dressing materials.

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P6

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DEEP EUTECTIC SOLVENTS AS PLASTICIZER AND ACTIVE AGENT IN CHITOSAN-BASED FILMS

Traditional packaging materials such as plastics, paper/cardboard or glass do not affect the properties of the packaged products. On the other hand, modern packaging should perform additional functions to maintain and even improve the quality of packaged food. Such packaging is called active packaging [1].

In recent years, many scientific papers have been published on active food packaging systems. The presented study focuses on creating new chitosan-based films that incorporate a deep eutectic solvent (DES). Choline chloride and various hydrogen bond donors were utilized as a plasticizer, and the active properties of the DES integrated into the chitosan matrix were investigated. To examine the structures and characteristics of the resulting Ch and Ch/DES films, various techniques were employed, including FT-IR spectroscopy, SEM, swelling tests, WVTR, TGA, mechanical testing, and analysis of active properties.

The Ch/DES films achieved mechanical properties comparable to conventional PE films, such as a tensile strength of up to 26 MPa and elongation at break of up to 210%. This synthesis approach represents a step towards replacing traditional packaging materials with environmentally friendly alternatives, developed based on the principles of green chemistry.

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P7

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ADSORPTION OF DIFFERENT HEAVY METAL IONS AND OXYANIONS ONTO CHITOSAN

In lakes and rivers, there are increasingly different inputs of contaminants. For example, as a result of the closure of many opencast mines that had been in operation for decades and their flooding, the groundwater is rising again and flushing out metallic constituents (iron salts and iron hydroxide) from naturally occurring soil layers. The weathering and oxidation of pyrite releases oxidation products, mainly iron ions and sulfate ions, which are known as acid mine drainage. In order to protect flora and fauna, for aesthetic reasons, and to maintain and regenerate the quality of surface waters, water treatment is of great relevance.

The biopolymer chitosan is a very efficient adsorber material for the removal of heavy metal ions and oxyanions from aqueous solutions. Due to the solubility properties of chitosan, it can be used as both a adsorber and a flocculant for water treatment.

An example is given of iron and sulfate ion removal from surface waters in mining regions using chitosan. By flocculation and adsorption processes with chitosan, finest foreign components are removed from the water.

In addition the adsorption capacity of different heavy metal ions onto chitosan was investigated in dependence on their corresponding anions by batch and column experiments.

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equal viscosity in dissolved and solid state - Insights by various complimentary methods. International Journal of Biological Macromolecules 171 (2021) 242-261

P8

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THE USE OF CHITIN FOR THE REMOVAL OF NITRATES AND PHOSPHATES FROM GREENHOUSE WASTEWATER

The subject of the research was the effectiveness of nitrates V and orthophosphates removal from greenhouse wastewater using chitin as a sorbent. The chitin used in the research came from snow crab shells and was in the form of flakes with a diameter of 2-3 mm. The wastewater tested in the work came from soilless cultivation of tomatoes. They were collected from September 1, 2022 to October 31, 2022 and then averaged. The greenhouse effluent parameters were as follows :N-NO_3 600.9 mg/L, P-PO_4 90.5 mg/L, SO_4^{2-} 605.0 mg/L, Cl^- 0.7 mg/L, Ca^{2+} 745.0 mg/L, Mg^{2+} 140.0 mg/L, K^+ 550.0 mg/L, hardness 113° dH and pH 6.0.

The scope of the research included, among others: FTIR analysis and determination of the pHPZC of the sorbent, research on the influence of pH on the efficiency of nutrient sorption from wastewater, the kinetics of nitrate and orthophosphate sorption (description of experimental data using pseudo-first and pseudo-second order models as well as the intramolecular diffusion model) and studies on the effect of the dose on the percentage of nutrients removal from wastewater.

The FTIR analysis confirmed the saccharide structure of the sorbent. Peaks characteristic for amino and acetamide groups were noticed in the spectrum. The pHPZC value determined for chitin was 7.26. Sorption efficiency of both nutrients from wastewater was highest at pH 3. At pH > 8, spontaneous precipitation of orthophosphates occurred with calcium and magnesium ions present in wastewater. The sorption kinetics of nitrates and orthophosphates on chitin was best described by a pseudo-second order model. Sorption of nutrients on the tested sorbent proceeded in three phases, differing in intensity and duration. Sorption equilibrium time on chitin for nitrates and orthophosphates was 210 min and 240 min, respectively. The chitin dose of 20 g/L guaranteed the removal of 92% of orthophosphates and 57% of nitrates from greenhouse wastewater.

P9

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WHAT ABOUT THESE WOUNDS - WHAT DRESSING TO CHOOSE AND HOW THEY DIFFER?

Introduction

Currently on the market there are about 6k MD products for wound management [1-4]. The effectiveness of treatment as well as the comfort and health of the patient depend on the choice of the appropriate wound dressing. Most health organizations, that prepare national recommendations and guidelines for wound care [5-10], agree that there is no single agreed set of criteria for assessing the effectiveness and quality of wound dressings. However, the features that an ideal dressing should meet are well established [1-4,11-13].

Dressing materials belong to the group of medical devices (MD). This means that in order to be able to introduce such a device to the market, the manufacturer or its authorized representative, independently or with the participation of a notified body, must classify such a device in accordance with the requirements of the Medical Devices Act, the Regulation of the Minister of Health and also guarantee that such a device meets a number of normative and legal requirements contained in the new requirements of the Medical Device Regulation (MDR). Nevertheless, the differences in the properties of individual materials are huge, even among dressings from the same group.

In the work carried out as part of the project „O rany, co na te rany, czyli jaki opatrunek wybrać i czym właściwie się one różnią?", commercial products of wound dressings from all key groups and types of dressing materials (intended for hard-to-heal wounds) selected in consultation with experts form Polish Wound Management Association (PTLR), were analysed.

Materials and Methods

Free swell absorptive capacity was tested according to PN-EN 13726-1:2005.

Antibacterial activity assessment was carried out in line with AATCC Test Method 147-2011 Antibacterial Activity Assessment of Textile Materials:Parallel Streak Method, against Escherichia coli ATCC 11 229, Staphylococcus aureus ATCC 6538, Candida albicans ATCC 10259.

Quantitative chemical analysis was performed on the basis of series of standards PN-EN ISO 1833 and Regulation (EU) No 1007/2011, Annex VIII.

Microscope images were taken using Motic SMZ-143-N2GG stereo microscope equipped with Motic Moticam 2500 camera.

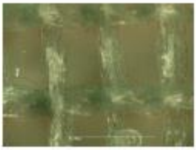
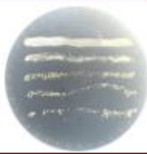
Results and Discussion

Determination of the antibacterial activity of samples of dressings containing diffuse active substance revealed that not all of the dressings exhibit antibacterial properties, in performed test method (table 2).

Table 1. Free swell absorptive capacity of wound dressings

Free swell ab.capacity, g/100cm²	**Suprasorb A+Ag**	**Aquacel Ag Extra**	**Sorbact Gel Dressing**	**Medisorb SILVER**	**DibuCell Active**
	81,596 ± 3,544	25,304 ± 0,991	0,108 ± 1,204	32,374 ±1,273	1,424 ± 0,175
Dressing type	calcium alginate + silver alginate fibers with ionic silver	sodium carboxy methyl cellulose fibers (Hydrofiber Technology) + ionic silver	hydrogel binding bacteria and fungi	non-woven with metallic silver	DIBUSHIELD PRO-HEAL™ biopolymer
percentage composition of dressings	46% viscose / 31% cotton / 23% PA	77% cellulose / 23% PA	tbd	59% viscose / 20% polyester / 12% PU / 9% non-textile elements	tbd

Table 2. Antibacterial activity assessment of wound dressings

Microscope image	*E. coli*	*S. aureus*	*C.albicans*
			
Suprasorb A+Ag	+	+	+
			
Aquacel Ag Extra	+	+	-
			
Sorbact Gel Dressing	-	-	-
			
Medisorb SILVER	-	-	-

Conclusions

Antibacterial activity of wound dressing can be assessed using multiple of different method. Failure of some of the samples in AATCC Test Method 147-2011 testifies to that active agent do not diffuse to the external environment. In order to check antibacterial activity of commercial products further antibacterial test will be carried out. As for the free swell absorptive capacity it corresponds strongly with the structure and raw materials composition of the tested dressings.

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P10

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CHITOSAN-BASED MATRIX AS A CARRIER FOR BACTERIOPHAGES

Hard-to-heal wounds are one of the most common problems in clinical practice, especially in patients with various diseases (mainly diabetes), but also during post-operative convalescence or post-traumatic treatment at various stages. Wound healing is a dynamic and complex process where infection prevention is key. Many important changes occur during healing: from inflammation, to cell migration, angiogenesis and cell matrix synthesis, to collagen deposition and reepithelialization.

Chitosan, thanks to its bactericidal effect on Gram-positive and Gram-negative bacteria as well as anti-inflammatory and haemostatic properties, is an excellent candidate for designing dressings for the treatment of hard-to-heal wounds. The great advantage of this biopolymer is its ability to be chemically modified, which allows for the production of various functional forms depending on the needs and later use. Chitosan can be an excellent polymer matrix for active antibacterial compounds, antibiotics and more. Selected bacteriophages may be an innovative approach to antibacterial therapy in dressings. This work is about preparation and characteristics of chitosan-based dressing material in the form of a film with the addition of lytic phages. We investigated a method for producing a dressing based on microcrystalline chitosan (CCH) and thermoplastic chitosan serving as a template for phage immobilization. The release of phages and the antibacterial properties of the prepared biocomposite were tested on the model bacterial strain *Pseudomonas aeruginosa* PAO1.

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ELECTROPHORETIC DEPOSITION AND CHARACTERIZATION OF RGD PEPTIDE-FUNCTIONALIZED CHITOSAN COATINGS ON TI13NB13ZR ALLOY

Metallic materials for long-term load-bearing implants still do not fully meet their requirements. Post-operative bacterial infections are one of the frequently occurring reasons for failed implant procedures. The problem is to provide implant surfaces with high antimicrobial activity while maintaining strong osteointegration properties [1,2].

This study aimed to modify the surface of Ti13Nb13Zr alloy by electrophoretic deposition (EPD) of a chitosan coating with a covalently attached Arg-Gly-Asp peptide (RGD). Two different approaches were used to prepare the EPD suspension for coating deposition - hydroxyacetic acid or carbon dioxide saturation. The coatings were deposited using a voltage of 10 V for 1 min. Techniques such as SEM, EDS, FTIR, and XPS were used to determine the physical and chemical properties of the prepared surfaces. In addition, the wettability of these surfaces was investigated using the falling drop method, and the adhesion of the coatings to the metallic substrate was evaluated by scratch test. The surfaces were also tested for antimicrobial activity using *E. coli* and *S. Aureus* bacterial strains. The cytotoxicity of the coatings in contact with human osteoblast cells (hFOB 1.19) was evaluated using MTT and LDH assays.

Chitosan coatings with attached RGD peptide on a Ti13Nb13Zr alloy substrate were successfully deposited by electrophoresis using various suspension preparation methods. Spectroscopic studies confirmed that the peptide did not detach from the chitosan chain during electrophoretic deposition. Both coatings demonstrated highly efficient antimicrobial activity, especially for Gram-positive bacteria. Cytotoxicity tests (MTT and LDH) showed high osteointegration properties of the modified surfaces compared to the bare Ti13Nb13Zr substrate. However, the adhesion of the coatings should be improved.

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DEVELOPMENT OF THERMOPLASTIC CHITOSAN ENRICHED WITH UNIQUE BIOMODIFIERS INTENDED FOR PROCESSING BY EXTRUSION

Recently, in the world research in the field of polymers, a trend has been observed towards obtaining modern environmentally friendly materials and thus replacing polyolefins produced from petroleum with natural macromolecules from renewable sources. The largest group of polymers synthesized in nature are polysaccharides, including: cellulose, starch, chitin and chitosan. An important advantage of synthetic polymers obtained from petroleum from the point of view of processing is their thermoplasticity, which enables the ease of manufacturing various types of functional forms from these macromolecules using the melt method. In the world, more and more attempts are being made to modify the physicochemical polysaccharides in order to conduct heat treatment without their degradation. This process leads to the plasticization of the polymer and makes it possible to process it by melt molding. The production of unique derivatives of natural polymers with improved thermal stability will allow to increase the scope of application of these polymers in the future, e.g. in the packaging industry, medicine as occlusive dressings, tissue engineering and agriculture [1].

The chitosan used in our research shows high chemical reactivity and biological activity due to the presence of protonated amino groups and primary and secondary hydroxyl groups. Important physico-chemical parameters that have a significant impact on the chemical purity and the value of parameters related to the technological usefulness of this polymer are the average molar mass and the degree of deacetylation. Due to the fact that the decomposition temperature of chitosan is below the melting point, it restricts its use in some industries, e.g. as food packaging material, agricultural cover or as a bone fixation agent [2]. In order to plasticize chitosan and increase its processing on a larger scale using the extrusion technique, research is being conducted on the synthesis of appropriate biomodifiers. The work presents the results of research related to the production of plasticized chitosan in the form of granulate with the assumed parameters based on SEM, FTIR, DSC and TGA analysis. For selected systems of chitosan/biomodifier mixtures, tests of physico-mechanical properties and microbiological purity were carried out.

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RELATION BETWEEN CHITOSAN FILM COMPOSITION, OVERALL MIGRATION, AND SWELLING IN VARIOUS FOOD SIMULANTS

The food packaging sector constitutes a broad industrial area where different polymeric materials are applied. Among different polymers devoted to this purpose, polyethylene terephthalate (PET), polyvinyl chloride (PVC), polyethylene (PE), polypropylene (PP), and polystyrene (PS) are mainly applied. These compounds possess excellent barrier properties, mechanical perfomance, and thermal and oxidative degradation tolerance. However, the polymeric packages made of these polymers do not degrade quickly. The global awareness of plastic pollution has led to the development of environmentally friendly technologies for food packaging, such as biodegradable polymers, edible films and coatings, and active or smart packaging [1].

Different polymers meet the requirements of packaging materials being in agreement with the "green chemistry" idea, i.e., polysaccharides (starch, cellulose, chitosan), proteins, and their derivatives. These biodegradable polymers are recognized as GRAS, Generally Recognized As Safe, and are allowed to be in contact with food products. However, some of the physicochemical properties of these polymers still need to be improved, like mechanical resistance, elasticity, barrier properties, and antioxidative/antimicrobial characteristics.

The modification of the properties mentioned above can be obtained by using different additives. When the modification process is applied, the possible migration of additives into the food products must be tested to ensure the package's safety to the food-consumer health.

The aim of the research was the preparation of a plasticized chitosan package with improved antioxidative properties. The polymeric films containing an equimolar deep eutectic mixture (choline chloride and citric acid) and the natural aqueous olive-wastes extract were prepared. The effect of the extract content on the overall migration in three different food simulants (10% ethanol (hydrophilic food), 3% acetic acid (acidic and hydrophilic food), and isooctane (hydrophobic food)) was evaluated. The migration process was discussed in the context of the swelling phenomenon, the film's molecular structure, the possible crosslinking process, and the hydrophilic/hydrophobic nature of the additive and the external medium. Furthermore, the antioxidative characteristic of the films was also evaluated with the DPPH method.

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PROTECTION OF THE ESOPHAGEAL MUCOSA WITH CHITOSAN GELS

The problem of acidic reflux is still not effectivelly resolved, as evidenced by articles in the available literature. Gastroesophageal reflux disease continues to be a common condition. It is estimated that up to half of adults have single symptoms of the disease. An important aspect of the protective effect of gels is their sufficiently long time to adhere to the mucosa. The conducted research indicates the possibility of obtaining preparations helping to solve this problem. Tested hydrogels are designed to protect the esophageal mucosa against harmful factors [1-5].

The aim on the work was to investigate the influence dextran on the properties of chitosan-containing gels.

In the initial part of the experiment, the influence of chitosan on the physicochemical properties of the tested gels was examined. The formulations were prepared with various pH and rheological properties. On the basis of performed investigations in vitro, it may be assumed that the gels will remain at the site of application in the form of a layer coating the mucous membrane of the oesophagus and protecting it against an irritating effect of gastric content backflow. Gels show the adhesion and the ability to cover the surface of the apparatus simulating the conditions in the esophagus. The texture tests showed the effect of dextran concentration on the adhesion work of the tested gels. All investigation gels contained chitosan. Due to their adhesive properties, the tested gels should stay on the esophageal mucosa for a long time and protect it against the adverse effects of gastric contents. The wide range of pH of the investigated gels enables selection of a preparation with optimal pH for the esophagus. Laboratory tests require clinical confirmation.

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STUDIES ON CHITOSAN GELS WITH POLYVINYL ALCOHOL PROTECTING THE ESOPHAGEAL MUCOSA

Health problems related to the gastrointestinal tract also affect the youngest patients – children. Diagnosing gastrophageal reflux in children is a difficult task. For a long time, the so-called gold standard for diagnosing gastrophageal reflux was 24-hour esophageal pH-metry. Recently, a more modern method has been introduced using the phenomenon of resistance, i.e. esophageal impedance. This method is much more effective than pH-metry, because it allows to detect not only acid gastroesophageal reflux, but also episodes of weak acid or alkaline reflux. The conducted research indicates the possibility of obtaining preparations helping to solve this problem. Tested hydrogels are designed to protect the esophageal mucosa against harmful factors [1-6].

The aim on the work was to investigate the influence polyvinyl alcohol on the properties of chitosan-containing gels.

Taking into account the specificity of treatment of small patients, gels were prepared that meet the relevant requirements. An important aspect of treatment in such cases is the consistency of the preparation that allows easy swallowing. At the same time, the gel should thoroughly cover the esophageal mucosa to protect against the irritating effects of the regurgitated contents. All investigation gels contained chitosan. The formulations were prepared with various pH and rheological properties. The wide range of pH of the investigated gels enables selection of a preparation with optimal pH for the esophagus. Gels show the adhesion and the ability to cover the surface of the apparatus simulating the conditions in the esophagus. The texture tests showed the effect of polyvinyl alcohol concentration on the adhesion work of the tested gels. The investigated gels, thanks to their adhesive properties, should remain on the mucous membrane of the oesophagus for a prolonged time and protect it against the unfavourable effect of gastric content and alkaline reflux. Presented assumptions and investigations in vitro, what is the aim of subsequent studies.

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RESEARCH ON THE PHYSICOCHEMICAL PROPERTIES OF DERMATOLOGICAL APPLICATIONS IN THE PRESENCE OF CHITOSAN CONTAINING AN EXTRACT FROM *HUMULUS LUPULUS L.*

Traditional herbal medicines are widely researched as alternative treatments for many diseases to minimise the potential risk of adverse effects and antibiotic resistance. Numerous studies on hop Humulus lupulus L. extract revealed an active effect not only in the treatment of acne but also in other dermatological conditions [1]. Hop cone extract has documented high antibacterial, antioxidant, anti-collagenase, anti-inflammatory and even antifungal activity thanks to isolated active ingredients such as humulones, lupulones, isohumulones, xanthohumols, polyphenols and others. In order to use the hop extract in dermatology, an appropriate carrier had to be developed, which should be miscible with the hop extract and have the appropriate physicochemical properties to enable the most favorable bioavailability of the hop extract from the semi-solid drug formulation.

Therefore, research was undertaken to obtain the optimal qualitative and quantitative composition of a dermatological application for use in the treatment of inflammatory skin conditions. For this purpose, hop extract obtained from Humulus lupulus L. using a supercritical CO2 extraction method was introduced as an active ingredient into a selected o/w emulsion medium under the name Pentravan with and without the addition of chitosan [2,3].

Studies show that the presence of chitosan significantly affects the rheological properties of oitments, their dynamic viscosity, hardness, consistency, cohesiveness, and blurring time. Formulations on Pentavan substrate with the addition of 2% chitosan showed the best application possibilities. The applications were subjected to the dissolution testing of cohumulone which is an analogue of humulone contained in hop cone extract.

Based on the data analysis, it was shown that the concentration of chitosan in the formulations has a significant effect on the dissolution testing parametres of the active ingredient, and with an increase in its of concentrarion, the desired effect of prolonged release time of the active ingredient was obtained while maintaining the emulsifying properties of Pentravan.

Pharmaceutical availability studies of cohumulone from Pentravan vehicle with the addition of 2% chitosan showed a decrease in the average percentage of released active ingredient of 8.25 % cohumulone at a constant release rate of 0.00268 h-1 compared to the application formulation with Pentravan base without chitosan. The tested formulations provide a great range of possibilities for matching properties to the disorder and skin requirements.

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MICROWAVE-ASSITED EXTRACTION OF CHITOSAN FROM FILAMENTOUS FUNGAL BIOMASS

Filamentous fungi are widely used in biotechnology as a source of many useful compounds, including organic acids, enzymes, lipids, antibiotics or polysaccharides, which include chitin and chitosan. Chitosan is naturally present in the cell walls of filamentous fungi classified to the Ascomycetes, Basidiomycetes, Deuteromycetes and Zygomycetes. The most common species from which chitosan is isolated are *Absidia* spp., *A. niger*, *Mucor rouxii* or *Rhizophus oryzae* [1-2].

Microwave radiation is commonly defined as the part of the electromagnetic spectrum with a wavelength of 1 mm to 1 m (or the equivalent frequency range of 300 GHz to 300 MHz). Microwaves are increasingly used in chemically and enzymatically catalyzed reactions. It is also used to support the extraction of various substances from microbial cells, including biopolymers [3-4]. Microwave-assisted extraction is an eco-friendly technique that offers many benefits, the most important of which are reduced extraction time and solvent consumption, as well as improved isolation efficiency of cellular components.

The aim of this study was to develop a method of chitosan extraction from the cells of three strains of filamentous fungi: *Mucor circinelloides*, *M. racemosus* and *M. hiemalis* using microwave energy. Chitosan extraction was carried out using a specialized CEM Discover SP-D microwave reactor. Microwave-assisted extraction of chitosan (200 W, 30 min, 25°C, 2500 Pa) allowed to obtain a higher yield of chitosan compared to that obtained in the conventional extraction process. It was found that the amount of biopolymer is 6.5, 9 and 8.5% higher for the *M. circinelloides*, *M. racemosus* and *M. hiemalis* strains, respectively.

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PATENTOLOGY OF CHITINOUS BIOMATERIALS

Chitin and chitosan are two important biopolymers that have garnered significant attention in diverse scientific and technological fields. Chitin, as a fundamental structural material found in various organisms, has attracted the interest of experts in biomedicine, materials science, and technology [1]. Additionally, cationic chitosan, derived from chitin, is extensively studied due to its unique properties, including biodegradability and biocompatibility. These attributes contribute to its broad applicability and patentability in different domains of modern science and technology. Notably, biomedical, material science, biotechnology, and chemical sectors have witnessed a substantial number of chitosan-related patents, while food, cosmetics, environmental protection, and agricultural fields have received relatively less attention [2].

This presentation offers a patentological overview of chitin and chitosan, emphasizing their significance as renewable bioactive materials that have propelled advancements in various applied science disciplines on a global scale. This study provides essential information for the advancement of patentology in relation to chitin and its derivatives. Additionally, the presentation provide valuable insights into the current state of chitosan research and its potential applications.

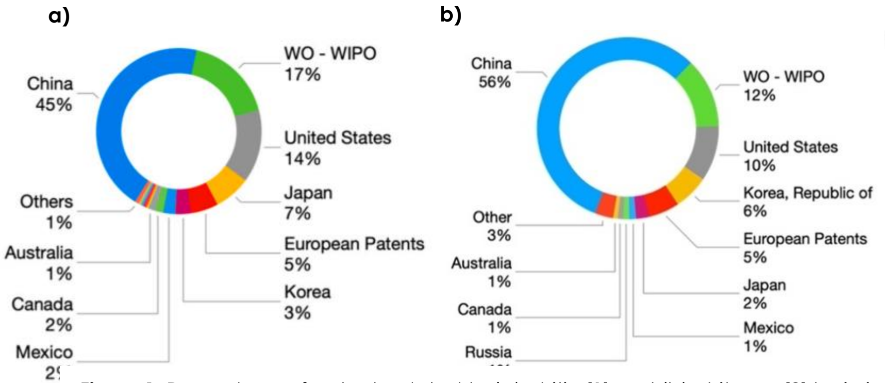

Figure 1. Percentage of patents related to (a) chitin [1] and (b) chitosan [2] by jurisdiction.

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CHITOSAN AND SYNTHETIC FUNGICIDES: SYNERGY OF ANTIFUNGAL ACTIVITY AGAINST *FUSARIUM GRAMINEARUM*

Fusarium graminearum is a fungal pathogen that causes Fusarium head blight disease in crop plants such as barley (*Hordeum vulgare* L.). Fungal infection, in addition to decreasing the total grain yield, also leads to the accumulation of mycotoxins, making it useless for food and feed. Conventional fungicides are the main method to protect plants from fungal pathogens. However, the use of such substances presents potential risks to the environment and the organisms that live in it.

Chitosan, which is a derivative of chitin, is considered a strong antifungal agent. Supposedly, the mechanism of antifungal action is based on disruption of fungal cell membranes, and subsequent destabilisation of intracellular metabolism, reviewed in [1]. Furthermore, in the case of plants, the application of chitosan stimulates biomass and induces genes functional in immune responses. Simultaneously, chitosan is biodegradable and considered safe for the environment. Such characteristics of chitosan make it perfect for large-scale applications related to plant protection. Currently, the known mechanism of antifungal action of chitosan, that is cell membrane disruption, could improve the antifungal efficiency of conventional fungicides. This might imply that simultaneous use of chitosan and conventional fungicide will lead to more efficient antifungal activity than each of the compounds used independently [2, 3].

The aim of our work is to test the synergistic activity of chitosan and conventional fungicides based on *Fusarium graminearum* infection of barley. Collection of 14 chitosan samples with different parameters (degree of deacetylation and molecular weight) were screened to select the sample with the strongest effect on *Fusarium graminearum* growth. Further, we selected several commercially important synthetic fungicides with different active substances such as thiophanate-methyl (Topsin), azoxystrobin (Amistar 250 SC), tebuconazole (Fungimat), metconazole (Micosar), and copper oxychloride (Miedzian Extra 350 SC). The antifungal activity of all substances and their combinations was analysed using *in vitro* assays (microtiter plates) and *in planta* setup (barley leaves inoculated with *F. graminearum* under controlled conditions). In our preliminary studies, we used Topsin, a fungicide containing thiophanate-methyl as an active ingredient, which antifungal activity is the result of inhibition of microtubule assembly. The fungicide, supplemented with 100 ppm chitosan, showed enhanced by almost 50% antifungal activity compared to the fungicide alone.

This strategy would potentially help to reduce the volumes of synthetic fungicides required for efficient plant protection. We assume that the results will support development of more ecological means of plant protection. It will also shed some light on chitosan's antifungal mode of action.

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MULBERRY SILKWORMY (*BOMBYX MORI*) AS AN ALTERNATIVE SOURCE OF CHITIN – ISOLATION AND ANALYSIS OF THE RESULTING POLYSACCHARIDE

Chitin, one of the most common polysaccharides, is seen as a very attractive biomaterial due to its physico-chemical and biological properties, with applications in areas such as medicine and the food industry. Currently, marine animals, primarily crustaceans, serve as the main source of chitin. However, due to current predictions of the chitin market and the general increase in interest in biopolymers, it has become necessary to explore alternative sources of this polysaccharide. Insects appear to be one of the more interesting sources of chitin as evidenced by the increased interest on the subject of extracting chitin from various insects, an example of such an insect being the mulberry silkworm (Bombyx mori). Silkworms have been known for centuries for the silk extracted from their cocoons, but it turns out that they can also be used as an interesting and attractive source of chitin, as evidenced by emerging publications on the subject. In this work, the extraction of chitin from different stages of the mulberry silkworm was carried out. The chemical extraction procedure used, included the following processes: Folch lipid extraction (chloroform/methanol mixture, 24 h at room temperature), demineralisation (2 h at 60°C, HCl solution), deproteinization (24 h at 60°C, NaOH solution) and decolourisation (30 min at 60°C, 35% H_2O_2).

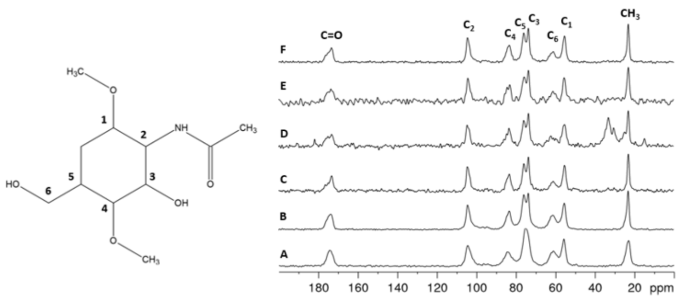

Figure 1. ^{13}C NMR spectra obtained for: (A) β-chitin standard sample, (B) a-chitin standard sample, (C) larval stage, (D) cocoon, (E) pupal residue, (F) adult form/imago.

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