24th Conference

of Polish Chitin Society

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

Tyniec, September 19-21st 2018

Polish Chitin Society

XXIV Conference

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

Conference place: "The Guest House" Benedictine Abbey in Tyniec

Scientific Committee: Prof. Małgorzata JAWORSKA, Ph.D.,D.Sc. – chairman

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Prof. Mirosława EL FRAY, Ph.D.,D.Sc.
Prof. Maria MUCHA, Ph.D.,D.Sc.
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Urszula FILIPKOWSKA, Ph.D.,D.Sc.

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Michał KACZMAREK, M.Sc.

Co-organizers: Institute of Biopolimers and Chemical Fibres, Lodz, Poland

We would like to inform that the organization of the XXIV Conference on "New Aspects on the Chemistry and Applications of Chitin and its Derivatives" is funded under contract 779/P-DUN/2018 from the resources of Ministry of Science and Higher Education allocatied for activities which disseminate the science.

Ministry of Science and Higher Education
Republic of Poland

XXIV Konferencja Polskiego Towarzystwa Chitynowego

„NOWE ASPEKTY W CHEMII I ZASTOSOWANIU CHITYNY I JEJ POCHODNYCH"

Tyniec, 19-21 września 2018 r.

Polskie Towarzystwo Chitynowe

XXIV Konferencja

„Nowe aspekty w chemii i zastosowaniu chityny i jej pochodnych"

Miejsce Konferencji: „Dom Gości" Opactwo Benedyktynów w Tyńcu

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Organizacja XXIV Konferencji Naukowej "Nowe aspekty w chemii i zastosowaniu chityny i jej pochodnych" - zadanie finansowane w ramach umowy 779/P-DUN/2018 ze środków Ministra Nauki i Szkolnictwa Wyższego przeznaczonych na działalność upowszechniającą naukę.

Ministerstwo Nauki
i Szkolnictwa Wyższego

Honorable quests,

We meet for 24th time on Conference „New Aspects in Chemistry and Applications of Chitin and Its Derivatives".
I do hope that our meeting will contribute as previously to the progress of Polish chitin and chitosan science.

I wish the fruitful debate to all participants of 24th Conference.

On behalf of the Polish Chitin Society

Prof. Małgorzata Jaworska

The President

Tyniec, September 19-21st 2018

Szanowni Państwo,

Po raz dwudziesty czwarty spotykamy się na Konferencji pt. „Nowe aspekty w chemii i zastosowaniu chityny i jej pochodnych".
Mam nadzieję, że tak jak poprzednio nasze spotkanie przyczyni się do rozwoju polskiej nauki w dziedzinie chityny i chitozanu.

Życzę wszystkim uczestnikom XXIV Konferencji owocnych obrad.

W imieniu Zarządu PTChit

Prof. nzw. dr hab. inż. Małgorzata Jaworska

Prezes

Tyniec, 19-21 września 2018 r.

SCHEDULE OF CONFERENCE

September 19th 2018 – Wednesday

16^{00}- 18^{00}	**Registration**
18^{00}- 22^{00}	**Welcome reception**
	Dinner

September 20th 2018 – Thursday

9^{00}-9^{10}		**Openning ceremony** **Prof. Małgorzata M. Jaworska, Ph.D.,D.Sc.**
9^{10}-9^{40}		**Prof Henryk Struszczyk prize-giving ceremony**
Session A		**Physico-chemical session**
Chairman		**Prof. Maria Mucha, Ph.D.,D.Sc.**
A 1	**9^{40}-10^{00}**	**George A.F. Roberts** DYE-CHITOSAN INTERACTIONS AT ACID pH
A 2	**10^{00}-10^{15}**	**Carmen Hernandez, Hiroyuki Saimoto, Shinsuke Ifuku, Keiko Shirai** STRUCTURAL CHARACTERIZATION OF BIOLOGICAL CHITIN NANOFIBERS OBTAINED BY ACID AND DEACETYLATED-SURFACE METHOD
A 3	**10^{15}-10^{30}**	**Barbara Kaker, Silvo Hribernik, Tamilselvan Mohan, Rupert Kargl, Karin Stana Kleinschek, Egon Pavlica, Shingjiang Jessie Lue, Mojca Božič** RENEWABLE CHITOSAN-GRAPHENE OXIDE NANOCOMPOSITES AS POTENCIAL MATERIAL FOR ETHANOL FUEL CELLS
A 4	**10^{30}-11^{45}**	**Urška Jančič, Silvo Hribernik, Tamilselvan Mohan, Rupert Kargl, Karin Stana Kleinschek, Mojca Božič** PHYSICOCHEMICAL PROPERTIES OF CHITOSAN-BASED FILMS DEPENDING ON THE pH OF CHITOSAN SOLUTION
10^{45}-11^{15}		**Coffee/tea break**

September 20th 2018 – Thursday

Session B		**Medical Session**
Chairman		**Prof. Barbara Kochańska, Ph.D.,D.Sc.**
B 1	**11^{15}- 11^{35}**	**Makoto Anraku, Shinsuke Ifuku, Daisuke Iohara, Masaki Otagiri, Fumitoshi Hirayama** CLINICAL APPLICATIONS OF CHITOSAN AND SURFACE-DEACETYLATED CHITIN NANOFIBERS ON OXIDATIVE STRESS RELATED DISEASES
B 2	**11^{35}- 11^{55}**	**Jialong Shen, Ahmed Nada, Nabil Abou Zeid, Samuel Hudson** SYNTHESIS AND CHARACTERIZATION OF HEMOSTATIC LAYERS FROM CHITOSAN AND OTHER BIOMATERIALS

September 20th 2018 – Thursday

Session 1		**Poster Session**
SP 1	**11^{55}- 12^{40}**	**Krzysztof Śmiechowski, Jan Żarłok, Jan Skiba** THE RESEARCH ON THE USE OF CHITOSAN IN LEATHER TANNING
SP 2		**Małgorzata Gnus, Roman Turczyn** COMPARISION OF THE SEPARATION PROPERTIES OF COMPOSITE CHITOSAN MEMBRANES CONTAINING METAL OXIDES IN DIFFERENT OXIDATION STATES
SP 3		**Katarzyna Nawrotek, Michał Tylman, Karolina Rudnicka, Justyna Gałkowska, Marek Wieczorek** ELECTRODEPOSITION MECHANISM OF CHITOSAN TUBULAR IMPLANTS INTENDED FOR ENHANCING AN INTRINSIC PATH OF AXON REGENERATION
SP 4		**Aleksandra Grzabka-Zasadzińska, Sławomir Borysiak** CHITOSAN COMPOSITES WITH NANOMETRIC CELLULOSE AND MODIFIED LAYERED SILICATE
SP 5		**Aleksandra Grzabka-Zasadzińska, Sławomir Borysiak,** NANOMETRIC CELLULOSE GRAFTED WITH DICARBOXYLIC ACIDS AS A FILLER FOR CHITOSAN
SP 6		**Georg Schierz, Klaus Boening, Heike Meissner, Mieszko Wieckiewicz , Katarzyna Walczak** ANTIFUGAL ACTIVITY OF CHITOSAN-SALT MODIFIED PMMA DENTURE MATERIALS
SP 7		**Maria Wiśniewska-Wrona, Ewa Kopania, Justyna Wietecha, Katarzyna Dziedziczak, Beata Pałys, Sylwia**

		Jagodzińska, Monika Owczarek, Magdalena Szalczyńska POSSIBLE APPLICATION OF CHITOSAN IN PROTECTION OF LEGUMINOUS PLANTS
SP 8		**Sylwia Jagodzińska, Beata Pałys, Dariusz Wawro** MICROSTRUCTURE OF CHITOSAN FILMS AND THE CONTACT ANGLE
SP 9		**Monika Owczarek, Monika Szkopiecka, Sylwia Jagodzińska, Marzena Dymel, Michał Kudra, Karolina Gzyra Jagieła, Patrycja Miros-Kudra** BIODEGRADABLE NONWOVEN WITH AN ACTIVE LAYER FOR COSMETIC PURPOSES
SP 10		**Anna Ilnicka, Jerzy P. Łukaszewicz** THERMAL CONVERSION OF CHITIN AND CHITOSAN TO THE NITROGEN-RICH CARBON FOAMS
SP 11		**Giedre Jakubauskaite, Lina Vaiciulyte-Funk, Vykintas Baublys, Vaida Tubelyte, Alona Oberemko, Murat Kaya, Povilas Mulercikas, Sonata Kazlauskaite** EVALUATION OF CHITOSAN ANTIBACTERIAL EFFECTS ON GROWTH OF FOOD PATHOGENIC BACTERIA
SP 12		**Alona Oberemko, Asier Martinez Salaberria, Rita Saule, Jalel Labidi, Gintautas Saulis, Vaida Tubelyte, Vykintas Baublys, Murat Kaya** PHYSICOCHEMICAL PROPERTIES AND IN VITRO CYTOTOXICITY STUDIES OF CHITOSAN AS A POTENTIAL AGENT FOR CANCER TREATMENT
SP 13		**Sonia Żółtowska-Aksamitowska, Christine Klinger, Iaroslav Petrenko, Tomasz Machałowski, Teofil Jesionowski, Marcin Wysokowski, Hermann Ehrlich** SPONGES (PORIFERA) AS SOURCE OF MORPHOLOGICALLY DEFINED CHITIN
12^{40} -13^{40}		**General Assemble of the Polish Chitin Society**
13^{40}-15^{00}		**Lunch break**
15^{00}-16^{00}		**Guided tour of the Abbey**
16^{00}-16^{30}		**Organ concert**
19^{00}- 23^{30}		**Conference dinner** Restaurant "*Tarasy Tynieckie*"

September 21st 2018 – Friday

Session 2		Poster Session
SP 14	**9^{30}- 10^{10}**	**Maria Doligalska**, **François-Mayeul Boudet**, **Marzena Laskowska** CHITOSAN ANTI-AMOEBA ACTIVITY TO AXENIC STAGE OF *ACANTHAMOEBA* SP.
SP 15		**Urszula Filipkowska**, **Tomasz Jóźwiak**, **Paula Bugajska**, **Małgorzata Kuczajowska-Zadrożna** THE INFLUENCE OF CHITOSAN CONTENT IN HYDROGEL BEADS ON THE SORPTION EFFECTIVENESS OF REACTIVE BLACK 5 DYE
SP 16		**Tomasz Jóźwiak**, **Urszula Filipkowska**, **Paula Bugajska**, **Małgorzata Kuczajowska-Zadrożna** THE INFLUENCE OF SALINITY ON THE REACTIVE BLACK 5 SORPTION EFFICIENCY ON HYDROGEL CHITOSAN SORBENTS
SP 17		**Maciej Galiński**, **Izabela Stępniak** CHITOSAN IN ELECTROCHEMICAL DOUBLE-LAYER CAPACITORS
SP 18		**Barbara Kochańska** THE POSSIBILITY OF CHITOSAN ASCORBATE APPLICATION IN DENTISTRY - THE DIRECTIONS OF INVESTIGATIONS
SP 19		**Maria Szcześniak**, **Bożena Grimling**, **Jan Meler**, **Sandra Biskup**, **Bożena Karolewicz** THE EFFECT OF SELECTED HYDROPHILIZING SUBSTANCES ON CHITOSAN BASED DERMATOLOGICAL HYDROGELS PROPERTIES
SP 20		**Katarzyna Małolepsza-Jarmołowska** THE PROPERTIES OF INTRAVAGINAL GLOBULES CONTAINING A LACTIC ACID-CHITOSAN COMPLEX
SP 21		**Katarzyna Małolepsza-Jarmołowska** PHARMACEUTICAL RESEARCH OF HYDROPHILIC GLOBULES CONTAINING A LACTIC ACID-CHITOSAN COMPLEX
SP 22		**Renata Czechowska-Biskup**, **Radosław Wach**, **Piotr Ulański** HYDROCHLORIC ACID AS A SOLVENT FOR CHITOSAN EXAMINATION OF PHYSICOCHEMICAL BEHAVIOR
SP 23		**Jan Meler**, **Bożena Grimling**, **Maria Szcześniak**, **Bożena Karolewicz**, **Paweł Biernat** ADSORPTION STUDY OF DROTAVERINE ON CHITOSAN IN AN *IN VITRO* PHARMACEUTICAL MODEL
SP 24		**Bożena Grimling**, **Jan Meler**, **Maria Szcześniak**, **Karolina Barańska**, **Bożena Karolewicz** INFLUENCE OF CHITOSAN ON THE PHARMACEUTICAL PROPERTIES OF DENTAL APPLICATIONS

SP 25		**Katarzyna Struszczyk-Świta, Michał Kaczmarek, Mirosława Szczęsna-Antczak, Tadeusz Antczak** CHITOOLIGOSACCHARIDES SYNTHESIS USING ENZYMES OF MUCOR CIRCINELLOIDES IBT-83
SP 26		**Magdalena Gierszewska, Marta Ziegler-Borowska** DIALDEHYDE STARCH AS A NOVEL CHITOSAN CROSSLINKER

September 21st 2018 – Friday

Session C		**Biotechnological Session**
Chairman		**Prof. Maria Doligalska, Ph.D.,D.Sc.**
C 1	**10^{10}-10^{30}**	**Murat Kaya** THREE-DIMENSIONAL CHITIN ISOLATION AND POSSIBLE APPLICATIONS
C 2	**10^{30}-10^{45}**	**Jesus Rojas, Zaizy Rocha, Hugo Nájera, Humberto Gonzalez, Keiko Shirai** PURIFICATION OF β-N-ACETYLHEXOSAMINIDASE OF *LECANICILLIUM LECANII* AND ITS APPLICATION IN THE OLIGOSACCHARIDES PRODUCTION
C 3	**10^{45} –11^{00}**	**Michał Kaczmarek, Katarzyna Struszczyk-Świta, Mirosława Szczęsna-Antczak, Maurycy Daroch, Tadeusz Antczak** CHITIN DEACETYLASES FROM *MUCOR CIRCINELLOIDES* IBT-83 AND THEIR USE IN THE SYNTHESIS OF A MULTI-ENZYME COMPLEX
11^{00}-11^{30}		**Coffee/tea break**

September 21st 2018– Friday

Session D		**Physico-chemical session**
Chairman		**Katarzyna Małolepsza-Jarmołowska, Ph.D.,D.Sc.**
D 1	**11^{30}-11^{45}**	**Katarzyna Pieklarz, Zofia Modrzejewska, Michał Tylman** STRUCTURAL CHARACTERISTICS OF THERMOSENSITIVE COMPOSITIONS BASED ON CHITOSAN
D 2	**11^{45}-12^{00}**	**Ewelina Chrzanowska, Magdalena Gierszewska, Wojciech Kujawski** NOVEL NANOCOMPOSITE CHITOSAN MEMBRANES ON POLYAMIDE-6 SUPPORT FOR PERVAPORATION
12^{00}-12^{30}		**Closing of the conference**
12^{30}-13^{30}		**Lunch**

PHYSICO-CHEMICAL SESSION

George AF Roberts
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DYE-CHITOSAN INTERACTIONS AT ACID PH

An understanding of the forces involved in the interaction between anionic dyes and chitin or chitosan is important both for applications, such as water purification [1, 2], and analysis [3]. Currently there are three attractive forces recognised as important in ionic dye-ionic substrate systems, including chitosan-acid dye systems: van der Waals' (London) forces, hydrogen bonding and hydrophobic bonding [3]. The role of electrostatic forces in the interaction is more controversial for as stated by Peters [4] "Clearly the affinity of an anion cannot be attributed to electrostatic interaction between the charged groups in the dye and those in the substrate since the adsorption properties of the dye acids would be similar to those of HCl", while Rattee and Brewer argued [5] in respect of coulombic interactions in dye sorption that "No distinction is made between ions of the same charge although these might be as simple as a chloride ion or as complex as an azo dye".

Protonated chitin and chitosan interact strongly with low molecular weight anionic acid dyes that have no affinity for the unprotonated forms of these substrates [6]. Obviously the $-NH_3^+$ groups of the former substrates are critical to the dye-substrate interaction but this cannot be simple ion pair formation as otherwise simple inorganic anions such as Cl^- would compete effectively with anionic dyes for the $-NH_3^+$ dye sites, and this is not observed in practice even when the molar concentration of Cl^- is many times greater than that of the dye. Neither can it be due to hydrophobic interaction between dye and chitin or chitosan as otherwise there would be an appreciable uptake of the dye at neutral pH. A study of the effect of inorganic anions on uptake of dye shows that the effectiveness of these anions increases with increase in size of the hydrated ion e.g. $Cl^{-1} < Br^{-1} < I^{-1} < SCN^{-1}$, which is the opposite order to that expected with Bjerrum-type ion pairing. Based on this a possible solution, which enables ionic sites on polymeric substrates to not only distinguish between dye ions and simple inorganic ions but also between different inorganic ions, is proposed.

References:

[1] M. Kuczajowska-Zadrożna, U. Filipkowska, T. Jóźwiak, P. Szymczyk, The use of polysaccharides for Acid Red 18 anionic dye removal from aqueous solutions. Progress on Chemistry and Applications of Chitin and its Derivatives, XXII (2017) 106-117.

[2] V.M. Esquerdo, T.R.S. Cadaval Jr., G.L. Dotto, L.A.A. Pinto, Chitosan scaffold as an alternative adsorbent for the removal of hazardous food dyes from aqueous solutions. Journal of Colloid and Interface Science, 424 (2014) 7-15

[3] Determination of the degree of N-acetylation of chitin and chitosan, G.A.F. Roberts in "Chitin Handbook", R.A.A. Muzzarelli and M.G. Peter (Eds.), European Chitin Society, Lyon, 1997.

[4] R.H. Peters, "Textile Chemistry: Volume III - The Physical Chemistry of Dyeing", p.245, Elsevier, London, 1975.

[5] I.D. Rattee, M.M. Brewer, "The Physical Chemistry of Dye Adsorption" p. 14, Academic Press, London, 1974.

[6] G.G. Maghami and G.A.F. Roberts, Studies on the adsorption of anionic dyes on chitosan. Makromol. Chemie, 188 (1988) 2239-2243.

Carmen Hernandez [1], Hiroyuki Saimoto [2], Shinsuke Ifuku [2], Keiko Shirai [1]

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[2] – Tottori University, Graduate School of Engineering, Koyamacho-Minami 4-101, Tottori city, Tottori Prefecture, 680-8550, Japan.

STRUCTURAL CHARACTERIZATION OF BIOLOGICAL CHITIN NANOFIBERS OBTAINED BY ACID AND DEACETYLATED-SURFACE METHOD

Chitin (Ch), is the main component of crustacean shell, fungal cell walls, and insect cuticle. An innovative process of Ch obtaining is by lactic acid fermentation, minerals are solubilized in situ and proteases remove protein from the shells [1,5]. In addition, it allows obtaining Ch with high molecular weight and crystallinity index (I_{CR}) [2]. The main constraint of Ch applications is its null solubility in water and common organic solvents. Therefore, it has been proposed several physical and chemical modifications to improve solubility in aqueous media [3]. In this regard, several authors have proposed the preparation of nanofibers (NFs) because the nanometric structure allows a high surface-to-volume ratio and can be dispersed homogeneously in water [4].

Ch was obtained from the lactic acid fermentation processing of 1 ton of shrimp and purified by the method of Cira et al. [5]. NFs were obtained by two methods, acid [6] and surface-deacetylation [7]. Chs (1% w/v) was dissolved in an acetic acid solution (0.08 N), pretreated in a grinder twice (MKCA6-3, Masuko Sangyo) at 1500 rpm. Then, the dispersion of each sample was mechanically treated for 1, 5, 10 and 30 cycles by the high-pressure water jet (HPWJ) system (Star Burst Mini, HJP-25001S, Sugino Machine). They were observed by scanning electron microscopy (JSM-6700F JEOL) (Fig. 1). For both Ch, most of the fibers become shorter and thinner that facilitated fibrillation in NFs attributed to the electrostatic repulsions. NFs significantly decreased the width from 28 to 8.6 nm for the acid method, while for the surface-deacetylated method was 13.5 to 7.3 nm at 0 and 30 cycles respectively. The viscosity of the NFs was determined in a Brookfield DV-E digital equipment (Middleboro, MA) at 27°C. This was affected by the thickness and length since the thin and short nanofibers are often entangled and increase the viscosity. Therefore, a maximum viscosity was observed for the acid method at ten cycles (6770 Cp), while for the surface-deacetylated method was in the first cycle (7220 Cp). X-ray diffractograms were obtained (Ultima IV equipment, Rigaku at 40 kV and 40 mA). The I_{CR} did not change significantly with the acid method and HPWJ cycles,

however, decreased with the deacetylated method at 1, 5, 10 and 30 cycles with I_{CR} of 62±0.3%, 58.5±0.8%, 56.8±0.3%, 55±0.8%, respectively. NFs obtained by both methods have nanometric sizes with different structural characteristics that can be used in various industrial applications due to their water solubility.

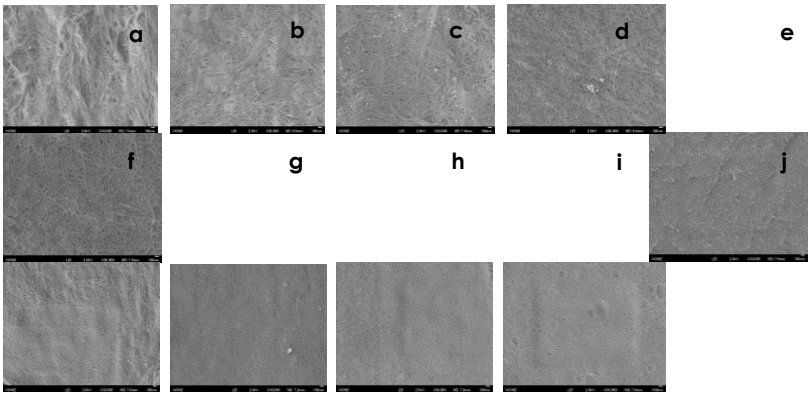

Figure 1. SEM of NFs after cycles of HPWJ with acid (upper) and surface-deacetylated method (below), at 0 (a;f), 1(b;g), 5(c;h), 10(d;i) and 30 (e;j) cycles.

Acknowledgements:

The experimental work was carried out with funding of CONACYT (Project No. 237292).

References:

[1] K. Shirai, M. Plascencia, L. Cira, S. Huerta, IMPI no. 247295. Int. Cl.8:A23J1/04; C08B37/08; C22B26/20, 2000.

[2] N. Pacheco, M. Gonzalez, M. Gimeno, E. Barzana, S. Trombotto, L. David, K. Shirai, Structural characterization of chitin and chitosan obtained by biological and chemical methods. Biomacromolecules, 12 (2011) 3285–3290.

[3] J. Kumirska, M. Czerwicka, Z. Kaczynski, A. Bychowska, K. Brzozowski, J. Thoeming, et al., Application of spectroscopic methods for structural analysis of chitin and chitosan. Marine Drugs, 8 (2010) 1567–1636.

[4] S. Ifuku, M. Iwasaki, M. Morimoto, H. Saimoto, Graft polymerization of acrylic acid onto chitin nanofiber to improve dispersibility in basic water. Carbohydrate Polymer, 90 (2012) 623 - 627.

[5] L. Cira, S. Huerta, G. Hall, K. Shirai, Pilot scale lactic acid fermentation of shrimp wastes for chitin recovery. Process Biochemistry, 37 (2002) 1359-1366.

[6] S. Ifuku, A. Ikuta, M. Egusa, H. Kaminaka, H. Izawa, S. Morimoto, H. Saimoto, Preparation of high-strength transparent chitosan film reinforced with

surface-deacetylated chitin nanofibers, Carbohydrate Polymers, 98 (2013) 1198– 1202.

[7] Y. Fan, T. Saito, A. Isogai, Individual chitin nano-whiskers prepared from partially deacetylated a-chitin by fibril surface cationization. Carbohydrate Polymers, 79 (2010) 1046–1051.

Barbara Kaker [1], Silvo Hribernik [1,2], Tamilselvan Mohan [1], Rupert Kargl [1,2,3], Karin Stana Kleinschek [1,2,3], Egon Pavlica [4], Shingjiang Jessie Lue [5], Mojca Božič [1,2]

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[5] – Chang Gung University, Department of Chemical and Materials Engineering, Kwei-shan, Taoyuan 33302, Taiwan

RENEWABLE CHITOSAN-GRAPHENE OXIDE NANOCOMPOSITES AS POTENCIAL MATERIAL FOR ETHANOL FUEL CELLS

A direct alkaline ethanol fuel cell (DAEFC) directly converts the chemical energy of liquid ethanol to electricity. The most important feature of such power source is the usage of on alkaline electrolyte membrane, which enables the use of Pt-free catalysts, achieving faster reaction kinetics and employing environmentally-friendly polysaccharide materials. DAEFCs also have an advantage of a simple, compact system with high specific energy. Thus, this type of cell can be a good alternative for powering portable and mobile electronic devices [1]. The usage of chitosan polymer (CS) as an alkaline membrane material is quite novel and it has been shown to be a suitable polymer for application in fuel cells. The additional incorporation of graphene oxide (GO) provides the enhancement of mechanical properties and ionic conductivity of the membrane, as well as ethanol permeability reduction. In addition, CS-based membranes are often modified with quaternary ammonium groups, to introduce higher amount of charged groups [2].

Quaternized CS composite membranes were produced by dispersion of graphene oxide (GO) and benzyltrimethylammonium chloride (BTMAC) in neutralized chitosan polymer solution. For membrane formation the solution casting technique was used with further drying and post-surface neutralization with alkali. The obtained membranes were characterized in terms of morphology (scanning electron (SEM) and atomic force microscope (AFM)), hydrophilicity/hydrophobicity was determined by contact angle measurements, ionic conductivity by alternating current impedance spectroscopy and direct ethanol fuel cell performance was evaluated. In addition, alkali uptake and ethanol permeability were determined. SEM images show the morphology of

pristine CS, CS-0.01 wt.% GO and quaternized membrane (Figure 1). The microstructure changes and uniform dispersion of GO sheets can be observed. We obtained relatively hydrophobic membranes (contact angles around 99 °) with high ionic conductivity up to 74 (± 10) mS/cm. Membrane performance in DAEFC exhibited a peak power density of 70 mW/cm2 and an open-circuit voltage of 0.8 V at 80 °C. The results revealed the potential of novel membranes for ethanol fuel cell application.

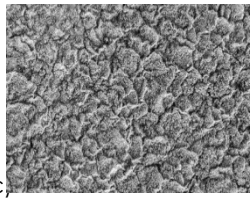
a)  b)  c) 

Figure 1. SEM images of CS composite membranes: a) pristine CS, b) CS-0.01 wt.% GO and c) CS-0.01 wt.% - 0,01 wt.% BTMAC.

Acknowledgements:

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References:

[1] Y. S. Li, T. S. Zhao*, Z. X. Liang, Effect of polymer binders in anode catalyst layer on performance of alkaline direct ethanol fuel cells, Journal of Power Sources, 190 (2009) 223-229.

[2] L. García-Cruz, et al., High Performance of Alkaline Anion-Exchange Membranes Based on Chitosan/Poly (vinyl) Alcohol Doped with Graphene Oxide for the Electrooxidation of Primary Alcohols, Journal of Carbon Research, 2 (2016) 1-19.

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PHYSICOCHEMICAL PROPERTIES OF CHITOSAN-BASED FILMS DEPENDING ON THE PH OF CHITOSAN SOLUTION

Increased use of petroleum-based synthetic polymers has led to accumulation of plastic waste and ecological problems. Therefore, the use of biodegradable polymers of renewable resources are considered as the most advantageous method to face up to this problem. Among the biodegradable films, a considerable attention has been given to chitosan - natural polymer obtained mainly from shellfish processing waste by deacetylation of chitin. Chitosan has reactive amino and hydroxyl groups. Due to the amino groups, it is a cationic polyelectrolyte soluble in weakly acidic solutions with a charge density dependent on the pH and the degree of deacetylation. It has been applied in many fields because of its unique properties such as nontoxicity, biodegradability, biocompatibility, antimicrobial activity, heavy metal adsorption effect, antioxidation effect and bioadhesive characteristics [1]. Due to its excellent film forming ability it has been especially used as edible films and coatings. However, chitosan films are highly permeable to water vapor and show poor mechanical properties what limits their use. These limitations could be monitored by changing pH of the film forming solution, used solvents, concentration of chitosan or the addition of plasticizing agents, organic/inorganic molecules and/or natural/ synthetic polymers [2].

The aim of this study was to prepare chitosan-based films using simple solution casting method and investigate the pH effect of chitosan film-forming solution on the microstructure, chemical structure, hydrophilicity, crystallinity, mechanical performance, water vapor and oxygen barrier properties of the films before and after treatment with sodium hydroxide. The chitosan-based films were prepared from chitosan solution at different pH (2, 4 and 6). Films were afterwards treated with NaOH and compared to one without NaOH treatment. The AFM and SEM images (Figure 1) of the films showed that with increasing pH surface

roughness decreased and films became smoother. Mechanical and oxygen barrier properties were also improved by increasing pH of the chitosan solution. The chitosan-based films at pH 6 before and/or after treatment with NaOH showed promising results in increasing hydrophobicity and crystallinity. Post film treatment with NaOH improved examined physicochemical properties due to re-packaging of chitosan polymer chains into regular structure and formation of the new hydrogen bonds.

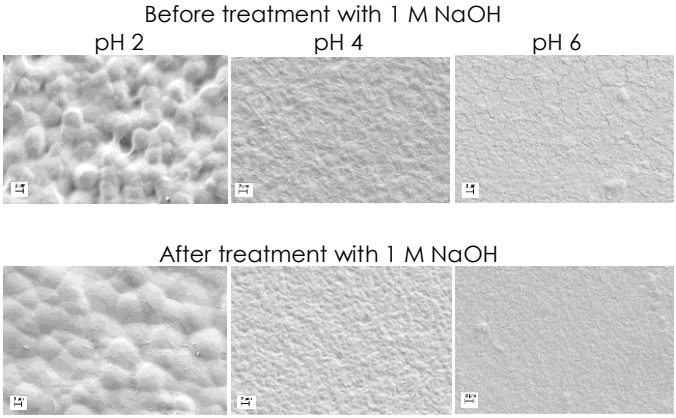

Figure 1: SEM images of 1.0 wt.% chitosan-based films before and after treatment with NaOH.

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References:

[1] N. Nady, S. H. Kandil, Novel Blend for Producing Porous Chitosan-Based Films Suitable for Biomedical Applications, Membranes, 1 (2018) 1-18.

[2] H. Wang, J. Qian, F. Ding, Emerging Chitosan-Based Films for Food Packaging Applications, Journal of Agricultural Food Chemistry, 66 (2018) 395−413.

MEDICAL SESSION

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CLINICAL APPLICATIONS OF CHITOSAN AND SURFACE-DEACETYLATED CHITIN NANOFIBERS ON OXIDATIVE STRESS RELATED DISEASES

Chitosan has been proposed to be a suitable functional material for treating oxidative stress-related diseases, because of its biocompatibility, biodegradability, non-toxicity, adsorption properties and antioxidant activities. The antioxidant properties of chitosan derivatives are subjects of considerable interest and these properties have been evaluated in in vivo studies. In human subjects or model rats with the metabolic syndrome, chitosan has a high degree of antioxidant activity as well as antilipidemic effects. Furthermore, in humans or model rats with chronic renal failure (CRF), chitosan has been reported to have a high antioxidant activity as well as reno-protective effects. From these results, we hypothesize that chitosan reduces the levels of lipids and/or uremic toxins that induce the production of reactive oxygen species in the intestinal tract, and, in the case of the metabolic syndrome or CRF, chitosan inhibits the subsequent development of oxidative stress in the systemic circulation. Thus, the pleiotropic effects of chitosan might lead to the development of new, more effective methods for the treatment of lifestyle diseases such as the metabolic syndrome or CRF [1]. We recently prepared surface-deacetylated chitin nanofibers (SDAC-NFs) by subjecting crab exoskeletons to a mechanical treatment crabs, followed by partial deacetylation of the amide groups that were located on the surface of the resulting chitin nanofibers. SDAC-NFs have attracted considerable interest in medical fields, because of their various bioactivities. In fact, SDAC-NFs are attractive, because their surface properties, as well as the macroscopic properties, of the fiber can be altered by chemically modifying the amino groups on the surface or by electrostatic interactions between the cationic amino groups on the surface and secondary components that carry an anionic charge, thus endowing the fibers with a variety of physicochemical and biological functions. In studies using CRF rats, the ingestion of SDACNFs over a 4 week period resulted in a significant decrease in the levels of uremic toxins and oxidative stress, compared with the levels of surface-deacetylated chitin (SDAC). Further, we prepared a gel starting from SDACNFs by taking advantage of electrostatic interactions with sulfobutyl

ether β-cyclodextrin (SBE-β-CD). Among the various CDs and derivatives that were tested, SBE-β-CD formed a stiff, non-fluid elastic gel, whereas other gels prepared from neutral CDs formed weak, rather fluid gels. SBE-β-CD is known to form inclusion complexes with various drug molecules and its ability to solubilize poorly water-soluble drugs is higher than that of the parent compound, β-CD, due to the presence of a hydrophobic butyl moiety. It therefore appears that a SDACNFs/SBE-β-CD elastic gel would be useful for preparing homogeneous high-content gels that can carry drugs that are poorly water soluble. Therefore, we report on the effect of a SDACNFs/SBE-β-CD elastic gel including prednisolone (PD) on an experimental model of ulcerous colitis as another oxidative stress related disease [2].

From these results, we conclude that the treatment of oxidative stress related diseases with chitosan and chitosan derivatives such as SDACNFs represents an efficient strategy. The pleiotropic effect of chitosan or SDC-NF has the potential for future applications in a wide variety of pharmaceutical fields.

Acknowledgements:

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References:

[1] M. Anraku et al, Antioxidant activities of chitosans and its derivatives in in vitro and in vivo studies (Review), Carbohydrate Polymers, 199 (2018) 141-9.

[2] M. Anraku et al, surface-deacetylated chitin nanofibers reinforced with a sulfobutyl ether β-cyclodextrin gel loaded with prednisolone as potential therapy for inflammatory bowel disease., Carbohydrate Polymers, 174 (2017) 1087-94.

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SYNTHESIS AND CHARACTERIZATION OF HEMOSTATIC LAYERS FROM CHITOSAN AND OTHER BIOMATERIALS

Uncontrolled hemorrhages are the major cause of fatality during traumatic events where standard medical treatments are usually not readily available. Therefore, a comprehensive trauma care product, which is effective in all aspects of the wound healing processes and that can be easily applied in the absence of trained medical personnel, is much sought after. However, none can be found in the current market that meets all the requirements including hemostatic, anti-inflammatory, anti-microbial, etc. We proposed to fabricate a laminated multifunctional medical textile using non-toxic biomaterials. Each layer serves certain functions along the wound healing processes, and out of which hemostatic layer is the most critical.

New hemostatic material chitosan iodoacetamides (CI) with varying degree of substitution (DS) were synthesized using carbodiimide chemistry and were characterized in terms of their DS using 1H and 13C NMR, FITR, SEM-EDS, elemental analysis, iodine content analysis, and conductometry titration. The effects of synthetic conditions were evaluated including the choice of solvent, pH of the reaction medium, reaction time and the molecular weights of the starting chitosan materials. Their hemostatic properties were evaluated using Erythrocyte Sedimentation Rate (ESR) method and were compared with regard to their iodine content and weight loading [1]. The active hemostatic mechanism of this material is believed to be outside of the regular blood coagulation cascade as was evidenced by the much faster ESR in the presence of CIs for citrated horse whole blood. Iodide is reactive towards several amino acid moieties, such as L-cysteine, lysine, methionine, and L-histidine, and could potentially form covalent crosslinks with proteins naturally found in blood. In order to have a complete effective hemostasis, a second mechanism can be incorporated as was exemplified by electrospun gelatin mat loaded with tranexamic acid (TA), a hemostatic medication. Preliminary work on a pain relief layer containing Capsaicin, will also be described.

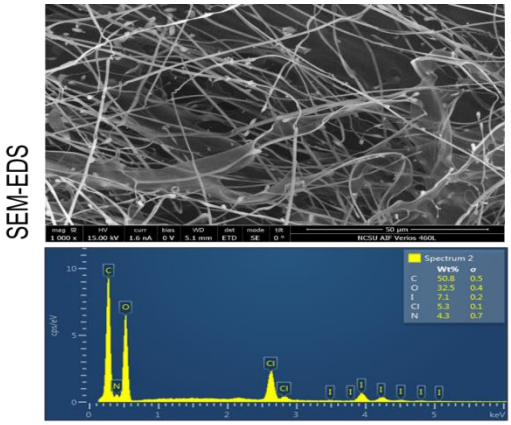

Figure 1. SEM image showing the micron to submicron fibrous structures of lyophilized CI sponge (top), and EDS elemental mapping (bottom).

Acknowledgements:

Funded by USAID via the U.S.-Egypt Science and Technology Joint Fund.

References:

[1] A.M. Abdelgawad, M.E. El-Naggar, S.M. Hudson, O.J. Rojas, Fabrication and characterization of bactericidal thiol-chitosan and chitosan iodoacetamide nanofibres, International Journal of Biological Macromolecules, 94 (2017) 96–105. doi:10.1016/j.ijbiomac.2016.07.061.

POSTER SESSION

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THE RESEARCH ON THE USE OF CHITOSAN IN LEATHER TANNING

The production of bovine leather for the shoe upper in the world is now about 16 billion square feet per year according to the FAO. This means that over 10 billion kg of raw hides are processed. More than 80% of them are chromed tanned leather. Every year the tanning industry uses about 100,000 tons of chromium as the form of a tanning agent. The tanning industry produces both the leather, chromium wastes and the leather waste containing chromium. The leather contains more than 3% (even 3.5-4.5% Cr_2O_3) of chromium based on the dry weight. Chromium is a heavy metal that is known to be carcinogenic (Cr^{+6}). Changes in the leather production technology, that eliminate or reduce the amount of chromium used, are therefore very important. Chitosan was used with a degree of deacylation more than 90% in studies. In the research these bovine hides were 2.5 mm thick. Chitosan was used as a tanning and retanning agent. The obtained leather were tested in the field of hydrothermal resistance. The tests also included the assessment of the technological operation, including the exhaustion of the bath. The results indicate limited possibilities of using chitosan in the selected technological leather processes.

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COMPARISION OF THE SEPARATION PROPERTIES OF COMPOSITE CHITOSAN MEMBRANES CONTAINING METAL OXIDES IN DIFFERENT OXIDATION STATES

Nowadays, one of the ways to improve the properties of polymer membranes is preparation of composites - organic-inorganic materials, which exhibit characteristic of both used components. However, it is hard to predict how the obtained material will behave.

The precence of metal oxides in polymer matrix essentially influence on physicochemical properties of obtained materials, which is confirmed by numerous literature reports. Knowledge of the impact of individual types of fillings on the material allows predicting the behavior of the material, direct the tests in the right way and finally get the material with the desired properties. Unfortunately, in the literature, single oxides are often studied on various materials, which makes it difficult to predict their behavior in relation to another polymer matrix. Moreover, comparison of literature data from different research groups with a unequal test conditions does not bring the satisfactory informations.

This work is a continuation of the research cycle devoted to the determination of the influence of metal oxides on chitosan membranes separation properties in pervaporative dehydration of ethyl alcohol. The aim of this work was determined the effect of metal oxides on the material's separation properties. For this purpose, pairs of metal oxides, where the metal ion was in a different oxidation state, were used. In tests were used chromium, manganese and iron oxides with which a series of membranes were made and separation capacities were tested. Based on the results of the ethanol dehydration process influence of metal oxides on separation process was determined.

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ELECTRODEPOSITION MECHANISM OF CHITOSAN TUBULAR IMPLANTS INTENDED FOR ENHANCING AN INTRINSIC PATH OF AXON REGENERATION

The Frequent injuries resulting from a wide variety of trauma (e.g. car accidents, lacerations, crash injuries) result in a large number of patients suffering from peripheral nerve dysfunctions [1, 2]. Both complications connected with autologous nerve grafts and shortage of peripheral nerve donors forced scientists to look for another solutions. High hope is placed into polymer hydrogels [3]. Designing three-dimensional tubular materials made of chitosan is still a challenging task [4, 5]. Availability of such forms is highly desired by peripheral nerve tissue engineering. Chitosan is a bio-based polymer with molecular structure similar to the one of ECM glycosmainoglycans. Chitosan-based implants have been shown to positively influence neuronal cell adhesion, cell migration, and axonal growth [6].

A new method for fabrication of chitosan-based hydrogel implants intended to serve as supporting scaffold in peripheral nervous tissue regeneration was elaborated. The method is based on electrophoretic deposition from the solution of chitosan and hydroxyapatite. Influence of initial solution composition (chitosan hydroxyapatite), process parameters (applied voltage) as well as time on resulting structure was evaluated. In order to determine the optimal composition of the initial chitosan solution as well as chitosan characteristics (e.g., molecular weight and degree of deacetylation) resulting in conduits meeting the requirements imposed on peripheral nerve implants, we employed the different methods assessing their physical (e.g., tensile strength assessment), chemical (e.g., Fourier transform infrared spectroscopy), and biological (e.g., MTT reduction assay, confocal microscopy) properties. The study showed great dependence of the examined parameters on structural and mechanical properties of the resulting implants. Because, the obtained structures show biocompatibility when contacting with a mouse hippocampal cell line (mHippoE-18), we further plan to test their application potential on an animal model (e.g. sciatic nerve injury model).

Acknowledgements:

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References:

[1] S. Jonsson, R. Wiberg, A.M. McGrath, L.N. Novikov, M. Wiberg, L.N. Novikova, P.J. Kingham, Effect of Delayed Peripheral Nerve Repair on Nerve Regeneration, Schwann Cell Function and Target Muscle Recovery, 2013, PLoS ONE, 8(2): e56484. doi:10.1371/journal.pone.0056484.

[2] W. Liu, Y. Ren, A. Bossert, X. Wang, S. Dayawansa, J. Tong, X. He, D.H. Smith, H.A. Gelbard, J.H. Huang, Allotransplanted Neurons Used to Repair Peripheral Nerve Injury Do Not Elicit Overt Immunogenicity, 2012, PLoS ONE 7(2): e31675. doi:10.1371/journal.pone.0031675.

[3] A.R. Nectow, K.G. Marra, D.L. Kaplan, Biomaterials for the development of peripheral nerve guidance conduits, Tissue Engineering Part B: Reviews, 18 (2012) 40-50.

[4] S. Itoh, I. Yamaguchi, M. Suzuki, S. Ichinose, K. Takakuda, H. Kobayashi, K. Shinomiya, J. Tanaka, Hydroxyapatite-coated tendon chitosan tubes with adsorbed laminin peptides facilitate nerve regeneration in vivo, Brain Research, 993 (2003) 111-123.

[5] K. Haastert-Talini, S. Geuna, L.B. Dahlin, C. Meyer, L. Stenberg, T. Freier, C. Heimann, C. Barwig, L.F.V. Pinto, S. Raimondo, G. Gambarotta, S.R. Samy, N. Sousa, A.J. Salgado, A. Ratzka, S. Wrobel, S. Grothe, Chitosan tubes of varying degrees of acetylation for bridging peripheral nerve defects, Biomaterials, 34 (2013) 9886-9904.

[6] Y. Yuan, P. Zhang, Y. Yang, X. Wang, X. Gu, The interaction of Schwann cells with chitosan membranes and fibers in vitro, Biomaterials, 25 (2005) 4273-4278.

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CHITOSAN COMPOSITES WITH NANOMETRIC CELLULOSE AND MODIFIED LAYERED SILICATE

Both nanometric cellulose (CNC) and chitosan are polysaccharides that due to their chemical similarity are used to form composites with some interesting properties (e.g. high sorption properties). Theoretically, these components can be combined without additional modifications but in fact there are some limitations that make the modification process necessary. The main issue limiting wider applications of chitosan/CNC composites is aggregation of CNC and low elasticity of such films [1,2]. Brittleness of these composites is usually reduced using plasticizing agent, mainly glycerol [2]. However, its excessive use can negatively affect mechanical properties of composites [3]. Layered silicates of nanometric size are already widely used as a filler for composites. They are known to improve not only mechanical, thermal but also barrier properties of composite materials. What is more, it was proved that addition of small amounts of silicates (2%) can induce plasticizing effect and increase values of elongation at break parameter [4].

It is believed that introducing into chitosan two fillers with completely different chemical compositions and different structures - CNC and modified layered silicates - will result in a synergistic effect. The resultant composite materials are thought to exhibit improved thermal and mechanical properties.

In this research modified layered silicate and nanometric cellulose were used as fillers for chitosan composites. Fourier-transform infrared spectra (FTIR) were recorded to assess the effectiveness of modification of layered silicate. Wide angle X-ray scattering (WAXS) and thermogravimetric analysis (TG) techniques were used to evaluate supermolecular structure and thermal stability of fillers. Mechanical tests were conducted to define mechanical properties of produced composites.

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References:

[1] D. Klemm, F. Kramer, S. Moritz, T. Lindström, M. Ankerfors, D. Gray, A. Dorris, Nanocelluloses: a new family of nature-based materials, Angewandte Chemie International Edition, 50 (2011) 5438–5466.

[2] H.M.C. Azeredo, L.H. Mattoso, R.J. Avena-Bustillos, G.C. Filho, M.L. Munford, D. Wood, T.H. McHugh, Nanocellulose Reinforced Chitosan Composite Films as Affected by Nanofiller Loading and Plasticizer Content, Journal of Food Science, 75 (2010) N1-N7.

[3] D. Dehnad, Z. Emam-Djomeh, H. Mirzaei, S.M. Jafari, S. Dadashi, Optimization of physical and mechanical properties for chitosan–nanocellulose biocomposites, Carbohydrate Polymers, 105 (2014) 222-228.

[4] I. Norazura, J. Margaret, P. Rajarathinam, Mechanical and thermal properties of melt processed PLA/organoclay nanocomposites, IOP Conference Series, Materials Science and Engineering, 191 (2017) 012005.

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NANOMETRIC CELLULOSE GRAFTED WITH DICARBOXYLIC ACIDS AS A FILLER FOR CHITOSAN

Nanometric cellulose (CNC) combines the most important features of both cellulosic materials and nanometric fillers. It exhibits some remarkable characteristics such as biocompatibility, biodegradability, low thermal expansion, gas impermeability or good mechanical properties [1]. Such attractive physicochemical features of CNC make it an ideal filler for biopolymers, including chitosan. Composite systems containing CNC can be used in the packaging industry (films for food products, paper coatings), in the chemical industry (catalysts, adsorbents), as well as in medicine (carriers of active substances, filaments) [2]. However, it was shown that lack of homogeneity and low adhesion at the interface between the two composite phases are the biggest problems while using CNC as a filler for polymeric matrices [3]. Strong tendency of cellulose to agglomerate and to form intramolecular hydrogen bonds can be overcome by chemical modification of CNC. Hydroxyl groups present in cellulose make it possible to modify its structure. The most commonly used modifying agents are carboxylic acids, carboxylic acids anhydrites, and acid chlorides. It was already shown that the modification of nanometric cellulose with acid chlorides is responsible for its better dispersion in polymer matrix [4].

The aim of this work was to chemically modify nanometric cellulose with dicarboxylic acids and determine influence of this modification on macroscopic properties of chitosan/CNC composites. For that reason nanometric cellulose was grafted with dicarboxylic acids with different chain lengths. Infrared spectroscopy (FTIR) spectra were recorded to confirm effectiveness of dicarboxylic acid grafting. Wide angle X-ray scattering (WAXS) technique was used to assess supermolecular structure of modified nanometric celluloses. Modified celluloses were subsequently used as fillers for chitosan matrix. The obtained chitosan/CNC composites were subjected to mechanical tests so to define the influence of the chain length of dicarboxylic acid on tensile properties of chitosan composites.

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This research was supported by the grant of Poznan University of Technology no. 03/32/DSPB/0803.

References:

[1] D. Trache, M.H. Hussin, M.K.M. Haafiz, V.K. Thakur, Recent progress in cellulose nanocrystals: sources and production. Nanoscale, 9 (2017) 1763-1786.

[2] T. Abitbol, A. Rivkin, Y. Cao, Y. Nevo, E. Abraham, T. Ben-Shalom, et al. Nanocellulose, a tiny fiber with huge applications. Current Opinion in Biotechnology, 39 (2016) 76-88.

[3] S. Borysiak, A. Grząbka-Zasadzińska, Influence of the polymorphism of cellulose on the formation of nanocrystals and their application in chitosan/nanocellulose composites, Journal of Applied Polymer Science, 133 (2016) 42864-42873

[4] A. Junior de Menezes, G. Siqueira, A. Curvelo, A. Dufresne, Extrusion and characterization of functionalized cellulose whiskers reinforced polyethylene nanocomposites, Polymer, 50 (2009) 4552-4563

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ANTIFUGAL ACTIVITY OF CHITOSAN-SALT MODIFIED PMMA DENTURE BASE MATERIAL

Purpose: Over the last years, the biopolymer chitosan found an increased attention in dentistry especially due to its antimicrobial and hemostatic properties [1]. Especially the antimicrobial properties may be of interest for treating oral mucosal infections as denture stomatitis common by denture wearers [2]. This in vitro study investigates the antifungal activity of chitosan-salt modified PMMA denture base material.

Materials and Methods: Cylindrical PMMA (n=72, Ø12.75 mm, height 6 mm, Palapress, Heraeus Group, Hanau, Germany) specimens modified with chitosan-salts (Heppe Medical Chitosan GmbH, Halle, Germany) were manufactured. Chitosan-HCl (n=32) and chitosan-glutamate (n=32) in concentrations 0.1%, 0.3%, 1% and 3% (n=8 each salt and concentration) were added to PMMA resin. As control group PMMA cylinder without chitosan-salts were produced (n=8). After manufacturing, all specimens were stored for 7 days in aqua dest. at 37 °C . After that the specimens were incubated with C. albicans for 24h at 37 °C. For the analysis the C. albicans cells were stained with calcofluor white and evaluated by fluorescence microscopy. Kruskal-Wallis and U-test (Bonferroni adjusted) were used for statistical analysis ($p<0.05$).

Results: The lowest C. albicans cell counts per 1 square centimeter in the chitosan-HCl group were found on PMMA specimens modified with 1% of chitosan-HCl (median=13600, min=1700, max=29800) and in the chitosan-glutamate group on specimens modified with 1% of chitosan-glutamate (median=3500, min=1700, max=66300). The highest number of fungal cells was counted on PMMA specimens modified with 3% of chitosan-glutamate (median=341000, min=59900, max=720100). PMMA modification with 3% chitosan-glutamate showed significant higher fungal cell counts ($p<0.02$) compared to control group without chitosan-salt. Other chitosan-salts modifications showed no significant differences compared to control group.

Conclusions: In this in vitro study the modification of PMMA denture base material with chitosan-salts showed no differences compared to pure PMMA. Modification of denture base material by adding of chitosan-HCl or chitosan-glutamate to PMMA seems to by

unfavorable for developing of PMMA denture base material with antifungal properties.

Acknowledgements:

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References:

[1] M. Rinaudo, Chitin and chitosan: Properties and applications, Progress in Polymer Science, 31 (2006) 603-632.

[2] Gendreau, L.; Loewy, Z.G. Epidemiology and etiology of denture stomatitis, Journal of Prosthodontics, 20 (2011) 251-260

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POSSIBLE APPLICATION OF CHITOSAN IN PROTECTION OF LEGUMINOUS PLANTS

Chitosan, a polysaccharide that is a product of chitin deacetylation, has high biological activity, which in plants treated with chitosan salt solutions, manifests itself in a number of immune reactions such as: induction of synthesis of chitinase and beta-glucanase enzymes [1], increased calose production and change in cell membrane permeability [2]. These are the elements of induced immunity of plants against pathogens such as fungi, bacteria and viruses. Based on the conducted studies, it has been found that in relation to some pathogens chitosan has a direct activity manifested by the inhibition of their growth in vitro [3-4].

The ongoing BIOSOYCOAT project is aimed at development of a protective coating for legume seeds against adverse weather conditions at the time of sowing. It was assumed that chitosan will be one of the components of the coating. Therefore, as part of the conducted research, the toxic effect on soil microorganisms of mono- and bilayer films based on chitosan as well as their physical and mechanical properties, morphology of the surface and susceptibility to biodegradation (by microbiological degradation) have been assessed. Biodegradation tests were carried out in the accredited own Biodegradation Laboratory in accordance with a research procedure based on ISO standard [5]. The project includes studies on the dynamics of biological deterioration of films under the influence of microorganisms naturally occurring in the soil. The degree of biodegradation of test samples was assessed by weight loss of the coating under controlled temperature and humidity conditions. Ecotoxicity tests were carried out according to the own test procedure, based on the PN-EN ISO standard [6]. In the accredited own Metrological Laboratory the strength parameters for selected polymer films constituting the coating were assessed according to PN-ISO standards [7, 8]. The rate of water vapor permeation through the films and contact angle by the sitting drop method were examined along with dynamics of the processes. The contact angle has been determined in accordance with the European Pharmacopoeia 8.0 p.2.9.45 [9].

Based on the obtained results, it can be concluded that monolayer films are dissolved within 2 minutes of contact with water.

In the case of bilayer films after 45 s, water is absorbed by the polymer and enclosed in its structure. The developed coatings in the form of single- and double-layer films are permeable to water vapour. The evaluation of this parameter is necessary from the point of view of germination capacity of seeds and the method of their storage. The ecotoxicity studies have shown that the produced films constituting the seed coating are not harmful to the soil microflora. On the other hand, organic compounds released from the films into the soil environment during their biological decomposition may be an additional source of nutrient compounds necessary for feeding soil microorganisms.

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References:

[1] F. Mauch, B. Mauch-Bani, T. Boller, Antifungal hydrolases in pea tissue. II Inhibition of fungal growth by combination of chitinase and β-1,3-glucanase. Plant Physiol, 88 (1988) 936 -942.

[2] H. Kohle, D.H. Young, H. Kauss, Physiological changes in suspension - cultured soybean cells elicited by treatment with chitosan, Plant Science Letters, 33 (1984) 221-223.

[3] A.T. Wojdyła, L.B. Orlikowski, Chitozan w zwalczaniu grzybów doglebowych i nalistnych, Progress in Plant Protection/ Postępy w ochronie roślin, 37 (1997) 300-305.

[4] A. Niekraszewicz, M. Wiśniewska-Wrona, E. Kopania, L. Orlikowski, H. Pospieszny, K. Krawczyk, Biopolymer compositions for ecological protection and growth stimulation of plants, Progress on Chemistry and application of Chitin and Its Derivatives, Polish Chitin Society, vol. XVII, ISSN 1896-5644 (2012) 145-157.

[5] PN ISO 11266: Jakość gleby. Zasady prowadzenia badań laboratoryjnych nad biodegradacją związków organicznych w glebie w warunkach tlenowych.

[6] PN-EN ISO 4833-1:2013-12 „Mikrobiologia łańcucha żywnościowego. Horyzontalna metoda oznaczania liczby drobnoustrojów. Część I " Oznaczanie liczby metodą posiewu wgłębnego w temp. 30 C. Add new reference.

[7] PN-ISO 4593:1999 "Tworzywa sztuczne - Folie i płyty - Oznaczanie grubości metodą skaningu mechanicznego".

[8] PN-EN ISO 527-3:1998 "Tworzywa sztuczne-Oznaczanie właściwości mechanicznych przy statycznym rozciąganiu - Warunki badań folii i płyt".

[9] European Pharmacopoeia 8.0: 2014 vol. I, p.2. 9.45 "Wettability of porous solids including powders"

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MICROSTRUCTURE OF CHITOSAN FILMS AND THE CONTACT ANGLE

At present, there is a strong emphasis on the use of natural polymers for obtaining environmentally friendly products. The production of chitosan or celluse films as packaging films is very desirable. These films must have appropriate mechanical and barrier properties. The surface microstructure is also very important. An important physical property characterizing surfaces of polymeric materials is its wettability [1], which effects, for example, its susceptibility to printing. The contact angle is a measure of the wettability of the film, it is the angle between the tangent and the drop at the point of contact with the surface of the film.

The aim of the research was to show the influence of the surface microstructure of chitosan films on the vallue of the contact angle. Films without plasticizing additives made of chitosan and chitosan acetate were selected for the tests. The results of test for cellulose films are presented for comparison. In the contact angle test, a sappy drop method was used based on the European Pharmacopoeia [2]. In order to determine the contact angle Θ the device STEFI AB Lorentzen-Wettre with Delta Optical Smart 5MP PRO microscope camera and the DLT- Cam Viewer computer program was used. Measuring liquid: glycerin, volume of drop: 5 µl, test conditions: temperature 23±1°C; humidity: 50±2%.

Due to the method of films production, the contact angle measurements were made on both the top of bottom layers of the film. In the case of chitosan film or chitosan acetate, the method of preparation slightly affected the difference in wettability between the top and bottom of the film. This was confirmed by the SEM pictures of the surface of the obtained films. On the other hand, in the case of cellulose films wettability varies greatly depending on the side of the film (top, bottom). Both chitosan and cellulose films are characterized by a homogeneous structure. There are differences in the microstructure between the top and the bottom of the sample in the cellulose film. This affects the value of the contact angle and thus the wettability of the surface.

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using a modern Delta Optical SZ-430T stereoscopic microscope of flat textiles products and films"

References:

[1] R. J. Goud, R.R. Stromberg, Eds.: Surface and Colloid Science, vol. 11, Plenum Press, New York 1979

[2] European Pharmacopoeia 8.0: 2014 vol. I, p.2. 9.45 "Wettability of porous solids including powders"

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BIODEGRADABLE NONWOVEN WITH AN ACTIVE LAYER FOR COSMETIC PURPOSES

Polymeric products enriched with natural additives such as chitosan, hyaluronic acid, plant hydrolates or collagen may find wide applications in cosmetic industry, due to the growing interest of consumers in natural cosmetics. The enviromental aspects is also beginning to play an important role in cosmetics. Therefore, as part of the research work, a biodegradable nonwoven fabric was designed with natural additives constituting the active layer for cosmetics purpose.

A synthesis of a biodegradable aliphatic - aromatic copolyester of poly(succinate-co-glutarate-co-adipate-co-terephthalate1,4-butylene) was carried out in order to obtain a polymer with specific physico-chemical properties, capable of processing into spun-bond nonwovens. Next, the active layer was deposited on the base nonwoven using padding method. For the obtained product microbiological, mechanical [1-2] as well as biodegradability properties in a compost environment [3] have been assessed. Antimicrobial activity tests were carried out on S. aureus and E. coli [4] and the microbiological purity [5] of the produced nonwovens has been assessed. On the basis of microbiological tests, the most favorable composition of the active layer was selected. The test of mechanical properties were carried out, with particular consideration of functional properties such as absorption capacities [6] and flexural rigidity [7]. Due to the good mechanical properties, the aliphatic-aromatic copolyester nonwoven can act as a cosmetics base. It is also biodegradable what is now an important feature due to environment concern. The selected active layer containing a mixture of sodium hyaluronates, bitter orange flowers hydrolate (Neroli) and collagen has good microbiological and mechanical properties. Collagen, hyaluronic acids and bitter orange floral water have nourishing properties and are widely used in cosmetics and if applied on nonwoven fabric can perfectly fulfill a cosmetic function, e.g. in the form of an active mask. The variant produced with the active layer (collagen, hydrolate, HA) was compared with nonwoven fabric with applied layer of chitosan lactate in terms fo the above-mentioned properties.

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References:

[1] PN-EN ISO 9073-2:2002 "Textiles-Test metods for nonwovens- Part 2: Determination of thickness".
[2] PN-EN 29073-3:1994 "Textiles-Test metods for nonwovens- Part 3: Determination of tensiles strength and elongation".
[3] Research procedure no. 2 developed at IBWCh "Determination of the degree of degradation of plastics and textiles in simulated composting conditions on a laboratory scale. Determination of mass loss " based on PN-EN ISO 20200: 2007, PN-EN ISO 14045: 2005, PN-EN ISO 14806: 2010.
[4] PN-EN ISO 20743:2013-10 Textiles - Determination of antibacterial activity of textile products.
[5] Pharmacopeia Polish edition X (2014), Volume I, point 2.6.12. Microbiological testing of non-sterile products (microbiological quantitative studies).
[6] PN-EN ISO 9073-6:2005 "Textiles-Test metods for nonwovens- Part 6: Absorption".
[7] PN-EN ISO 9073-7:2011 "Textiles-Test metods for nonwovens- Part Determination of bending lenght".

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THERMAL CONVERSION OF CHITIN AND CHITOSAN TO THE NITROGEN-RICH CARBON FOAMS

Marine-derived feedstocks offers important benefits, such as abundance, morphological and structural variety, and the presence of multiple elements, including nitrogen and carbon. Therefore, these renewal resources may be useful for obtaining N- and C-containing materials that can be manufactured by various methods, such as pyrolysis processes supported by means of chemical and physical activators. Generally, synthesis concept relies on an efficient transfer of nitrogen and carbon from marine feedstock to the final product. The manufacturing procedure influences some crucial properties of nitrogen-doped carbon materials, such as pore structure and the chemical composition of the surface. These research is given on the relationship between carbon materials manufacturing from chitin and chitosan and the elemental content of nitrogen, together with a description of the chemical bonding of nitrogen atoms at the surface. The contribution describes the first ever study on the synthesis of nitrogen-rich carbon foams (N-CFOs) from chitin and chitosan by their thermal decomposition in the presence of a solid and removable nano-particles (template). N-CFOs were synthesized by carbonization of chitin and chitosan as the carbon phase precursors with two alternative hard templates of $CaCO_3$ (added as nano-sized powder) or Na_2CO_3 (in situ synthetized as nano-crystallites). The carbonization process was carried out in the temperature range from 600°C to 900°C under the flow of nitrogen. The results show that the textural and chemical properties of the N-CFOs could be controlled in a wide range by changing the template admission mode, the type of precursor and/or activation temperature. The obtained N-CFOs were either meso- or microporous matrixes (open porosity) with an exceptionally high nitrogen content. The high N-content remained even for samples carbonized at the highest temperature of 900°C. The walls of the obtained N-C-FOs were molecularly thin.

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EVALUATION OF CHITOSAN ANTIBACTERIAL EFFECTS ON GROWTH OF FOOD PATHOGENIC BACTERIA

The goal of this research was to evaluate chitosans antibacterial effect on the growth of pathogenic bacteria. 5 different preparations of chitosan were used for the research: PFANNENSCHMIDT-HAMBURG® and Nexira health® (crustacean taxonomic group) as well as preparations extracted from *Cervimunida johni* organism (crustacean taxonomic group), *Hylobius abietis* (insect taxonomic group) organism, *Boletus bovinus* (fungi taxonomic group) organism. Methods used in this research: chitosan extraction, chitosan sample preparation, preparation of bacterial solutions, contamination of microorganism nutrient medium, agar diffusion method. Chitosans physicochemical analysis was performed, each chitosan preparations were evaluated in different concentrations based on their ability to supress the growth of pathogenic bacteria. Field of application: chitosan preparations can be adapted to be used to ensure microbiological safety of food by adding them into food products or using them in edible wafers and plain packaging because of their antibacterial effects on foods pathogenic bacteria. Antibacterial effect research methodology was optimized for examining pathogenic food bacteria by applying 1,5·108 CFU/ml and 2 g/l chitosan concentration. It was found that chitosan, that was extracted from *Hylobius abietis* organisms, expressed the most potent inhibitory effect on all tested bacteria (17,18±1,54 mm), the least potent was the commercial chitosan Nexira health® (13,00±4,36 mm). The antibacterial effect of chitosan was pronounced the most on *L. monocytogenes* bacteria (17,50±0,57-21,5±0,57 mm) and S. typhimurium bacteria (15,50±0,19-21,00±0,24) and the least sensitive to chitosans effects was the S. aureus bacteria (0,00-13,5±0,60 mm).]

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PHYSICOCHEMICAL PROPERTIES AND IN VITRO CYTOTOXICITY STUDIES OF CHITOSAN AS A POTENTIAL AGENT FOR CANCER TREATMENT

The mechanisms responsible for the cytotoxic effects of chitosan on cancer cells are still unclear. The goal of this study was to evaluate the physicochemical and cytotoxic properties of chitosan in vitro on mouse hepatoma MH-22A (tumor cells) and Chinese hamster ovary (CHO, non-cancerous) cells cultivated on films with different chitosan concentrations and cultivated on Petri dishes after preincubation with chitosan solutions.

The chitins and chitosans of different origin (crustacean, insect, mushroom) were characterized by attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR), elemental analyses (EA), nuclear magnetic resonance spectroscopy (NMR), thermogravimetric analyses (TGA) and X-ray diffraction (XRD). To measure plating efficiency (cloning efficiency), cells were plated at low densities on chitosan films or after preincubation with chitosan solutions cells were plated on Petri dishes and allowed to grow. To see the colonies clearly, they were stained with crystal violet. To reveal the rupture of cell membrane (morphologic expression of necrosis) cells were stained with Trypan Blue solution after 40 and 100 min of chitosan treatment.

The dry weight chitin contents of the mushroom species were determined as 8.5% for *L. laccata* and 6.2% for B. bovinus. Chitosan yields of the chitins isolated from *B. bovinus* and *L. laccata* were 70.9% and 64.3%, respectively. ATR-FTIR spectra analysis demonstrated the characteristic bands of the chitin and chitosan molecules. While, the maximum degradation temperatures of *B. bovinus* and *L. laccata* chitins were found to be 320°C and 363°C by TGA, the maximum degradation temperatures of *B. bovinus* and *L. laccata* chitosans were recorded as 317°C and 309°C, respectively. The crystallinity index values of *B. bovinus* and *L. laccata* chitins were calculated as 85% and 78%, respectively according to the XRD results. Degree of acetylation (DA) determined by EA and NMR was found to be 90±5 and 92±4 for chitin from *L. laccata*, 93±5 and 94±4 for chitin from *B. bovinus*, respectively. Values

of cell viability indicating the rupture of cell membrane (morphologic expression of necrosis) after 100 min of incubation were 95 ± 5 % for cells without of chitosan, 78 ± 4 % for cancerous cells MH-22A with chitosan solution (1g/L), 70 ± 5 % for normal CHO cells with chitosan solution (1g/L). Also, morphologic features of early process of apoptosis were observed (cell shrinkage, pyknosis and vacuolization). The number of cell colonies after 1 h incubation with chitosan solution (1g/L) and subsequent cultivation was lower ($p < 0.05$) than for non-treated cells (31 ± 2 % for cancerous cells MH-22A, 21 ± 2 % for normal CHO cells).

The results of this study revealed that mushroom species *B. bovinus* and *L. laccata* may be used as a potential chitin source with appropriate physicochemical properties (chemical structure, thermal stability, crystallinity index, degree of acetylation) for possible biomedical application. This study clearly revealed that chitosan solutions can act cytotoxically ($p < 0.05$) on both tumor and normal cells inducing necrosis and apoptosis.

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SPONGES (*PORIFERA*) AS SOURCE OF MORPHOLOGICALLY DEFINED CHITIN

Marine demosponges (*Porifera: Demospongiae*) are recognized as first metazoans, which have developed over millions of years of evolution effective survival strategies based on unique metabolic pathways to produce both biologically active secondary metabolites and biopolymer-based stiff skeletons with unique 3D architecture. The sophisticated architecture of such chitinous skeletons opens the way for their recent applications as adsorbents, as well as scaffolds for biomedicine and biomimetics [1]. This study has the ambitious goal to highlight the sponges as the alternative sources of naturally prestructured chitinous scaffolds. Special attention is paid in those demosponge species, which can be cultivated at large scales using marine farming conditions and used as a renewable source of unique chitin scaffolds. Special attention has been paid to the non-Verongiid demosponges *Mycale euplectellioides (Heteroscleromorpha: Poecilosclerida: Mycalidae); Acarnus wolffgangi* and *Echinoclathria gibbosacollected*. We present here a detailed study of the isolation of chitin from the skeleton of these sponges, as well as its identification using diverse bioanalytical tools. Calcofluor white staining, Fourier-transform Infrared Spcetcroscopy (FTIR), electrospray ionization mass spectrometry (ESI-MS), scanning electron microscopy (SEM), and fluorescence microscopy, as well as a chitinase digestion assay were applied in order to confirm with strong evidence the finding of a-chitin in the isolated skeletons [2,3].

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References:

[1] M. Wysokowski, I. Petrenko, A.L. Stelling, D. Stawski, T. Jesionowski, H. Ehrlich, Poriferan chitin as a versatile template for extreme biomimetics, Polymers, 7 (2015) 235-265.

[2] H. Ehrlich, L.A. Shaala, D.T.A. Youssef, S. Żółtowska-Aksamitowska, M. Tsurkan, R. Galli, H. Meissner, M. Wysokowski, I. Petrenko, K.R. Taabachnick, V.N. Ivanenko, N. Bechmmann, Y. Joseph, T. Jesionowski, Poriferan chitin as a versatile template for extreme biomimetics. PloS One 13 (2018) e0195803.

[3] S. Żółtowska-Aksamitowska, L.A. Shaala, D.T.A. Youssef, S.S. Elhady, M.V. Tsurkan, I. Petrenko, M. Wysokowski, K.T. Tabachnick, H. Meissner, V.N. Ivanenko, N. Bechmann, Y. Joseph, T. Jesionowski, H. Ehrlich, First Report on Chitin in a Non-Verongiid Marine Demosponge: The Mycale euplectellioides Case. Marine Drugs 16 (2018) 68.

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CHITOSAN ANTI-AMOEBA ACTIVITY TO AXENIC STAGE OF *ACANTHAMOEBA SP*.

The genus *Acathamoeba*, comprises pathogenic free-living protozoa known to be present worldwide in water, soil and even the air. The amoeba cause infection of the central nervous system, lungs, sinuses and skin in humans, leading to death, particularly in immunocompromised patients [1]. *Acanthamoeba* is best known as a causal agent of keratitis, an inflammatory disorder of the eye found in contact lens wearers [2]. Free-living amoeba actively feed on bacteria, thus contributing to the structure of the microbial community in the natural environment [3].

Due to its biocompatibility, biodegradability and bioactivity, chitosan has been the object of interest for applications in agriculture, biomedicine, biotechnology and the food industry [4]. To activate its anti-microbial activity, the polymer must be dissolved in an acid solution, where the molecular weight (MW) of the polymer and its concentration influence its properties. The potential pathogenic activity of the *Acanthoamoeba* and their beneficial influence on soil structure make them valuable candidates for evaluating the potential of chitosan as an agent for treating amoebal infection in patients and its harmful effect on soil life.

The aim of study was to evaluate the anti-amoeba activity of different molecular weights and concentrations of chitosan solution against an *in vitro* culture of axenic *Acanthamoeba* sp.

The cytotoxic properties of high and low molecular weight chitosan were investigated employing *in vitro* systems. Low Molecular Weight; LMW; Mw= 50-190 kDa, DD >75%, Poly(D-glucosamine) Sigma,. 448869) and High Molecular Weight; HMW; Mw= 190-375 kDa, DD >75%, Sigma, 419419) chitosan were used. The amoebae were bred on a liquid medium. Chitosan was added at concentrations of 1, 10 and 100 µg/ml into a culture containing 2×10^5 amoebae. The number of amoebae was evaluated in a Burcker cell-counting chamber after seven days of culture. The control group was a culture of amoebae not treated with chitosan or Adypic acid, a solvent of chitosan. The vital condition of the amoebae was evaluated using Rhodamine 123 accumulation.

Acanthamoeba growth *in vitro* was reduced in a manner dependent on the molecular weight and concentration of the applied chitosan solution. The LMW chitosan inhibited the growth of amoeba culture more effectively than the HMW chitosan. The amoeba changed shape after exposure to chitosan; flat, moving and fully vacuolated cells dominated in control culture, but spheroid cells and cysts in the samples with HMW chitosan. Spheroid and adherent trophozoites, and a few cysts were also present in the samples treated with LMW. The amoebas exposed to LMW accumulated rhodamine in their vacuoles to a greater degree than those exposed to HMW. Rh123 was present in the cytoplasm in the HMW-treated cells, but was found in the cytoplasm in controls.

Our findings confirm that chitosan possesses anti-protozoan activity [5] also against *Acanthamoeba* trophozoites; however, the mechanism of these effects still remains to be identified. *Acanthamoeba* number reduced in the culture, while the number of cysts increased only in control samples treated with adipic acid. Proliferation was inhibited and the amoebae died when exposed to chitosan. Following exposure to the environment, polymeric materials such as chitosans can undergo changes in chemical structure, and demonstrate variations in their biochemical or physical properties which allow them to induce cytotoxicity by generation reactive oxygen species. In free-living amoebas, one crucial survival strategy may be based around the modulation of oxidative metabolisms, while another may involve the disruption of the cell by the free amino groups present in chitosan [6]. Our findings fully confirm those of previous studies that both HMW and LMW chitosan deacetylated to a greater degree than 75% express anti-protozoan activity when dissolved in adipic acid at various concentrations.

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References:

[1] F.L. Schuster, G.S. Visvesvara, Free-living amoebae as opportunistic and non-opportunistic pathogens of humans and animals. International Journal of Parasitology, 34 (2004) 1001–1027.

[2] J.R. Verani, L.A. Lorick, J.S. Yoder, M.J. Beach, C.R. Braden, J.M. Roberts, C.S. Conover, S. Chen, K.A. McConnell, D.C. Chang, B.J. Park, D.B. Jones, G.S. Visvesvara, S.L. Roy, National outbreak of Acanthamoeba keratitis associated with use of a contact lens solution, United States. Emerging Infective Diseases, 15 (2009) 1236–1242.

[3] R. Rønn, A.E. McCaig, B.S. Griffiths, Prosser JI: Impact of protozoan grazing on bacterial community structure in soil microcosms. Applied of Environmental Microbiology, 68 (2002) 6094-105.

[4] T. Wu, S. Zivanovic, F.A. Draughon, W.S. Conway, C.E. Sams, Physicochemical properties and bioactivity of fungal chitin and chitosan. Journal of Agriculture Food Chemistry, 53 (2005) 3888-3894.
[5] M. Tavassoli, A. Imani, H. Tajik, M. Moradi, S. Pourseyed, Novel in Vitro Efficiency of Chitosan Biomolecule against Trichomonas gallinae . Iranian Journal of Parasitology, 7 (2012) 92-96.
[6] Y. Andres, L. Giraud, C. Gerente, P. Le Cloirec, Antibacterial effects of chitosan powder: mechanisms of action. Environmental Technology, 28 (2007)1357-1363.

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THE INFLUENCE OF CHITOSAN CONTENT IN HYDROGEL BEADS ON THE SORPTION EFFECTIVENESS OF REACTIVE BLACK 5 DYE

In the work the influence of chitosan content in hydrogel beads on the sorption effectiveness of Reactive Black 5 (RB5) dye from aqueous solutions has been presented. The amount of dry mass of chitosan in tested hydrogel chitosan sorbent was 2 up to 10 %. The range of study on sorbents included: determining the influence of pH (pH 4-11) on the RB5 sorption effectiveness on chitosan hydrogels, research on the RB5 sorption kinetics and research on sorption capacity of the tested chitosan sorbents in relation to RB5. The optimal pH of the RB5 sorption as well as pHPZC of the tested sorbents were both determined. The experimental data form the research on sorption kinetics were fit to pseudo-first, pseudo-second order model as well as to the model of intramolecular diffusion. The maximum sorption capacity of tested sorbents dependant from the content of chitosan dry mass in hydrogel beads has been determined. The obtained data was fit to Langmuir 1, Langmuir 2 and Freundlich model.

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THE INFLUENCE OF SALINITY ON THE REACTIVE BLACK 5 SORPTION EFFICIENCY ON HYDROGEL CHITOSAN SORBENTS

In the work the influence of salinity (NaCl/ Na2SO4) on sorption effectiveness of the Reactive Black 5 (RB5) dye from aqueous solutions on chitosan in the form of hydrogen beads has been researched.

In the first part of the study the influence of pH (pH 4-11) on the effectiveness of the RB5 sorption form the solutions containing sodium chloride and sodium sulphate (0,10 – 1,00 M) has been determined along with determining the optimal pH of the dye sorption. In the second part the research on the kinetics of the RB5 sorption in the solutions of NaCl and Na2SO4 (0,02-1,00 M) has been carried out. The RB5 sorption equilibrium time in relation to salinity has been determined. Experimental data was fit to pseudo-first , pseudo-second order model as well as to the model of intramolecular diffusion. In the third part of the study the maximal sorption capacity of chitosan in relation to RB5 depending on the concentration of sodium chloride and sodium sulphate (0,10-1,00 M) was determined. The obtained data was described with sorption isotherms: Langmuir 1, Langmuir 2 and Freundlich.

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CHITOSAN IN ELECTOCHEMICAL DOUBLE-LAYER CAPACITORS

Electrochemical Double-Layer Capacitors or Supercapacitors (EDLCs) are the energy storage devices that can store energy at the electrode/electrolyte interface. Carbon materials with the very high specific surface are used as an electrode materials.[1-2]. Different kinds of electrolytes: aqueous, organic and ionic liquids are proposed [3].

Over the resent years, the interest in biopolymers and materials obtained from renewable sources is enormous[4-6]. This is due to the desire to replace the commonly used materials, in most environmentally unfriendly, by natural origin materials, that can reduce the environmental impact.

In this project our attention has focused on chitosan as material obtained from renewable sources which it was used as a component for EDLC.

In the first case, chitosan was used as a binder for electroactive material. In the second case, the chitosan membrane was obtained and it was tested as the separator and the polymer electrolyte in capacitor cells.

Chitosan (CS) (Sigma Aldrich, ca. 80% deacetylation degree) was dissolved in 2% acetic acid, frozen and dried by the lyophilisation and immersed in the ethanol-water solution. Membranes of different thickness were prepared and tested as a separator in EDLC cell. Several concentrations of the CS were prepared to obtain different structures the freeze-casting method.

In the case of testing CS as a binder commercial carbon material (Active Carbon, Super 30) was dispersed in chitosan solution with different concentration rates, froze and dried. Finally, sponge-like structures were obtained and used as electroactive materials for EDLC preparation. As electrolytes, 2M Lithium Acetate and 1M Lithium Sulfate solutions were used.

The results suggest the chitosan can be used as a replacement for commercial applied materials for EDLC technology.

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References:

[1] A.Gonzalez, E.Goikolea, J.A. Barrena, and R. Mysyk, Renew Sustain. Energy Rev., 2016, 58, 1189-1206.

[2] Electrochemical Supercapacitors, Scientific Fundamentals and Techological Application, B.E. Conway, Kluwer Academic, New York 1999.

[3] M. Galinski, A. Lewandowski and I. Stepniak, Electrochim. Acta, 2006, 51, 5567–5580.

[4] M. N.V. Ravi Kumar, React. Funct. Polym., 2000, 46, 1–27.

[5] X. Shen, J. L. Shamshina, P. Berton, G. Gurau and R. D. Rogers, Green Chem., 2015, 18, 53–75

[6] C. K. S. Pillai, W. Paul and C. P. Sharma, Prog. Polym. Sci., 2009, 34, 641–678.

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THE POSSIBILITY OF CHITOSAN ASCORBATE APPLICATION IN DENTISTRY - THE DIRECTIONS OF INVESTIGATIONS

The possibility of chitosan ascorbate (ChA) application in dentistry were investigated in Department of Conservative Dentistry of Medical University of Gdansk from1988. The investigations were led in the co-operation from Sea Fisheries Institute (Gdynia, Poland) and Department of Oral Microbiology of Medical University of Gdansk (prof. A. Kędzia). Chitosan ascorbate was prepared in the Sea Fisheries Institute (dr. A. Wojtasz-Pająk). Chitosan was obtained from krill chitin. The degree of deacetylation was between 60 - 75%. The ratio of ascorbic acid to chitosan was 1:1 (percentage by weight). Investigations were multidirectional and had the interdisciplinary character.

First investigations concerned the possibility of the application of the ChA as a dressing that could be used in the oral cavity [1-2]. The purpose of these studies was to examine haemostatic and hygroscopic properties of ChA. The film – forming properties of ChA as well as the degree of adhesion to soft tissues were studied. The ChA application in conservative dentistry and prosthetics during various dental operations within the region of marginal gingiva were studied too. The results of these research suggested that ChA can be useful as a sort of dressing having of unique haemostatic, drying and protective proprieties [1-2].

The second part of research related to the biodegradation of ChA [3-4]. The aim of these in vitro studies was to examine degradation of ChA and N-acetyl-D-glucosamine formation during incubation in various experimental environments. The obtained data suggested that under in vitro conditions, human saliva stimulated the ChA biodegradation and the N-acetyl-D-glucosamine release. The progress of ChA biodegradation in saliva was slow [3-4].

The third part of research has focused on the antimicrobial activity of ChA on oral microbiota [5-8]. The aim of these studies was to determine the activity of ChA to obligate anaerobes (i. a. strains of *Porphyromonas spp, Propionibacterium spp, Prevotella spp, Bacteroides spp,* Eubacterium *spp, Peptostreptococcus spp, Fusobacterium spp*), facultative anaerobes (strains of *Streptococcus spp*), capnophilic bacteria and microaerophiles (i. a. strains of *Campylobacter spp, Eikenella corrodens, Helicobacter pylori, Aggregatibacter actinomycetemcomitans, Prevotella intermedia*) and the strains of yeast-like fungi of *Candida*, *Geotrichum* and *Rhodotorula* genera. The minimal

inhibitory concentration (MIC), defined as the minimal ChA concentration that inhibited growth of studied microorganisms, was determined by means of the plate dilution technique in agar. ChA was dissolved in sterile distilled water at the following concentrations: 4.0, 2.0, 1.5, 1.0, 0.75, 0.5, 0.25, 0.12, 0.06, 0.03 and 0.01 mg/ml. In each experimental series, the growth of strains was checked on the culture medium without ChA. The microorganisms were isolated from infections within the oral cavity (i. a. dental pulp inflammation, gingivitis, periodontitis, oral mucous membrane inflammation and ulceration). The obtained data indicated that ChA exhibited differential antibacterial and antifungal activity [5-8].

References:

[1] Haemostatic properties of chitosan and its application in stomatology, B. Kochańska, E. Witek, J. Śramkiewicz, in „Chitin World", Monograph, Z.S. Karnicki, A. Wojtasz-Pajak, M.M. Brzeski, P. Bykowski (eds), Wirtschaftsverlag NW, Bremerhaven, 1994, 520-529.

[2] JEvaluation of chitosan ascorbate application as a multifunctional dressing during dental operation within the region of dental cervix, B. Kochańska, J. Śramkiewicz, in "Chitosan *per os*: from dietary supplement to drug carrier", R.A.A. Muzzarelli (ed), Grottammare, 2000, 257-264.

[3] Biodegradation of chitosan in saliva: In vitro studies, B. Kochańska, in "Progress on Chemistry and application of chitin and its derivatives. Monograph", H. Struszczyk (ed), PTCHit, Łódź, 1997, III, 103-108.

[4] Release of N-acetyl-D-glucosamine from chitosan in saliva, B. Kochańska, in "Advances in Chitin Science", M. G. Peter, A. Domard , R. A. A. Muzzarelli (eds), University of Potsdam, 2000, 4, 291-294.

[5] Effect of chitosan ascorbate on bacteria isolated from the oral cavity, B. Kochańska, A. Kędzia, in "Chitosan in pharmacy and chemistry", R. A. A. Muzzarelli, C. Muzzarelli (eds), Ancona, 2002, 171-176.

[6] Susceptibility of oral Streptococci to chitosan ascorbate, B. Kochańska, A. Kędzia, A. Wojtasz-Pająk, in "Progress on chemistry and application of chitin and its derivatives: Monograph", H. Struszczyk (ed), Łódź, 2002, VIII, 107-111.

[7] B. Kochańska, A. Kędzia, A. Gębska, Sensitivity to chitosan ascorbate microaerophilic bacteria isolated from infections of oral cavity, Progress on Chemistry and Application of Chitin and its Derivatives, 21 (2016) 109-113.

[8] Susceptibility of *Candida, Geotrichum* and *Rhodotorula* to chitosan ascorbate, B. Kochańska, A. Kędzia, in "Chitosan in pharmacy and chemistry", R. A. A. Muzzarelli, C. Muzzarelli (eds), Ancona, 2002, 165-170.

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THE EFFECT OF SELECTED HYDROPHILIZING SUBSTANCES ON CHITOSAN BASED DERMATOLOGICAL HYDROGELS PROPERTIES

Hydrogels have a wide range of applications, especially as modern dressings. They are used in the treatment of ulcers, bedsores, burn wounds, in rheumatic diseases and diseases characterized by excessive pigmentation of the skin. They provide optimal temperature, pH and humidity, so the wound heals faster. Thanks to their properties such as bio-adhesiveness, bacteriostaticity and biocompatibility they ensure long-term skin contact. Gels spread well on the surface of the skin, do not adhereto the wound , reduce the formation of scars , their change is painless and comfortable. Dressings of this type are produced on the basis of methylcellulose, Carbopol or chitosanamount of hydrophilizing agents. All these properties are useful in creating a drug form that allows optimal contact with the skin.

References:

[1] W. Mozalewska, R. Czechowska-Biskup, A.K. Olejnik, R.A. Wach, P. Ulański, J.M. Rosiaka, Chitosan-containing hydrogel wound dressings prepared by radiation technique. Radiation Physics and Chemistry, 134 (2017) 1-7.

[2] K. Bialik-Wąs, B. Tyliszczak, E. Wilk, K. Pielichowski, Preparation of innovative hydrogel wound dressings based on poly(acrylic acid). Chemik, 67 (2013) 99-104.

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THE PROPERTIES OF INTRAVAGINAL GLOBULES CONTAINING A LACTIC ACID-CHITOSAN COMPLEX

Constant contact between the drug form and the vaginal mucosa during the patient's daily activities is the basic condition for successful therapy. This condition can be met by forms of the drug with high adhesion to the vaginal mucosa. Available literature does not inform about a significant progress in the effectiveness of treatment of bacterialvaginosis [1-3]. Continuing the implementation of research on the problem of treatment of vaginal pH disorders, intravaginal globules were examined as vaginal drug form which carries the lactic acid complexes with chitosan. Application of the globules passing applications in natural conditions in the gel is designed to obtain a physiological pH of the vaginal environment.

Aim of this work was to study the pharmaceutical properties of globules for gynecological, passing under natural conditions in the gel covering the vaginal mucosa. In an effort to solve the problem, the effect of hydroxypropylmethylcellulose and hydrophilizing substances such as glycerol, 1,2-propylene glycol and polyoxyethylene glycol 200 on the properties of beads was investigated. Formulations were prepared with varying pH and rheological properties.

As a result of the research was obtained preparations with different pH values including the physiological range. Globules show the adhesion of the gel covering the surface of the apparatus simulates the conditions in the vagina. The gels obtained from the globules were characterized by the thixotropy and specific dynamic viscosity.

References:

[1] G.G.G. Donders, P.G. Larson, J.J. Platz-Christensen, A. Hallen, van der W. Meijden, P. Wolner-Hanssen, Variability in diagnosis of clue cells, lactobacillary grading and white blood cells in vaginal wet smears with conventional bright light and phase contrast microscopy, European Journal of Obstetrics & Gynegology and Reproduction Biology, 145 (2009) 109-112.

[2] J.M. Denney, J.F. Culhane, Bacterial vaginosis: A problematic infection from both a perinatal and neonatal perspective, Seminars in Fetal & Neonatal Medicine, 14 (2009) 200-203.

[3] M. Brandt, C. Abels, T. May, K. Lohmann, I. Schmidts-Winkler, U.B. Hoyme, Intravaginally applied metronidazole is as effective as orally applied in the treatment of bacterial vaginosis, but exhibits significantly less side effects, European Journal of Obstetrics & Gynegology and Reproduction Biology, 141 (2008) 158-162.

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PHARMACEUTICAL RESEARCH OF HYDROPHILIC GLOBULES CONTAINING A LACTIC ACID-CHITOSAN COMPLEX

Insufficient duration of the drug's contact with vaginal mucosa does not provide adequate pH conditioning the phisiological biocenosis of the organ. Traditional therapeutic schemes recommended by world treatment centres and health organizations do not bring satisfactory results [1-3]. The conducted research and obtained positive results encourage further research possibilities in the field of optimization of pharmaceutical properties of the examined gynecological preparations.

Aim of this study was to investigate the influence of selected polymers on the physico-chemical properties of globules for gynecological purposes. Assuming the above assumptions tested impact of polyvinylpyrrolidone K-15, polyvinylpyrrolidone K-30 and polyvinylpyrrolidone K-90 in the properties of the globules. The study was prepared formulations with different pH and rheological properties. The device simulating the conditions in the vagina, studied the adhesion and movement of the gel on the mucosa of the organ. Globules passed in gels were examined for their properties. In vitro studies have demonstrated that the gels obtained from the globules are maintained at the application site. Globules afer application of the apparatus simulates the natural conditions in the gel passes and cover the surface. As a result of studies, the dynamic viscosity of the gels obtained from globules. The test shows thixotropical properties of gels. A wide range of pH of the gels allows the selection of the optimum formulation.

References:

[1] G.G.G. Donders, P.G. Larson, J.J. Platz-Christensen, A. Hallen, van der W. Meijden, P. Wolner-Hanssen, Variability in diagnosis of clue cells, lactobacillary grading and white blood cells in vaginal wet smears with conventional bright light and phase contrast microscopy, European Journal of Obstetrics & Gynegology and Reproduction Biology, 145 (2009) 109-112.

[2] J.R. Schwebke, New concepts in the Etiology of Bacterial Vaginosis, Current Infectious Disease Reports, 11 (2009) 143-147.

[3] M. Brandt, C. Abels, T. May, K. Lohmann, I. Schmidts-Winkler, U.B. Hoyme, Intravaginally applied metronidazole is as effective as orally applied in the treatment of bacterial vaginosis, but exhibits significantly less side effects, European Journal of Obstetrics & Gynegology and Reproduction Biology, 141 (2008) 158-162.

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HYDROCHLORIC ACID AS A SOLVENT FOR CHITOSAN EXAMINATION OF PHYSICOCHEMICAL BEHAVIOR

Successful accomplishments in basic research on chitosan and practical exploitation of this biopolymer require knowledge of the average molecular weight and the degree of deacetylation. These two most important intrinsic parameters of chitosan determine not only the simplest properties, such as the solubility or viscosity of its solutions, but also in many cases the biological activity of this polysaccharide.

Measurements of average molecular weight by viscometry, gel permeation chromatography and light scattering require molecular dissolution of chitosan in a suitable solvent. Numerous systems used for dissolving chitosan consisting of water, acid and salt are known from the literature [1-3]. The presence of salt in the solution causes shielding of charges along the polymer chains and subsequently the shrinkage of the chains into coil form, as a preferred conformation for molecular weight measurements. A few years ago we have studied the possibility of using a simpler, aqueous two-component solvent, in which the chitosan chains would attain the coil conformation desired in the molecular weight measurements. Simplification of the measurement procedure and reducing the number of factors may have a positive influence on the quality and accuracy of measurement results. Preliminary results have shown that the salt, usually added as the third ingredient of the solvent, can be successfully replaced by using excess, relative to the stoichiometric amount, of a strong acid (e.g. hydrochloric acid) [4]. Application of such a solvent could facilitate the study of the properties of chitosan and would eliminate one of the sources of error (salt concentration). Measurements were done by viscometric and static light scattering (SLS) methods.

Current study focused on physicochemical behavior of chitosan in a new solvent, i.e. inorganic acid aqueous solution (HCl) and (for comparison) in the classical three-component systems, i.e. inorganic acid with salt, while keeping equal ionic strength of both solvents.

Studies of aqueous chitosan solutions indicate that strong monoprotic acids (HCl, HClO4), used in mall excess to the stoichiometric amount of amino groups in the chain, behave like small-molecule electrolytes. This leads to partial shielding of electrostatic interactions between positively charged groups on polymer chain. Physicochemical properties of chitosan in the solution of 0.1 mol dm-3 hydrochloric acid

(such as radius of gyration Rg, hydrodynamic radius Rh, second virial coefficient A2 and solution viscosity) are comparable to those determined in the systems with the same ionic strength, containing acid with the addition of salt.

The values of Rg/Rh ratios and the constants in the Mark-Houwink equation indicate that the chitosan chains in the 0.1 molar aqueous hydrochloric acid solution assume the characteristic structure of the coil in a good solvent. It therefore seems that the proposed, simple two-component solvent can effectively substitute more complex three-component classical solvents for measurement of chitosans molecular weights by the viscosimetric method and laser light scattering.

References:

[1] M. Rinaudo, M. Milas, P.L. Dung, Characterization of chitosan. Influence of ionic strength and degree of acetylation on chain expansion, International Journal of Biological Macromolecules, 15, 1993, 281.

[2] G. Berth, H. Dautzenberg, The degree of acetylation of chitosans and its effect on the chain conformation in aqueous solution, Carbohydrate Polymers, 47, 2002, 39

[3] P. Sorlier, C. Viton, A. Domard, Relation between solution properties and degree of acetylation of chitosan: role of aging, Biomacromolecules, 3, 2002, 1336.

[4] R. Czechowska-Biskup, A. Wojtasz-Pająk, J. Sikorski, A. Henke, P. Ulański, J.M. Rosiak, Aqueous solutions of hydrochloric acid as simple solvents of chitosan for viscosity- and light-scattering-based molecular weight determination. Progress on Chemistry and Application of Chitin and its Derivatives, vol. XIII (Ed. M. Jaworska), ISBN 83-918969-5-1, Polish Chitin Society, Kraków 2007, 87.

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ADSORPTION STUDY OF DROTAVERINE ON CHITOSAN IN AN IN VITRO PHARMACEUTICAL MODEL

During slimming therapy, slimming preparations containing chitosan are recommended and used. In some cases, they may be used in parallel with medications containing diastolic ingredients, for example during pain treatment or in abdominal contractions.

The aim of the study was to determine the binding capacity of drotaverine depending on various physicochemical factors that may be present in the gastrointestinal tract and then transferred to an in vitro model of the gastrointestinal tract specially created for this purpose for modification according to Polish Pharmacopoeia XI.

The adsorption of drotaverine was studied using a dynamic method adopted to a biopharmaceutical model imitating in vivo conditions. The amount of chitosan adsorbed drotaverine was calculated from the difference in concentrations of the examined drotaverine before and after sorption. The results of measurements of the amount of the combined drug were used to calculate the percentage of the absorbed dose.

The obtained results confirm that drotaverine is adsorbed by chitosan in the model regardless of the values of the pH ranges used, and the binding capacity depends on the pH of the environment and viscosity.

The concentration of the drug used, as well as the type of chitosan used and the substances present in the gastrointestinal tract, affect the adsorption rate. The average amount of adsorption of the drug depending on the dose of drug used in the chitosan-substance system ranged from 59 to 100%. The greatest adsorption can be observed when the reaction of the environment is above pH 6.4 (the polymer above this Ph begins to precipitate to solid form).

Analysis of the obtained results shows that the addition of chitosan as a slimming supplement reduces the amount of drotaverine used and has a very significant impact on its bioavailability and pharmacological effects.

References:

[1] Polskie Towarzystwo Farmaceutyczne: Farmakopea Polska VIII. Warszawa (2008): Urząd Rejestracji Produktów Leczniczych, Wyrobów Medycznych i Produktów Biobójczych, 3491.

[2] J. Meler, J. Pluta, The effect of auxiliary substances the activity of lipase pancreatic biopharmaceutical patternelof digestive tract. In: Progress of Chemistry and Application of Chitin and its Derivatives. Vol. X, 131-137, (ed.: H. Struszczyk), Polish Chitin Society, Łódź 2004

[3] J. H. Ryu, S. Hong, H. Lee, Bio-inspired adhesive catechol-conjugated chitosan for biomedical applications: A mini review, Acta Biomaterialia. 27 (2015) 101–15.

[4] D.W. Lee, et al., "Strong adhesion and cohesion of chitosan in aqueous solutions", Langmuir, 29 (2013) 14222–14229.

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INFLUENCE OF CHITOSAN ON THE PHARMACEUTICAL PROPERTIES OF DENTAL APPLICATIONS

The aim of the work was to develop the composition of a bioadhesive film containing benzydamine hydrochloride. The therapeutic substance was incorporated in bioadhesive polymers: chitosan and guar gum in various quantitative ratios.

For this purpose, the technology of making films containing various quantitative ratios of chitosan into guar gum was developed: 1: 0.25; 1: 0.50; 1: 0.75; 1: 1 and 1.2: 0.50. Film production technology was developed, which was poured into molds and subjected to evaporation by hot air drying.

The films were subjected to organoleptic and microscopic analysis their structure and mechanical properties were assessed. Studies have shown that the quantitative ratio of chitosan to guar gum significantly affects the mechanical properties of films, dynamic viscosity of the gels, texture, elasticity, tensile strength, swelling of the tested carriers. Formulations with the best parameters were used to obtain mucoadhesive films with the addition of benzydamine hydrochloride.
Based on the study, it was proved that the amount of chitosan added to the formulation reduces the quantity of released substance and slows down the process. The films containing higher content of guar gum show also the highest tensile strength and the lowest elongation. Guar gum addition did not affect the water vapor permeability, solubility, and moisture of films. The research led to conclusion that the quantitative relation of chitosan and guar gum in the media impacts the mechanical properties and release parameters of the drug dosage form.

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CHITOOLIGOSACCHARIDES SYNTHESIS USING ENZYMES OF *MUCOR CIRCINELLOIDES* IBT-83

In our previous research we have showed that mycelium of Mucor circinelloides IBT-83 is a promising source of chitin and chitosan hydrolysing enzymes [1-2]. They can be used for the production of chitosan oligomers of different polymerization degrees: low-molecular weight chitosan, chitooligosaccharides and D-glucosamine (GlcN). The intracellular chitosanase isolated from *M. circinelloides* IBT-83 was partially purified and characterised [2]. Moreover, it has been proven that this strain produces intracellular chitin deacetylase [3].

As part of our ongoing research, we have confirmed that the enzymes contained in the cell-free extract of *Mucor circinelloides* IBT-83 are capable to catalyze the synthesis of hetero-dimers composed of D-glucosamine (GlcN) and N-acetyl-D-glucosamine (GlcNAc) efficiently.
The reaction mixture contained 100 mg of GlcN and 350 mg of GlcNAc were mixed in 100 ml of acetate buffer (pH of 5.5) containing 15% of ammonium sulfate. Optimum time and temperature of synthesis reaction was found to be 45 h and 50°C, respectively. Synthesis reaction was not observed when only GlcN or GlcNAc was present in the reaction mixture.

References:

[1] K. Struszczyk-Świta et all, Scale-up of PUF-immobilized fungal chitosanase and lipase preparation production. Preparative Biochemistry and biotechnology, 47 (2017) 909-917.

[2] K. Struszczyk et all, Isolation and purification of Mucor circinelloides intracellular chitosanolytic enzymes. Carbohydrate Polymers 78(1) (2009)16-24.

[3] B.M. Kaczmarek et all, Isolation, molecular cloning and characterisation of two genes coding chitin deacetylase from Mucor circinelloides IBT-83. Progress on Chemistry and Application of Chitin and its Derivatives 21 (2016) 93-103

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DIALDEHYDE STARCH AS A NOVEL CHITOSAN CROSSLINKER

Chitosan is a natural biopolymer obtained e.g. from seafood wastes, a copolymer of β-(1→4)-linked-2-acetamido-2-deoxy-β-D-glucopyranose and 2-amino-2-deoxy-β-D-glucopyranose. The high interest in polymer, aboundant in the natural environment, is a consequence of its unique properties [1, 2], i.e. biodegrability, biocompatibility, bioactivity and non-toxicity which all result in its wide application in medicine. However it is also well-known that chitosan isn't resistant to acidic pH. Among different methods of increasing chitosan's resistance, crosslinking is one of the most widely chosen procedures [3].

Among different chitosan crosslinkers glutaraldehyde (GA), genipin, glyoxal are the most commonly used substances. Dialdehydes, however, are considered to be toxic or mutagenic in their unreacted form what can limit the application of crosslinked materials, especially in biomedicine and pharmaceutics. Thus in the present study application of a new crosslinker - a starch derivative, containing aldehyde functional groups - has been proposed.

A series of dialdehyde derivatives was obtained by oxidation process of potato starch (Fig. 1). Aldehyde groups' content in dialdehyde starch (DAS) was determined.

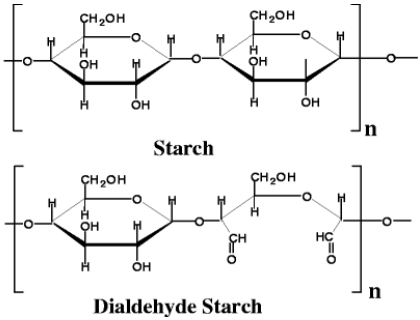

Fig. 1. Chemical structure of starch and its dialdehyde derivative

Chitosan films crosslinked with DAS were obtained. To confirm the chemical structure of prepared membranes Fourier transform infrared spectroscopy (FTIR) was applied. The surface morphology of modified films was studied by scanning electron microscopy (SEM) and atomic force microscopy (AFM). Changes in surface hydrophilicity were characterized by contact angle measurements. Swelling characteristic in different media was also investigated.

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References:

[1] C. Chatelet, O. Damour, A. Domard, Influence of the degree of acetylation on some biological properties of chitosan films. Biomaterials, 22 (2001) 261-268.

[2] M.H. Struszczyk, Chitin and Chitosan. Part II. Applications of chitosan. Polimery, 6 (2002) 396-403.

[3] E. Guibal, Interactions of metal ions with chitosan-based sorbents: a review. Separation and Purification Technology, 38 (2004) 43-74.

BIOTECHNOLOGICAL SESSION

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THREE-DIMENSIONAL CHITIN ISOLATION AND POSSIBLE APPLICATIONS

As is know very well, chitin and its derivatives have many applications in industry thanks to their biodegradable, biocompatible and non-toxic properties. The used chitin or its derivatives in these applications areas are in dust form or the films solved in acetic acid solutions. But here in the present study, I will give information about isolation of three-dimensional (3D) chitin from diverse organisms by keeping the original shape of the organisms or any part of the organism. Thanks to this kind of intact chitin isolation, we have certain shape of chitin films and specific surface characteristics for further application. I will give information about possible application areas of the obtained three-dimensional chitin materials. Also it is very important to have your valuable opinions to make colloboration by using the obtained 3D chitin materials.

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PURIFICATION OF B-N-ACETYLHEXOSAMINIDASE OF *LECANICILLIUM LECANII* AND ITS APPLICATION IN THE OLIGOSACCHARIDES PRODUCTION

Introduction

β-N-acetylhexosaminidase (Nhase) are chitinolytic enzymes capable of hydrolysis and transglycosylation activity (TGA). TGA allows synthesizing of oligosaccharides (COS) with specific degree polymerization (DP) and acetylation which until now have been difficult to obtain by other methods since it represents the disadvantage of reducing the acetylation degree [2]. Therefore, this work aimed to study the COS production through transglycosylation activity of purified Nhase of *L. lecanii*.

Results and Discussion

Nhase produced by L. lecanii in submerged culture purified by salting out with ammonium sulfate (ASP), size exclusion chromatography (SEC) and anion exchange chromatography (AEC), obtaining a yield of 39.5% with a specific activity of 82.29 U /mg of protein. Several temperatures and pH ranges characterized the hydrolytic activity, finding the optimal conditions of temperature at 37 ° C and pH of 6. As well, pH 6, 7 and 8 tested TGA using Glucose, Mannose, N-acetyl-D-lactosamine and N-acetyl-D-glucosamine as donor substrates [2]. Thin layer chromatography (TLC) and mass spectrometry (MALDI-TOF) analyzed the reaction products [1]. The Nhase produced at pH 7 higher concentration of COS than pH 6 and 8. COS presented DP between 2-6 with acetylated fraction (FA) of 1-4.

Conclusions

Purification of the Nhase from *L. lecanii* was carried out efficiently obtaining a high yield. Nhase displayed both activities hydrolytic and transglycosylating. The TGA allowed the synthesis of COS with high F_A and DP.

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References:

[1] A. Gal, "Separation and identification of monosaccharides from biological materials by thin-layer chromatography", Analytical Biochemistry, 24 (1968) 452-461.

[2] C. Li, W. Huang, L. Wang, "Chemoenzymatic synthesis of N-linked neoglycoproteins through a chitinase-catalyzed transglycosylation", Bioorganic and Medicinal Chemistry, 16 (2008) 8366-8372.

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CHITIN DEACETYLASES FROM *MUCOR CIRCINELLOIDES* IBT-83 AND THEIR USE IN THE SYNTHESIS OF A MULTI-ENZYME COMPLEX

Chitosan, the N-deacetylated derivative of chitin, is characterised by good solubility in acidic solutions among therange of other unique properties, therefore it is widely used in numerous industries [1]. In the recent years, there has been much interest in the products of chitin and/or chitosan hydrolysis, especially linear chitooligosaccharides (COS). COSare soluble in solutions with the neutral pH. Due to their low molecular weight and decreased viscosity they can be successfully applied, for example, as carriers for gene transfection and drug delivery matrices [2]. At present, chitosan and COS are obtained as a result of environmentally unfriendly thermo-chemical and/or physical processes which are very difficult to control [3].

The aim of our project is to create a first, polycistronic, eukaryotic system enabling simultaneous expression of three genes coding chitin- and chitosanolytic enzymes, which are crucial for efficient modification of chitin and its derivatives. The functioning of this multi-gene system will be based on conservative, viral 2A self-processing sequences enabling polycistronic expression under the control of one promoter [4]. In our previous research, we have heterologously expressed two isoforms of chitin deacetylase (ChDaI and ChDaII) from *Mucor circinelloides* IBT-83 [5]. Activity of one of the chitin deacetylase isoforms (ChDaII) was confirmed after expression in methylotrophic yeast Pichia pastoris. After biochemical characterisation, the gene encoding ChDaII and other genes comprising multienzymatic complex (encoding chitosanase and chitinase) were used for the synthesis of polycistronic system pCHITB03. The proper functioning of A2 self-processing sequences was confirmed after expression of pCHITB03 in P. pastoris. The sequences of obtained heterologous proteins were analysed by LC MS/MS.

Our approach will significantly reduce the cost of producing enzyme preparations, making biotechnological methods competitive to conventional physical and chemical processes. In addition, the presenceof all enzymes in one reaction mixture can significantly increase the yield of the production of defined chitin derivatives.

References:

[1] C. T. G. V. M. T. Pires, J. A. P. Vilela, C. Airoldi. The Effect of Chitin Alkaline Deacetylation at Different Condition on Particle Properties, Procedia Chemistry, 9 (2014) 220-225.

[2] I. Hamed, F. Ozogu, J.M. Regenstein. Industril applications of crustacean by products (chitin, chitosa and chitooligosaccharides): A review, Trands in Food and Science Technology, 48 (2015) 40-50 Publisher, City year.

[3] G.A. Roberts, Thirty Years of Progress in Chitin and Chitosan, Progress in the Chemistry and Application of Chitin and its Derivatives, 13 (2008) 7-15.

[4] M. Geier, P. Fauland, T. Vogl, A. Glieder, Compact multi-enzyme pathway in P. Pastoris, Chemical communications, 51 (2015) 1643-1646.

[5] M.B. Kaczmarek, K. Struszczyk-Swita, T. Florczak, M. Szczesna-Antczak, T. Antczak, Isolation, molecular clonning and characterisation of two genes coding chitin deacetylase from Mucor circinelloides IBT-83, , Progress in the Chemistry and Application of Chitin and its Derivatives, 16 (2016) 93-103.

PHYSICO-CHEMICAL SESSION

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STRUCTURAL CHARACTERISTICS OF THERMOSENSITIVE COMPOSITIONS BASED ON CHITOSAN

In the past few years, there has been an avalanche development of nanotechnology sciences, among which nanomedicine has a special position, referring to diagnostic and research activities and therapeutic treatment using nanoparticles, including graphene and graphene oxide [1]. Graphene oxide (GO) is an oxidized form of graphene that has in its structure characteristic carboxyl, hydroxyl and epoxy functional group (Fig.1.).

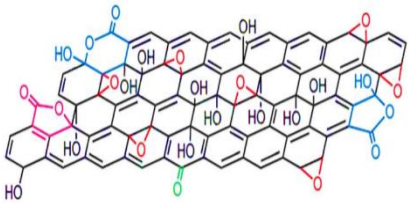

Figure 1. Structure of GO [2]

Due to thr non-typical biological properties (biocompatibility with the tissues and organs, bacteriostatic and antiseptic features) and high solubility in physiological solutions - hydrophilicity, there have been many reports in the literature regarding the possibility of using GO in medicine.

The aim of the research was to perform a structural analysis of chitosan - graphene oxide nanocomposites, with a view to its potential use as injectable scaffold for regeneration of damaged tissues. In this study, two different type of chitosan thermosensitive hydrogels were used: pure gel and gel containing GO with a concentration of 0.028 to 0.1 mg GO/ml chit.salt.

The structural characterics of hydrogels were based on the analysis of FTIR spectra, images from polarizing (Fig.2.) and scanning electron microscope.

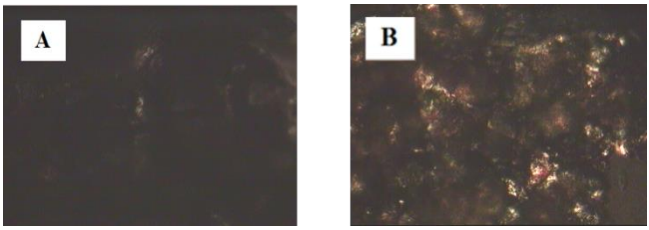

Figure 2. Images from the polarizing microscopy: A - chitosan lactate gel with 0.1 mg GO/ml chit. salt; B - chitosan chloride gel with 0.1 mg GO/ml chit. salt

Analyzing the above images, we can conclude that the image for chitosan chloride gel corresponds to a typical crystal structure. We can observe the characteristic golden areas.

In addition, a preliminary biological study was carried out - the survival of osteoblast cells of the Saos-2 line was analyzed on the surface of pure chitosan hydrogel and GO enriched systems.

Based on obtained results, it can be state that the thermosensitive chitosan gels formed at physiological body temperature can contain nano-ingredients in their structure. All analyzed systems exhibit thermogel features for which temperature increase initiates the sol-gel phase transformation. Thus, chitosan-graphene oxide nanocomposites are very promising base for biomaterial fabrication for regenerative medicine applications.

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References:

[1] R.Wojnicz, Nanomedycyna jako fundament medycyny personalizowanej, Kardiologia Polska, 69 (2011) 1107-1108.

[2] R. Kumar Singh, R. Kumar, D. Pratap Singh, Graphene oxide: strategies for synthesis, reduction and frontier applications, RSC Advances, 6 (2016) 64993-65011

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NOVEL NANOCOMPOSITE CHITOSAN MEMBRANES ON POLYAMIDE-6 SUPPORT FOR PERVAPORATION

Chitosan (Ch), a main chitin derivative, is regarded as one of the most promising membrane materials for ultrafiltration, reverse osmosis and pervaporation. Pervaporation (PV) is a membrane technique used in separation of azeotropic or close-boiling mixtures [1,2]. In PV separation of water-ethanol mixtures, chitosan is regarded as one of the most effective water-permselective material since chitosan membranes show high separation factors. Due to its film-forming property Ch can be used to prepare homogenous membranes as well as the skin layer of the composite membranes.

Thermal stability, hardness, and gas barrier properties of Ch films are not favourable enough to meet a wide range of applications. According to the literature survey, Ch membranes can be modified using various fillers and supports. In recent years, natural polymer/clay composites have attracted considerable interest because they combine the structure as well as the physical and chemical properties of inorganic and organic materials [3]. Natural organic clays have become pivotal in composite manufacturing due to several factors which include their renewable nature, wide availability, relatively low cost and the ease of surface functionalization. A number of reports presented in literature describe the use of clay (e.g. montmorillonite and its derivatives etc.) to reinforce and improve physical, mechanical and structural properties of chitosan films [3]. Another approach is the formation of thin selective Ch layer on different porous supports.

In the present study novel chitosan nanocomposite membranes deposited on asymmetric polyamide-6 (PA6) support were fabricated for the pervaporative separation of azeotropic mixtures. PA6 films were obtained by the phase inversion method. Subsequently, chitosan/clay mixtures were applied using a casting knife with 0.3 mm slit. Scanning electron microscopy (SEM) and atomic force microscopy (AFM) were applied to determine the morphology of membranes' surface. Separation properties of membranes in pervaporation of water/alcohol mixtures were also examined. Binary water/ethanol and water/isopropanol solutions of different composition were used as a feed. All PV experiments were performed at 30 °C. The separation efficiency was described in terms of permeation fluxes, separation factor and pervaporation separation index.

It was found that all investigated membranes were selective towards water. The Pervaporation Separation Index (PSI) for water was the highest in the case of polyamide-6/chitosan membrane filled with 3 wt.% modified Cloisite 30B clay (Fig. 1).

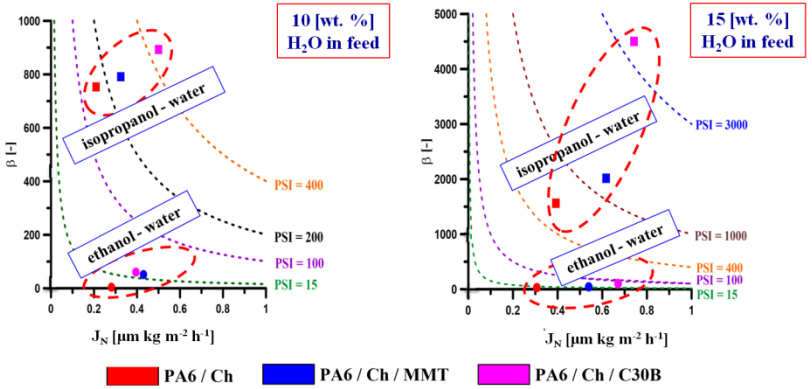

Fig. 1. Performance of PA6 supported chitosan membranes in separation of water/alcohol mixtures.

Acknowledgements:

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References:

[1] S.M. Zhang, E. Drioli, Pervaporation membranes (review). Separation Science and Technology, 30 (1995) 1-31.

[2] D. Xu, S. Hein, K. Wang, Chitosan membranes in separation applications. Materials Science and Technology, 24 (2008) 1076-1087.

[3] K. Lewandowska, A. Sionkowska, B. Kaczmarek, B. Furtos, Characterization of chitosan composites with various clays. International Journal of Biological Macromolecules, 65 (2014) 534-541.

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[1] Smith J, Jones M, Houghton L; (1999) Future of health insurance. N Engl J Med 965, 325–329.

[2] Slifka MK, Whitton JL; (2000) Clinical implications of dysregulated cytokine production. J Mol Med. Doi:10.1007/s001090000086

[3] South J, Blass B; (2001) The future of modern genomics. Blackwell, London.

[4] Brown B, Aaron M; (2001) The politics of nature. In: Smith J (ed), The rise of modern genomics, 3rd edn. Wiley, New York, 230-236.

[5] Wolfrom ML, Szarek WA; (1972) Halogen Derivatives. In: Pigman W, Horton D (eds), The Carbohydrates, Chemistry and Biochemistry, Vol. 1A, 2nd ed, Academic Press, New York, 239-251.

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