1st International Conference
on Chemo and BioInformatics
ICCBIKG 2021

ICCBIKG

1st International Conference on
Chemo and BioInformatics

BOOK OF PROCEEDINGS

October 26-27th, 2021,
Kragujevac, Serbia

www.iccbikg.kg.ac.rs

Ministry of Education, Science and Technological Development of the Republic of Serbia

SHIMADZU
Excellence in Science

KEFO
SINCE 1949

Alfamed

UNI-CHEM

SUPERLAB
Your Lab - Our Passion

ART WINE

ICCBIKG
1st International Conference on Chemo and BioInformatics

1st International Conference on Chemo and BioInformatics
ICCBIKG 2021

BOOK OF PROCEEDINGS

October 26-27, 2021
Kragujevac, Serbia

Sponsored by

Ministry of Education, Science and Technological Development of the Republic of Serbia

SUPERLAB
Your Lab - Our Passion

SHIMADZU
Excellence in Science

KEFO
SINCE 1949

ART WINE

UNI-CHEM

Alfamed

1st International Conference on Chemo and BioInformatics, Kragujevac, October 26-27, 2021 Serbia

Editors:

Professor Zoran Marković

Professor Nenad Filipović

Technical Editors:

Vladimir Simić

Izudin Redžepović

Nikola Srećković

Illustrations:

Igor Stanković, „Vector Alchemist“ d.o.o.

Publisher:

Institute for Information Technologies, University of Kragujevac, Serbia, Jovana Cvijića bb, 2021

Press:

„Grafo Ink“, Kragujevac

Impression:

120 copies

CIP - Каталогизација у публикацији - Народна библиотека Србије, Београд

54:004(048)(0.034.2)
57+61]:004(082)(0.034.2)

INTERNATIONAL Conference on Chemo and BioInformatics (1 ; 2021 ; Kragujevac) Book of Proceedings [Elektronski izvor] / 1st International Conference on Chemo and BioInformatics, ICCBIKG 2021, October 26-27, 2021 Kragujevac, Serbia ; [editors Zoran Marković, Nenad Filipović]. - Kragujevac : University, Institute for Information Technologies, 2021 (Kragujevac : Grafo Ink). - 1 USB fleš memorija ; 3 x 2 x 1 cm

Sistemski zahtevi: Nisu navedeni. - Nasl. sa naslovne strane dokumenta. - Tiraž 120. - Bibliografija uz svaki rad.

ISBN 978-86-82172-01-7

a) Хемија - Информациона технологија - Зборници b) Биомедицина - Информациона технологија - Зборници

COBISS.SR-ID 48894473

Organized by

- Institute for Information Technologies, Organizer

- Faculty of Science, University of Kragujevac, Suborganizer

- Faculty of Engineering, University of Kragujevac, Suborganizer

- University of Kragujevac, Supporting organization

- The Ministry of Education, Science and Technological Development of The Republic of Serbia, Supporting organization

Committees

International Organizing Committee:

Chairman:	Prof. Zoran Marković (Serbia)
Vice-chairmans:	Prof. Zlatan Car (Croatia)
	Prof. Carlos Silva Lopez (Spain)

Members:

Dr Dejan Milenković (Serbia), Dr Dubravka Živković (Serbia), Dr Biljana Šmit (Serbia), Dr Miljan Milošević (Serbia), Dr Edina Avdović (Serbia), Dr Aleksandar Ostojić (Serbia), Dr Verica Jevtić (Serbia), Dr Milan Kovačević (Serbia), Dr Dragana Šeklić (Serbia), Dr Sanja Matić (Serbia), Dr Dušica Simijonović (Serbia), Dr Aleksandar Nikolić (Serbia), Dr Tatjana Miladinović (Serbia), Dr Saša Ćuković (Serbia), Dr Biljana Glišić (Serbia), Dr Vladimir Petrović (Serbia), Dr Andrija Ćirić (Serbia), Dr Nenad Janković (Serbia).

International Scientific Committee:

Chairman:	Prof. Nenad Filipović (Serbia)
Vice-chairmans:	Prof. Claudio Santi (Italy)
	Prof. Goran Kaluđerović (Germany)

Members:

Prof. Zoran Marković (Serbia), Prof. Ivan Gutman (Serbia), Prof. Miloš Kojić (USA), Prof. Velimir Popsavin (Serbia), Prof. Miloš Đuran (Serbia), Prof. Nenad Kostić (USA), Prof. Ljiljana Kolar-Anić (Serbia), Prof. Svetlana Marković (Serbia), Prof. Snežana Zarić (Serbia), Prof. Marija Stanić (Serbia), Prof. Biljana Petrović (Serbia), Prof. Dobrica Milovanović (Serbia), Prof. Miroslav Živković (Serbia), Prof. Nenad Grujović (Serbia), Prof. Dragoslav Nikezić (Serbia), Prof. Zlatan Car (Crotaia), Prof. Ivan Potočňák (Slovakia), Prof. Luciano Saso (Italy), Prof. Dražen Vikić-Topić (Croatia), Prof. Bakhtiyor Rasulev (USA), Prof. Erik Klein (Slovakia), Prof. Viktor Stefov (Macedonia), Prof. Svetlana Simova (Bulgaria), Prof. Enver Karahmet (Bosnia and Herzegovina), Prof. Themis Exarchos (Greece), Prof. Carlos Silva Lopez (Spain), dr. sc. Mario Vazdar (Czech Republic), Prof. Arturas Ziemys (USA), Prof. Jasmina Dimitrić-Marković (Serbia), Prof. Snežana Bogosavljević Bošković (Serbia), Prof. Jasmina Stevanović (Serbia).

Local Executive Committee:

Chairman:	Dr Dejan Milenković (Serbia)
Vice-chairmans:	Dr Jelena Đorović Jovanović (Serbia)
	Dr Jelena Katanić Stanković (Serbia)

Members:

Dr Darko Ašanin (Serbia), Dr Emina Mrkalić (Serbia), Žiko Milanović (Serbia), Vladimir Simić (Serbia), Bogdan Milićević (Serbia), Aleksandar Milovanović (Serbia), Nevena Veselinović (Serbia), Izudin Redžepović (Serbia), Nikola Srećković (Serbia).

doi:10.46793/ICCBI21.125M

EFFECT OF CROSSLINKER AMOUNT ON HYBRID HYDROGELS SWELLING AND DRUG RELEASE

Maja D. Markovic[1], Vesna V. Panic[1], Julijana D. Tadic[2,3], Rada V. Pjanovic[3]

[1] Innovation Center of Faculty of Technology and Metallurgy,
University of Belgrade, Karnegijeva 4, 11000 Belgrade, Serbia
e-mail: mmarkovic@tmf.bg.ac.rs
[2] Vinča Institute of Nuclear Sciences – National Institute of the Republic of Serbia, University of Belgrade, Mike Petrovića Alasa 12-14, 11001 Belgrade, Serbia
[3] Faculty of Technology and Metallurgy,
University of Belgrade, Karnegijeva 4, 11000 Belgrade, Serbia
e-mail:vpanic@tmf.bg.ac.rs , jtadic@tmf.bg.ac.rs, rada@tmf.bg.ac.rs

Abstract:

Targeted drug delivery is powerful tool which researchers use to achieve safer and more efficient therapy of many diseases, including various types of cancer. Many chemotherapeutics are poorly water-soluble, so their encapsulation and targeted delivery remain quite challenge. Hydrogels based on poly(methacrylic acid) (PMAA) are widely investigated for targeted drug delivery due to their pH sensitivity, non-toxicity and biocompatibility. Still, due to the PMAA highly hydrophilic nature, PMAA can only be used for encapsulation and targeted delivery of water-soluble drugs. Our previous research was directed towards overcoming this limitation: PMAA was modified with amphiphilic protein – casein and poorly-water soluble model drug – caffeine – was encapsulated (PMAC). Present study is focused on investigation how variation of amount of one of the most important hydrogels network parameter such as crosslinker affect PMAC swelling properties and caffeine release. The group of hybrid hydrogels – PMAC – was synthesized with various amount of crosslinker: 0.4mol%, 0.8mol%, 1.6mol% and 3.2mol% with respect to methacrylic acid. Swelling behavior of hybrid hydrogels and caffeine release was investigated in two environments which simulated human stomach and intestines. Obtained results showed that targeted delivery of poorly water-soluble model drug was achieved and that its release can be prolonged up to 24h. Also, kinetic of poorly water-soluble drug release can be easily modified only by changing crosslinker amount. PMAC hybrid hydrogels have huge potential for targeted delivery of poorly water-soluble active substances.

Key words: poly(methacrylic acid), casein, crosslinking, hydrogels swelling, drug release

1. Introduction

Researchers are faced with a lot of challenges in their attempts to find new or to improve existing drug delivery systems to achieve safer and more efficient therapy of various diseases. This is especially significant in cancer therapy, since many anticancer drugs are poorly water soluble and have severe side effects [1]. Encapsulation and targeted delivery of these drugs can improve their bioavailability, decrease side effects and reduce the number of required therapeutic dosages. Hydrogels based on poly(methacrylic acid) (PMAA) are widely used for targeted drug delivery. These extraordinary materials are pH sensitive, biocompatible, non-toxic and their morphology is similar to human tissues [2]. Their highly hydrophilic nature, however, limits their application only to encapsulation and targeted delivery of water-soluble drugs. In our previous research, we overcome this limitation by modifying PMAA with amphiphilic casein, which enabled encapsulation of poorly water soluble model drug - caffeine (PMAC) [3, 4]. Casein is non-toxic, biocompatible, pH sensitive milk protein, which use is

approved by Food and Drug Administration (FDA)[3]. This study is focused on the investigation how crosslinker amount affects swelling of these hybrid PMAC hydrogels and caffeine release, because degree of crosslinking of the hydrogels network are one of the most important parameters which affect the hydrogels properties and consequently drug release.

2. Materials and methods

2.1 Materials

Methacrylic acid (99.5%) and caffeine were purchased from Merck (Germany). Sodium caseinate was supplied from Lactoprot Deutschland GmbH (Germany). The crosslinker N,N'-methylenebisacrylamide (p.a.) (MBA) and sodium hydroxide (p.a.) were supplied from Aldrich Chemical Co. (USA). The initiator, 2,2'-azobis-[2-(2-imidazolin-2-yl)propane] dihydrochloride (99.8%) was purchased from Wako Pure Chemical Industries (Japan). Monobasic sodium phosphate (anhydrous) and dibasic sodium phosphate (anhydrous) was supplied from Centrohem (Serbia). Hydrochloric acid (37%) was supplied from Zorka Pharma (Serbia). All chemicals were used as received.

2.2 Samples preparation

The process of the samples preparation and characterization are described in details in our previous research [3, 4]. In this study we varied the amount of crosslinker. Briefly: after 4 ml of MAA and 0.2 g of caffeine dissolution in distilled water (see Table 1. for Feed composition), sodium hydroxide was added in order to completely neutralized MAA. Then, the temperature of reaction mixture was elevated to 60ºC and during vigorously stirring of the reaction mixture 4 g of casein was added. After casein dissolution, a certain amount of crosslinker (Table 1.) was added and dissolved, followed by the addition and dissolution of the initiator (0.9 cm^3 of 1wt% aqueous solution). Then, the reaction mixture was quickly poured in glass molds and left in the air oven at 60°C for 5h after which disc shaped samples were cut and dried at room temperature. Synthetized samples were denoted as PMAC-xM-0.2, where xM represents the amount of crosslinker (2M, 4M and 8M = 0.8mol%, 1.6mol% and 3.2mol% of crosslinker, respectively).

Table 1. Feed composition

Samples	Distilled water (ml)	Crosslinker amount (mol%*)
PMAC-0.2	6.20	0.4
PMAC-2M-0.2	3.30	0.8
PMAC-4M-0.2	8.93	1.6
PMAC-8M-0.2	8.81	3.2

*with respect to the MAA amount

2.3 Samples swelling

The experiments of the samples swelling were conducted at 37ºC in two environments with different pH values: 0.1M HCl with pH of 1 (as simulation of human stomach) and phosphate buffer with pH of 6.8 - PB 6.8 (as simulation of human intestines). The mass of each sample was first measured (MO, g) and then each sample was immersed in each environment. At previously defined time intervals sample was removed from the environment, its mass was measured (MT, g) and then the sample was immersed again. The experiment was conducted until equilibrium state was reached (i.e. until mass of the sample did not change). The swelling degree (SD) was calculated according to the following equation:

$$SD = \frac{MT-MO}{MO} \quad (1)$$

2.4 Drug release from the samples

The drug release process was investigated in the same environments and at the same experimental conditions as the process of the samples swelling. At predetermined time intervals 3 ml of the solution was colected and UV analyzed at 273 nm, after which the solution was returned back. Each experiment

was conducted three times and mean value of absorbance was used for further determination of concentration of drug released in the environment.

3. Results and discussion

Samples with various amount of crosslinker – MBA– were synthetized in order to investigate how the degree of crosslinking of samples network affect the process of samples swelling and drug release. The curves of the process of the samples swelling in two environments with different pH values – 0.1M HCl and PB 6.8 – are presented in Fig. 1 a) and b). The values of equilibrium swelling degree (SDeq) of the samples are presented in Table 2.

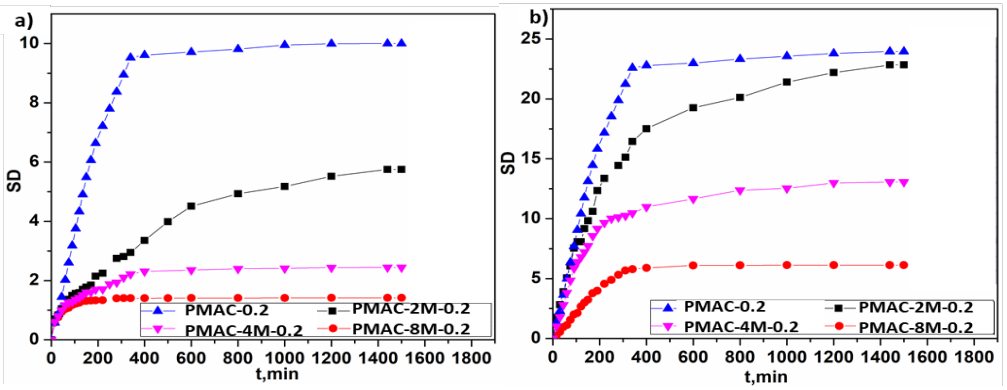

Fig. 1. The curves of samples swelling in: a) 0.1M HCl and b) PB 6.8

As it can be seen in Fig. 1 a) and b), the samples swell more in PB 6.8 than in 0.1M HCl. The values of SDeq are around five times higher in PB 6.8 than in 0.1M HCl. This specific pH- dependent swelling behavior of the samples can be explained by deproteinization of the carboxylic groups in environments with pH value higher than pKa of PMAA and pI of casein (such as PB 6.8) [2, 5, 6]. Consequently, negative charges on polymeric chains are generated, which further leads to the repulsion between polymeric chains and samples swelling.

Table 2. The values of equilibrium swelling degree (SDeq) of the samples

Samples	0.1M HCl	PB 6.8
PMAC-0.2	12.4	23.9
PMAC-2M-0.2	5.75	22.8
PMAC-4M-0.2	2.44	13.1
PMAC-8M-0.2	1.42	6.12

Based on the swelling curves of the samples presented in Fig. 1. a) and b) and the results presented in Table 2. it can be concluded that the values of SDeq decrease with increase of crosslinker amount. These results are in accordance with the fact that hydrogels with higher degree of crosslinking of network have smaller pores and more rigid polymers chains [6].

The profiles of drug release from the samples in 0.1M HCl and PB 6.8 are presented in Fig. 2. a) and b), respectively. All samples release higher amount of drug in PB 6.8 than in 0.1M HCl due to the specific pH dependent swelling behavior. The profiles of drug release showed that increase of crosslinker amount led to decrease of the drug release rate. Increase of crosslinker amount led to the decrease of the diameter of the pores of the hydrogels network, which prolonged drug diffusion from the samples [7]. Drug amount which was released from the sample with 3.2 mol% of crosslinker (PMAC-8M-0.2) in PB 6.8 was around four times less than drug amount released from the referent sample with 0.4 mol% of crosslinker (PMAC-0.2).

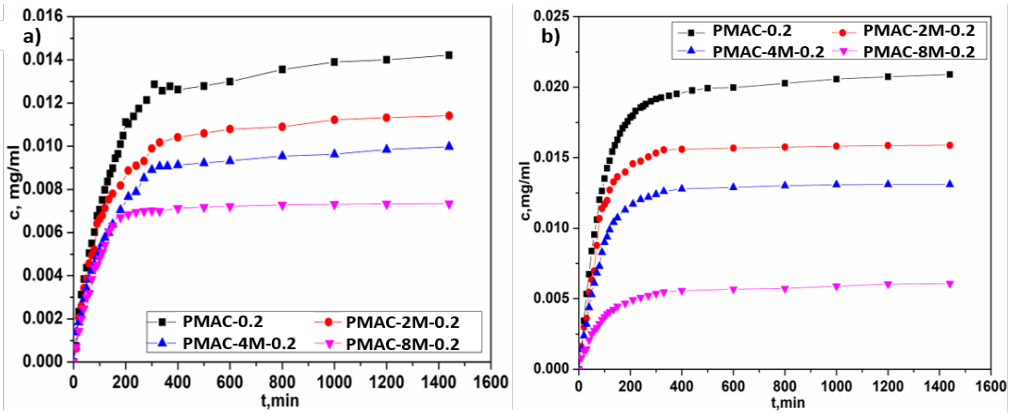

Fig. 2. The profiles of drug release from the samples in: a) 0.1M HCl and b) PB 6.8

4. Conclusions

In this paper, the group of the samples based on poly(methacrulic acid) and casein with encapsulated poorly water soluble model drug – caffeine – were prepared. The amount of crosslinker was varied in order to investigate how the degree of crosslinking of the hydrogels network affect swelling behavior of the samples and drug release. The increase of the crosslinker amount led to the decrease of the samples swelling and consequently to the decrease of the rate of drug release. Presented results showed that drug release kinetics could easily be tuned just by changing crosslinker amount. Present study gives good fundamentals for future investigation of targeted delivery of certain poorly water-soluble drugs (such as chemotherapy agents) which is of great importance in cancer therapy.

Acknowledgement

This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Contract Nos. 451-03-9/2021-14/200287 and 451-03-9/2021-14/200017).

References

[1] S. Singh, U.N. Dash, M. Talukdar, *Solubility enhancement and study of molecular interactions of poorly soluble ibuprofen in presence of urea, a hydrotropic agent*, Materials Today: Proceedings 30 (2020) 246-253.

[2] M.D. Markovic, P.M. Spasojevic, S.I. Seslija, I.G. Popovic, D.N. Veljovic, R.V. Pjanovic, V.V. Panic, *Casein-poly(methacrylic acid) hybrid soft networks with easy tunable properties*, European Polymer Journal 113 (2019) 276-288.

[3] M.D. Markovic, V.V. Panic, S.I. Seslija, P.M. Spasojevic, V.D. Ugrinovic, N.M. Boskovic-Vragolovic, R.V. Pjanovic, *Modification of hydrophilic polymer network to design a carrier for a poorly water-soluble substance*, Polymer Engineering & Science 60(10) (2020) 2496-2510.

[4] M.D. Markovic, V.V. Panic, S.I. Seslija, A.D. Milivojevic, P.M. Spasojevic, N.M. Boskovic-Vragolovic, R.V. Pjanovic, *Novel strategy for encapsulation and targeted delivery of poorly water-soluble active substances*, Polymer Engineering & Science 60(8) (2020) 2008-2022.

[5] S.Z.M. Rasib, Z. Ahmad, A. Khan, H.M. Akil, M.B.H. Othman, Z.A.A. Hamid, F. Ullah, *Synthesis and evaluation on pH- and temperature-responsive chitosan-p(MAA-co-NIPAM) hydrogels*, Int J Biol Macromol 108 (2018) 367-375.

[6] V. Panic, B. Adnadjevic, S. Velickovic, J. Jovanovic, *The effects of the synthesis parameters on the xerogels structures and on the swelling parameters of the poly(methacrylic acid) hydrogels*, Chemical Engineering Journal 156(1) (2010) 206-214.

[7] T. Ukmar, U. Maver, O. Planinšek, V. Kaučič, M. Gaberšček, A. Godec, *Understanding controlled drug release from mesoporous silicates: Theory and experiment*, Journal of Controlled Release 155(3) (2011) 409-417.