CONTROLLED RELEASE FROM LAYERED NANOSTRUCTURED MATERIALS DRIVEN BY CAPILLARY FORCES

[1,2]Anna SUDAKOVA, [5]Duc Anh NGUYEN, [3]Le MINH HA, [2]Jiri SMILEK, [4]Thi Phuong NGO, [1]Jan ZIDEK

[1]CEITEC – Central European Institute of Technology, Brno, Czech Republic, EU, Anna.Sudakova@ceitec.vutbr.cz, jan.zidek@ceitec.vutbr.cz

[2] Institute of Physical and Applied Chemistry, Faculty of Chemistry, Brno, Czech Republic, EU, smilek@fch.vut.cz

[3]Institute of Natural Products Chemistry (INPC), Vietnam Academy of Science and Technology (VAST), Nghia Do, Cau Giay, Hanoi, Viet Nam, halm2vn@gmail.com

[4]Phenikaa University Nano Institute (PHENA), Phenikaa University, Hanoi, Vietnam, thuy.nguyenthithu@phenikaa-uni.edu.vn

[5]Center for Research and Technology transfer, Vietnam Academy of Science and Technology (VAST), Nghia Do, Cau Giay, Hanoi, Viet Nam, ducanh221100@gmail.com

https://doi.org/10.37904/nanocon.2022.4608

Abstract

Controlled release of active compounds has been investigated in current medical research and there were developed many approaches, how to control it. For instance, complexation of active compound, encapsulation in matrix, or release from gels. However, there are still opportunities to improve its mechanisms.

This contribution presents potentially promising method of controlled release, which is based on two modifications of standard mechanisms: i. release from nanostructured layered material, ii. release forced by capillarity. That mechanism allows us to design systems with gradual release of the active substance.

The release of indigotin dye was performed from materials where layers of non-woven chitosan fabric alternate with gelatine layers. Active compound was incorporated in each layer. Gradual wetting of layers is caused by capillarity effect. The controlled release can be observed visually, e.g. by optical spectroscopy. In the future, the colour compound will be substituted by medical active substances, such as antibiotics etc.

The final systems will be composed of natural and biocompatible materials, such as polysaccharides and proteins. The active compound will be in synergy with the chitosan matrix. Chitosan is a unique natural polymer with positive charge on the surface, it exhibits mucoadhesive and antibacterial properties. Gelatine was chosen because it is a protein with a negative charge on its surface, which forms a polyionic mixture with positively charged chitosan.

Such material will improve the current practice in the field of medical materials. It will allow us to design patches treated with an active substance that could improve the therapeutic effect.

Keywords: Chitosan, nanostructured gel, controlled release, capillary force, layered material

1. INTRODUCTION

The study of controlled release of active particles and drugs is gaining importance nowadays. This is because it can result in an optimal dosage of the drug in the desired location, not only for delivery over longer period of

time and one-time application [1]. A major problem here is primarily the burst effect, which is characterized by the rapid release of a certain amount of active substance from the structure in presence of a medium.

This goal is to develop a material composed of nanolayers of non-woven fabric in combination with chitosan hydrogel for the controlled release of an active substance and the polypropylene non-woven fabric will control the release by two methods, the first is multilayer release and the second is by capillary force.

A non-woven polypropylene fabric was used to prepare the layered material, which was glued together by using a collagen solution. Negatively charged indigotin dye was incorporated into this material, which was used to observe the release from the material. The polypropylene non-woven fabric [2] was chosen because it is a material with large pores (approximately10 μm), which exhibits excellent thermal and chemical stability, and at the same time retains its flexibility.

Collagen [3] is a part of larger glycoprotein family, which consist of 27 types of collagen. This linear polymer is a component of extracellular matrices. In its primary structure occurs amino acid glycine in every third position. Higher structure consist of three polypeptide chains folded into triple helices. Gelatine [4] is a product of collagen hydrolysis. In the preparation of the nanolayered material, a gelatine solution is used, it connects the layers of the non-woven fabric by creating a hydrogel, where the individual fibres are connected by covalent bonds.

2. PRINICPLE AND METHOD

2.1. Preparation of layered material

A non-woven polypropylene fabric was chosen for the preparation of the layered material. The initial, squares of non-woven fabric with a size of approximately 4×4 cm were cut out. A concentration series of the blue dye indigotin was prepared with the concentration of 50; 25; 12.5; 6.25; 3.1; 1.6 and 0.8 wt. %. The gelatine solution was prepared at a concentration of 3 wt. %. The layered material was assembled by applying the most concentrated solution of indigotin to one layer of non-woven fabric, followed by a layer of gelatine and two more layers of pure non-woven fabric, which were bonded together by gelatine.

2.2. Release of the indigotin dye

The layered material was treated by indigotin. It was hermetically sealed from water from side in order to prevent the diffusion from side. The diffusion is enabled only from the front. The release was observed to controlled amount of water. The release was observer during 96 hours, the release of the indigotin dye from the layered material into water environment.

2.3. Diffusion through the layered material

The diffusion of the dye through the layered material was also monitored using diffusion cells. Measurement using diffusion cells [5] is a very easy method for determining the diffusion coefficient through our layered material. The apparatus consisted of two cells, receiving and source, where one was filled with 60 ml of distilled water and the other with 60 ml of methylene blue solution (0.01 g/l). Prepared layered materials were inserted between these cells. Layered materials with the number of layers 1, 2 and 8 were prepared for this measurement. A methylene blue solution with a concentration of 0,01 g/l was prepared for diffusion through the material in the diffusion cells. The experiment ran for 24 hours, after which samples were taken from the receiving cells for absorbance measurement on a UV-VIS spectrophotometer.

2.4. UV-VIS spectrophotometry

By measuring UV-VIS spectrophotometry, the diffusion coefficient for layered materials with a different number of layers was determined. A Hitachi U-3900H Spectrophotometer was used. The measured wavelength range

was from 900 to 300 nm. First, the calibration curve of methylene blue solutions, which had concentrations of 0.01; 0.007; 0.005; 0.003 and 0.001 g·dm^{-3}. From these measurements, the absorbance measured at 665 nm was selected the values were related to the concentration and a graph and linear regression equation were plotted. Using the regression equation and the obtained absorbance values, the concentrations in the receiving and source cells were calculated, as well as the concentration of methylene blue retained in the layered material. The following equation was used to calculate the diffusion coefficient.

$$\mathrm{D} \cdot t = -\frac{1}{\beta} \cdot \ln\left(\frac{(c_{sc} - c_{rc})_t}{(c_{sc} - C_{rc})_0}\right) \quad (1)$$

Where:

D - diffusion coefficient (m^2·s^{-1})

β - diffusion cell constant (m^{-2})

c_{sc} - concentration in source cell (g·dm^{-3})

c_{rc} - concentration in receiving cell (g·dm^{-3})

2.5. Atomic Force Microscopy

Atomic force microscopy (AFM) is a method belonging to the group of tunnelling microscopy, which is standardly used to image samples at the nano (micro) level [6]. The scanning principle is based on the measurement of attractive and repulsive forces that are applied between the measuring tip and the sample. In practice, atomic force microscopy is mainly used for imaging the surfaces of samples in aqueous environment, as well as in air. Another possible use is the measurement of the mechanical and adhesive properties of the samples. As a part of this project, this device will be used to investigate the structure of the layered material and at the same time to measure the mechanical and adhesion properties of the material.

3. RESULTS AND DISCUSSION

Subsequently, another layer of non-woven fabric was covered with a dye of a lower concentration. Next, the procedure was repeated up to the layers with the lowest dye concentration (**Figure 1**).

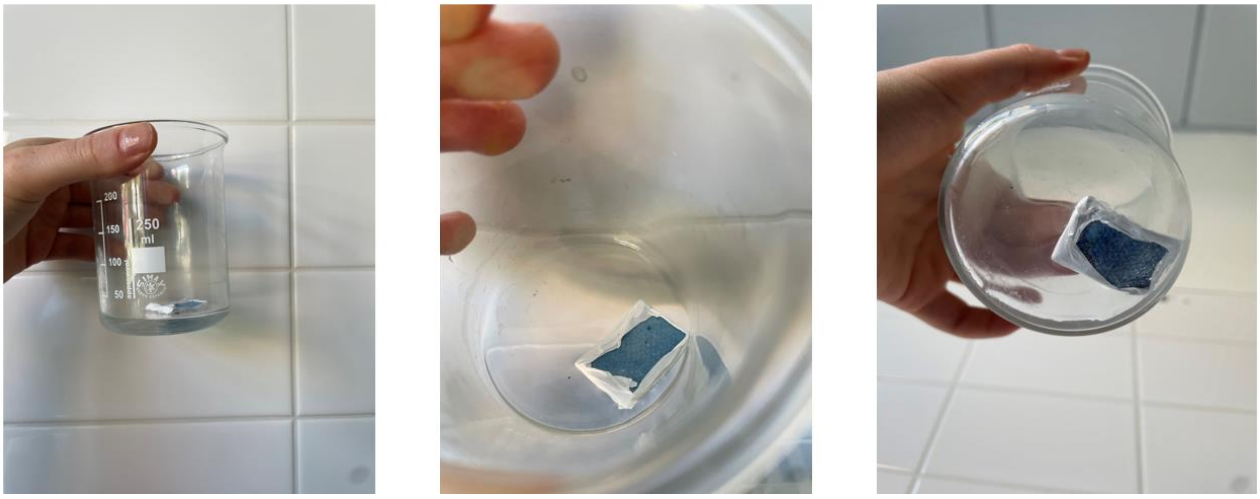

Figure 1 Layered material of a non-woven polypropylene fabric with gradient of concentration of indigotin

3.1. Release of the indigotin dye

During the next 96 hours, the release of the indigotin dye from the layered material into water environment was observed. In the first 25 minutes, a larger amount of dye as observed to be released from the lower layers of the material (**Figure 2**), this is so called burst effect.

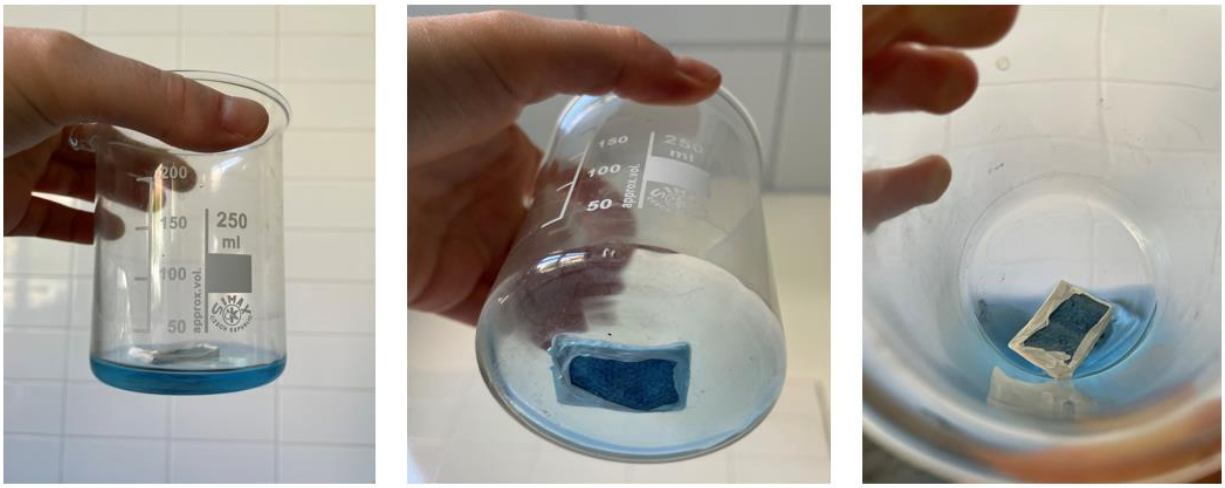

Figure 2 Released indigotin dye from layered material after 25 minutes

Subsequently, the water was replaced with fresh water and the experiment continued. Further release of the dye continued at time intervals. Release of the dye was not as concentrated a in the beginning and each subsequent release was of approximately the same concentration every time (**Figure 3**).

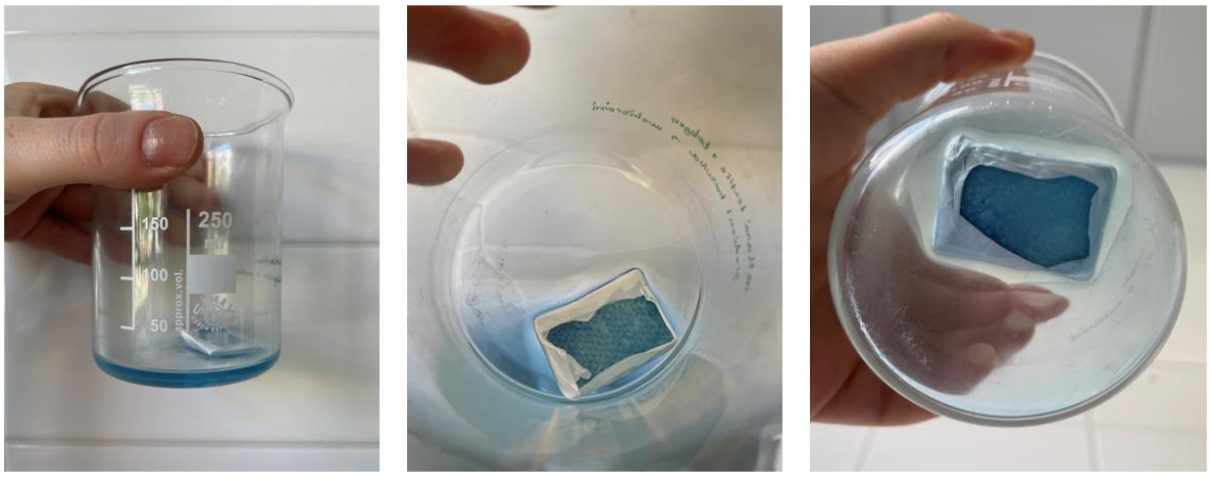

Figure 3 Released indigotin dye from layered material after 24 hours

The total dye release time from the layered non-woven fabric was 96 hours. During this process, the dye was first released into the water from the lowest layers, where the concentration was highest. After a longer period of time, water penetrated into the higher layer of the material, thereby starting the diffusion of the dye through the layers of polypropylene non-woven fabric and chitosan into the lower layers. This caused an equalization of the lower layers of the material, thereby still releasing approximately the same concentration of dye.

During the measurement of the release from the layered material with dye gradient, it was found that after a short time, a larger amount of dye is released from the material. This was caused by the release of the dye from the bottom layer that was not coated with the gelatin layer. The subsequent release was slowed down

because water had to penetrate the material first. Only then did diffusion through the material, the dye had to pass through the layers of the non-woven fabric, but the diffusion was influenced the most by the presence of the gelatin, which had much smaller pores.

3.2. UV-VIS spectrophotometry

The measurements revealed not only the diffusion coefficient (*D*) but also the concentration of methylene blue (c_m), which remained retained in the layered material (**Table 1**).

Table 1 Diffusion coefficient and concentration of methylene blue retained in layered material

number of layers	D ($m^2 \cdot s^{-1}$)	c_m ($g \cdot dm^{-3}$)
1	$6.9 \cdot 10^{-10}$	$2.6 \cdot 10^{-3}$
2	$5.9 \cdot 10^{-10}$	$1.3 \cdot 10^{-3}$
8	$9.9 \cdot 10^{-11}$	$1.2 \cdot 10^{-3}$

It was found that the diffusion coefficient decreases linearly with increasing number of layers. During the calculation of the concentration of methylene blue that remained in the layered material, it is possible to observe that only one layer of polypropylene non-woven fabric captured a larger amount of the used dye. The other two material used between the layers contained gelatin, the retention results were very similar. With the increasing number of layers, it is possible to observe that the concentration of the capture dye in the material decreases and, at the same time, the release time also increases.

4. CONCLUSION

This article deals with the solution of the problem of controlled release of active substances from the layered material of the non-woven polypropylene alternating with gelatin. So far, the release of active substances if mainly controlled by first-order kinetics, i.e. it is dependent on the concentration of the active substance from the material. There is above all a big problem in the so-called burst effect, during which a certain amount of active substance is quickly released from the material. The problem is solved here by releasing the indigotin dye from the layered material into the water environment. Furthermore, the penetration of methylene blue through the prepared materials is measured by diffusion cells and UV-VIS spectrometer. During the measurement, it was concluded that the so-called burst effect can still be observed in the material, but the gelatin in the material can slow down the release enough to last several days. Thanks to measurement on diffusion cells, it was found that the diffusion coefficient decreases with an increasing number of layers. At the same time, we can also observe that there is a absorption of the dye into the layered material. The result show that by creating a layered material with a sufficient number of individual layers, the release time of the active substance will be extended and at the same time the concentration retained in the material will be reduced. The advantage of this method will be material with the active substance that could be applied repeatedly, or two active substances at the same could be incorporated into it with gradual excretion.

ACKNOWLEDGEMENTS

This research was conducted under the project TM03000061 with financial support from the Technology Agency of the Czech Republic.

REFERENCES

[1] HUANG, Xiao a Christopher S BRAZEL. On the importance and mechanisms of burst release in matrix-controlled drug delivery systems. *Journal of Controlled Release*. 2001, vol. 73, no. 2-3, pp. 121-136. ISSN 01683659. Available from: https://doi.org/10.1016/S0168-3659(01)00248-6.

[2] KONG-YIN, Zhao, Chen TIAN, Li DONG-YING, Zhang XIN-XIN, Song HUAN-YU a Wei JUN-FU. Preparation and characterization of mesoporous calcium silicate grafted polypropylene non-woven fabric. *Materials Letters*. 2015, vol. 141, pp. 110-113. ISSN 0167577X. Available from: https://doi.org/10.1016/j.matlet.2014.11.055.

[3] BIRK, David E. a Peter BRUCKNER. Collagen Suprastructures. *Collagen*. Berlin, Heidelberg: Springer Berlin Heidelberg, 2005, 2005-4-12, pp. 185-205. Topics in Current Chemistry. ISBN 978-3-540-23272-8. Available from: https://doi.org/10.1007/b103823.

[4] GÓMEZ-GUILLÉN, M.C., B. GIMÉNEZ, M.E. LÓPEZ-CABALLERO a M.P. MONTERO. Functional and bioactive properties of collagen and gelatin from alternative sources: A review. *Food Hydrocolloids*. 2011, vol. 25, no. 8, pp. 1813-1827. ISSN 0268005X. Available from: https://doi.org/10.1016/j.foodhyd.2011.02.007.

[5] SMILEK, Jiri, Petr SEDLACEK, Marcela LASTUVKOVA, Michal KALINA a Martina KLUCAKOVA. Transport of Organic Compounds Through Porous Systems Containing Humic Acids. Bulletin of Environmental Contamination and Toxicology. [online]. 2017, vol. 98, no. 3, pp. 373-377 [Accessed 2022-09-29]. ISSN 0007-4861. Available from: https://doi.org/10.1007/s00128-016-1926-0.

[6] How AFM Works. *Park Systems* [online]. Korea: Park Systems, 2020.