

THE APPLICATION OF CONDUCTIVITY MEASUREMENTS FOR PRELIMINARY ASSESSMENTS OF CHLORHEXIDINE AND LIDOCAINE HYDROCHLORIDE RELEASE FROM METHYLCELLULOSE GEL AT VARIOUS TEMPERATURES

Witold Musiał^{1,2)}, Vanja Kokol¹⁾, Bojana Voncina¹⁾

¹⁾Department for Textile Materials and Design

University of Maribor, Slovenia

^{1,2)}Chair and Department of Pharmaceutical Technology

Wroclaw Medical University, Poland

Summary

For the evaluation of conductivity measurements in the control and monitoring of release process, high number of conductivity measurements was performed. The measurements were done for the compositions of chlorhexidine with methylcellulose, and lidocaine hydrochloride with methylcellulose. Chlorhexidine, a very slightly soluble substance is released from the methylcellulose bead in the amounts ca. 30%-70%, and it depends of temperature of the release process. The lidocaine hydrochloride at the same time is released from methylcellulose formulation in 70-100%.

The conductivity in the donor compartment at the start point, and in the acceptor compartment at the termination point, reflect the released amounts of the drug. This study confirms the possibility of application of conductivity measurements for the preliminary assessments of the kinetics of release of soluble and practically insoluble substances from the nonionic polymeric matrix.

Key words: conductivity, release, methylcellulose, lidocaine hydrochloride, chlorhexidine

INTRODUCTION

New drug forms are developed with different kinds of polymers. The polymers may bind active substances and prolong the release in the place of application, i.e. on the skin or on the mucus. The polymers are swelling in aqueous environment and the viscosity of resulting systems is high. Since several years there is an increase observed in the research on new gels and microgels, characterized as thermo-sensitive factors. When an increase or decrease in environmental temperature around specified critical value is performed the conformation of a macromolecule is changing, and also the changes of the release are observed, gradual or rapid.

Application of this kind of polymers may be used i.a. in a delay of drug release, or for drug-targeting to specified tissues. This should also diminish the possibility of eventual adverse drug reactions. Additionally in the release kinetics assessments the temperature has to be controlled in a special manner, when the thermo-sensitive polymers are applied.

Between most used macromolecules, applied in research, there are derivatives of N-isopropylacrylamide [1]. They were studied, i.a., as factors enabling the drug targeting of anticancers and products of advanced biotechnology [2]. In this contexts, the analysis of thermal properties of known and widely applied polymer - methylcellulose seems to be interesting, as actually also thermosensitivity of methylcellulose was evidenced in the ophthalmic drugs technology [3] (Fig. 1).

The measurements of drug release kinetics require complicated methods, and developed research apparatus. However, the amount of the drug, which is moved from donor compartment to the acceptor compartment may be reflected by the conductivity of ions resulting from the dissociation of the active substance molecule [4].

The important factor, which influences the possibility of conductivity measurement is the solubility of the analyzed substances. An example of a very low soluble substance is chlorhexidine base, whereas the lidocaine hydrochloride has

much higher solubility. The additional factors are the viscosity and molar weight of the active compound [5].

For the assessments were chosen substances with high application potential and opposite solubility in water. Chlorhexidine is a known antiseptic, and since many years is applied in stomatology and dermatology. Numerous attempts to apply it in various drug formulations, confirm its high value in infectious conditions of the oral cave, and in the prophylaxis and medication of teeth caries. Very low water solubility, which is ca. 0,008% [6] is characteristic for this compound.

The lidocaine hydrochloride has much higher values of water solubility - ca. 68% [7]. It is used in local anesthesia of the skin and mucous membranes, in various diseases and during surgical interventions (Table 1).

For the evaluation of conductivity measurements in the control and monitoring of release process, high number of conductivity measurements was performed. The measurements were done for the compositions of chlorhexidine with methylcellulose, and lidocaine hydrochloride with methylcellulose.

MATERIAL AND METHODS

Applied polymer and compositions of the formulations

Methylcellulose is a polymer widely applied in the pharmaceutical technology, and this polymer is very good characterized [8]. The methylcellulose was used to prepare formulations with lidocaine hydrochloride and chlorhexidine. The concentration of lidocaine hydrochloride and chlorhexidine was 0,125%, and equal quantities of methylcellulose were given to the formulations - according to the attached Table 2.

The release of the active substance

The release of the active substances was performed according to the European Pharmacopoeia, however the temperature was modified, as one of the topics of the study was to evaluate the influence of temperature on the drug release [9]. In this study temperatures of 22°C, 32°C, oraz 42°C were maintained. The first temperature

parameter was introduced to the study, as the shelf-conditions are specified in this temperature. The second temperature parameter is the standardized skin surface temperature, important for the patient, when applying the drug onto the skin. Both chlorhexidine and lidocaine hydrochloride are applied in this conditions. The 42°C temperature is a limit value - highest for the patho-physiological conditions [10].

The donor compartment, of 25cm³ volume, was prepared from the dialysis membrane, closed by lab clips, without air inside. In the acceptor compartment the deionized water was used, with conductivity not higher than 10 µS/cm. The release vessels were tightly closed and the air was removed from the device, with the pressurized nitrogen.

In the release the cumulated amounts of active substance after 6 h were evaluated - the topical dose interval was equal to four application daily of the semisolid drug on the skin. The active substances were assessed by the UV spectrophotometric method - TECAN Infinite 2000. The percentage extinction coefficient in 0.01 mol/L HCl solution was 225.11 by the $p > 0.999$, at 250 nm for chlorhexidine. For the lidocaine the percentage extinction coefficient was 207.35 ($p > 0.999$), at 254 nm. The method gave linear effects from concentration of 1 to 18 µg/mL.

The conductivity assessments

The conductivity was assessed in donor compartment and acceptor compartment before release start, and after 6 h, at specified before temperatures. Also the conductivity of pure solutions of lidocaine hydrochloride and chlorhexidine was assessed in concentrations applied in the donor and acceptor compartment. On the Scheme the measurements rules were presented. The Seven Multi Metler Toledo, and conductivity segment TDS/SAL/resistivity connected to conductivity sensor InLab 730, NTC, 0,001-1000 mS/cm was used - the temperature range -5 °C – 100 °C.

RESULTS

Release rates

The cumulated amounts of lidocaine hydrochloride and chlorhexidine in acceptor compartment, after specified period, were various, as it is presented on the respective graphs on the Fig. 2 and 3. Between this systems the higher amounts of released drug were identified in the case of lidocaine hydrochloride - the cumulated amount after 6 h was 99,12; 70,68; 69,64%, for the temperatures 22, 32 and 45°C respectively. For the comparison the cumulated amounts of chlorhexidine released from the donor compartment were 28,99; 29,04 and 63,87% respectively.

CONDUCTIVITY

As it is presented in the tables 3 and 4, the conductivity of methylcellulose preparations was between 127,88 $\mu\text{S}/\text{cm}$ and 3019,61 $\mu\text{S}/\text{cm}$. The parts in the system conductivity had the active compound, the macromolecule, the water. With the release process, the content of chlorhexidine or lidocaine hydrochloride decreased in the donor compartment, and increased in the acceptor compartment. Also the respective conductivity decreased and increased in the same manner. After 24 h the conductivity in the donor compartment was between 54,53 and 110,75 $\mu\text{S}/\text{cm}$, depending of the active substance and temperature of the release process. The conductivity of chlorhexidine system containing the methylcellulose at the beginning of the process was 127,88 $\mu\text{S}/\text{cm}$, and after the release process termination, the value was ca. 54,53 $\mu\text{S}/\text{cm}$, at 22 °C. In the acceptor compartment, after finishing the process, the conductivity was 32,11 and 65,12 $\mu\text{S}/\text{cm}$ at 22 °C. Respective values of the conductivity in higher temperatures - 32 °C i 42 °C in donor compartment are presented in the table 3. The values of conductivity for the acceptor compartment are gathered in the table 4.

DISCUSSION

According to the achieved data, the release process is faster in the case of lidocaine hydrochloride preparations, comparing to the chlorhexidine, even if the temperature is the same. As it was observed for the preparations with chlorhexidine, with the increase of temperature, between 22 and 32 °C the increase in the released amount was rather slight, whereas when the temperature raised for the next 10°C, the increase in released amount was significant - Fig. 2. The topic of drug binding, and types of the respective bonds in the course of the interaction with cellulose is extensively studied [11].

The increasing temperature influences the amounts of solute chlorhexidine, thus the temperature increase accelerates the dissolution process. As it is presented on the Fig. 4, the preparation of chlorhexidine with methylcellulose is a suspension, and in the preparation of lidocaine hydrochloride with methylcellulose the active substance is in molecular dispersion. From our data we can conclude, that the increase to 42°C influences significantly the solubility of chlorhexidine in the methylcellulose medium.

In different way proceeded the kinetics of lidocaine hydrochloride from the methylcellulose bead. The assessed in the acceptor compartment amount of lidocaine hydrochloride was highest at 22°C, and increase of the temperature resulted in decrease of lidocaine hydrochloride observed in the acceptor compartment. However the increase from 32 °C to 42 °C did not influence significantly the process - graph on Fig. 3.

In the assessments of release kinetics some approach was made to the conductivity assays, considering the release of microbial organophosphorous compounds [12]. In this study the correlation in two aspects was performed. The amount of released active substance was compared to the conductivity in the donor compartment before the release assessments. Also the relation between the released amount and the conductivity in acceptor compartment after release termination was studied. Depending of the temperature there are significant correlation in the temperatures 22 °C and 32 °C, whereas at 42 °C the relation was rather weak - Fig. 5.

According to general rules, the amount of the active substance in the donor compartment is decreasing and in the acceptor compartment is increasing. Thus also

the conductivity should change the values in proportional way. Actually in the case of chlorhexidine the concentrations and conductivity were lower, comparing to that of lidocaine hydrochloride - Fig. 6.

To perform more detailed study of the changes in the drug release from methylcellulose bead, the actual values of conductivity in the acceptor compartment (A2a) were compared to that calculated according to the dissolution of amounts released from the donor compartment (A2t) (Tab. 5).

As it is presented in the table, the ratio of the value of conductivity measured in the acceptor compartment and the calculated value, was near the 1,0. The minimal increase beyond the 1,0 may be explained by the dissolution of the molecule, according to the Ostwald rule. However for the preparation of chlorhexidine the ratio A2a/A2t is between 19,05 and 37,98 $\mu\text{S}/\text{cm}$. As the chlorhexidine is characterized by very low solubility, the conductivity measured in the donor compartment reflects this amount of chlorhexidine, which is in equilibrium with the sediment.

Thus the theoretical, calculated value of the conductivity in the acceptor compartment, evaluated from the initial conductivity in the donor compartment is much lower, than the actual conductivity values taken from the measurements.

As an additional parameter the ratio D1/D2 in % was applied. It enables the comparison of the donor compartments at the initial stage (D1) and after termination of the release (D2). The D1/D2% factor had values 41%-73% for the preparation of chlorhexidine with methylcellulose. The lidocaine hydrochloride-methylcellulose preparations were characterized by much lower values of this factor - around 3%.

CONCLUSIONS

We can conclude from this assessments that chlorhexidine, a very slightly soluble substance is released from the methylcellulose bead in the amounts ca. 30%-70%, and it depends of temperature of the release process. The lidocaine hydrochloride at the same time is released from methylcellulose formulation in 70-100%.

The conductivity in the donor compartment at the start point, and in the acceptor compartment at the termination point, reflect the released amounts of the drug.

This study confirms the possibility of application of conductivity measurements for the preliminary assessments of the kinetics of release of soluble and practically insoluble substances from the nonionic polymeric matrix.

The limitations of this method are connected to the proper characterization of the solubility of analyzed compound, and evaluation of molar mass of this compound in the interpretation.

The method will be further developed, to determine the optimal study conditions. This should assure good additional characteristics of polymers during the release research.

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Address of the authors

¹Department for Textile Materials and Design

University of Maribor

Smetanova Str. 17, 2000 Maribor, Slovenia

Tel: +386 2 220-7500; Fax: +386 2 220-7990

e-mail: bvoncina@uni-mb.sl

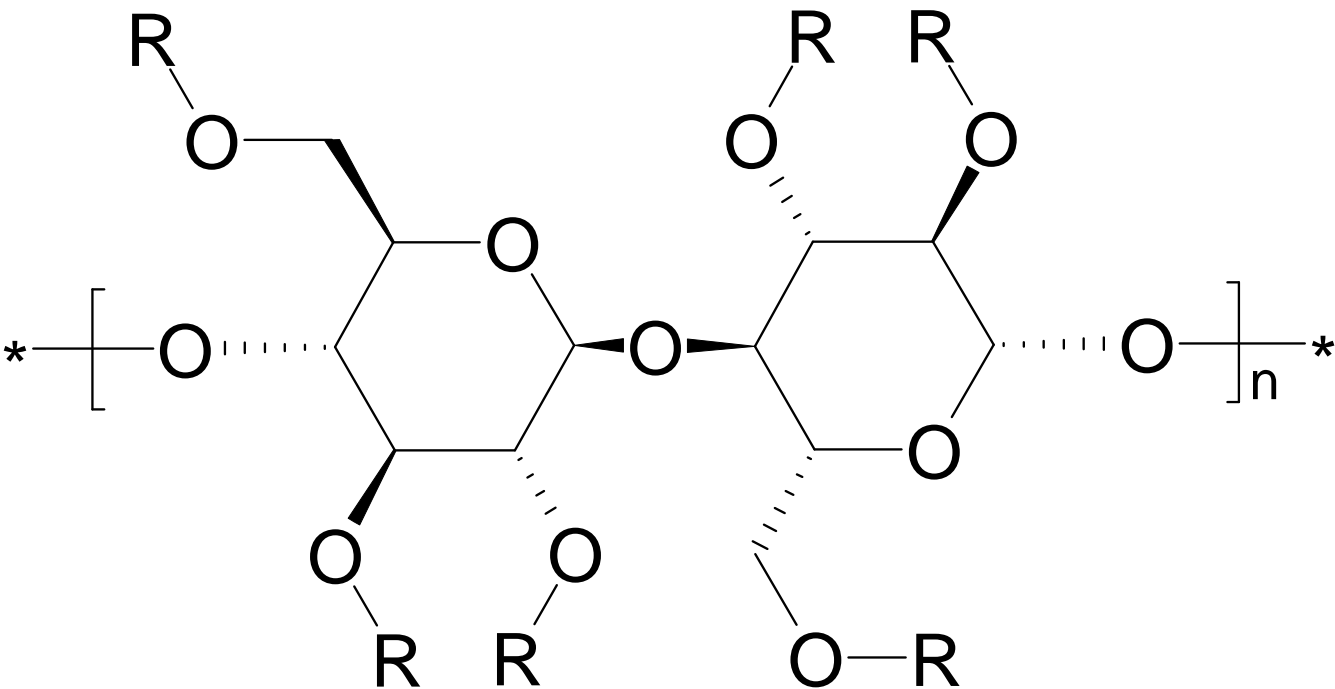
^{1,2} Katedra i Zakład Technologii Postaci Leku

Akademia Medyczna we Wrocławiu

ul. Szewska 38, 50-139 Wrocław, Poland

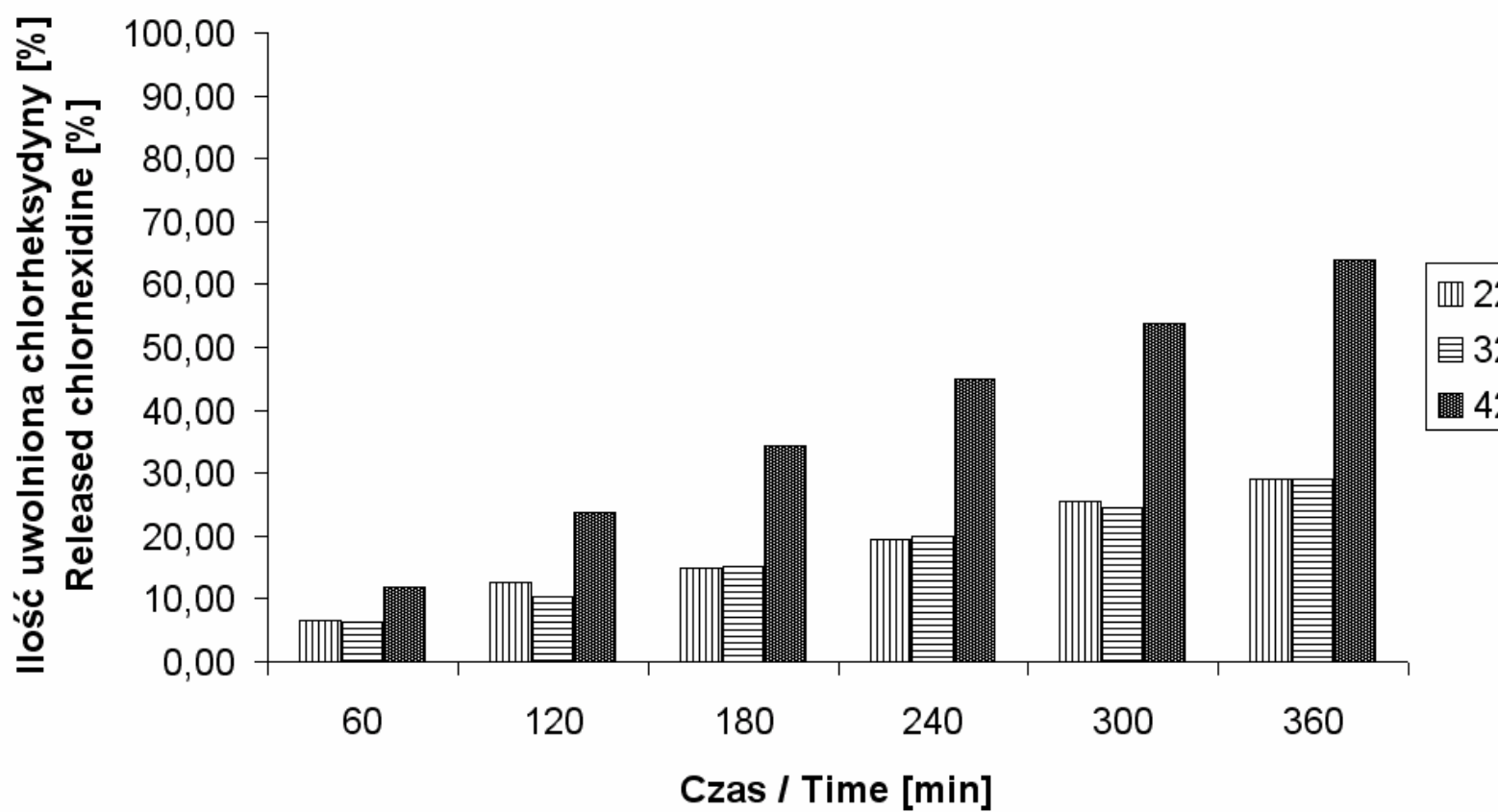
Tel: +48 71 784-03-15; Fax: +48 71 784-03-17

e-mail: witold@ktpl.am.wroc.pl



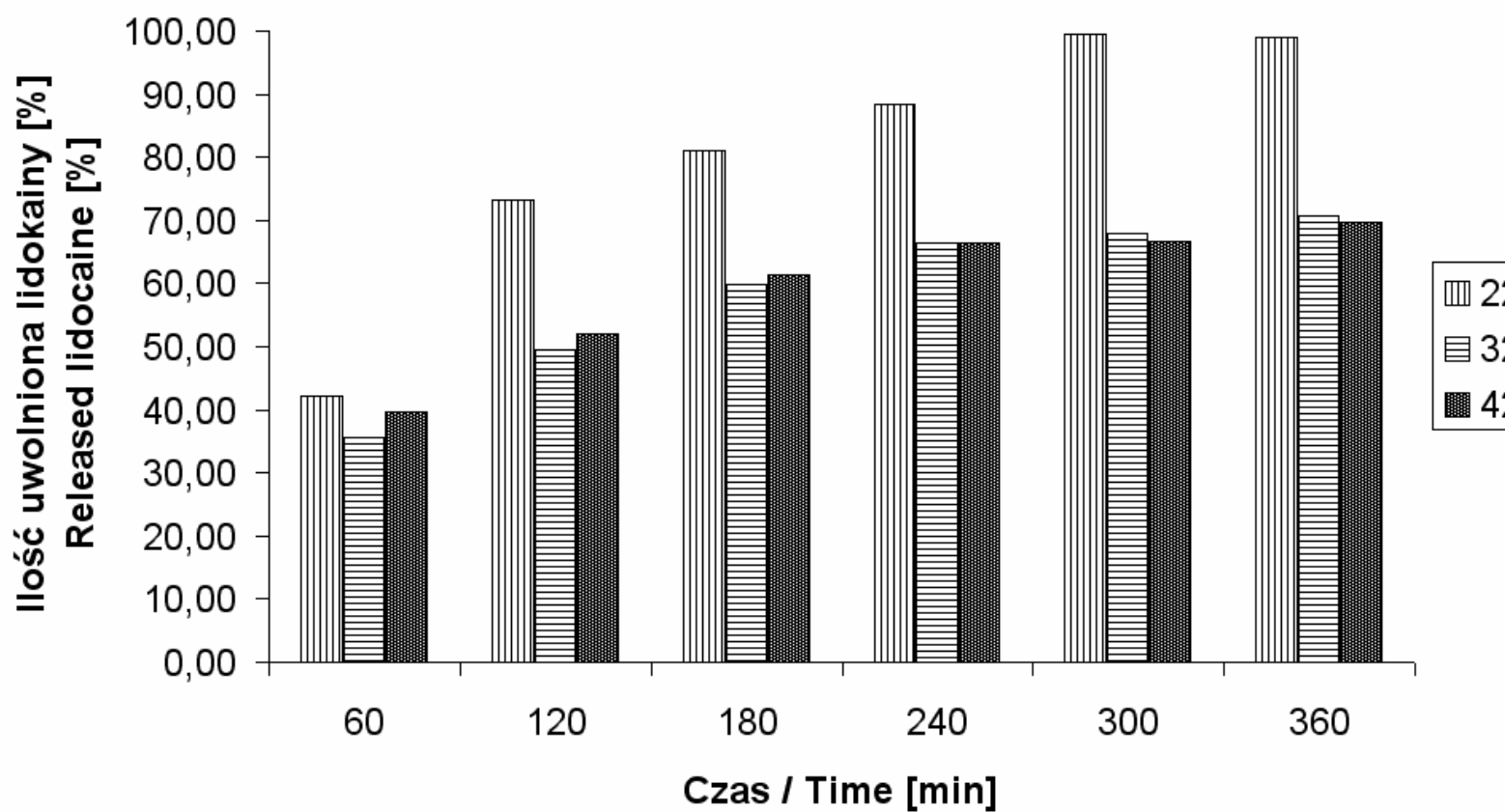
Ryc. 1. Struktura metylocelulozy, R=CH₃ lub R=H, stosunek CH₃/H około 60%

Fig. 1. The structure of methylcellulose, R=CH₃ or R=H, the ratio CH₃/H is in the range of 60%



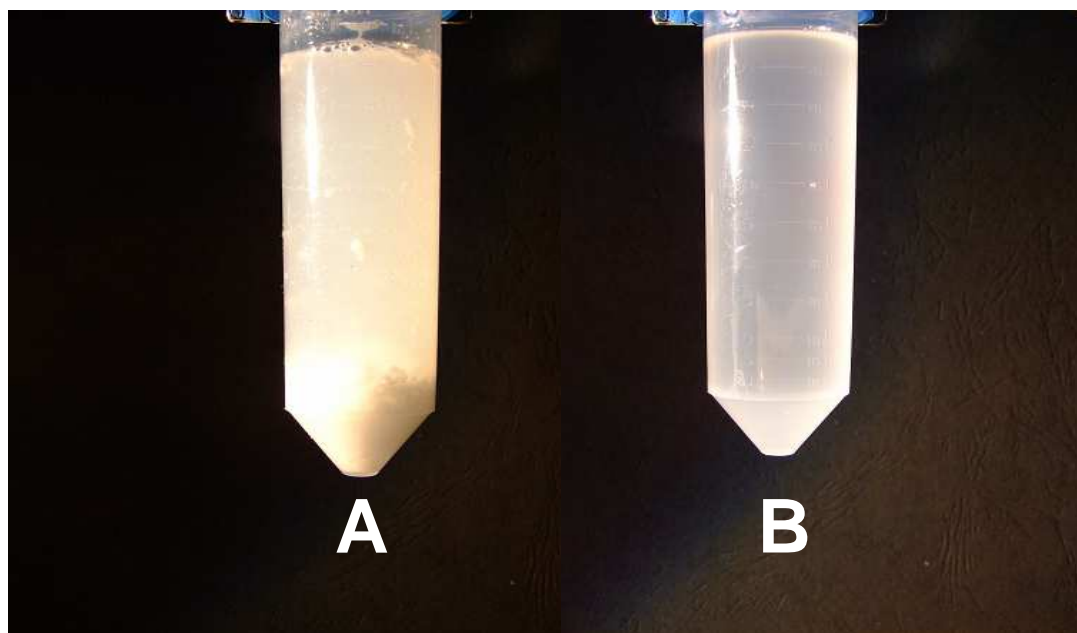
Ryc. 2. Stężenie uwolnionej chlorheksydyny po 1, 2, 3, 4, 5 i 6 godzinach procesu

Fig. 2. The percentage of released chlorhexidine after 1, 2, 3, 4, 5, and 6 hours]



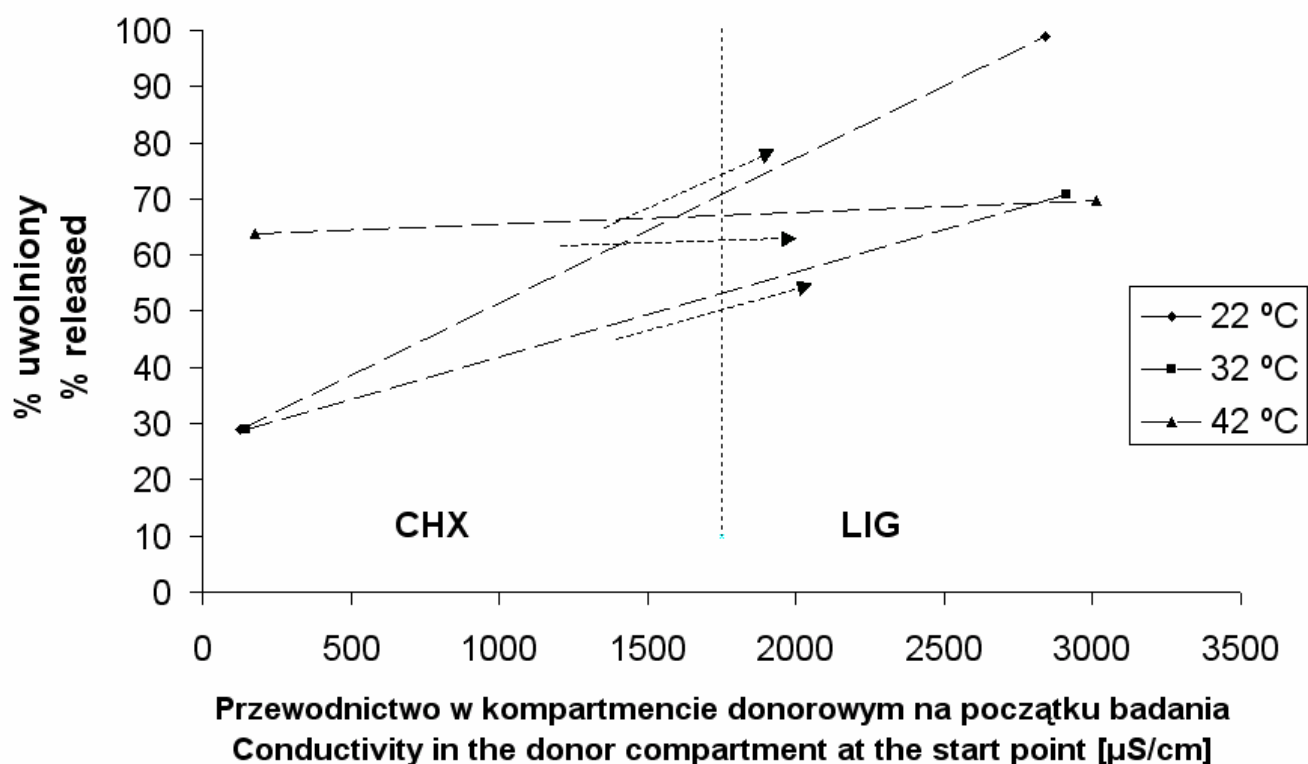
Ryc. 3. Stężenie uwolnionego chlorowodorku lidokainy po 1, 2, 3, 4, 5 i 6 godzinach procesu

Fig. 3. The percentage of released lidocaine hydrochloride after 1, 2, 3, 4, 5, and 6 hours



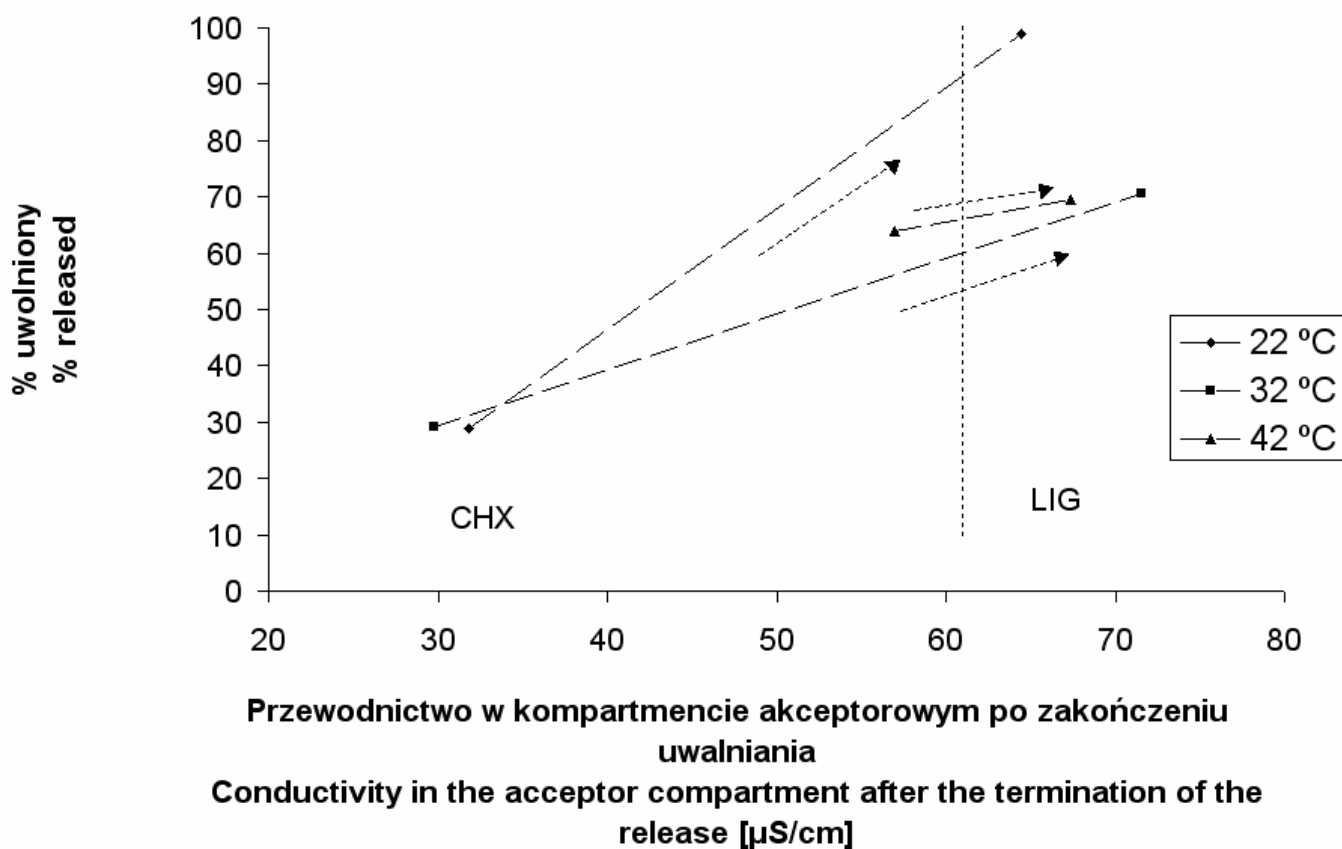
Ryc. 4. Makrofotografie badanych próbek: połączenie metylocelulozy i chlorheksydyny (A) oraz połączenie metylocelulozy i lidokainy (B)

Fig. 4. Visualized formulations of chlorhexidine (A) and lidocaine (B) with methylcellulose



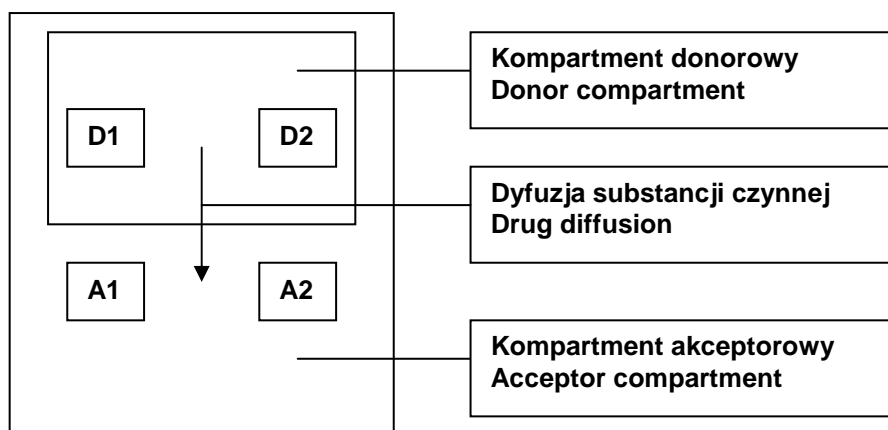
Ryc. 5. Wpływ temperatury na wartość przewodnictwa badanych próbek, jako czynnika predyktywnego we wstępnej ocenie kinetyki uwalniania substancji czynnej z podłoża metylocelulozowego. Punkty po lewej stronie wykresu odpowiadają preparatom chlorheksydy (CHX), po prawej - lidokainy (LID).

Fig. 5. The influence of temperature on the conductivity in donor compartment, correlated with temperature, and released percentage of chlorhexidine (left part of the graph, CHX) and lidocaine (right part of the graph, LID)



Ryc. 6. Wpływ temperatury na wartość przewodnictwa w kompartmencie akceptorowym, jako czynnika predyktywnego we wstępnej ocenie kinetyki uwalniania substancji czynnej z podłoża metylocelulozowego. Punkty po lewej stronie wykresu odpowiadają preparatom chlorheksydyny (CHX), po prawej - lidokainy (LID).

Fig. 6. The influence of temperature on the conductivity in acceptor compartment, correlated with temperature, and released percentage of chlorhexidine (left part of the graph, CHX) and lidocaine (right part of the graph, LID)



Schemat. Sposób pomiaru przewodnictwa w kompartmentach donorowym (D) i akceptorowym (A). Cyfra "1" oznacza stan na początku badania, a cyfra "2" stan po zakończeniu badania

Scheme. System of evaluated donor (D) and acceptor (A) compartments. The "1" denominates the beginning of the process, whereas the "2" denominates the end of the process

Tabela 1. Porównanie rozpuszczalności chlorheksydyny i chlorowodorku lidokainy

Table 1. The comparison of solubility of lidocaine hydrochloride and chlorhexidine

Substancja Active compound	Rozpuszczalność w wodzie Solubility in water	Bibliografia Reference
Chlorheksydyna, zasada Chlorhexidine, base	ca. 0,008 %	[6]
Lidokaina, chlorowodorek Lidocaine, hydrochloride	ca. 68 %	[7]

Tabela 2. Skład badanych systemów

Table 2. The composition of assessed formulations

Formulacja Formulation	MC	LID	CHX	Woda Water
MC-LID	0,125	0,125	-	25,750
MC-CHX	0,125	-	0,125	25,750

MC - metyloceluloza, methylcellulose, LID - chlorowodorek lidokainy, lidocaine hydrochloride, CHX - chlorheksydyna, chlorhexidine, MC-LIG, MC-CHX - odpowiednie formułacje, respective formulations

Tabela 3. Temperatura i przewodnictwo w kompartmentcie donorowym

Table 3. Temperature and conductivity in donor compartment

Typ preparatu Formulation	Temperatura Temperature [°C]	Przewodnictwo - początek procesu Conductivity - start point [μS/cm]	SD	Przewodnictwo - koniec procesu Conductivity - finish [μS/cm]	SD
MC-CHX	22	127,88	0,18	54,53	0,55
	32	144,15	0,43	107,72	1,40
	42	176,19	0,48	110,75	0,67
MC-LIG	22	2852,33	3,71	85,21	0,32
	32	2922,98	12,31	91,43	0,21
	42	3019,61	38,19	92,82	0,19

SD-standard deviation

Tabela 4. Temperatura i przewodnictwo w kompartmentie akceptorowym

Table 4. Temperature and conductivity in acceptor compartment

Typ preparatu Formulation	Temperatura Temperature [°C]	Przewodnictwo po zakończeniu procesu Conductivity after finish [μ S/cm]	SD
MC-CHX	22	32,11	0,38
	32	28,92	1,30
	42	57,99	1,41
MC-LIG	22	65,12	0,33
	32	70,98	0,91
	42	67,45	1,54

Tabela 5. Uzupełniający parametr oceny badanych formułacji - stosunek A2a/A2t.

Szczegóły w tekście

Table 5. The additional parameter - A2a/A2t ratio - details in the text

Typ preparatu Formulation	Temperatura Temperature [°C]	A2a/A2t	SD	D2/D1	SD
MC-CHX	22	20	0,21	42	0,51
	32	34	2,43	73	1,15
	42	38	1,18	62	0,50
MC-LIG	22	1	0,01	3	0,01
	32	1	0,01	3	0,01
	42	1	0,02	3	0,04

SD - standard deviation