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Antibacterial effect and tetracycline release of poly(Ɛ-caprolactone) matrices obtained by iodine polymerization

Efeito antibacteriano e liberação de tetraciclina de matrizes de poli(Ɛ-caprolactona) obtidas por polimerização de iodo

Efecto antibacteriano y liberación de tetraciclinas de matrices de poli(Ɛ-caprolactona) obtenidas por polimerización con yodo

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Lidiane Gomes da Silva
ORCID: https://orcid.org/0000-0003-0424-4275
Centro Universitário de Itajubá, Brasil
E-mail: g.lidiane@yahoo.com.br
Amanda de Carvalho Pereira Moraes
ORCID: https://orcid.org/0000-0002-5661-9174
Faculdade de Medicina de Itajubá, Brasil
E-mail: amandinhafar@yahoo.com.br
Patricia Capellato
ORCID: https://orcid.org/0000-0002-6397-5820
Universidade Federal de Itajubá, Brasil
E-mail: pat_capellato@yahoo.com.br
Gilza Carla Ribeiro
ORCID: https//orcid.org/0000-0002-1724-893X
Universidade Federal de Itajubá, Brasil
E-mail: gilza.carla@zipmail.com.br
Ana Angélica Martins Costa
ORCID: https//orcid.org/0000-0002-7771-8091
Universidade Federal de Itajubá, Brasil
E-mail: anamartins_0410@hotmail.com
Álvaro Antônio Alencar de Queiroz
ORCID: https://orcid.org/0000-0002-1769-7022
Universidade Federal de Itajubá, Brasil
E-mail: alencar@unifei.edu.br
Daniela Sachs
ORCID: https://orcid.org/0000-0002-3767-2455
Universidade Federal de Itajubá, Brasil
E-mail:danisachs@unifei.edu.br

Abstract
The development of devices to controlled the release of drugs are in constant technological innovation. The aim is to improve the release of drugs on target areas. Poly (ε-caprolactone) (PCL) has been widely investigated because of degradation rate, biocompatibility, availability, no toxicity, cost and good adhesion to a large number of drugs. Thus, in the present study was associated polymer PCL with antibiotics tetracycline as local delivery system. PCL was obtained by ring-opening polymerization of monomer Ɛ-caprolactone (Ɛ-CL). The samples were characterized by fourier transformed infrared (FTIR), differential scanning calorimetric (DSC), thermogravimetric analyses (TGA) and X-ray diffraction analysis (X-rays). Likewise, was investigated antimicrobial activity against gram-positive bacteria (S. aureus) and gram-negative bacteria (E. coli, P. mirabilis, P. aeruginosa and K. pneumoniae). According to the results, the antibiotics tetracycline has been successfully incorporated to PCL matrices. They release tetracycline in the ideal rates and shows antibacterial activity. So, this material has a potential to been used in implants for drug release.
Keywords: Poly (ε-caprolactone); Bioresorbable polymer; Drug delivery; Antimicrobial activity.

Resumo
O desenvolvimento de dispositivos para controle da liberação de medicamentos está em constante inovação tecnológica. O objetivo é melhorar a liberação de medicamentos nas áreas-alvo. A poli(ε-caprolactona) (PCL) tem sido amplamente investigada devido à taxa de degradação, biocompatibilidade, disponibilidade, ausência de toxicidade, custo e boa adesão a um grande número de fármacos. Assim, no presente estudo foi associado o polímero PCL com o antibiótico tetraciclina como sistema de liberação local. PCL foi obtido por polimerização por abertura de

anel do monômero Ɛ-caprolactona (Ɛ-CL). As amostras foram caracterizadas por infravermelho transformado de Fourier (FTIR), calorimetria de varredura diferencial (DSC), análise termogravimétrica (TGA) e análise de difração de raios-X (raios-X). Da mesma forma, foi investigada a atividade antimicrobiana contra bactérias gram-positivas (S. aureus) e bactérias gram-negativas (E. coli, P. mirabilis, P. aeruginosa e K. pneumoniae). De acordo com os resultados, o antibiótico tetraciclina foi incorporado com sucesso às matrizes PCL. Liberam tetraciclina nas taxas ideais e apresentam atividade antibacteriana. Assim, este material tem potencial para ser utilizado em implantes para liberação de fármacos.
Palavras-chave: Poli(ε-caprolactona); Polímero bioabsorvível; Drug delivery; Atividade antimicroba.

Resumen
El desarrollo de dispositivos para la liberación controlada de fármacos está en constante innovación tecnológica. El objetivo es mejorar la liberación de fármacos en las áreas objetivo. La poli(ε-caprolactona) (PCL) ha sido ampliamente investigada debido a su tasa de degradación, biocompatibilidad, disponibilidad, ausencia de toxicidad, costo y buena adhesión a una gran cantidad de fármacos. Por lo tanto, en el presente estudio se asoció el polímero PCL con los antibióticos tetraciclina como sistema de administración local. La PCL se obtuvo mediante polimerización con apertura de anillo del monómero Ɛ-caprolactona (Ɛ-CL). Las muestras se caracterizaron por infrarrojo transformado de Fourier (FTIR), análisis calorimétrico diferencial de barrido (DSC), análisis termogravimétrico (TGA) y análisis de difracción de rayos X (rayos X). Asimismo, se investigó la actividad antimicrobiana frente a bacterias grampositivas (S. aureus) y gramnegativas (E. coli, P. mirabilis, P. aeruginosa y K. pneumoniae). Según los resultados, el antibiótico tetraciclina se ha incorporado con éxito a las matrices de PCL. Liberan tetraciclina en las tasas ideales y muestran actividad antibacteriana. Por lo tanto, este material tiene potencial para ser utilizado en implantes para la liberación de fármacos.
Palabras clave: Poli (ε-caprolactona); Polímero bioabsorbible; Administración de fármacos; Actividad antimicrobiana.

1. Introduction

Nowadays, there are major health problem associated with pathogenic bacteria infection. This leads to a wide range of problems including the high morbidity and mortality rates of people and the high cost to the public health system on patient management. Although there are several options antimicrobial therapy researches still search for a better way to eliminate the infection(Thomas et al., 2008; Yeh et al., 2020) .

Despite comprehensive drug efficiency advances, the administration faces a several collateral effects on oral administration. So, studies have been looking to development of controlled release of drugs to may enhance the medicine's effectiveness because they can be just on target organ (Wsoo et al., 2020).

The development of devices to controlled the release are in constant technological innovation. The aim is to improve the release of drugs, becoming increasingly specialized. Likewise, with focus on achieving the targets where the product really needs to act without damaging other areas of the human body. Studies in controlled drug delivery using the polymer matrix as the PCL has been widely developed due to its long degradation and its good adhesion to a large number of drugs (R. P. Verma, 2020).

In the last decades polymer has quicker the interest on biomedical field. Among many polymers the Poli(ε-caprolactone) (PCL) present some properties that make it vantage to been used like biomaterial. PCL is biocompatible with human body and presents no toxicity. The chemical mechanical and biological properties are attractive and the price is relatively inexpensive (Ezhilarasu et al., 2019; Fereshteh et al., 2015; Souza et al., 2018). Degradation time rate of PCL is quite long, thus it is used mainly in the replacement of hard tissues in the body and has a good adherence with a wide drug making PCL a good polymer for works in drug delivery (de Arruda Almeida et al., 2004; Pathak et al., 2018; Rezk et al., 2019).

Other important fact is that PCL is approved for human uses for principal regimentation world agencies including FDA (Food and Drug Administration) and EMEA (European Medicines Agency) (Zupančič et al., 2018). PCL can be used as films, fiber, sponge and capsule, making possible many order biomedical applications like treatment of wounds, in orthopedic and odontology fields (screws and pins) and widely used in tissue engineering field (Cui et al., 2010; Kuznetsov et al., 2018;

Lü et al., 2012). In addition, PCL has compatibility with a wide range of drugs which provides homogeneous distribution of lipophilic drugs in the carrier matrix due to its hydrophobic nature (Malikmammadov et al., 2018; Nagiah et al., 2020).

Manoukian (2017) observed the drug release was sustained up to 120 days and Kim, 2016 developed a PCL implant that release drugs for over 6 months. It shows the possibility of polymeric matrices application on dressing, tissue engineering and drug administration (J. Kim et al., 2016; Manoukian et al., 2018).

The tetracycline (Tr) is a family of broad-spectrum antibiotics widely used against Gram-positive and Gram-negative bacteria, and can avoid many of the common antibiotic-resistance mechanisms that inactivate other tetracyclines. Besides that, antimicrobial activity, they block bone resorption, present tolerability and clinical safety. Nowadays, its antibiotics have been used on drug delivery system in combination with PCl due to good adherence (Grossen et al., 2017; Iman et al., 2020; Kaur & Singh, 2019; Y. Kim et al., 2019; Xie et al., 2018).

In this study was investigated the polymer poly(ε-caprolactone) (PCL) to produce matrices for drug delivering system. The ring-opening polymerization of monomer Ɛ-caprolactone (Ɛ-CL) was promoted by iodine to render poly(Ɛ-CL) to obtained the PCL synthesized. The reaction is carried out under mild conditions (lower temperature) as compared with conventional thermal methods for ring opening of Ɛ-CL polymerization(De Queiroz et al., 2002). The antibiotics tetracycline was incorporated to PCl. The antimicrobial activity of matrices associated with tetracycline was evaluated by diffusion tests.

2. Methodology

Chemical synthesis

The syntheses of Poli (Ɛ-caprolatone, PCL) were obtained according to Queiroz (2002). To produce the matrices was mixed 1g of PCL powder and 50 mL of chloroform under constant agitation at room temperature to totally dissolution of PCL. Then, 50 mg of tetracycline was added to the system keeping under constant agitation for 120 seconds. The solution stayed at a dryer for 48 hours resulted in PCL associated to tetracycline (PCL-TCs). In order to obtained final shape of matrices PCL-TCs stainless steel 316 NL tablet was pressed under 2,5 ton for 2 minutes to making matrices with 1 mm of thick and 10 mm of diameter. The material was divided into 2 groups: Group 1- PCL and group 2- PCL-TCs.

Physical Chemestry characterization

The characterized was performed by fourier transformed infrared (FTIR, Perking Elmer, Spectrum 100), differential scanning calorimetric (DSC, Shimadzu DSC-60), thermogravimetric analyses (TGA, Mettler TG 50) and X-ray diffraction (DRX-diffraction Panalytical, modelo X'Pert PRO).

Tumescence test

The tumescence test was performed on three matrices at the same weight, thick and diameter. Each matrices were dried on vacuum for 48 hours at 30°C. After dried were deposited in a 1:1 PBS solution for 24 hours at 37°C, and measured the weight of matrices to investigate the water absorption.

Tetracycline (Tr) delivery

The analysis of transport mechanism to control the release of tetracycline of polymeric matrices are the key for development of drug delivering systems. Thus, the in-vitro release of tetracycline of polymeric matrices was evaluated. The matrices (discs with diameter 10 mm and 1 mm of thickness) were immersed in falcon tubes with 10 mL of PBS at pH 7.4 at 37 °C in the dark. On the determinate times was withdrawal 1 mL in concentrations 1:5. In order to quantify the amount of tetracycline delivery was measured UV absorbance (276 nm) in spectroscopy of UV- visible (Varian, model Cary 50). The

weight of tetracycline was measured using calibration equation 1, for that it was diluted tetracycline solution for concentrations ranging 1,5 and 24 mg/mL at PBS. The experiments were triplicated performed.

$$Cn=(Abn+0,0099)/32,2179;\ n\geq 55 \quad (1)$$

The results were analyzed like cumulative percentage of liberated tetracycline in function of time. As it was removed aliquots of solution each time, it was necessary correct the obtained weight, obtained using equation 2.

$$Fn=Cn(1+0,1n);\ n\geq 55 \quad (2)$$

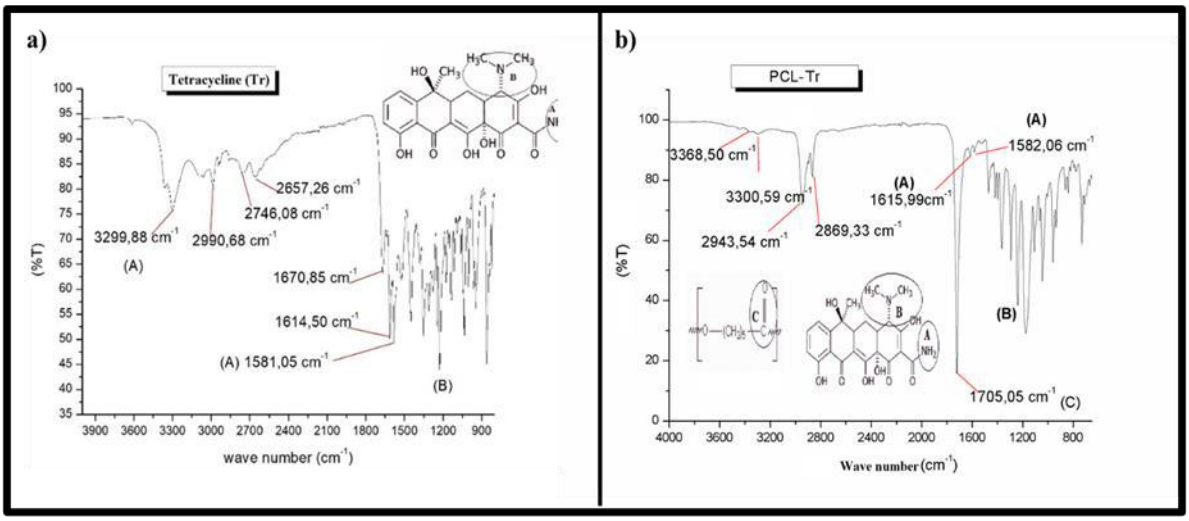
Antimicrobial activity

The disk diffusion tests were used to evaluate the antimicrobial potential of the matrices according to previous studies (Patricia Capellato et al., 2020). Briefly, the zone inhibition of bacterial growth indicates the magnitude of antimicrobial activity. Five reference strains [American Type Culture Collection (ATCC)] Staphylococcus aureus (ATCC 6538), Escherichia coli (ATCC 8739), Klebsiella pneumoniae (ATCC 13883), Proteus mirabilis (ATCC 25933) and Pseudomonas aeruginosa (ATCC 9027) were used in this study. Bacterial strains were removed from preserved culture plates and suspended in sterile saline solution (0.9% sodium chloride, NaCl). Standard bacterial suspension was standardized on 108 cells mL-1 using the MacFarland 0.5 scale and seeded on the surface of the Muller-Hinton agar plates with the of sterile swabs. Matrices on group 1 -PCL and group2- PCL-TCs were placed under bacterial culture and incubated for 24 h at 37 °C. Inhibition zone formed around the matrices was measured according to CLSI (formerly NCCLS) standards for disc sensitivity diffusion tests. All tests were performed in triplicate, the diameters of the inhibition zone formed around the matrices were measured in millimeters. The data were submitted to statistical analysis of variance (ANOVA) test to verify the existence of significant differences between the antimicrobial activity of the matrices. In the case of significance, the Tukey post-test was performed. The data were expressed as mean ± standard error of the mean with n=3 and significance level adjusted for $P < 0.05$ (Patrícia Capellato, Cláudia Eliana Bruno Marino, Gilbert Silva, Lucas Victor Benjamim Vasconcelos, Rodrigo Perito Cardoso, Kayam Hamdar, 2020).

3. Results and Discussion

Figure 1a) and 1b) shows the FTIR spectrum of a) Tetracycline and b) PCL-TCs, respectively. Figure 1 b) shows the spectra absorption characteristics of Tr. The absorption band 1705,05cm-1 in Figure 1 b) correspond to the vibrations of the carbonyl group (C=O) present in spectra. The absorption band at 1615 cm-1 e 1582,06cm-1 corresponds to vibrations of the carbonyl (C=O) and N-H respective. It was observed the absorption band at 3299 cm-1 in Figure 1a) is with less intensity at PCL-TCs (Figure 1b)) in the region 3368,50cm-1 suggesting partial interaction between the Tr and PCL probably by forming hydrogen bonds. These results are similar to studies development for Leypold (2003) and Bartzatt (2001) (Bartzatt et al., 2001; Leypold et al., 2003).

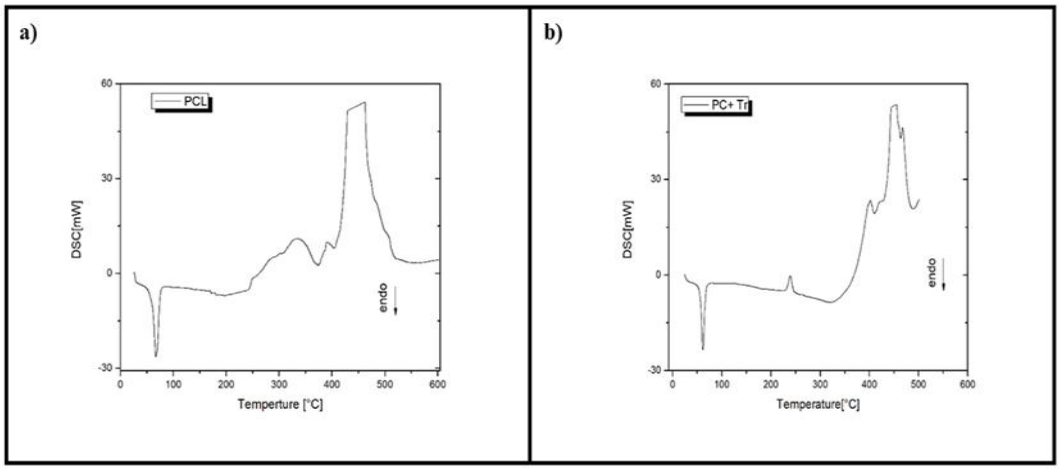
Figure 1. Infrared spectrum by Fourier Transform PLC and Tr performed in a Perkin Elmer Spectrum 100 in the wavelength range of 400-3900 cm-1. a) Tetracycline and b) PCL-TCs.



Source: Authors.

The Figure 2 shows the DSC curves of the thermal analysis. This characterization provides important context information in pharmaceutics. They confirm the purity of the active principles used, compatibility of the polymer with the drug and the stability of pharmaceutical dosage forms. It is observed from Figure 2 a) and b) an endothermic peak at 58°C corresponding to the melting point of the material, and an exothermic point at 450°C corresponding to degradation point. This can indicate the polymer matrix (PCL) does not change its melting temperature and principal characteristics (Liechty et al., 2010).

Figure 2 - Differential Scanning Calorimetry (DSC) of a) PCLand b) PCL-TCs.



Source: Authors.

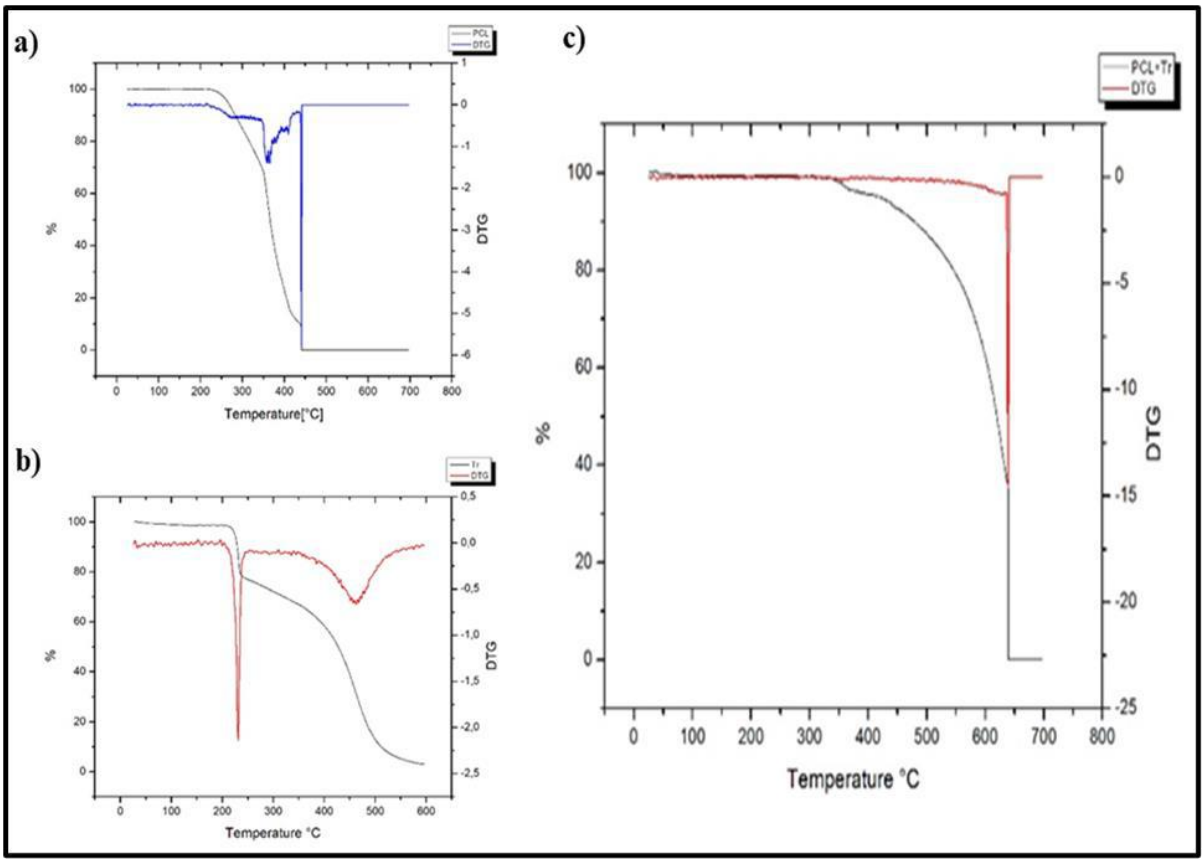
The Figure 3a), b) and c) present the Thermogravimetric Curve (TG) of PCL, Tr and PCL-TCs matrix respectively. The temperature at 350 °C in PCL Figure 3a) shows a decrease in weight of 20%, corresponding of initial degradation of carbon. The Figure 3b at 250°C present a decrease in weight of 20% which marks the start of degradation of the drug Tr and carbides formed on both corners. Thermogravimetric Analysis of both PCL and Tr showed at 400°C and 450°C a drop of 80

mass% that can be attributed transition pyrolysis of hydrocarbons formed. Also, Figure 3c) the thermal properties of the polymeric matrix (PCL) prevailed, occurring a weight loss at 400 °C of 10% corresponding to the degradation of Tr, and an increase of temperature degradation at 650°C (Puoci et al., 2008). Through the melting enthalpy obtained by DSC chart of the polymer matrix PCL and PCL-TCs. It was the crystallinity index using Equation 3 for the PCL and PCL-TCs matrix, getting the results of 57.9% and 52.3%. As melting enthalpy of PCL and ΔH_f^0 enthalpy of fusion of 100% crystalline PCL polymer (ΔH_f^0 PCL = 146.0 J / g) (Fernandes et al., 1999).

$$X_c = \frac{|\Delta H_f|}{\Delta H_f^0} \times 100 \quad (3)$$

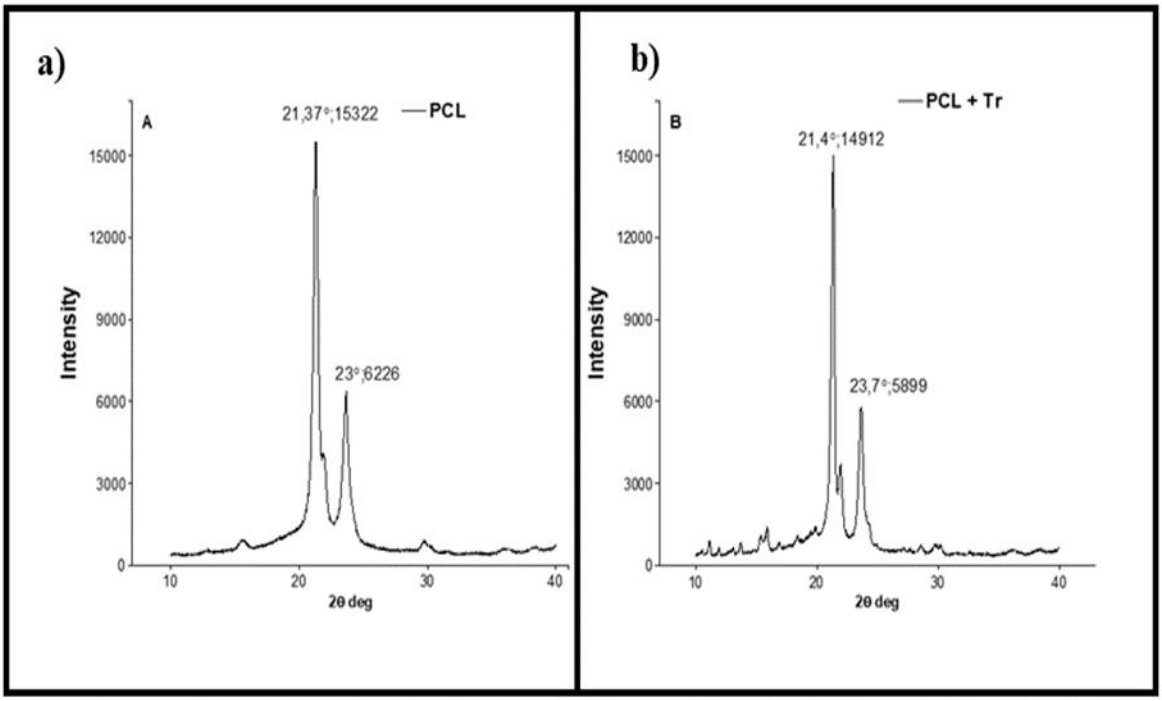
The XRD diffractograms of the PCL and PCL-TCs can be observed at Figure 4 a) and b), respectively. The PCL as a matrix has well-defined peaks approximately 21°and 23 ° characterizing the material as crystalline orthorhombic form in planes 110 and 200 (Agarwal & Speyerer, 2010). It is observed that the angles of diffraction 2θ of the polymer matrix with drug PCL and PCL-TCs suffered no displacement while maintaining the crystallinity of the PCL matrix. Analyzing the polymer matrix have the crystallinity 52.3%. This a characteristic of semi-crystalline materials confirmed by X-ray diffraction (Figure 4) and the thermogravimetric analysis (Figure 3) (Ma & Moulton, 2011).

Figure 3 - Thermogravimetric Analysis (TGA) of a) PCL, b) Tetracycline and c) PCL--Tr.



Source: Authors.

Figure 4 - X- Ray Diffractogram of a) PCL and b) PCL-TCs.



Source: Authors.

The hydrophobic tests were performed with the matrices at 35°C, conditioned for 24 hours in PBS pH 6 solution. Initially measured masses of the three samples, after 24 hours was removed excess water, and measured again. Table 1 presents the data from this test, and observing the data the matrix that best absorbed water had a percentage of 20% confirmed the hydrophobic behavior of the material.

Table 1 - Study of water swelling at PBS solution for 24 h.

	Matrix 1	**Matrix 2**	**Matrix 3**
Dry weight (mg)	68,9	51,7	79,1
Water weight (mg)	10	13	13
Water absorption (%)	14	20	16

Source: Authors.

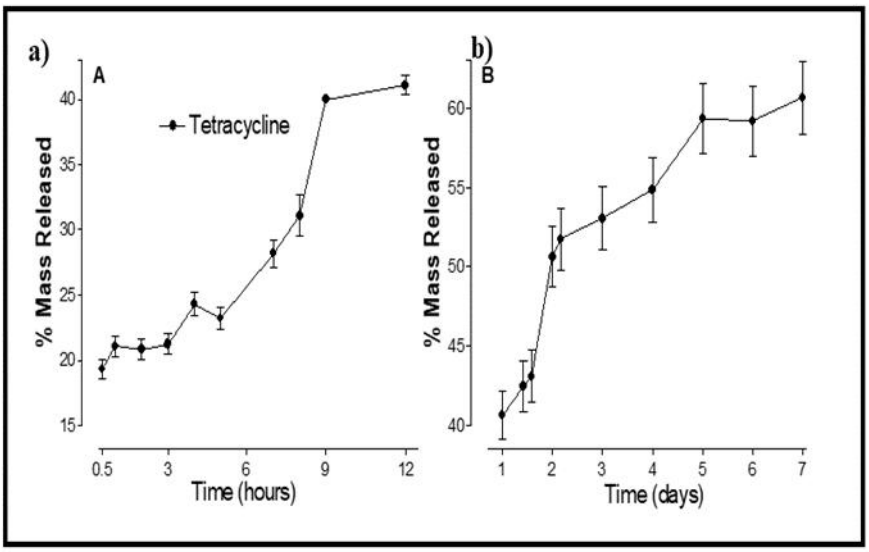
The release of tetracycline from the polymer matrix PCL is the type Fickian (2nd Fick's law, mass transportation) according with the mathematical model of Higuchi, where that release occurs only in one direction (to the matrix solution) not modifying the concentration of drug remaining in the polymer matrix (Schlesinger et al., 2015; Siepmann & Peppas, 2012).

Such a mechanism explains the release of Swelling of the matrix, drug diffusivity constant, greater than the solubility of the matrix drug(Wang et al., 2009). Even if the matrix is a hydrophobic material (maximum water uptake of 20% and does not undergo swelling) when associated with tetracycline acquires a surface hydrophilicity and swelling fact that may be explained due to hydrophilicity of tetracycline which is dispersed in the matrix surface.

Tetracycline release was investigated by the cumulative release of the drug for 168 h in PBS of pH 6.0 and showing

two distinct stages, as shown in Figure 5. In firth stage it was observed a burst-release at the first 12 h (Figure 5 a) of testing when tetracycline-loaded PCL matrices were immersed in PBS at 37°C, resulting in a rapid loss of around 40% of the drug content. Burst release is generally considered to result from dissolution of drug located at or close to the surface of the delivery device. In the second stage (after 7 days) the release becomes constant on average, 60% of the initial mass of tetracycline is released, showed sustained release (Figure 5 b) (Macedo et al., 2020; L. T. Verma et al., 2020).

Figure 5. Cumulative release of the tetracycline for 10 days in PBS of pH 6.0. a) shows tetracycline release for 12h and b) tetracycline release for 7 days.



Source: Authors.

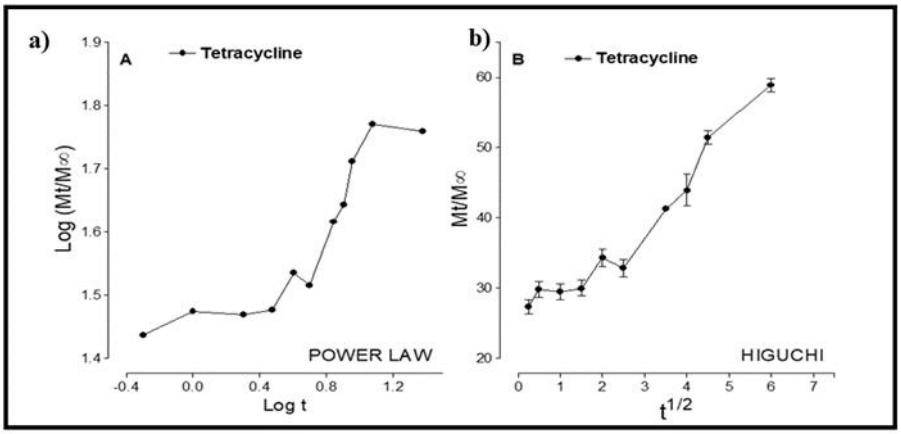
In order to determine if the release is by diffusion, swelling, erosion or even an addition of one or more cases the data were analyzed according to the mathematical model, Figure 6 the power law equation 4.

$$\frac{M_t}{M_{(\inf)}} = kt^n \xrightarrow{\log} \log\left(\frac{M_t}{M_{(\inf)}}\right) = n\log t + \log k \quad 4)$$

The graph of Figure 6 identified mathematical model which best fits for release studied by analyzing the constant n. This coefficient approached 0.5 therefore the best mathematical model of Higuchi. The mathematical model describes the Higuchi release mechanism Fickian, diffusion of drug from the polymeric matrix.

Besides, the crystallinity has a influence on material degradation. Then, the rate of degradation is proportionally to crystallinity of the material. The mechanisms of degradation of the polymer (blend or pure) have an indirectly affect in the release of the encapsulated or embedded to it drug. The release depends to the properties of the hydrophilic or lipophilic the drug. The distribution of the lipophilic drug is uniformly on the surface of the polymer matrix while the hydrophilic drug is concentrated in the surface matrix. Researches indicate that hydrophilic drugs are released more rapidly than lipophilic (Macedo et al., 2020).

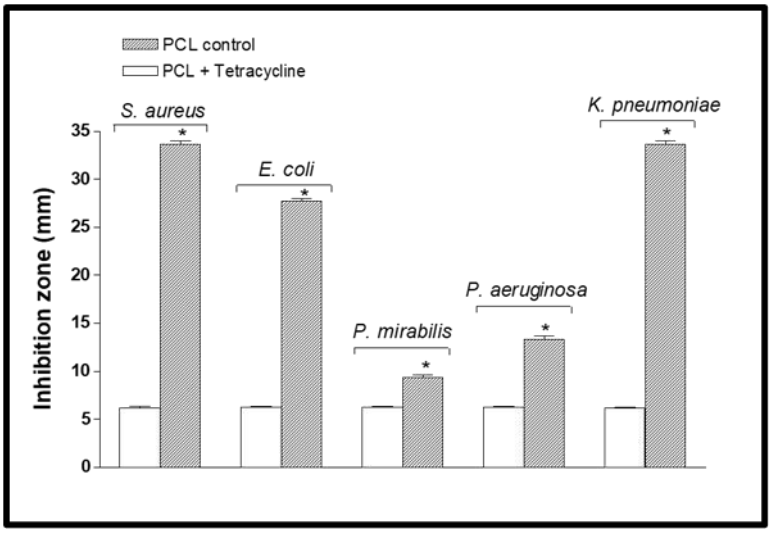
Figure 6 - a) Power low and b) Higuchi model for release perform.



Source: Authors.

The antibacterial activity of tetracycline associated polymeric matrices was evaluated (Figure 7). The disc diffusion method was based on the diffusion of an antimicrobial agent present in the disc inserted on the medium (agar) of the microorganism. The test investigates the inhibition zone causing reduction of bacterial growth. Figure 7 shows the PCL-TCs significantly induced inhibition zone formation against the five different bacteria (01 gram-positive bacteria and 4 gram-negative bacteria). The result is an antibacterial activity against one gram-positive bacteria and 4 gram-negative bacteria (P. Capellato et al., 2018).

Figure 7. Evaluation of antimicrobial activity of PCL (PCL control) and PCL- Tr (PCL+tetracycline) against Gram-negative and Gram-positive bacteria. PCL control and PCL tetracycline inhibition zone (mm). The values were expressed as mean ± standard error of the mean with n = 3, with significance *P<0,05 when compared with PCL control.



Source: Authors.

4. Conclusion

The development of devices to controlled the release are in constant technological innovation to improve the release

of drugs. PCL present a long degradation and its good adhesion to a large number of drugs. In this study was investigated the polymer PCL associated with antibiotics tetracycline as local delivery system.

According to the results, the antibiotics tetracycline has been successfully incorporated to PCL matrices. They release tetracycline in the ideal rates and shows antibacterial activity. The initial burst of tetracycline match with the critical first hours of implantation minimalizing the risks of complications. So, this material has a potential to been used in implants for drug release. For future works the authors suggest in vivo studies.

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References

Agarwal, S., & Speyerer, C. (2010). Degradable blends of semi-crystalline and amorphous branched poly(caprolactone): Effect of microstructure on blend properties. *Polymer*, *51*(5), 1024–1032. https://doi.org/10.1016/j.polymer.2010.01.020

Bartzatt, R., Koziol, K., Benish, T., & Stoddard, J. (2001). Synthesis and analysis of ethylated tetracycline, an antibiotic derivative that inhibits the growth of tetracycline-resistant XL1-Blue bacteria. *Biotechnology and Applied Biochemistry*, *33*(2), 65. https://doi.org/10.1042/BA20000079

Capellato, P., Claro, A. P. R. A., Silva, G., & Zavaglia, C. A. C. (2018). Antimicrobial effect of TiO2 nanotubes coating for dental implant. *Dental Materials*, *34*, e21. https://doi.org/10.1016/j.dental.2018.08.043

Capellato, P., Camargo, S. E. A., & Sachs, D. (2020). Biological Response to Nanosurface Modification on Metallic Biomaterials. *Current Osteoporosis Reports*, 1–6. https://doi.org/10.1007/s11914-020-00635-x

Cui, W., Zhou, Y., & Chang, J. (2010). Electrospun nanofibrous materials for tissue engineering and drug delivery. *Science and Technology of Advanced Materials*, *11*(1), 014108. https://doi.org/10.1088/1468-6996/11/1/014108

de Arruda Almeida, K., de Queiroz, A. A. A., Higa, O. Z., Abraham, G. A., & San Román, J. (2004). Macroporous poly(ϵ-caprolactone) with antimicrobial activity obtained by iodine polymerization. *Journal of Biomedical Materials Research Part A*, *68A*(3), 473–478. https://doi.org/10.1002/jbm.a.20085

De Queiroz, A. A. A., França, É. J., Abraham, G. A., & Román, J. S. (2002). Ring-opening polymerization of ϵ-caprolactone by iodine charge-transfer complex. *Journal of Polymer Science Part B: Polymer Physics*, *40*(8), 714–722. https://doi.org/10.1002/polb.10133

Ezhilarasu, H., Ramalingam, R., Dhand, C., Lakshminarayanan, R., Sadiq, A., Gandhimathi, C., Ramakrishna, S., Bay, B. H., Venugopal, J. R., & Srinivasan, D. K. (2019). Biocompatible aloe vera and tetracycline hydrochloride loaded hybrid nanofibrous scaffolds for skin tissue engineering. *International Journal of Molecular Sciences*, *20*(20). https://doi.org/10.3390/ijms20205174

Fereshteh, Z., Nooeaid, P., Fathi, M., Bagri, A., & Boccaccini, A. R. (2015). Mechanical properties and drug release behavior of PCL/zein coated 45S5 bioactive glass scaffolds for bone tissue engineering application. *Data in Brief*, *4*, 524–528. https://doi.org/10.1016/j.dib.2015.07.013

Fernandes, N. S., Da Silva Carvalho Filho, M. A., Mendes, R. A., & Ionashiro, M. (1999). Thermal Decomposition of Some Chemotherapic Substances. *Journal of the Brazilian Chemical Society*, *10*(6), 459–462. https://doi.org/10.1590/S0103-50531999000600007

Grossen, P., Witzigmann, D., Sieber, S., & Huwyler, J. (2017). PEG-PCL-based nanomedicines: A biodegradable drug delivery system and its application. In *Journal of Controlled Release* (Vol. 260, pp. 46–60). Elsevier B.V. https://doi.org/10.1016/j.jconrel.2017.05.028

Iman, M., Barati, A., & Safari, S. (2020). Characterization, in vitro antibacterial activity, and toxicity for rat of tetracycline in a nanocomposite hydrogel based on PEG and cellulose. *Cellulose*, *27*(1), 347–356. https://doi.org/10.1007/s10570-019-02783-5

Kaur, M., & Singh, K. (2019). Review on titanium and titanium based alloys as biomaterials for orthopaedic applications. *Materials Science and Engineering: C*, *102*, 844–862. https://doi.org/10.1016/J.MSEC.2019.04.064

Kim, J., Kudisch, M., Mudumba, S., Asada, H., Aya-Shibuya, E., Bhisitkul, R. B., & Desai, T. A. (2016). Biocompatibility and pharmacokinetic analysis of an intracameral polycaprolactone drug delivery implant for glaucoma. *Investigative Ophthalmology and Visual Science*, *57*(10), 4341–4346. https://doi.org/10.1167/iovs.16-19585

Kim, Y., Kim, J., Lee, H., Shin, W. R., Lee, S., Lee, J., Park, J. Il, Jhun, B. H., Kim, Y. H., Yi, S. J., & Kim, K. (2019). Tetracycline analogs inhibit osteoclast differentiation by suppressing MMP-9-Mediated Histone H3 cleavage. *International Journal of Molecular Sciences*, *20*(16). https://doi.org/10.3390/ijms20164038

Kuznetsov, K. A., Stepanova, A. O., Kvon, R. I., Douglas, T. E. L., Kuznetsov, N. A., Chernonosova, V. S., Zaporozhchenko, I. A., Kharkova, M. V., Romanova, I. V., Karpenko, A. A., & Laktionov, P. P. (2018). Electrospun produced 3D matrices for covering of vascular stents: Paclitaxel release depending on fiber structure and composition of the external environment. *Materials*, *11*(11). https://doi.org/10.3390/ma11112176

Leypold, C. F., Reiher, M., Brehm, G., Schmitt, M. O., Schneider, S., Matousek, P., & Towrie, M. (2003). Tetracycline and derivatives - Assignment of IR and

Raman spectra via DFT calculations. *Physical Chemistry Chemical Physics*, *5*(6), 1149–1157. https://doi.org/10.1039/b210522e

Liechty, W. B., Kryscio, D. R., Slaughter, B. V., & Peppas, N. A. (2010). Polymers for Drug Delivery Systems. *Annual Review of Chemical and Biomolecular Engineering*, *1*(1), 149–173. https://doi.org/10.1146/annurev-chembioeng-073009-100847

Lü, L.-X., Wang, Y.-Y., Mao, X., Xiao, Z.-D., & Huang, N.-P. (2012). The effects of PHBV electrospun fibers with different diameters and orientations on growth behavior of bone-marrow-derived mesenchymal stem cells. *Biomedical Materials*, *7*(1), 015002. https://doi.org/10.1088/1748-6041/7/1/015002

Ma, Z., & Moulton, B. (2011). Recent advances of discrete coordination complexes and coordination polymers in drug delivery. In *Coordination Chemistry Reviews* (Vol. 255, Issues 15–16, pp. 1623–1641). Elsevier. https://doi.org/10.1016/j.ccr.2011.01.031

Macedo, A. S., Castro, P. M., Roque, L., Thomé, N. G., Reis, C. P., Pintado, M. E., & Fonte, P. (2020). Novel and revisited approaches in nanoparticle systems for buccal drug delivery. In *Journal of Controlled Release* (Vol. 320, pp. 125–141). Elsevier B.V. https://doi.org/10.1016/j.jconrel.2020.01.006

Malikmammadov, E., Tanir, T. E., Kiziltay, A., Hasirci, V., & Hasirci, N. (2018). PCL and PCL-based materials in biomedical applications. *Journal of Biomaterials Science, Polymer Edition*, *29*(7–9), 863–893. https://doi.org/10.1080/09205063.2017.1394711

Manoukian, O. S., Arul, M. R., Sardashti, N., Stedman, T., James, R., Rudraiah, S., & Kumbar, S. G. (2018). Biodegradable polymeric injectable implants for long-term delivery of contraceptive drugs. *Journal of Applied Polymer Science*, *135*(14). https://doi.org/10.1002/app.46068

Nagiah, N., Murdock, C. J., Bhattacharjee, M., Nair, L., & Laurencin, C. T. (2020). Development of Tripolymeric Triaxial Electrospun Fibrous Matrices for Dual Drug Delivery Applications. *Scientific Reports*, *10*(1), 1–11. https://doi.org/10.1038/s41598-020-57412-0

Pathak, M., Coombes, A. G. A., Ryu, B. M., Cabot, P. J., Turner, M. S., Palmer, C., Wang, D., & Steadman, K. J. (2018). Sustained Simultaneous Delivery of Metronidazole and Doxycycline From Polycaprolactone Matrices Designed for Intravaginal Treatment of Pelvic Inflammatory Disease. *Journal of Pharmaceutical Sciences*, *107*(3), 863–869. https://doi.org/10.1016/j.xphs.2017.09.033

Capellato, P., Marino, C. E. B., Silva, G., Vasconcelos, L. V. B., Cardoso, R. P., & Kayam Hamdar, D. S. (2020). Surface treatment with silver particles isles on Titanium cp: study of antimicrobial activity. *Research, Society and Development*, *9*(4). https://doi.org/http://dx.doi.org/10.33448/rsd-v9i4.2662

Puoci, F., Iemma, F., & Picci, N. (2008). Stimuli-Responsive Molecularly Imprinted Polymers for Drug Delivery: A Review. *Current Drug Delivery*, *5*(2), 85–96. https://doi.org/10.2174/156720108783954888

Rezk, A. I., Lee, J. Y., Son, B. C., Park, C. H., & Kim, C. S. (2019). Bi-layered nanofibers membrane loaded with titanium oxide and tetracycline as controlled drug delivery system for wound dressing applications. *Polymers*, *11*(10). https://doi.org/10.3390/polym11101602

Schlesinger, E., Ciaccio, N., & Desai, T. A. (2015). Polycaprolactone thin-film drug delivery systems: Empirical and predictive models for device design. *Materials Science and Engineering C*, *57*, 232–239. https://doi.org/10.1016/j.msec.2015.07.027

Siepmann, J., & Peppas, N. A. (2012). Modeling of drug release from delivery systems based on hydroxypropyl methylcellulose (HPMC). In *Advanced Drug Delivery Reviews* (Vol. 64, Issue SUPPL., pp. 163–174). Elsevier. https://doi.org/10.1016/j.addr.2012.09.028

Souza, S. O. L., Cotrim, M. A. P., Oréfice, R. L., Carvalho, S. G., Dutra, J. A. P., de Paula Careta, F., Resende, J. A., & Villanova, J. C. O. (2018). Electrospun poly(ε-caprolactone) matrices containing silver sulfadiazine complexed with β-cyclodextrin as a new pharmaceutical dosage form to wound healing: preliminary physicochemical and biological evaluation. *Journal of Materials Science: Materials in Medicine*, *29*(5). https://doi.org/10.1007/s10856-018-6079-8

Thomas, M. V., Jarboe, G., & Frazer, R. Q. (2008). Infection control in the dental office. *Dental Clinics of North America*, *52*(3), 609–628. https://doi.org/10.1016/J.CDEN.2008.02.002

Verma, L. T., Singh, N., Gorain, B., Choudhury, H., Tambuwala, M. M., Kesharwani, P., & Shukla, R. (2020). Recent advances in self-assembled nanoparticles for drug delivery. *Current Drug Delivery*, *17*. https://doi.org/10.2174/1567201817666200210122340

Verma, R. P. (2020). Titanium based biomaterial for bone implants: A mini review. *Materials Today: Proceedings*, *26*, 3148–3151. https://doi.org/10.1016/J.MATPR.2020.02.649

Wang, X., Wang, Y., Wei, K., Zhao, N., Zhang, S., & Chen, J. (2009). Drug distribution within poly(ε-caprolactone) microspheres and in vitro release. *Journal of Materials Processing Technology*, *209*(1), 348–354. https://doi.org/10.1016/J.JMATPROTEC.2008.02.004

Wsoo, M. A., Shahir, S., Mohd Bohari, S. P., Nayan, N. H. M., & Razak, S. I. A. (2020). A review on the properties of electrospun cellulose acetate and its application in drug delivery systems: A new perspective. In *Carbohydrate Research* (Vol. 491, p. 107978). Elsevier Ltd. https://doi.org/10.1016/j.carres.2020.107978

Xie, Y., Liu, C., Huang, H., Huang, J., Deng, A., Zou, P., & Tan, X. (2018). Bone-targeted delivery of simvastatin-loaded PEG-PLGA micelles conjugated with tetracycline for osteoporosis treatment. *Drug Delivery and Translational Research*, *8*(5), 1090–1102. https://doi.org/10.1007/s13346-018-0561-1

Yeh, Y.-C., Huang, T.-H., Yang, S.-C., Chen, C.-C., & Fang, J.-Y. (2020). Nano-Based Drug Delivery or Targeting to Eradicate Bacteria for Infection Mitigation: A Review of Recent Advances. *Frontiers in Chemistry*, *8*, 286. https://doi.org/10.3389/fchem.2020.00286

Zupančič, Š., Preem, L., Kristl, J., Putrinš, M., Tenson, T., Kocbek, P., & Kogermann, K. (2018). Impact of PCL nanofiber mat structural properties on hydrophilic drug release and antibacterial activity on periodontal pathogens. *European Journal of Pharmaceutical Sciences*, *122*, 347–358. https://doi.org/10.1016/j.ejps.2018.07.024