Evaluation of Electrospun Nanofibrous Structures for Drug Release Application

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Abstract

Biopolymers show the excellent biodegradability and efficient release sustainability for encapsulated drugs. In particular, electrospun polymer or composite fibre mats provide greater benefits owing to their competitive release properties and large specific surface area. This research work focused on electrospun nanofibres derived from poly(ε-caprolactone) (PCL), poly(lactic acid) (PLA) and PCL/ magnetic nanoparticles (MPs) solutions by carrying a therapeutic compound tetracycline hydrochloride (TCH) with the potential use for medical applications. The material systems were examined to evaluate how composite constituents affected the surface morphology with the aim of drug release control. It has been found that the fibre diameter decreased considerably with the addition of TCH drug. The average fibre diameter was also reduced with additional MPs due to enhanced solution conductivity. Furthermore, Fourier transform infrared spectroscopy (FTIR) proved the successful encapsulation of TCH drug. Over short-term periods, the TCH release from PCL nanofibres was higher than PCL/ MPs and PLA nanofibres; whereas, on a long-term run, TCH release from PCL became slower owing to its high degree of crystallinity. The TCH release kinetics of PCL/ TCH nanofibres were better estimated by Zeng model when compared with PLA/TCH counterparts.

Keywords: Electrospinning; Magnetic nanoparticles; Nanocomposites; Drug release; Release kinetics.

Introduction

Electrospinning is a material fabrication technique that produces functional and economical three-dimensional fibrous structures (Heydarkhan-Hagvall et al., 2008). The electrospun fibres result in the formation of a fabric network that has a high porosity, small pore size and large surface area to volume ratio (Liang et al., 2007). In recent years, this technique has been utilised for many biomedical applications particularly in controlling drug delivery systems (Kenawy et al., 2009; Haroosh et al., 2014). Biopolymers have been frequently employed for multiple purposes in the pharmaceutical applications such as tissue engineering and drug delivery. A several biodegradable polymers has been fabricated for drug delivery, such as poly (lactic acid) (PLA) (Noor Azman et al., 2013, Dong et al., 2014b, Dong et al., 2014a), poly (ε-caprolactone) (PCL) (Haroosh et al., 2012), poly (glycolic acid) (PGA), poly (lactide-co-glycolide) (PLGA) and polyurethane (PU) as well as natural proteins including collagen, gelatin and elastin(Heydarkhan-Hagvall et al., 2008). Poly(ε-caprolactone) (PCL) is a semi-crystalline polymer that is broadly used in tissue scaffolding owing to its good drug permeability and slow biodegradability (Haroosh et al., 2013a). Poly(lactic acid) (PLA) is typically employed in diverse biomedical purposes, benefiting from its biodegradability and biocompatibility (Dong et al., 2011; Dong et al., 2013). Magnetic nanoparticles such as Fe_3O_4 are

inorganic materials with low toxicity, super-paramagnetic properties, good biocompatibility and better stability in the atmosphere than metal nanoparticles. Tetracycline hydrochloride (TCH) is an antibiotic utilised for the curing and avoidance of bacterial infections in the burn, injury and surgery, in addition to numerous wound healing areas (Haroosh et al., 2013b). The objective of this study is to evaluate the fabrication, structural analysis and TCH release of PLA/TCH, PCL/ MPs and PCL/ MPs nanofibres and further to fabricate a novel carrier structures for the efficient drug delivery. The mathematical modelling has also been employed for drug release kinetics.

Materials and Fabrication

Materials

PLA 3051D of *MW* = 93,500 g/mol was supplied by Nature Works, USA. PCL of *MW* = 80,000 g/mol, iron sulfate, iron nitrate, tetracycline hydrochloride (TCH, $C_{22}H_{24}N_2O_8{\cdot}HCl$, MW=480.9 g/mol), phosphate buffer solution (PBS), chloroform and methanol were purchased from Sigma-Aldrich Ltd, Australia and used without any purification. Iron sulfate and iron nitrate were used to synthesise MPs whilst TCH and PBS acted as a model drug and a medium of drug release, respectively. Chloroform and methanol were employed as solvents for PLA and PCL.

Electrospinning

Electrospinning was carried out using 8 wt%/v PLA solution and 8 wt%/v PCL solution. The solvent used in all cases was a mixture of chloroform and methanol (volume ratio: 2:1). The MPs were synthesised from iron sulfate ($FeSO_4$) and iron nitrate ($Fe(NO_3)_3$) and were added at 0.1 wt%/v to the PCL solution, subjected to 2 h ultrasonication. 5wt% TCH solution was then mixed with 0.1wt%/v MPs for 2h in methanol by using the similar ultrasonication process and then added to PLA, PCL and PCL/MPs solutions. For electrospinning, the solution was transferred to a 10ml syringe mounted onto a syringe pump (A Fusion 100 syringe pump, Chemyx Inc. Stafford, TX USA) with a 20G metallic needle (0.584 mm inner diameter). The flow rate of polymer solution was set at 2 ml/h, and the applied positive voltage was 25 kV. The electrospinning process was conducted at 24°C. The resulting fibres were collected on a ground collector covered by a flat aluminium foil. The distance between the needle tip and the target was 13 cm.

In Vitro Drug release Study

The drug-loaded fibre mat sample (2 × 2 cm^2) was incubated in a rotary shaker at 37 °C in 20 ml PBS (pH=7.4). After the required incubation time for drug release, the sample was transferred to a 20 ml fresh buffer solution and the released TCH amount in the buffer solution was determined.

Zeng model has been used to interpret the drug release mechanism (Zeng et al., 2011). The equation for Zeng model is relatively complex as shown in equations 1 and 2.

$$\frac{M_t}{M_\infty} = \frac{K_{off}}{K_{on}+K_{off}}\left(1-e^{-K_S t}\right)+\frac{K_{on}}{K_{on}+K_{off}}\left(1-e^{-K_{off} t}\right) \quad (1)$$

$$\Delta G = -k_B T\left(\frac{K_{on}}{K_{off}}\right) \quad (2)$$

K_{on} is the rate constant of association for non-dispersed drug molecules in the system that need to be disassociated from carriers prior to release. On the other hand, K_{off} is the rate constant of disassociation accordingly and K_S is a constant proportional to the surface-to-volume ratio of carriers for the enhancement of drug release. ΔG is the free energy difference between the free and bound state while k_B is the Boltzmann's constant is the absolute temperature that is assumed to be 300 K.

Characterisation Techniques

Solution viscosity was evaluated with a Visco 88 portable viscometer from Malvern Instruments (UK). The nanofibre morphology was studied via an EVO 40XVP scanning electron microscope (SEM) (Germany) at an accelerating voltage of 5 kV. Before SEM observation, the samples were sputter-coated with platinum. Fibre diameter was calculated from SEM images by using an image analysis tool in Zeiss smart SEM software. For each sample, measurements were made from a minimum of 150 fibres from multiple scanned SEM images at a sampling rate of 15 fibres per image. The amount of TCH present in the release buffer was determined by a UV–vis spectrophotometer (JascoV-67) at the wavelength of 360 nm. Fourier transform infrared spectroscopy (FTIR) was performed in a Spectrum 100 FTIR Spectrometer Perkin Elmer (Japan). Spectra were recorded in range of 4000–550 cm^{-1} with 4 cm^{-1} resolution with an attenuated total reflectance (ATR) technique.

Results and Discussion

Fibre Morphology

To investigate the effect of nanoparticles and drug on the morphology, PCL, PCL/MPs, PCL/MPs/TCH, PLA and PLA/TCH nanofibres were fabricated. As illustrated in Figure 1, electrospun naofibres produced from PCL and PLA have the average fibre diameters of (368 ± 15) nm and (510 ± 20) nm respectively. The addition of MPs to PCL enhanced electrical conductivity, obviously owing to the increased iron content. The PCL/MPs composite system appears to create fibrous structures with smaller fibre diameters (316 ± 20) nm than electrospun PCL fibres.

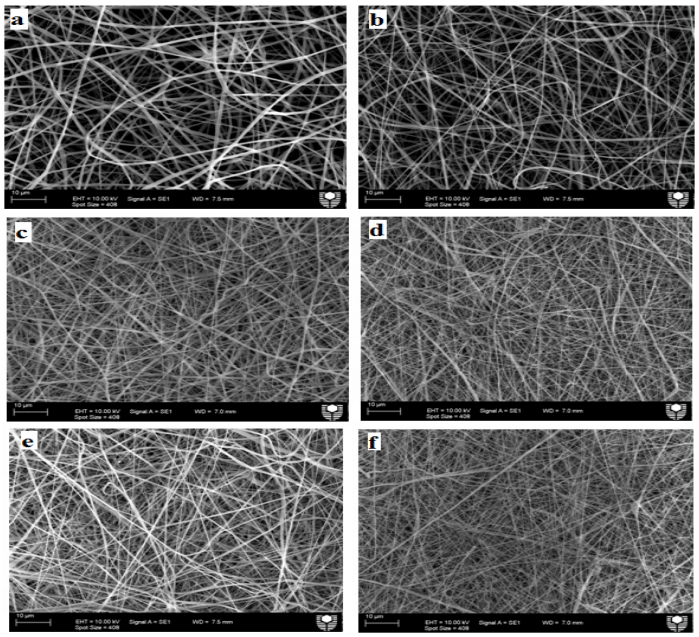

Figure 1. SEM micrographs of electrospun naofibres : (a) PLA, (b) PLA/TCH, (c) PCL, (d) PCL/TCH, (e) PCL/MPs, and (f) PCL/ MPs/ TCH. The scale bars represent 10 µm.

On the other hand, it is clear that the fibre diameter was reduced significantly from (368 ± 15) nm to (279 ± 15) nm for PCL, from (316 ± 20) nm to (225 ± 15) nm for PCL/MPs and from (510 ± 20) nm to (397 ± 20) nm for PLA accordingly when adding TCH drug. This decline in nanofibre diameters can be attributed to amphoteric molecules in the TCH drug with several ionisable functions groups (Sassman and Lee, 2005), which in turn leads to increasing the solution electrical conductivity.

FTIR Evaluation

Figure 2 shows the FTIR spectra of PLA, PLA/ TCH, PCL, PCL/TCH and PCL/ MPs/ TCH. The spectra exhibit initially carbonyl stretching (C=O) band at 1722 cm^{-1} for PCL nanofibres and at 1750 cm^{-1} for PLA nanofibres. The peaks at 1453 cm^{-1} and 1382 cm^{-1} specify a C-H deformation of PLA. In addition, many peaks represent C–C and C–O stretching in the range from 1240 cm^{-1} to 840 cm^{-1} for PLA and PCL. With respect to PCL/MPs composites, it is difficult to indicate the effect of MPs on the bonds, which may be due to the small amount of MPs. The spectrum of TCH is also hard to be allocated for the band shifting. However, the main noticeable change in the infrared spectra is that the weak bands of TCH at 1615 cm^{-1} and 1580 cm^{-1} were detected, and these are assigned to C=O stretching in ring A and C=O stretching in ring C, respectively. This observation proves the successful encapsulation of TCH drug.

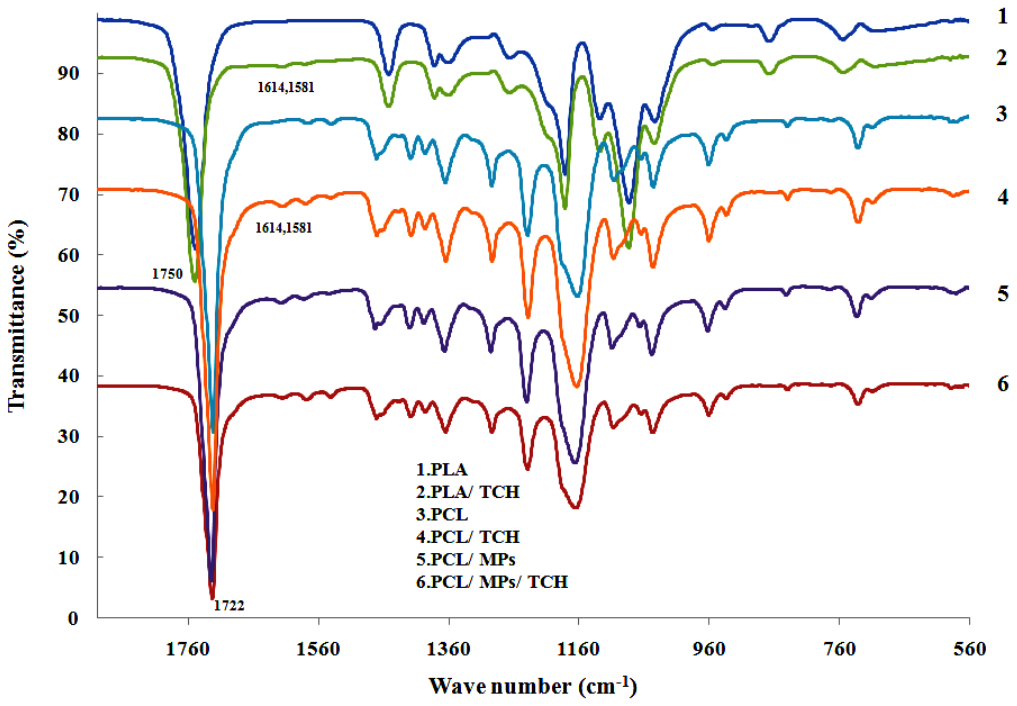

Figure 2. FTIR spectra for selected material samples showing relative FTIR peaks.

In Vitro Drug Release

TCH release profiles from electrospun PLA, PCL and PCL/ MPs composite fibre mats are reported in Figure 3. Similarly, both PCL and PCL/ MPs fabric samples show the fast drug release behaviour. In the initial 25 min, 66% drug was released from PCL samples and 58% from PCL/ MPs relative to 34% from PLA. The initial higher TCH release from PCL nanofibres accounts for a relatively high degree of crystallinity of PCL 59.1% compared with 53.7% and 11.3% for PCL/ MPs and PLA nanofibres,

respectively. The high crystallinity of nanofibres easily restricts the mobility of drug molecules to stay on the fibre surface areas. In addition, rapid release is associated with the electrospinning process, which includes quick solvent evaporation and highly ionic interactions leading to the mobility of drug molecules on the surfaces of electrospun fibres. Nevertheless, TCH release from PCL starts to become slower than that from PCL/ MPs nanofibres after 25h. The incorporation of MPs leads to the increased fibre porosity, resulting in the acceleration of TCH release. In addition, the small diameters of fibrous structures of PCL/ MPs/ TCH compared with PCL/ TCH nanofibres tend to facilitate the increase of the surface area, thus leading to short diffusion distances and fast TCH release. On the other hand, TCH release from PLA starts to become faster than that from PCL and PCL/ MPs nanofibres after 50 h and 110 h, respectively. In the long term of drug release, the semi-crystalline PCL may hinder the water penetration inside nanofibres, and thus the absorption of water occurs slowly compared with amorphous PLA. Moreover, the low crystallinity of PLA can accelerate the fibre degradation. The amorphous regions are easily degraded as opposed to the crystalline regions due to the random and less tightly packed arrangement of molecular chains.

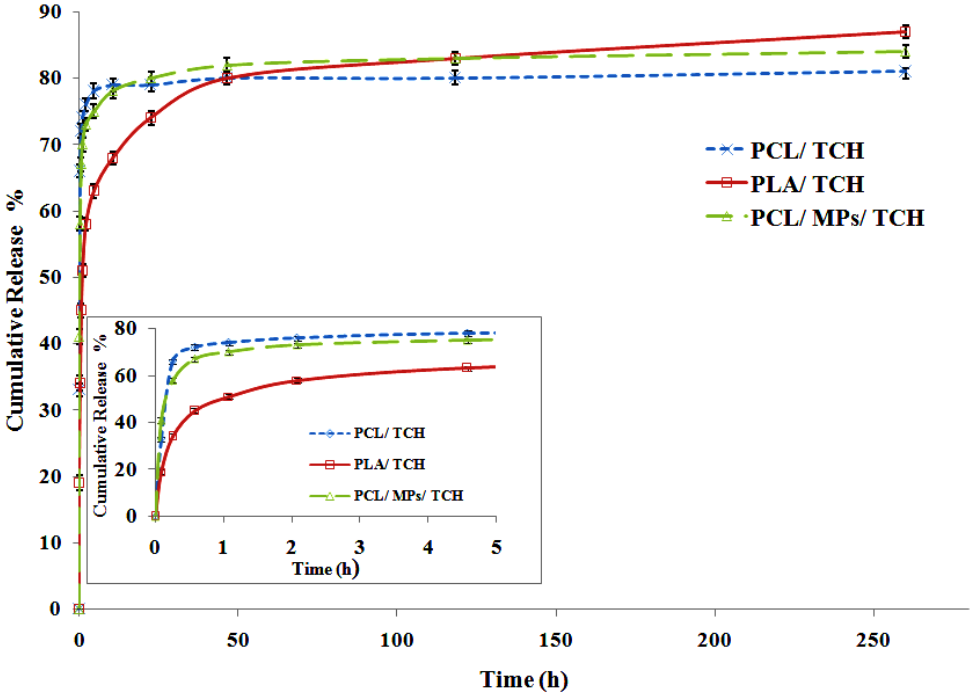

Figure 3. TCH release profiles from PLA, PCL and PCL/ MPs.

Release Kinetics

The release kinetics of TCH drug was investigated by fitting Zeng model (Zeng et al., 2011) to the experimental release data, as demonstrated in Figure 4. The addition of MPs to PCL causes a little decrease in ΔG values from 4.94×10^{-21} J to 4.19×10^{-21} J, indicating that there is a slight enhancement in the interaction between the PCL nanofibres and TCH. However, such enhancement just adjusts the TCH at the first stage, and the drug release from PCL/ MPs/ TCH was fast than that from PCL/TCH after 25h. This perception may be due to the formation of small fibre diameters by adding MPs to accelerate the TCH release. Moreover, increased K_s values reflect the enhancement of fibre pore size, which in turn leads to the fast TCH release. On the other hand, PLA/TCH exhibits a lower ΔG than PCL/ TCH (4.94×10^{-21} J vs. 2.0×10^{-21}), suggesting that PLA partially decrease the release rate of TCH due to their improved interaction. The TCH release kinetics are not completely described by this

model because it does not take into account the erosion/biodegradation and dimensional alteration of PLA/TCH and PCL/ TCH nanofibres.

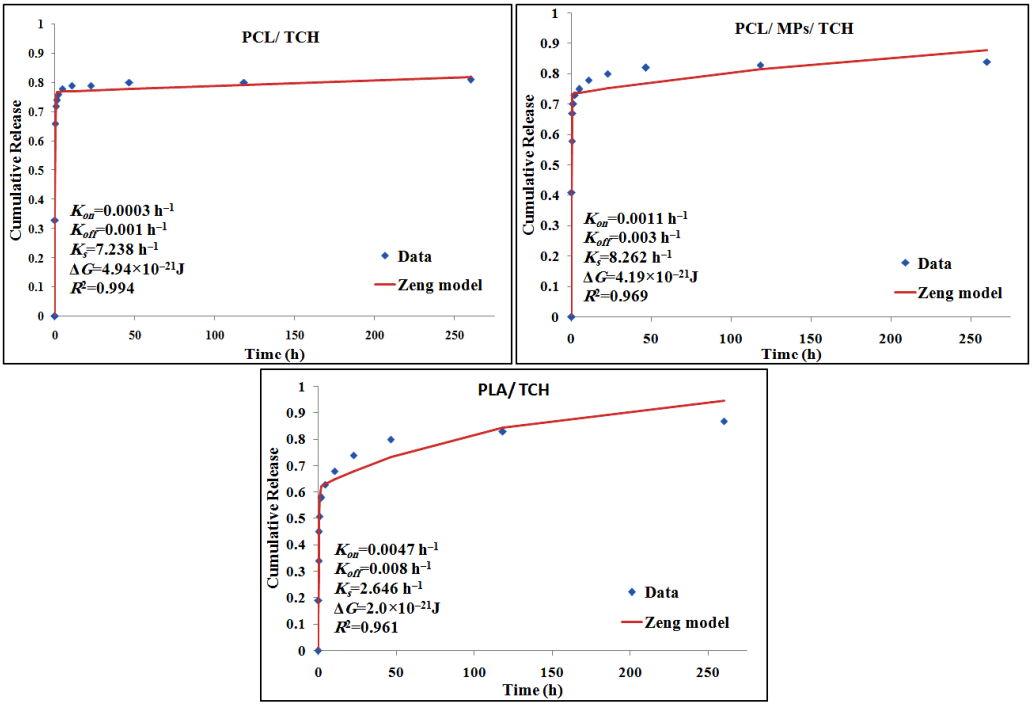

Figure 4. Model fitting with the release data based on PCL/ TCH, PCL/ MPs/ TCH and PLA/ TCH nanofibres.

Conclusions

The addition of MPs to PCL and TCH drug enhances the electrical conductivity of solution, thus leading to the reduction of average fibre diameters. FTIR spectra examination demonstrates a successful encapsulation of TCH within the fibre mats. PCL and PCL/ MPs nanofibres show fast drug release behaviour compared with PLA nanofibres after 25 min. TCH release from PCL nanofibres begins to be slower than that from PCL/ MPs nanofibres after 25h. In comparison, TCH release from PLA appears to be faster than those from PCL and PCL/ MPs nanofibres after 50 h and 110 h respectively. Zeng model indicates minor enhancement in the interaction between PCL nanofibres and TCH with the additional MPs. Besides, such model also shows that PLA partially decreases the release rate of TCH due to their higher interaction with TCH than PCL.

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Biography

Hazim Haroosh is currently a PhD student at the Chemical Engineering Department, Curtin University. He received B.Sc. and M.Sc. degrees in Chemical Engineering from Tikrit University, Iraq. He has

worked in the State Company for Drug Industries and Medical Appliances, Samara (SDI), Iraq since November 2002. He has hands-on experience in quality control and management, project planning and environmental impact assessment. He was the Director of the Wastewater Treatment Department, and was also appointed as the Assistant Chief Engineer. He is a member of the Iraqi Engineers Association. He published a book chapter, 10 journal papers and 13 conference papers in electrospun nanofibres and advanced composites. He has been working as a Tutor since 2012, for Engineering Foundations: Design and Processes 100 at Curtin University. In addition, He was also involved in the Workshops for Engineering Foundations: Principles and Communications 100, as well as for Engineering Materials 100.

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