

Chapter Title: Monoaxial electrospinning

Book Title: Nanofibres in Drug Delivery

Book Author(s): Gareth R. Williams, Bahijja T. Raimi-Abraham and C. J. Luo

Published by: UCL Press. (2018)

Stable URL: <https://www.jstor.org/stable/j.ctv550dd1.7>

---

JSTOR is a not-for-profit service that helps scholars, researchers, and students discover, use, and build upon a wide range of content in a trusted digital archive. We use information technology and tools to increase productivity and facilitate new forms of scholarship. For more information about JSTOR, please contact support@jstor.org.

Your use of the JSTOR archive indicates your acceptance of the Terms & Conditions of Use, available at

<https://about.jstor.org/terms>



This book is licensed under a Creative Commons Attribution 4.0 International License (CC BY 4.0). To view a copy of this license, visit  
<https://creativecommons.org/licenses/by/4.0/>.



UCL Press is collaborating with JSTOR to digitize, preserve and extend access to *Nanofibres in Drug Delivery*

JSTOR

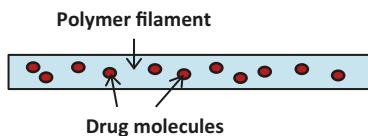
# 3

## Monoaxial electrospinning

### 3.1 Introduction

Monoaxial (or uniaxial) electrospinning is the simplest route by which fibres can be made using the electrohydrodynamic technique. To employ this to generate drug delivery systems, a mixed liquid (solution, emulsion or suspension) of a polymer and a drug in a (typically volatile) solvent is first prepared. This is dispensed through a single-bore blunt-end needle (often made of a solvent-resistant metal such as stainless steel) towards a metal collector (such as a stainless steel plate or an aluminium sheet), with a high electrical potential difference (commonly 5–20 kV for monoaxial single-liquid electrospinning) applied between the two. The fibres collected comprise monolithic systems, with the drug most usually evenly dispersed throughout the polymer, as depicted in Figure 3.1. Because the electrospinning process is very rapid, with the time between the solution exiting the spinneret and reaching the collector being well under a second, the arrangement of the molecules in the solution is propagated into the solid state. Thus, electrospun fibres typically take the form of amorphous solid dispersions (ASDs), as described in section 1.3.4.

The first report of electrospun fibres being used in the drug delivery context came from Kenawy *et al.*<sup>1</sup> These researchers generated fibres loaded with tetracycline hydrochloride using poly(lactic acid) (PLA), poly(ethylene-co-vinyl acetate) (PEVA) or a blend of the two as the filament-forming polymer matrix. Since then, a vast number of studies have been reported in which fibres from electrospinning are explored for drug delivery: a Web of Science search for ‘electrospinning



**Figure 3.1** A schematic illustration of an electrospun fibre from monoaxial spinning.

AND drug delivery' on 28 June 2017 gave 1547 hits. This interest has arisen because there are a number of advantages of using electrospun nanofibres in drug delivery. These include: (1) high drug loadings (up to 60%) and encapsulation efficiency (up to 100%);<sup>2</sup> (2) a wide range of polymers (> 100) can be spun;<sup>3</sup> (3) drug release can be modulated through the choice of polymer (hydrophilic or hydrophobic, molecular weight);<sup>4</sup> (4) the fibres have high specific surface areas ( $10\text{--}100 \text{ m}^2 \text{ g}^{-1}$ );<sup>5</sup> and (5) the process is simple to perform and cost-effective, on the laboratory scale at least.<sup>6</sup>

In this chapter, we will review the major applications of fibres from monoaxial spinning in drug delivery.

## 3.2 Experimental considerations

As was discussed in section 2.5, a number of solution and processing parameters need to be controlled for successful electrospinning.

### 3.2.1 Solution parameters

The molecular weight and concentration of the polymer(s) and the choice of solvent are important because they control the viscosity, surface tension and conductivity of the spinning solution, and thus how it behaves in electrospinning. The spinnable viscosity range varies with the polymer and solvent, but should be at least around 120 cP. In very-low-viscosity solutions, there will be insufficient polymer chain entanglements to produce fibres.<sup>7</sup> In contrast, if the viscosity is too high then the surface tension cannot easily be overcome. In both cases, the result can be droplets or particles forming rather than fibres.

The solvent used must be capable of dissolving the polymer of interest at an appropriate concentration to form fibres, and must possess a suitable volatility. A low-volatility solvent like water (boiling point (b.p.) 100°C) may fail to evaporate completely over the distance

between the spinneret and the collector. When the fibres form, they will hence contain residual water owing to this incomplete evaporation. The residue solvent will subsequently evaporate from the fibres upon storage, resulting in ribbon-like (flattened) fibres, wrinkles on the fibre surface or fused fibres. On the other hand, a high-volatility solvent may evaporate very quickly, leading to larger fibre diameters (less time for elongation before solidification) and clogging of the spinneret (due to drying of the liquid at the spinneret before jetting, or drying of the Taylor cone during jetting). Solvents commonly used for electrospinning include ethanol (b.p. 78°C), chloroform (61°C), dichloromethane (40°C) and hexafluoroisopropanol (58°C).

Mixtures of miscible solvents can be used to ensure that sufficient polymer can be dissolved to give a solution of appropriate viscosity and volatility with a suitable dielectric constant range to allow fibre formation. For instance, less volatile solvents such as dimethyl formamide (DMF: b.p. 153°C) and dimethylacetamide (b.p. 165°C) are frequently used in combination with more volatile solvents like acetone (b.p. 56°C), dichloromethane (b.p. 40°C), tetrahydrofuran (b.p. 66°C) or methanol (b.p. 64°C). However, care must be taken because using a mixture of solvents with very different volatilities can result in porous fibre structures, as reported by Katsogiannis *et al.* for organic solvent mixtures with dimethyl sulfoxide (DMSO).<sup>8</sup> DMSO evaporates much more slowly than the organic solvents used, which results in its incorporation into the fibres. The remaining DMSO will eventually evaporate, yielding porous fibres.

It is also important to take into account the surface tension of the solution. Solvents with very high surface tensions (e.g. water) can result in instability arising during the spinning process, and a broad range of fibre diameters in the products and/or the generation of electrosprayed particles rather than fibres. If necessary, a surfactant can be added to reduce the surface tension, but this will be incorporated into the fibres produced and may require post-spinning removal.

### 3.2.2 Processing parameters

The applied voltage needs to be sufficient to overcome the surface tension of the liquids being processed, giving a clear Taylor cone and emitting a polymer jet from the latter. The range of voltages suitable for monoaxial electrospinning in a conventional benchtop apparatus is typically between 5 and 25 kV.

The distance between the needle and collector, the temperature, the humidity and the surrounding air flow can exert a major influence on

the process, as discussed in section 2.5.3. It will be important to monitor both temperature and humidity during the experimental process to ensure reproducibility.

The distance between the spinneret and collector is usually in the range of 10–20 cm. It is possible to place the collector further away, but problems arise because the charged polymer jet seeks the nearest grounded surface on which to discharge. Thus, higher distances can lead to much of the fibre product landing in places other than the collector. If the distance is too short, there will be insufficient time for solvent evaporation and the fibres may fuse or become wrinkled when the residual solvent in the fibre subsequently diffuses out on the collector.

Finally, we should note that these parameters are not mutually exclusive, and that varying one will inevitably have an effect on others (for instance, an increased temperature will result in reduced surface tension and viscosity of the solution). This means that, although the electrospinning process is conceptually simple, its implementation requires some optimisation. A good understanding of the fundamentals of electrospinning (as described in Chapter 2) can significantly facilitate the optimisation process.

### 3.2.3 The spinneret

The spinneret for monoaxial spinning most commonly comprises a simple narrow-bore, blunt-end metal needle. The diameter of this needle can vary, but most commonly researchers work with internal diameters below 1 mm, with around 0.4–0.8 mm probably being most usual. This translates to needles of gauge 18–22. In general, this simple spinneret design can be used to achieve successful spinning. A blunt end rather than a tapered end for the needle exit is important as the size and size distribution of the products increase with an increase in needle tip angle. Blunt-ended needles can be procured commercially, or a disposable medical hypodermic needle can be adapted by cutting off the bevelled edge of the needle with a pair of scissors and using a filing paper to smooth the end. However, it should be noted that there will be some interactions between the solvent and polymer molecules in the solution and the metal surface of the spinneret. There will exist some attractive forces between these (e.g. between polar groups in the polymer and the electropositive metal surface), which can act counter to the drawing force of the electric field and can pull the polymer solution back into the spinneret. It has been found that coating the spinneret exterior in a non-conducting and non-stick polymer such as

Teflon can reduce these interactions.<sup>9</sup> As a result, the electrical energy can be more efficiently used to elongate and narrow the polymer jet, and narrower fibres can be produced. In addition, strong attractive forces between the polymer jet and a metal spinneret can result in fibres or other solid material becoming attached to the needle, leading to lower yields and potentially to blocking of the exit orifice. This effect too can be ameliorated using a Teflon coating. An epoxy coating can also be used to similar ends.<sup>10</sup>

### 3.2.4 Polymer choice

The choice of polymer will be largely dependent on the intended application. For instance, if a fast-dissolving drug delivery system is required, then a rapidly dissolving hydrophilic polymer such as poly(vinyl pyrrolidone) (PVP) or poly(ethylene oxide) (PEO) should be selected. For extended release, a slow-dissolving polymer (e.g. poly( $\epsilon$ -caprolactone) (PCL) or poly(lactic-co-glycolic acid) (PLGA)) or a water-insoluble system (e.g. ethyl cellulose (EC)) can be employed. This will be discussed in more detail in subsequent sections of this chapter.

Consideration must also be given to compatibility between the polymer and the drug of interest. For instance, if a hydrophilic polymer is being used to form fibres but the drug is very hydrophobic, phase separation is likely to occur, to minimise any hydrophobic/hydrophilic contacts. The fibres will thus be unstable, and their functional performance will change upon storage. Selecting a polymer which can form intermolecular interactions (such as hydrogen bonding, van der Waals forces) with the drug can help to prevent this. The literature refers often to *component compatibility*, by which it means the possibility of forming such interactions to prevent phase segregation and encourage long-term stability. Infrared spectroscopy can be helpful to identify intermolecular interactions, which are indicated by small shifts in peak positions (see section 1.4.4).

A solution of the polymer at an appropriate concentration will need to be prepared for spinning. The concentration range required will vary widely depending on the polymer and solvent (see description of the critical concentration  $c_e$  in section 2.5). By way of example, PEOs with  $M_w \leq 300$  kDa in chloroform or ethanol solvent can be spun from solutions of 3–4% w/v, while very-high-molecular-weight PVP solutions in ethanol ( $M_w = 1300$  kDa) are typically 6–10% w/v,<sup>11</sup> and for the naturally occurring polymer shellac, very high concentrations of around 80% w/v are needed.

### 3.2.5 Starting experimental work

When beginning a new electrospinning experiment, the literature will be an invaluable source of guidance as to suitable experimental parameters to use. Working from these precedents, the best place to start is to prepare a range of polymer solutions in the solvent of interest (i.e. one that dissolves both the polymer and drug at appropriate concentrations). If the best solvent to use is not known, then a solvent screen to explore the solubility of the polymer in a range of solvents commonly used for electrospinning (e.g. acetone, ethanol, dichloromethane, DMF, chloroform, methanol) can be helpful. In the authors' personal experience, we have found that very many materials dissolve in the solvent hexafluoroisopropanol. However, this solvent is both expensive and toxic; if it is to be used then care must be taken to ensure there is no residual solvent in the fibres produced.

If a single solvent does not prove satisfactory – for instance, because the volatility or dielectric constant is inappropriate – then solvent mixtures may offer the opportunity to modulate the solution properties. If a solution can be prepared which is a viscous liquid but flows (that is, it is slightly sticky, like honey), then it is suitable for further study. If the polymer concentration is too high, then a gel will be obtained – this will not flow under gravity, and will not be spinnable.

Once a series of solutions has been prepared, electrospinning can begin. The solution should be loaded into a syringe, taking care to avoid any air bubbles. The spinneret is then fitted to the syringe, which is mounted on to the syringe pump. For viscous solutions, the experimenter may find it preferable to use a syringe with a Luer lock tip, which connects with the needle in a more secure fashion than slip tip syringes.

The solution should be dispensed slowly from the needle; 0.5–6 mL h<sup>-1</sup> is a generally appropriate range. As an approximate guide to flow rate, when a droplet forms at the tip, wipe it away; if the droplet is immediately replaced then the flow rate is in an appropriate range for spinning.

The high-voltage power supply should now be connected to the needle with the mains power remaining switched off for safety, and the collector grounded. Next, increase the voltage slowly from zero until the Taylor cone can be seen, and a long jet of polymer is ejected from it. The process should now be monitored for a few minutes to see if the cone jet remains stable and continuous. If so, then optimisation of the processing parameters can begin in order to fine-tune the morphology and diameter of the fibres. If not, then the polymer concentration will need

to be adjusted. This procedure is depicted in a video article by Leach and co-workers.<sup>12</sup>

It is important to take care when increasing the voltage beyond 18 kV. If the voltage is too high then the flow rate feeding the solution into the needle may not keep up with the electric field drawing the solution out of the needle, and the Taylor cone may disappear into the spinneret (see section 2.5.2 for a detailed description of the effect of processing parameters on the electrospun product).<sup>13</sup>

The next stage is to look at the fibres formed. To do this, we recommend collecting for around 5 s to 1 min on a glass slide or aluminium sheet, and then examining this under a microscope (see section 1.4.1 on nanofibre characterisation by microscopy). By systematically varying the voltage, flow rate and spinneret-to-collector distance from the initially identified parameters it should be possible to produce high-quality monodisperse fibres with a uniform morphology and diameter distribution and no beading or other defects (see sections 2.4 and 2.5).

### 3.3 Fibre properties

By varying the material and processing parameters, it is possible to obtain control of the surface morphology and porosity of the fibres generated. Their surface is generally smooth if the materials and processing parameters are optimised. A high electrical potential or low concentration tends to result in beaded structures with considerably rougher surfaces, however.<sup>14</sup> In very concentrated solutions, as well as the main population of fibres, a secondary population of smaller fibres is often seen.<sup>15</sup> The fibre diameter can be controlled by tuning the solution and processing parameters (e.g. polymer concentration, flow rate, voltage), as described in section 2.5.

Two types of pores are possible with the fibre mats generated by electrospinning. Pores may form on or within the fibres themselves, and there will also be pores between the layers of fibres forming the mat (unless the fibres are electrospun with a very high degree of alignment such as by direct writing melt electrospinning; see Chapter 6 for more details).<sup>15b, 16</sup> A knowledge of pore size and porosity can be important, because in many cases it will affect the performance of the formulation. The pore size will control the size of substances which can pass through the fibre mat (individual molecules will always be able to pass through, but cells require pores of tens of microns to permeate into the mat). The porosity will affect the diffusion rate across the fibre mat, governing for instance how

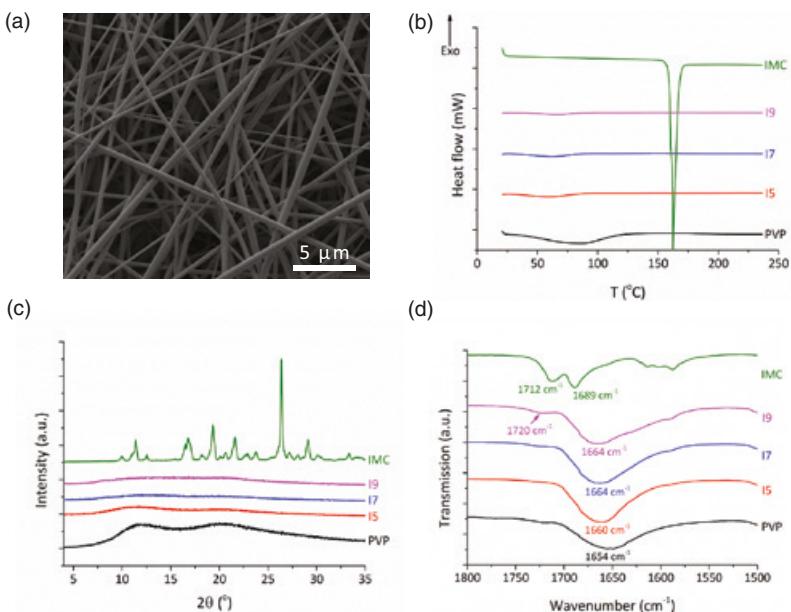
rapidly water can penetrate into the aggregate of fibres and interact with the polymer and drug molecules.<sup>15b</sup> All these properties will influence the drug release profiles observed. If it is necessary to increase the porosity of the fibre mat, porogens (e.g. salt and clay particles) can be added to the polymer solution, and then removed after electrospinning. This usually leads to micron-sized pores where the porogens were originally situated.<sup>17</sup>

### 3.4 Some typical results

After electrospinning, the standard series of characterisations to be performed will comprise an assessment of the fibres using electron microscopy (to visualise morphology), together with X-ray diffraction (XRD) and differential scanning calorimetry (DSC) to observe physical form. Infrared spectroscopy will be employed to look for any interactions between the components of the fibres, and then functional performance studies will be undertaken (see sections 1.4 and 2.8). A typical set of results from one of our studies is given in [Figure 3.2](#). These data are for PVP fibres loaded with indomethacin.<sup>18</sup> Materials were prepared with drug contents between 9.1% and 33.3% w/w in the final fibres. Scanning electron microscopy showed the fibres to have smooth cylindrical morphologies with the optimised parameters ([Figure 3.2\(a\)](#)).

DSC data ([Figure 3.2\(b\)](#)) show that the pure indomethacin powder is crystalline, because there is a large endothermic peak at 159°C which corresponds to melting. Pure PVP has a broad endotherm below around 125°C. This is not a melting event (melting endotherms are sharp peaks), but arises due to the evaporation of water: PVP is a very hygroscopic polymer, and will absorb water from the air. The fibres show no sharp melting endotherms in their DSC traces, with only very shallow endotherms below 100°C. The latter most likely occurs as a result of the fibres absorbing some water upon storage. Therefore, it can be concluded that the drug is amorphously distributed in the fibres. This should accelerate dissolution, since there will be no intermolecular interactions between indomethacin molecules.

The findings from DSC are confirmed by XRD ([Figure 3.2\(c\)](#)). The pattern for pure indomethacin contains many sharp peaks (Bragg reflections), because it is a crystalline material with the molecules arranged in a regular manner. In contrast, there are no sharp peaks in the patterns of either raw PVP or the drug-loaded fibres. This confirms them to be amorphous materials, with no long-range order. The XRD data thus agree well with the DSC findings.



**Figure 3.2** Exemplar data obtained on electrospun fibre formulations, as reported for poly(vinyl pyrrolidone) (PVP)/indomethacin systems by Lopez *et al.* (a) Scanning electron microscopy image of fibres with a 9.1% w/w drug content; (b) differential scanning calorimetry thermograms; (c) X-ray diffraction patterns; and (d) infrared spectra. IMC denotes pure indomethacin, and I5, I7 and I9 are fibres with 9.1%, 23.1% and 33.3% w/w drug loadings. (Adapted from Lopez, F. L.; Shearman, G. C.; Gaisford, S.; Williams, G. R. ‘Amorphous formulations of indomethacin and griseofulvin prepared by electrospinning.’ *Mol. Pharm.* 11 (2014): 4327–4338. This is an open access article published under a Creative Commons Attribution (CC-BY) License.)

Infrared spectra for the formulations are shown in Figure 3.2(d). Indomethacin shows carboxylate vibrations at 1689 and 1712 cm<sup>-1</sup>, while the C=O band for PVP can be observed at 1654 cm<sup>-1</sup>. In the formulations with 9.1% and 23.1% w/w drug contents, the carboxylate bands from the drug and PVP have merged into one broad band, at ca. 1660 or 1664 cm<sup>-1</sup>. This shift in peak positions can be attributed to intermolecular interactions between the two components of the fibres.

When the drug loading was raised to 33.3% w/v, a shoulder on the main peak at 1664 cm<sup>-1</sup> could be seen at 1720 cm<sup>-1</sup>. The observation

of this additional peak can be attributed to the increased drug content. Again, it can be seen that the position is shifted from that in the raw material, indicative of intermolecular interactions. Overall, therefore, it can be concluded that the PVP/indomethacin fibres exist as amorphous solid dispersions, stabilised by intermolecular interactions between the two constituents.

### 3.5 Fast-dissolving drug delivery systems

Fast-dissolving drug delivery systems (FD-DDSs) were initially developed in the late 1970s and have attracted a great deal of investment from the pharmaceutical industry.<sup>19</sup> Rather than releasing their drug cargo in the stomach or small intestine like most oral formulations, FD-DDSs either dissolve or disintegrate in the mouth in a few seconds. Since their drug cargo is released directly and rapidly in the mouth, where there are large numbers of blood vessels, the drug can quickly reach the systemic circulation. Thus, FD-DDSs can enhance bioavailability and give rapid onset of action.<sup>20</sup> This can be very beneficial for a number of situations, for instance with children or the elderly (who might struggle to swallow large tablets), or where very rapid relief of symptoms is required. Unlike tablets, which can be difficult to swallow, FD-DDSs can be applied universally without requiring any water to aid in swallowing.

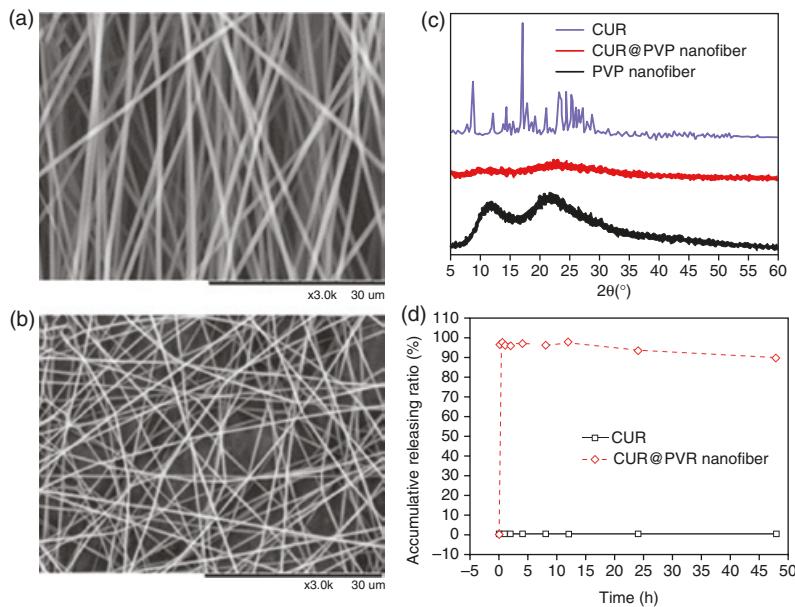
#### 3.5.1 Electrospun fast-dissolving drug delivery systems

Electrospun fibres have been widely explored as FD-DDSs. They have a number of properties which make them very suitable for this application. First, the drug is typically amorphously dispersed in the fibre, usually as an ASD. This means that there is no lattice energy barrier to dissolution, because the drug molecules are randomly arranged in the polymer matrix with no drug–drug intermolecular interactions. The presence of long-chain polymer molecules in the fibre will provide steric hindrance to any recrystallisation occurring, and thus the ASDs can have long-term stability (see section 1.3.4). If the fibres are made of a water-soluble and fast-dissolving polymer, then they will dissolve very rapidly into an aqueous medium. They are aided in this by the high surface-area-to-volume ratio of electrospun fibre mats, coupled with their high porosity; a high specific surface area accelerates dissolution, and the high porosity makes water ingress into the centre of the fibre mat easy and rapid.

One of the first reports of an electrospun FD-DDS came from Zhu's group in 2009.<sup>21</sup> Fibres loaded with the non-steroidal anti-inflammatory drug (NSAID) ibuprofen were prepared based on PVP with a molecular weight of 58 kDa. Using a polymer concentration of 30% w/v, fibres could be electrospun with drug loadings of 20% and 33% w/w in the dried products. The fibres had smooth cylindrical morphologies, although at the higher loading some drug particles could be seen at the fibre surface. Analysis by DSC and XRD proved the fibres to be amorphous materials, with no evidence of any crystalline drug being present. When added to water, the fibres disintegrated into small aggregates in less than 10 s, and all the drug loading was freed into solution within 1 min. Since the fibre mats had thicknesses of less than 1 mm, it was proposed that they have great potential for use as oral fast-dissolving films. The mats are also flexible, and can be easily cut into different shapes suitable for sublingual (under the tongue) or buccal (to the cheek) delivery.

PVP is a particularly useful polymer for FD-DDSs, because it is very hydrophilic and dissolves rapidly into water. A range of drug-loaded PVP fibres have thus been reported, containing active ingredients as diverse as irbesartan (used for treating high blood pressure),<sup>22</sup> ketoprofen (an NSAID),<sup>23</sup> mebeverine hydrochloride (an antispasmodic drug also used for dental analgesia),<sup>24</sup> vitamin D,<sup>25</sup> isosorbide dinitrate (used for treating angina),<sup>26</sup> borneol<sup>27</sup> (a common ingredient in traditional Chinese medicine) and curcumin<sup>28</sup> (a natural product thought to have a range of health benefits). In all cases, the formulations behave similarly. Fibres are produced which are largely smooth and cylindrical, and contain the drug in the amorphous form (as demonstrated by XRD and DSC). The fibre mats disintegrate very rapidly (in a few seconds) and free all their drug loading into solution in a few minutes. Exemplar data for curcumin-loaded systems<sup>28</sup> are given in Figure 3.3.

Other water-soluble polymers are also suitable for use in FD-DDSs, and poly(vinyl alcohol) (PVA) fibres loaded with caffeine or riboflavin have been reported by Li *et al.*<sup>29</sup> As previously, the drug was found to be loaded in the fibres in the amorphous physical form, and the formulations released their drug cargo very rapidly (within 4 min). The fibres were found to perform much better than analogous cast films (made by pouring the drug/polymer solution into a Petri dish and allowing the solvent to evaporate slowly), with significantly faster release from the former. This can be attributed both to the higher surface area and porosity of the fibres, and also to the rapid nature of electrospinning preventing any crystallisation of the active ingredients. Crystallisation is much more likely when cast films are made, because the solvent evaporation rate is slow and



**Figure 3.3** Experimental data for curcumin (CUR)-loaded poly(vinyl pyrrolidone) (PVP) fibres. Scanning electron microscopy images of (a) pure PVP and (b) PVP/curcumin fibres show them to comprise smooth cylindrical entities; (c) X-ray diffraction (XRD) shows that, while the pure drug (blue) is crystalline with many Bragg reflections in its XRD pattern, the fibres (red and black) are amorphous systems; and (d) the fibres release their drug loading much more quickly than pure curcumin dissolves.  
(Reproduced with permission from Wang, C.; Ma, C.; Wu, Z.; Liang, H.; Yan, P.; Song, J.; Ma, N.; Zhao, Q. ‘Enhanced bioavailability and anticancer effect of curcumin-loaded electrospun nanofiber: *In vitro* and *in vivo* study.’ *Nanoscale Res. Lett.* 10 (2015): 439. This is an open access report published under the Creative Commons Attribution 4.0 International License.)

therefore there is time for molecular reorganisation to take place. PEO has also been explored for accelerating drug release, for instance for the beta-blocker carvedilol.<sup>30</sup>

### 3.5.2 Caveats

It is important to note that simply electrospinning fibres containing a drug and hydrophilic polymer will not necessarily result in the very fast release discussed here. If the drug loading is too high (above its solubility limit in the polymer) then crystallisation may occur, meaning that for at

least a portion of the drug there is an energy barrier to dissolution. This has been seen by Taepaiboon *et al.* when making PVA-based fibres.<sup>31</sup> The presence of some crystalline drug in the fibres led to their performing similarly to cast films in terms of drug release.

The molecular weight of the polymer and the release environment are also important. For instance, Ahmad's group prepared PVP fibres loaded with the NSAID indomethacin, using PVP with a molecular weight of 1.3 million Da.<sup>32</sup> Although drug release was accelerated compared to the raw indomethacin, this occurred over around 30 min, much slower than the systems discussed above. This can be ascribed to the high molecular weight of PVP used, which results in its molecules being very long. There will be extensive entanglements between the PVP molecules in the fibre, which will take time to unravel. This will lead to slower dissolution than with the lower-molecular-weight PVPs used above.

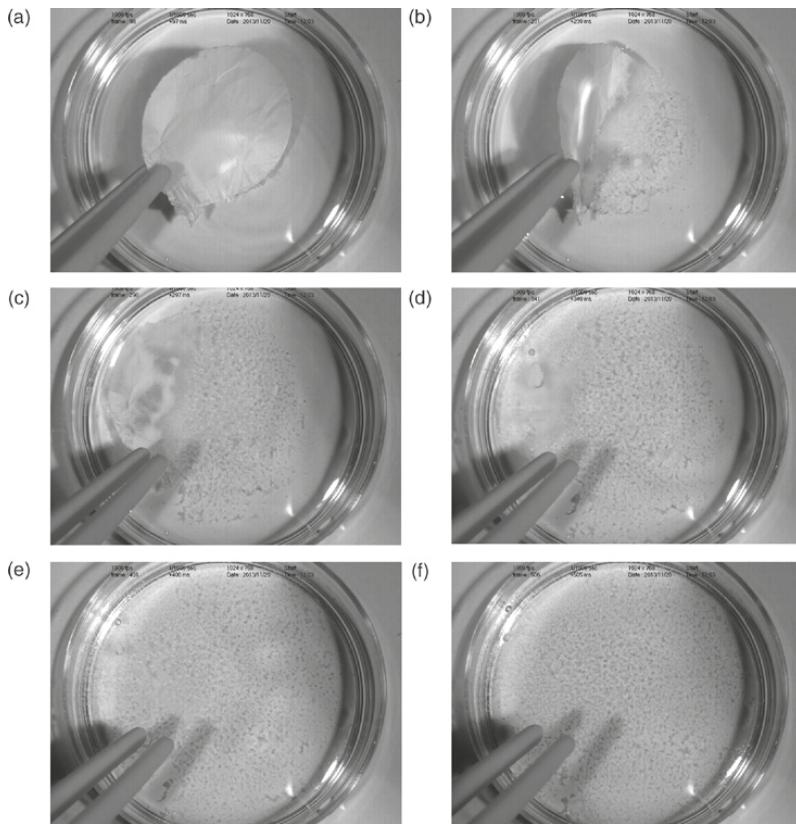
In another example, Baskakova *et al.* explored PVP-based fibres for drug delivery to the eye.<sup>33</sup> In an *in vitro* model of the eye, release occurred over more than 100 h. This can be ascribed to the low volumes of fluid in the eye (< 5 mL, as opposed to the 1 L scale typically used for dissolution testing; see section 1.4.5.1).

### 3.5.3 Multicomponent formulations

It is possible to prepare systems more complex than a simple binary mixture of drug and polymer. This can be helpful to overcome some of the problems associated with oral FD-DDSs. Since the drug is released in the mouth, it is important to consider the taste (palatability) of the formulation. Many drugs are bitter-tasting, and if the formulation tastes unpleasant then there is a risk a patient will not take the required medication.

In one approach to solve this issue, Samprasit *et al.* reported three-component fibres made of PVP, meloxicam (an NSAID) and a cyclodextrin (CD).<sup>34</sup> CDs are cup-like molecules with a hydrophilic exterior and hydrophobic interior. They can encapsulate hydrophobic molecules in their interior, thus hiding them from an external aqueous environment. CDs might hence be used to improve the solubility and mask the taste of hydrophobic active ingredients. Additional fibre formulations were prepared containing menthol and aspartate as taste-masking agents. While the CD alone was not sufficient to hide the bitter taste of meloxicam in palatability studies, the addition of menthol and aspartate was able to do so effectively.

Fibres may also be prepared containing combinations of drugs. For instance, Illangakoon *et al.* produced fibres of PVP loaded with paracetamol and caffeine (often used together to treat colds and influenza).<sup>19c</sup>



**Figure 3.4** Images taken with a high-speed camera depicting the rapid disintegration of paracetamol/caffeine-loaded poly(vinyl pyrrolidone) (PVP) fibres prepared by Illangakoon *et al.* Stills from a video are given (a) 0; (b) 133; (c) 200; (d) 243; (e) 303; and (f) 408 ms after the fibres were added to a Petri dish containing simulated saliva. (Reproduced with permission from Illangakoon, U. E.; Gill, H.; Shearman, G. C.; Parhizkar, M.; Mahalingam, S.; Chatterton, N. P.; Williams, G. R. ‘Fast dissolving paracetamol/caffeine nanofibers prepared by electrospinning.’ *Int. J. Pharm.* 477 (2014): 369–379. Copyright Elsevier 2014.<sup>19c</sup>)

The fibre mats disintegrated within 300 ms (Figure 3.4), and both drugs were fully released within 200 s. A raspberry flavouring was also incorporated into the fibres to help with taste masking. Vrbata *et al.* have similarly reported fibres containing both sumatriptan succinate and naproxen, which might be used as oral FD-DDSs to treat migraines.<sup>35</sup>

More complex systems can also be generated containing multiple excipients. One example of this comes from Yu *et al.*<sup>36</sup> PVP fibres were prepared loaded with the antioxidant ferulic acid (FA), sodium dodecyl sulfate and sucralose. Sucralose was added to mask the taste of the very bitter FA. Sodium dodecyl sulfate acts as a permeation enhancer, aiding the drug to pass through the mucosa in the mouth to reach the systemic circulation. The fibres could increase the FA permeation rate by more than 13-fold.

### 3.5.4 Stability

The stability of ASDs is a major concern for the pharmaceutical industry, because of the propensity for an amorphous system to convert to a crystalline material over time. It is vitally important that the medicine taken by a patient has the intended performance. This might well not be the case if there is recrystallisation during storage. The stability of electrospun FD-DDSs has been studied by several authors. Brettmann *et al.* prepared indomethacin/PVP fibres and found that they remained amorphous after being stored for 6 months in a desiccator at 40°C.<sup>37</sup> Illangakoon and co-workers determined that PVP fibres containing mebeverine hydrochloride remained as ASDs over 12 months' storage in a desiccator.<sup>24</sup>

Lopez and colleagues investigated in detail long-term stability for PVP-based fibres loaded with either indomethacin or griseofulvin (an antifungal agent).<sup>18</sup> These active ingredients were chosen because of their very different glass-forming behaviour. Indomethacin can be made amorphous very easily and forms stable glasses, but amorphous griseofulvin is known to convert rapidly to the crystalline state. Both drugs could be converted into ASDs with PVP, even with drug loadings of up to 33%. Drug release from all the composites was very rapid (< 10 min). In stability studies, there was some evidence for phase separation of the active ingredient and polymer after 8 months of storage in a desiccator, but no crystallisation was observed and the fibres remained amorphous after this time.

Nagy's team have also looked in detail at stability, in their case for poly(vinylpyrrolidone/vinyl acetate) (PVPVA) FD-DDSs loaded with itraconazole.<sup>38</sup> The amorphous form was preserved over 1 year at 25°C and 60% relative humidity (RH), but at 40°C and 75% RH some recrystallisation was observed over 3 months.

Overall, it appears that electrospun ASDs can retain their stability for a prolonged period of time. However, the hydroscopic nature of the polymers used for such systems can lead to recrystallisation at high

humidities, since the fibres will be able to absorb moisture. This will enhance molecular mobility, allowing the drug molecules to reorient themselves into regular arrangements and form crystals. Packaging the formulations under nitrogen should prevent this instability from arising, however, and is an eminently affordable option for industry (crisps are packaged under nitrogen to keep them fresh).

### 3.6 Extended-release systems

The United States Pharmacopoeia (USP) defines a modified-release dosage form as one where ‘the drug release characteristics of time course and/or location are chosen to accomplish therapeutic or convenience objectives not offered by conventional dosage forms such as solutions, ointments, or promptly dissolving dosage forms’.<sup>39</sup> Extended-release systems (also known as controlled-release, prolonged-release, or sustained or slow-release) free their drug cargo over a prolonged period of time in the body. This can be beneficial to avoid there being an excessively high drug loading in the systemic circulation, or for drugs which can be degraded in the blood. To achieve extended release with electrospun fibres, a polymer is chosen which dissolves or degrades slowly, or one which is insoluble. As for the FD-DDSs, it is usual that the fibres comprise ASDs, with the drug amorphously distributed in the fibres, but because the polymer itself is slow to dissolve or does not dissolve at all, drug release should be delayed.

Extended-release fibres may be prepared from non-biodegradable or biodegradable polymers.<sup>40</sup> The former do not degrade in the body, while the latter are broken down into small components over time. Biodegradable polyesters such as PCL, poly(glycolic acid) (PGA), PLA and PLGA have been widely studied.<sup>40</sup> Suitable choices of non-degradable polymers might be polyurethane, polycarbonate or nylon-6. Naturally occurring biopolymers (e.g. silk, collagen, chitosan, gelatin or alginate) are also appropriate. However, biopolymers are usually extracted from natural sources and invariably have batch-to-batch variations in molecular weight, purity, distribution of charged groups and crystallinity. This material inconsistency makes it difficult to generate reproducible electrospun products from different batches of material, and complicates practical applications.<sup>41</sup> In addition, owing to their strong inter- and intramolecular forces, many biopolymers only dissolve in a limited number of solvents that are unsuitable for electrospinning (e.g. polysaccharides such as cellulose and chitin, proteins such as collagen).

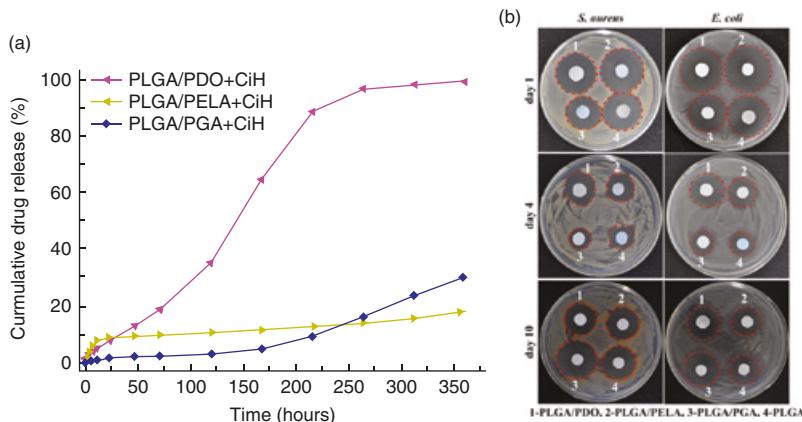
They may also form an ionic solution; when such solutions are electrified, the ionic groups generate high repulsive forces, disrupting continuous fibre formation and leading to defects (e.g. with chitosan and gelatin). Biopolymers are thus often mixed with a secondary polymer such as PEO or PVA to facilitate the electrospinning process.

### 3.6.1 Applications

Kenawy was the first to report an extended-release electrospun fibre system.<sup>1</sup> PLA, PEVA and blend fibres were prepared loaded with the antibiotic tetracycline hydrochloride, and as a result release could be extended over more than 120 h. Such systems have a range of applications, most likely in the form of wound dressings or implantable formulations. One obvious indication in which they might be used is the treatment of cancer, and a number of studies have been reported looking at electrospun fibres in this regard. PLGA fibres loaded with the anticancer drug paclitaxel have been shown to give release extended over 60 days, and to inhibit the proliferation of glioma cancer cells effectively.<sup>42</sup> Fibres made from a mixture of PLGA and PLA and loaded with cisplatin, another anticancer drug, also show promise for cancer treatments, with drug release occurring over at least 30 days, and reductions in cancer cell viability.<sup>43</sup> In addition, the inclusion of targeting molecules in the fibres can help target drug delivery to particular cell types; for instance, folic acid has been incorporated into fibres specifically to deliver viral vectors to cancer cells, while normal cells are unaffected.<sup>44</sup>

Other applications of extended-release fibres include in regenerative medicine. Puppi *et al.* electrospun PLGA fibres containing retinoic acid, with the aim of making scaffolds for tissue regeneration.<sup>45</sup> Their materials were seen to retain their morphology over 12 weeks, with continuous drug release during this time. Cells were able to proliferate on the fibre mats. PCL fibres loaded with heparin have been reported to release around 50% of their drug loading over 14 days, and to have potential for delivering heparin to the site of vascular injury to prevent stenosis (narrowing of the arteries).<sup>46</sup> More details on tissue-engineering applications are given in section 3.12.

The prevention of infection in wounds or after surgery is another area to have attracted attention. For instance, PCL fibres with potential applications in hernia repair have been produced containing antibiotics or antibacterial agents.<sup>47</sup> Zhang *et al.* made blends of PLGA and poly(dioxanone) (PDO), poly(ethylene glycol)-*b*-poly(D,L-lactide-co-glycolide) (PELA) or PGA loaded with ciprofloxacin hydrochloride, moxifloxacin or moxifloxacin



**Figure 3.5** Data obtained by Zhang *et al.* for poly(lactic-co-glycolic acid) (PLGA) blend fibres loaded with ciprofloxacin hydrochloride (CiH). (a) Variation in the blend of polymers used leads to distinctly different release properties, with drug release occurring over more than 200 h in some cases. (b) The CiH-loaded fibres are able to inhibit bacterial growth effectively over at least 10 days. PDO: poly(dioxanone); PELA: poly(ethylene glycol)-*b*-poly(D,L-lactide-co-glycolide); PGA: poly(glycolic acid). (Reproduced with permission from Zhang, Z.; Tang, J.; Wang, H.; Xia, Q.; Xu, S.; Han, C. C. ‘Controlled antibiotics release system through simple blended electrospun fibers for sustained antibacterial effects.’ *ACS Appl. Mater. Interfaces* 7 (2015): 26400–26404. Copyright American Chemical Society 2015.)

hydrochloride.<sup>48</sup> Selected data from this work are shown in Figure 3.5. The blend of polymers used permits control to be exercised on the release pattern, with some formulations able to give approximately linear drug release over more than 200 h. The fibres could also effectively inhibit bacterial growth *in vitro*.<sup>48</sup> These systems might effectively be used as an implant to prevent infection after abdominal surgery.

Other researchers have looked at wound healing. PCL and PLA fibres containing a vitamin D<sub>3</sub> derivative have been electrospun and found to free their drug cargo over around 10–15 days, potentially suitable for wound healing.<sup>49</sup> Polyvinylidene fluoride fibres have also been explored in this setting, and release of the active ingredient observed over 45–90 h.<sup>50</sup> In other work, Zamani and co-workers explored metronidazole benzoate-loaded PCL fibres for periodontal diseases (inflammation and degeneration of the gums).<sup>51</sup> Drug release was prolonged over 20 days.

Extended release over shorter time periods than those discussed above can also be useful. For instance, it takes approximately 1 day for a formulation taken orally to pass through the gastrointestinal tract, and so a system able to extend release over the 10–20-h time period could be beneficial for making once-daily medicines. Transdermal (through skin) delivery over this time period would also lead to potent formulations. Yu *et al.* have performed work in which poly(acrylonitrile) (PAN) fibres were loaded with acyclovir and explored as a potential transdermal drug delivery system.<sup>52</sup> As for the fibres discussed above, the drug was amorphously distributed in the polymer matrix at lower loadings (10% w/w), but at higher drug concentrations (up to 20% w/w) some crystalline material was observed. Release was sustained over 12 h. In other work, indomethacin-loaded fibres prepared from EC and zein (water-insoluble polymers) have been found to extend drug release over more than 20 h.<sup>53</sup> This type of system might thus be useful for oral administration.

Vaginal applications have also been proposed for extended-release electrospun fibres, as reviewed by Blakney *et al.*<sup>3b</sup>

### 3.6.2 Drawbacks and release mechanisms

One key drawback encountered with using monolithic fibres from monoaxial spinning for extended release is that, in the majority of cases, there is a burst of release at the start of the process. This is because of the fibre mat's very high surface-area-to-volume ratio, which results in a significant proportion of the drug molecules in the fibres being near to the surface. These can rapidly diffuse out into solution, even if the polymer matrix is slow-dissolving or insoluble. The burst is not a problem in the context of FD-DDSs where all the active ingredient is released very quickly, but with an extended-release formulation the rate of release (and thus the amount of drug reaching the bloodstream) will be much quicker immediately after the medicine has been taken than later on.

The burst release effect can be minimised with a low drug loading, but for clinical applications high doses (10–100 mg of drug per dose) are often required. These necessitate high drug loadings in the fibres, and present additional challenges for sustaining drug release. Higher drug loadings lead to larger amounts of the active ingredient at or near the surface. Many studies have been performed with low loadings (< 1% w/w), and thus the formulations prepared have limited clinical applications. For instance, Ball *et al.*<sup>3a</sup> made fibres from biodegradable polymers and explored them for extended release, with antibacterial applications in mind. Maraviroc, azidothymidine, acyclovir and

glycerylmonolaurate-loaded fibres were generated successfully, and the formulations were found to be non-toxic. Although the fibres provided sustained release in some cases, the drug loading was only 1% w/w, and so the formulations as they stand cannot directly be translated to the clinic.<sup>3a</sup>

If the burst release is a major problem for a particular system, there are several possible solutions. One is coaxial or multi-axial electrospinning, as will be discussed in Chapter 4. Another option is to bond the active ingredient of interest covalently to the polymer and then spin this into fibres, a concept demonstrated by Jalvandi and co-workers in the case of levofloxacin and chitosan.<sup>54</sup>

Natu *et al.* have investigated the effect of drug location and physical form on the release rate from fibres electrospun from slow-dissolving polymers, using acetazolamide and timolol maleate as hydrophobic and hydrophilic model drugs, respectively.<sup>55</sup> At low concentrations (below the drug solubility in the polymer: 1.16–1.55% w/w for acetazolamide; 0.86–0.88% w/w for timolol), the drug was amorphously distributed in the fibres, but crystalline material was observed when the drug content was greater than its solubility in the polymer. With the low concentrations, where the drug was well encapsulated in the fibres, there was only a small initial burst of release. However, at high concentrations (above the drug solubility in the polymer: 12.67% w/w for acetazolamide; 6.99–7.6% w/w for timolol) this burst was much greater, attributed to the presence of crystalline drug particles at the edge of the fibres.

The mechanism of drug release from extended-release fibres can be controlled by one or more of three processes: drug diffusion through the polymer, polymer degradation and drug dissolution into solution.<sup>2a</sup> This makes the details of the release process complex, and it can be difficult to predict how a particular system will behave. It would be intuitively expected that if drug release is controlled solely by the diffusion of the drug through the polymer, then larger-diameter fibres will release more slowly because there will be a longer path through which the drug must diffuse to reach solution. However, this is not always the case, and the diffusion distance is not the only factor which changes with fibre diameter. Verreck *et al.*<sup>56</sup> posited that smaller-diameter fibres might be more tightly packed, inhibiting the swelling of the fibre matrix and the ingress of water. Fibre diameter is typically controlled through formulation properties such as polymer concentration and solvent choice, but these can also affect the dispersion of drug in the fibre products (among other factors), which must be considered when developing monolithic extended-release fibres.<sup>2a</sup>

There are several reports in the literature confirming the importance of considering formulations on a case-by-case basis. Studies performed with PLA (molecular weight = 75–120 kDa) as the polymer matrix found that the rate of drug delivery for tetracycline became slower as the fibre diameter increased from  $220 \pm 60$  nm to  $360 \pm 70$  nm, and to  $830 \pm 280$  nm.<sup>57</sup> However, when the fibres were loaded with chlorotetracycline the release rate became faster with an increase in fibre diameter from 200 nm to 1.6  $\mu\text{m}$ . The opposite release behaviours were thought to be because multiple factors, including the fibres' swelling behaviour and the drug's solubility in the polymer matrix and the release medium, influenced the release profile.<sup>57</sup>

A wide variety of active ingredients can be mixed with a polymer and electrospun into fibres, as has been described above. However, most examples of drug release extending beyond seven days relate to macromolecules or hydrophobic small-molecule drugs.<sup>58</sup> These active ingredients have at least one of the following characteristics: poor solubility in aqueous media, large molecular size and/or favourable intermolecular interactions with the hydrophobic polymers typically used for extended-release formulations. All these characteristics help to retard the rate at which the drugs are freed from the polymer matrix. In contrast, hydrophilic small-molecule drugs are very difficult to formulate for extended release because they have high solubility in the release medium, and more favourable interactions with an aqueous environment than with the polymer in the fibres. These factors promote drug release from the fibres.

## 3.7 pH-controlled delivery

### 3.7.1 Oral administration

When a medicine is given orally, the variations in pH occurring in the intestinal tract offer the potential to provide delayed release. The pH of the stomach is acidic, typically at around 1–2, while that of the small intestine is close to neutral (with variations depending on the exact location). Thus, if a fibre is made of a pH-sensitive polymer, which is insoluble at low pH but soluble at neutral pH, then it might be expected that its drug loading would be released only in the small intestine. This is potentially very useful, because there are many drugs (such as the NSAIDs) which can cause serious irritation to the stomach, and others which are degraded by the acid or enzymes present.

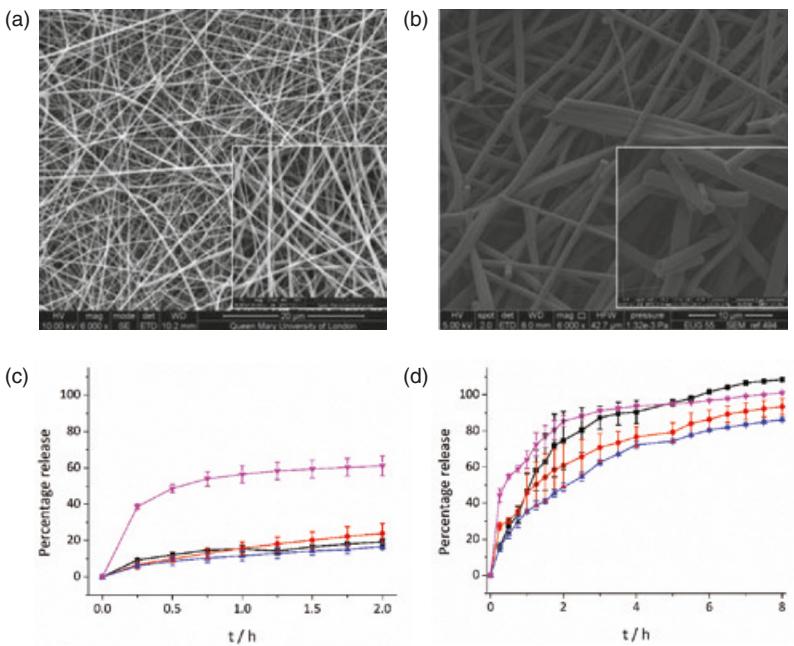
A number of researchers have explored these possibilities. In 2007, Wang *et al.* produced erythromycin-loaded fibres made of

hydroxypropylmethylcellulose phthalate (HPMCP).<sup>59</sup> Erythromycin (an antibiotic) is rendered inactive at low pH, and thus it is desirable to prevent it dissolving in the stomach. Since HPMCP is insoluble below pH 5.5, it was reasoned that fibres made of this polymer could have potential here. As expected, drug release was much slower at pH 1.2 than at 6.8.

The Eudragit polymers, a family of materials based on methacrylates, have also received extensive attention for electrospun pH-sensitive drug delivery systems. For instance, Eudragit L100-55 dissolves at pH 5.5, L100 at pH 6.0 and S100 at 7.0; below these pH values, the polymers are insoluble. Thus, targeting to the small intestine should be possible. The polymers can be successfully processed by electrospinning, with Shen *et al.* reporting Eudragit L100-55 fibres loaded with diclofenac sodium in 2011.<sup>60</sup> The formulations were subject to a dissolution test where the fibres were first placed in a pH 1.0 solution (to mimic the stomach) and then transferred to a pH 6.8 buffer (representative of the small intestine). Less than 3% of the diclofenac content was released at pH 1.0, while at pH 6.8 extended release over up to 5 h was observed. These findings suggested that the fibres have potential for targeting release to the small intestine. Similar results have been found by Akhgari and co-workers for indomethacin-loaded fibres.<sup>61</sup>

The work of Shen and Akhgari used an acidic drug (one which can be ionised through loss of a proton). Such active ingredients have solubility which increases with pH, and thus will always have a lesser tendency to be released from a formulation at low pH than under neutral conditions. Illangakoon *et al.* prepared Eudragit L100-55 fibres containing the basic drug mebeverine hydrochloride.<sup>24</sup> This will be more soluble at low pH, and thus one might expect a greater amount of release in conditions representative of the stomach than with an acidic drug. Smooth cylindrical fibres were obtained with drug loadings of 30% w/w or below, but when the loading was increased to 55% w/w the fibres were observed to break apart upon storage ([Figure 3.6\(a\) and \(b\)](#)). As for the FD-DDS materials discussed previously, the fibres comprise ASDs with no evidence for any crystalline drug being present.

After 2 h at pH 2 some 15–25% of the mebeverine loading had been freed into solution, as would be expected given the basic nature of mebeverine. However, the fibres release much less drug at this pH than a Eudragit/mebeverine physical mixture. At pH 6.8, extended release over ca. 8 hours was observed. Thus, using Eudragit-based fibres can be a suitable route to prepare delayed-release oral formulations even for basic drugs. However, it should be noted that the same burst release issue as



**Figure 3.6** Experimental data on mebeverine chloride-loaded Eudragit fibres prepared by electrospinning. Scanning electron microscopy images of the fibres with (a) 30% and (b) 55% w/w drug contents; (c) drug release at pH 2; and (d) the release profiles obtained at pH 6.8. Release data are shown for fibres containing 5 (■), 15 (●) and 30 (▲) % w/w mebeverine hydrochloride contents, together with a physical mixture of drug and Eudragit as a control (▼). (Reproduced with permission from Illangakoon, U. E.; Nazir, T.; Williams, G. R.; Chatterton, N. P. ‘Mebeverine-loaded electrospun nanofibers: Physicochemical characterization and dissolution studies.’ *J. Pharm. Sci.* 103 (2014): 283–292. Copyright Elsevier 2014.)

described in section 3.6.2 arises with these monolithic fibre systems. Further, some authors have observed significant amounts of release at low pH even with an acidic drug. Karthikeyan *et al.* reported fibres made from a blend of zein (a water-insoluble polymer) and Eudragit S100, loaded with aceclofenac (an acidic NSAID) and pantaprazole (a basic drug used to help prevent side effects when patients are taking NSAIDs for extended time periods).<sup>62</sup> While the fibres were able to prevent pantaprazole release effectively at pH 1, some 25% of the aceclofenac

was freed into solution at this pH. This may be a result of the way in which the fibres were prepared from a mixture of pantaprazole/Eudragit and aceclofenac/zein solutions, but nevertheless shows that simple monolithic fibres are not always effective for pH-targeted release.

### 3.7.2 Anticancer applications

Another route to impart pH sensitivity on electrospun fibres is to incorporate a pH-sensitive agent other than the polymer. Cui's group were able to prepare poly(L-lactic acid) fibres which released their drug cargo faster at lower pHs by the simple expedient of including sodium bicarbonate in the formulation.<sup>63</sup> The fibres contained 5-fluorouracil (5-FU), an anticancer drug, and were found to be able to inhibit the growth of human osteosarcoma cancer cells while being harmless to fibroblast cells. This is because the tumour microenvironment is somewhat more acidic than normal physiological pH. At pH 5.0, sodium bicarbonate will react with the protons present, forming carbon dioxide gas. As the gas leaves the fibres, it creates channels in them. These channels make it easier for water to enter the fibres to dissolve the drug, and for the drug to diffuse into solution. Thus, in this particular instance 5-FU is freed more rapidly in the slightly acidic pH conditions of the cancer environment, and cancer cell proliferation is inhibited over a 4-day *in vitro* culture. At the normal physiological pH (7.4), the sodium bicarbonate in the fibres does not react, and thus no pores are created and drug release is slower. Similar results have been reported for ibuprofen-loaded PLGA fibres containing sodium bicarbonate.<sup>64</sup>

Interactions between the drug and polymer may also be used to provide pH sensitivity, if these change with pH. Salehi *et al.* have reported such a system built on poly(*N*-isopropylacrylamide-co-methacrylic acid-co-vinylpyrrolidone) and the anticancer drug doxorubicin.<sup>65</sup> At pH 7.4, the polymer is ionised and favourable electrostatic interactions with the drug cause release to be slowed. At pH 5.4, representative of the cancer microenvironment, the polymer is uncharged, and thus these interactions are reduced and the drug releases more rapidly.

## 3.8 Pulsatile release

Pulsatile release refers to an on/off pattern of release, with bursts of release separated by periods where no further drug is freed from a formulation. This can be particularly beneficial for conditions which follow

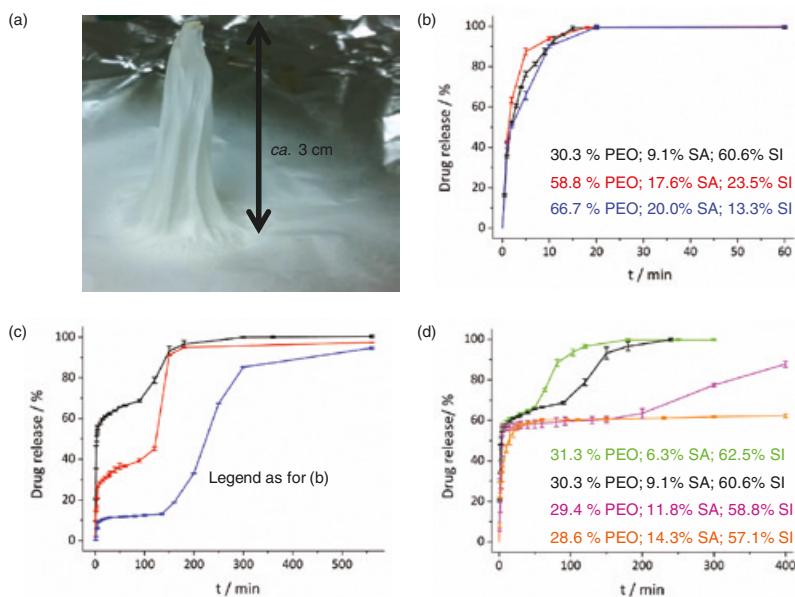
the body's natural circadian rhythms and are worse at particular times of day. Asthma attacks are more common late at night, for instance, while rheumatoid arthritis is worse in the morning. Electrospun fibres have not been much explored for pulsatile release, but there is one report of this modality being achieved.

Kaassis and co-workers made fibres from PEO, sodium alginate (a naturally occurring polysaccharide which is insoluble at low pH but freely soluble at pHs higher than around 5) and sodium ibuprofen.<sup>66</sup> While the other fibre systems discussed in this chapter are obtained as flat mats, in this study the fibres were found to form a 'mountain' shape growing up from the collector (Figure 3.7(a)). Evidence for crystalline sodium ibuprofen was found in XRD and DSC. At pH 6.8 (representative of the small intestine), the fibres free their drug loading within around 20 min (Figure 3.7(b)). However, at pH 3 (representative of the stomach in the fed state) clear pulsatile release is seen (Figure 3.7(c)), with two stages of release and a lag between them. The percentage of drug in the first stage can be controlled through the ibuprofen content of the fibres (Figure 3.7(c)), while the lag between them can be varied by altering the amount of alginate present (Figure 3.7(d)).

### 3.9 Multilayer materials

As discussed above, through variation of the polymer used to prepare electrospun fibres, it is possible to provide a range of drug delivery patterns. It is also possible to produce a mat made of multiple layers of monolithic fibres, with each layer made of a different material, to provide more complex release profiles. Blagbrough's team have used this approach extensively to prepare multilayer meshes for wound-healing purposes.

In the first such study, Alhusein *et al.* used solutions of either PCL or PEVA.<sup>67</sup> By sequentially electrospinning layers of each polymer on to the collector, they could produce a three-layered formulation with alternating PCL/PEVA/PCL layers. The antibiotic tetracycline hydrochloride was either incorporated into all three layers, or just in the central PEVA layer. Simple two-component fibre mats of PCL/tetracycline and PEVA/tetracycline were also prepared. While the PCL/tetracycline formulation released all its drug within 1 day, the PEVA/tetracycline fibres were able to extend this over 8 days. The three-layer PCL/PEVA/PCL system could extend tetracycline release over 14 days; when the drug was present in all three layers, a burst of 60% release was seen in the early stages of the experiment, but incorporating the drug only in the central layer reduced this to 10%, and permitted



**Figure 3.7** Pulsatile release from electrospun fibres comprising poly(ethylene oxide) (PEO), sodium alginate (SA) and sodium ibuprofen (SI), as reported by Kaassis *et al.*<sup>66</sup> (a) The mountain of fibres which was observed to form on the collector; (b) at pH 6.8 the fibres free their drug loading within 15 min; (c) at pH 3 pulsatile release is observed, with the amount of release in the first stage dictated by the ibuprofen content of the fibres; and (d) the lag between the two phases of release can be tuned by varying the sodium alginate content of the fibres. (Reproduced with permission from Kaassis, A. Y. A.; Young, N.; Sano, N.; Merchant, H. A.; Yu, D.-G.; Chatterton, N. P.; Williams, G. R. ‘Pulsatile drug release from electrospun poly(ethylene oxide)–sodium alginate blend nanofibres.’ *J. Mater. Chem. B* 2 (2014): 1400–1407. Copyright Royal Society of Chemistry 2014.)

an almost constant (zero-order) rate of release to be achieved. The PCL/PEVA/PCL system could also effectively inhibit bacterial growth.<sup>68</sup> Similar results were reported using three-layer systems with tetracycline solely in the central layer when these were fabricated from zein or zein/PCL blends.<sup>69</sup> The latter have been found to be potent in preventing biofilm formation, and to be compatible with human skin cells.<sup>70</sup>

This idea of depositing multiple fibre layers was also explored by Huang *et al.*<sup>71</sup> Using ketoprofen as a model drug, polymer/drug solutions

were prepared using PVP and EC. PVP/EC/PVP three-layer systems were produced, with ketoprofen present in each layer. Systems were generated with the different layers having varying thickness by varying the duration of time for which each was electrospun. The products comprised ASDs, and the three-layer system was found to give a two-phase (*biphasic*) drug release pattern. The PVP layers provided a burst of release at the start of the experiment, and then extended release ensued from the EC layer. Such formulations could be beneficial because the initial burst provides a *loading dose*, rapidly increasing the drug concentration in the plasma to therapeutic levels; the second, slow-release phase can then maintain the concentration in the effective range. Huang and colleagues were able to control the extent of the burst by varying the layer thickness.<sup>71</sup>

### 3.10 Thermoresponsive systems

A number of polymers are *thermosensitive*; that is, they change properties in response to temperature variation. Such polymers have been explored in electrospinning, with most attention paid to poly(*N*-isopropyl acrylamide) (PNIPAAm). This polymer is hydrophilic at room temperature, and can dissolve in water. However, at around 32°C it undergoes a chain-to-globule transition and becomes hydrophobic. The temperature at which this arises is known as the *lower critical solution temperature* (LCST). Although PNIPAAm can be electrospun alone,<sup>72</sup> it is more usually blended with carrier polymers such as PEO to facilitate the spinning process.<sup>73</sup> The thermosensitive properties of PNIPAAm are preserved after spinning, meaning that the fibres generated change their hydrophilicity in response to temperature.

Thermosensitive fibres have attracted some attention for drug delivery purposes. Fibres made from a blend of PNIPAAm and poly(2-acrylamido-2-methylpropanesulfonic acid) release a nifedipine cargo more rapidly below the LCST than above.<sup>74</sup> PNIPAAm/PEO blend fibres containing vitamin B<sub>12</sub> behave similarly,<sup>75</sup> as do PNIPAAm/EC formulations.<sup>76</sup> Although the exact behaviour can be tuned by varying the composition of the fibres, the *in vivo* applications of these particular formulations are not clear: a system able to control release in response to small variations in body temperature would be useful, but PNIPAAm cannot deliver this because its LCST is well below the physiological temperature range.

Where systems based on LCST polymers might be very useful, however, is in the treatment of wounds. When a wound dressing is changed, it

is common for some tissue to be pulled away from the wound: this results in what are known as secondary injuries. Thermosensitive materials can be used to control cell adhesion, because interactions between cells and the polymer are much stronger above the LCST than below it. Thus, a thermosensitive wound dressing could be very beneficial if the LCST was a little below body temperature. A local low-temperature treatment would drop the temperature of the skin below the LCST, reduce the strength of interactions between the dressing and the cells around the wound, and hence prevent secondary injuries when the dressing is removed. Electrospun fibres have many properties making them suitable for wound dressings, since their high porosity allows them to absorb any fluids exuded from the wound, and their web structure helps to facilitate cell growth.

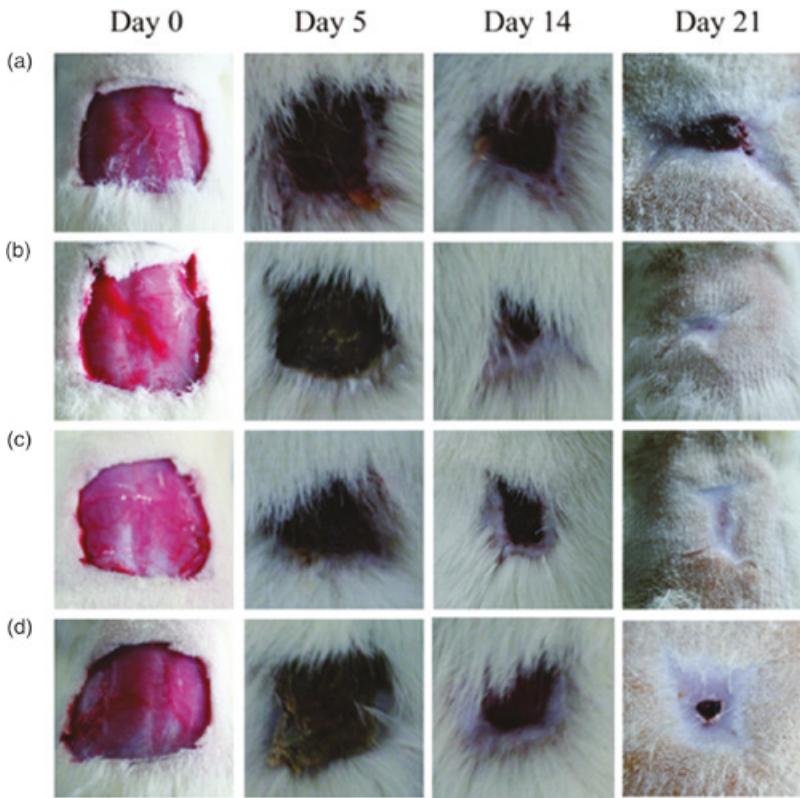
Thermoresponsive electrospun wound dressings have recently been reported by Li *et al.* using either PNIPAAm<sup>77</sup> or poly(di(ethylene glycol) methyl ether methacrylate)<sup>78</sup> as the thermosensitive polymer in a blend with poly(L-lactic acid-co-ε-caprolactone) (PLCL). The anti-bacterial drug ciprofloxacin was incorporated into the fibres to prevent infection. Ciprofloxacin was freed from the fibres over more than 200 h, and the formulations could effectively prevent bacterial growth. The thermosensitive mats were able to accelerate wound healing over fibres prepared from PLCL and ciprofloxacin alone (Figure 3.8).

## 3.11 Emulsion and suspension electrospinning

The above discussion has focused on electrospinning a solution of a drug and polymer. In addition, it is possible to process emulsions and suspensions, and the resultant fibres can have a number of beneficial properties.

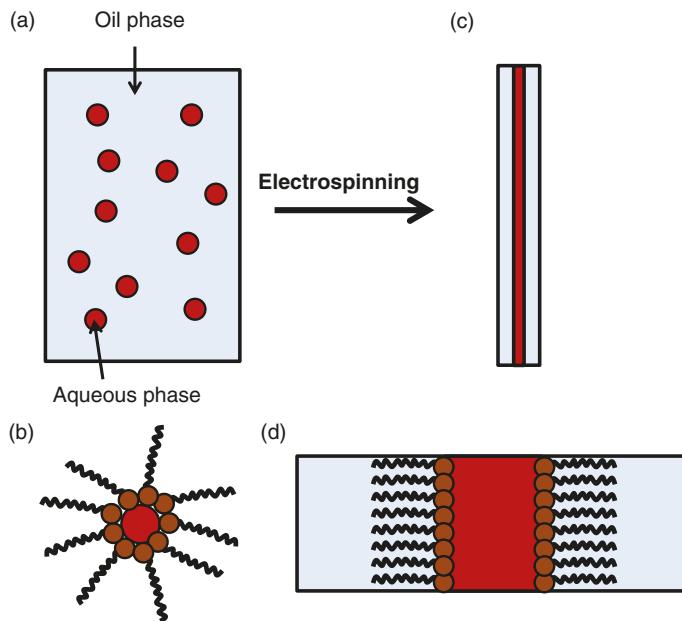
### 3.11.1 Emulsions

A range of fibres have been prepared using monoaxial emulsion electrospinning. For instance, Jing's group made a water-in-oil emulsion stabilised with the surfactant sodium dodecyl sulfate.<sup>79</sup> The hydrophilic active pharmaceutical ingredient (API) doxorubicin hydrochloride was present in the water phase. A block copolymer (poly(ethylene glycol)-poly(L-lactic acid)) dissolved in chloroform was used to form the fibres. This copolymer is amphiphilic and has both hydrophobic and hydrophilic sections, which means that it should be able to interact with both



**Figure 3.8** Images showing the healing of wounds inflicted on rats and treated with electrospun dressings. The wounds were treated with (a) a commercial gauze; (b) poly(*N*-isopropyl acrylamide) (PNIPAAm)/poly(*L*-lactide-co- $\epsilon$ -caprolactone) (PLCL)/ciprofloxacin fibres with a 1:1 mass ratio of the polymers; (c) PNIPAAm/PLCL/ciprofloxacin fibres with polymer mass ratio 1:2; and (d) PLCL/ciprofloxacin fibres. It is clear that the formulations containing PNIPAAm lead to faster wound healing. (Reproduced with permission from Li, H.; Williams, G. R.; Wu, J.; Wang, H.; Sun, X.; Zhu, L. M. 'Poly(*N*-isopropylacrylamide)/poly(*L*-lactic acid-co-caprolactone) fibers loaded with ciprofloxacin as wound dressing materials.' *Mater. Sci. Eng. C* 79 (2017): 245–254.<sup>77</sup> Copyright Elsevier 2017.)

parts of the emulsion. After electrospinning, the fibres were found to have an aqueous core/hydrophobic shell structure, despite having been produced from a monoaxial process. This was attributed to a separation of the hydrophobic and hydrophilic components of the fibres, with the surfactant at the interface. This spontaneous self-assembly of core/shell



**Figure 3.9** Schematic showing how a core/shell fibre can be generated from emulsion electrospinning. (a) A water-in-oil emulsion, with droplets of water dispersed throughout a continuous oil phase; (b) a close-up of a single water droplet, showing how a surfactant (an amphiphilic molecule with a hydrophilic head and hydrophobic tail) is used to stabilise the emulsion. Surfactant molecules will be arranged around the edge of the water droplets, with their hydrophilic head groups touching the droplet and their hydrophobic tails dangling into the oil phase. (c) After electrospinning a core shell fibre is formed, with (d) the surfactant molecules collected along the oil/water interface to minimise unfavourable interactions.

structures is very commonly seen in emulsion spinning. The mechanism underlying this is illustrated in Figure 3.9, and has been discussed in detail by Wang and Wang.<sup>80</sup>

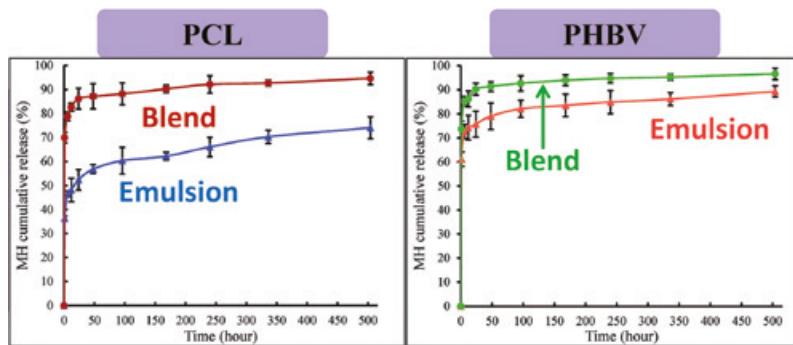
The presence of a water-soluble active ingredient in the core of fibres spun from water-in-oil emulsions has the benefit of ameliorating any initial burst release. This was shown for the model drugs cefradine and 5-FU: fibres prepared from emulsion spinning had reduced initial burst release and a slower release rate than analogous fibres made by standard monoaxial solution spinning.<sup>81</sup> A lack of burst release from emulsion spun fibres was also noted with doxorubicin hydrochloride by Xu *et al.*<sup>79</sup>

Further, emulsion electrospinning has been demonstrated to modulate fibre properties, for instance reducing the fibre diameters in comparison with monoaxial solution electrospinning.<sup>82</sup> Sy *et al.* observed that the mixing of phases of different rheological properties in an emulsion caused electrospinning to occur at a much lower range of liquid viscosity than that for a conventional monoaxial solution system.<sup>82</sup>

The emulsion approach has attracted particular attention for the production of protein-loaded fibres. The therapeutic efficacy of protein drugs is highly dependent on their three-dimensional structure (known as the tertiary structure), and this is easily degraded by the organic solvents commonly used for electrospinning. Incorporating them into water droplets in an organic continuous phase could be an effective route to prevent protein denaturation and degradation.<sup>83</sup> This was reported for the model protein bovine serum albumin (BSA) in 2008.<sup>84</sup> Core/shell fibres were obtained via emulsion electrospinning, and protein stability appeared to be preserved throughout the process.

Protein-loaded core/shell fibres prepared through emulsion spinning have been explored for a range of applications. These include the delivery of growth factors to encourage tissue regeneration. For instance, PLCL fibres containing vascular endothelial growth factor have been shown to have potential for cardiac regeneration.<sup>85</sup> Vascular endothelial growth factor was released over more than 28 days, and human bone marrow-derived mesenchymal stem cells could proliferate on the fibres over a 20-day culture period. Emulsion fibres carrying a protein payload have further been investigated in bone tissue engineering, *inter alia*.<sup>86</sup>

Another benefit of emulsion spinning is in incorporating APIs of opposing polarity to the polymer used. As discussed in section 3.2.4, if a hydrophobic drug is loaded in a hydrophilic polymer (or vice versa) there is a risk of phase separation occurring on storage. This can be a problem if the extended release of a hydrophilic drug is required, since most of the suitable polymers for this are hydrophobic. Similarly, since hydrophilic polymers are used for fast-dissolving systems, it can be difficult to incorporate hydrophobic entities into these. Emulsion electrospinning was used to overcome the latter issue in the case of the lipophilic API celecoxib.<sup>87</sup> Emulsion droplets could be seen in the fibres produced and the drug was amorphously distributed in the fibres, boding well for improved dissolution properties (although these were not tested). The situation of a hydrophilic drug in a hydrophobic polymer has been studied by Ramakrishna's team.<sup>88</sup> Emulsion and blend fibres of PCL or poly(3-hydroxybutyric acid-co-3-hydroxyvaleric acid) (PHBV) loaded with metformin hydrochloride or metoprolol tartrate were generated,



**Figure 3.10** The release of metformin hydrochloride (MH) from blend and emulsion poly( $\epsilon$ -caprolactone) (PCL) and poly(3-hydroxybutyric acid-co-3-hydroxyvaleric acid) (PHBV) fibres. Reduced burst release and slower rates are seen with the emulsion system. (Reproduced with permission from Hu, J.; Prabhakaran, M. P.; Tian, L.; Ding, X.; Ramakrishna, S. 'Drug-loaded emulsion electrospun nanofibers: characterization, drug release and *in vitro* biocompatibility.' *RSC Adv.* 5 (2015): 100256–100267.<sup>88</sup> Copyright Royal Society of Chemistry 2015.)

and the initial burst and release rate were both reduced in the emulsion system (Figure 3.10).

It is not always the case that fibres made by emulsion electrospinning are superior to those from monoaxial solution spinning, however. A study by Zhao *et al.* fabricated PCL fibres loaded with L-ascorbic acid-2-phosphate magnesium using standard blend spinning (using a co-dissolving solution of drug and polymer), as well as with emulsion and coaxial electrospinning.<sup>89</sup> In this instance, the optimum fibres spun from solutions were found to have better mechanical properties and a reduced burst of release than the emulsion analogue.

### 3.11.2 Suspensions

Suspensions of small (typically nanoscale) particles can be converted into fibres by electrospinning. For example,  $\text{CaCO}_3$  microparticles incorporating model drugs have been embedded in PLGA fibres by electrospinning.<sup>90</sup> The fibres appeared beaded owing to the presence of particles. However, it is possible to prepare relatively smooth cylindrical fibres with minimal beading, if the experimental conditions are appropriately optimised.<sup>91</sup>

Magnetic nanoparticles, such as those of  $\text{Fe}_3\text{O}_4$ , have attracted particular attention for inclusion into fibres, because of their potential in biosensing, imaging and cancer treatment (e.g. inducing magnetic hyperthermia to destroy cancer cells).<sup>92</sup> Wang *et al.* produced systems made of cellulose derivatives and containing  $\text{Fe}_3\text{O}_4$  nanoparticles as well as either indomethacin or aspirin.<sup>93</sup> This resulted in magnetic fibres, but the inclusion of nanoparticles did not affect drug release. Other inorganic materials to have been electrospun include hydroxyapatite (useful for bone repair applications),<sup>94</sup> clays,<sup>95</sup> drug-loaded metal hydroxide nanoparticles<sup>96</sup> and silicas.<sup>97</sup> Jalvandi *et al.* have shown that by covalently binding levofloxacin to mesoporous silica nanoparticles and then spinning these into PCL fibres, drug release can be slowed and the initial burst reduced, compared to PCL/levofloxacin fibres with no nanoparticles present.<sup>98</sup>

Protein nanoparticles can also be prepared to help prevent degradation. This has been demonstrated by first encapsulating bone morphogenetic protein-2 (BMP-2) in BSA nanoparticles, and then electrospinning these into fibres based on poly( $\epsilon$ -caprolactone-co-poly(ethylene glycol)).<sup>99</sup> The rationale for this is that the BMP-2 is fragile, and could become degraded during electrospinning if processed in solution. Incorporating it into the BSA particles can protect it from degradation. This approach was found to give formulations able to repair defects in the skull of rats.

Finally, although most commonly researchers work with mixed solutions of a drug and polymer to generate fibres in the form of ASDs, it is also possible to electrospin suspensions of API particles.<sup>100</sup> This could be attractive for making extended-release systems, for instance.

### 3.12 Tissue-engineering applications

Electrospun fibres have attracted much attention for tissue-engineering applications, where they are explored as scaffolds to replace or heal biological tissue. The structure of the mat mimics the structural component of the extracellular matrix, a scaffold or network of nanofibrous proteins and gels of polysaccharides secreted by cells to give mechanical support to surrounding cells. This, coupled with the porosity of the mats (which means that nutrients and cellular waste can diffuse in and out, and it is possible for cells to infiltrate into the scaffold), can encourage cell growth. The fibres can be loaded with promoters of cell growth if required, and other functional ingredients (e.g. to prevent infection and modulate the inflammatory response toward regeneration rather than repair) may also be incorporated. Some examples of this are discussed above in the context of drug delivery.

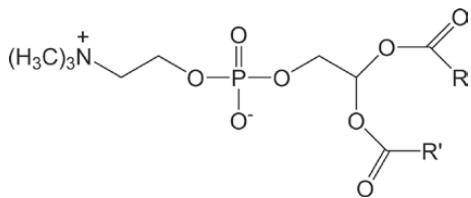
For tissue engineering, the release from the fibres of a functional molecule may well be important, but the mechanical properties are also vital (they must mimic the relevant native tissue). Increasingly, researchers are seeking to make *bioresorbable scaffolds*, materials which degrade slowly over time in the body and are replaced by the body's own tissue. There are myriad applications of such systems, including for instance in the treatment of congenital heart defects. For such resorbable scaffolds to be effective, the degradation time of the polymer and the rate at which it is replaced by native tissue will need to be carefully considered and controlled. A detailed discussion of electrospun fibres in tissue engineering lies outside the scope of this volume, but there are a number of recent reviews which summarise elegantly the state of the art.<sup>101</sup>

### 3.13 Using fibres as sacrificial templates

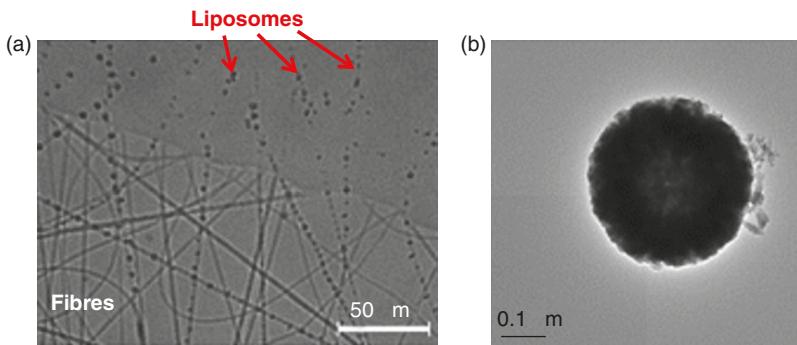
As well as being used directly for drug delivery purposes, electrospun fibres can be used as sacrificial templates to generate higher-order materials. Electrospinning is regarded as a *top-down* fabrication technique, because the structure of the macroscale spinneret is propagated through into solid fibres (that is, a macroscopic object is used to prepare smaller ones). Another way to prepare nanoscale objects is *bottom-up*, in which smaller building blocks (often atoms and molecules) are brought together to form aggregates (e.g. in molecular self-assembly). This is a powerful approach, but unfortunately it is highly time-consuming, low-yield and more expensive than the top-down route. It is also very difficult to ensure that the building blocks assemble in the desired configuration. For example, self-assembled nanofibres often take the form of discontinuous fragments of fibrils of different lengths.

Liposomes (artificial vesicles bounded by a lipid bilayer) are one type of nanoscale system typically prepared bottom-up. Liposomes have numerous applications, including the delivery of therapeutic and diagnostic agents. They self-assemble in aqueous media owing to their amphiphilic nature; the formation of liposomes minimises contact between hydrophobic and hydrophilic groups, and thus is thermodynamically favourable. However, liposomes are unstable over time and have a tendency to aggregate. Controlling the size of the liposomes formed can also be challenging. Often, templates are used to direct the self-assembly processes and drive them towards the desired structure. Electrospun fibres can be used as such templates.

Yu *et al.* were the first to report this,<sup>102</sup> making PVP fibres containing phosphatidyl choline (PC), an amphiphilic molecule with a polar head

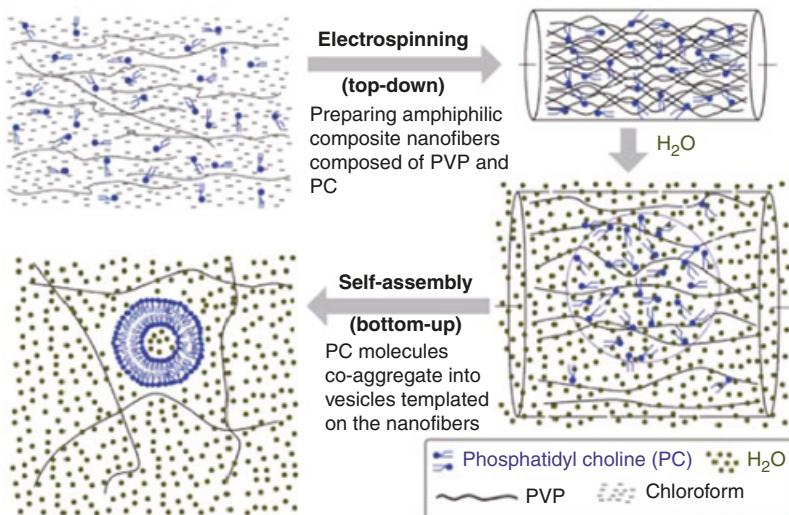


**Figure 3.11** The chemical structure of phosphatidyl choline. R and R' are long-chain fatty acid groups.



**Figure 3.12** The self-assembly of phosphatidyl choline liposomes from poly(vinyl pyrrolidone)/phosphatidyl choline fibres, as reported by Yu *et al.*<sup>102</sup> (a) An optical microscopy image obtained during the self-assembly process, and (b) a transmission electron microscopy image of one of the resultant liposomes. (Reproduced with permission from Yu, D.-G.; Branford-White, C.; Williams, G. R.; Bligh, S. W. A.; White, K.; Zhu, L.-M.; Chatterton, N. P. ‘Self-assembled liposomes from amphiphilic electrospun nanofibers.’ *Soft Matter* 7 (2011): 8239–8247. Copyright Royal Society of Chemistry 2011.)

group and hydrophobic hydrocarbon tail (Figure 3.11). Amorphous solid dispersions of PC in PVP were obtained from electrospinning. When the fibres were added to water, the PC molecules were observed to self-assemble into liposomes (Figure 3.12), the size of which could be tuned by varying the fibre composition. The authors attributed this to the PVP matrix confining the PC molecules in close proximity when the fibres are



**Figure 3.13** The proposed mechanism for liposome assembly from poly(vinyl pyrrolidone) (PVP)/phosphatidyl choline (PC) fibres.  
 (Reproduced with permission from Yu, D.-G.; Branford-White, C.; Williams, G. R.; Bligh, S. W. A.; White, K.; Zhu, L.-M.; Chatterton, N. P. ‘Self-assembled liposomes from amphiphilic electrospun nanofibres.’ *Soft Matter* 7 (2011): 8239–8247. Copyright Royal Society of Chemistry 2011.)

added to water, driving them to aggregate into liposomes (Figure 3.13). This approach is potentially beneficial over other liposome production techniques, since it does not require any controlled heating, cooling or agitation steps. Furthermore, it can ameliorate some of the stability issues which are encountered during the storage of liposomes, because the fibres can act as proto-liposomes. These liposome precursors can be stored ‘frozen’ in the solid-state nanofibres, endowing them with high stability, but can easily be converted to liquid suspensions *in situ* upon demand.<sup>102</sup>

This work was developed further by Song and co-workers, who developed magnetic liposomes via a similar route.<sup>103</sup> Magnetic iron oxide nanoparticles were included in the fibres along with PVP and PC, and magnetic liposomes formed spontaneously when the fibres were added to water. The liposome size could be controlled by varying the iron oxide content of the fibres. The magnetic properties of the nanoparticles were unaffected by both the electrospinning and self-assembly processes, and thus these materials might find applications in targeted drug delivery.

## 3.14 Conclusions

In this chapter, we have considered the uses of fibres prepared by monoaxial electrospinning in drug delivery. The experimental set-up required was first explained, and details given as to how to begin a new electrospinning process. The various different types of fibres that can be generated were then discussed. Fibres made by monoaxial electrospinning find applications as FD-DDSs able to provide very rapid release in the mouth, as extended-release systems to prolong release over time, and can be used to target drug delivery in response to particular temperature or pH conditions.

It is possible to prepare fibre mats made of multilayers of different drug-carrying polymers to give more complex or precisely controlled drug delivery patterns, and suspensions and emulsions can be processed in addition to solutions; this can be beneficial in preventing an initial burst release of drug and protecting proteins from degradation. Finally, the potential to use the fibres as templates to self-assemble higher-order objects was considered.

Monoaxial spinning is the simplest electrospinning technique and thus has received a great deal of attention in the literature, with many promising applications. However, there are some drawbacks – in particular the burst of drug release which is often seen at the beginning of experiments – and therefore in some instances more complex approaches are required. We will discuss these in [Chapters 4 and 5](#).

## 3.15 References

1. Kenawy, E.-R.; Bowlin, G. L.; Mansfield, K.; Layman, J.; Simpson, D. G.; Sanders, E. H.; Wnek, G. E. ‘Release of tetracycline hydrochloride from electrospun poly(ethylene-co-vinylacetate), poly(lactic acid), and a blend.’ *J. Control. Release* 81 (2002): 57–64.
2. (a) Chou, S.-F.; Carson, D.; Woodrow, K. A. ‘Current strategies for sustaining drug release from electrospun nanofibers.’ *J. Control. Release* 220 (2015): 584–591; (b) Krogstad, E. A.; Woodrow, K. A. ‘Manufacturing scale-up of electrospun poly(vinyl alcohol) fibers containing tenofovir for vaginal drug delivery.’ *Int. J. Pharm.* 475 (2014): 282–291; (c) Ball, C.; Woodrow, K. A. ‘Electrospun solid dispersions of maraviroc for rapid intravaginal preexposure prophylaxis of HIV.’ *Antimicrob. Agents Chemother.* 58 (2014): 4855–4865; (d) Blakney, A. K.; Krogstad, E. A.; Jiang, Y. H.; Woodrow, K. A. ‘Delivery of multipurpose prevention drug combinations

- from electrospun nanofibers using composite microarchitectures.' *Int. J. Nanomedicine* 9 (2014): 2967–2978.
- 3. (a) Ball, C.; Krogstad, E.; Chaowanachan, T.; Woodrow, K. A. 'Drug-eluting fibers for HIV-1 inhibition and contraception.' *PLoS ONE* 7 (2012): e49792; (b) Blakney, A. K.; Ball, C.; Krogstad, E. A.; Woodrow, K. A. 'Electrospun fibers for vaginal anti-HIV drug delivery.' *Antiviral Res.* 100 (2013): S9–S16.
  - 4. (a) Pelipenko, J.; Kocbek, P.; Kristl, J. 'Critical attributes of nanofibers: Preparation, drug loading, and tissue regeneration.' *Int. J. Pharm.* 484 (2015): 57–74; (b) Zupančič, Š.; Sinha-Ray, S.; Sinha-Ray, S.; Kristl, J.; Yarin, A. L. 'Long-term sustained ciprofloxacin release from PMMA and hydrophilic polymer blended nanofibers.' *Mol. Pharm.* 13 (2016): 295–305; (c) Falde, E. J.; Freedman, J. D.; Herrera, V. L. M.; Yohe, S. T.; Colson, Y. L.; Grinstaff, M. W. 'Layered superhydrophobic meshes for controlled drug release.' *J. Control. Release* 214 (2015): 23–29.
  - 5. Williams, G. R.; Chatterton, N. P.; Nazir, T.; Yu, D.-G.; Zhu, L.-M.; Branford-White, C. J. 'Electrospun nanofibers in drug delivery: Recent developments and perspectives.' *Therap. Deliv.* 3 (2012): 515–533.
  - 6. Krogstad, E. A.; Rathbone, M. J.; Woodrow, K. A., 'Vaginal drug delivery.' In *Focal Controlled Drug Delivery*, edited by Domb, A. J.; Khan, W., 607–651. Boston: Springer US, 2014.
  - 7. Chakraborty, S.; Liao, I. C.; Adler, A.; Leong, K. W. 'Electrohydrodynamics: A facile technique to fabricate drug delivery systems.' *Adv. Drug Deliv. Rev.* 61 (2009): 1043–1054.
  - 8. Katsogiannis, K. A. G.; Vladisavljević, G. T.; Georgiadou, S. 'Porous electrospun polycaprolactone (PCL) fibres by phase separation.' *Eur. Polym. J.* 69 (2015): 284–295.
  - 9. Xiang, Q.; Ma, Y.-M.; Yu, D.-G.; Jin, M.; Williams, G. R. 'Electrospinning using a Teflon-coated spinneret.' *Appl. Surf. Sci.* 284 (2013): 889–893.
  - 10. Wang, X.; Li, X.-Y.; Li, Y.; Zou, H.; Yu, D. G.; Cai, J.-S. 'Electrospun acetaminophen-loaded cellulose acetate nanofibers fabricated using an epoxy-coated spinneret.' *e-Polymers* 15 (2015): 311–315.
  - 11. Chuangchote, S.; Sagawa, T.; Yoshikawa, S. 'Electrospinning of poly(vinyl pyrrolidone): Effects of solvents on electrospinnability for the fabrication of poly(p-phenylene vinylene) and TiO<sub>2</sub> nanofibers.' *J. Appl. Polym. Sci.* 114 (2009): 2777–2791.
  - 12. Leach, M. K.; Z.Q., F.; Tuck, S. J.; Corey, J. M. 'Electrospinning fundamentals: Optimizing solution and apparatus parameters.' *J. Vis. Exp.* 47 (2011): e2494.
  - 13. Sill, T. J.; von Recum, H. A. 'Electrospinning: Applications in drug delivery and tissue engineering.' *Biomaterials* 29 (2008): 1989–2006.
  - 14. Acatay, K.; Simsek, E.; Ow-Yang, C.; Menceloglu, Y. Z. 'Tunable, superhydrophobically stable polymeric surfaces by electrospinning.' *Angew. Chem.* 116 (2004): 5322–5325.
  - 15. (a) Deitzel, J. M.; Kleinmeyer, J.; Harris, D.; Beck Tan, N. C. 'The effect of processing variables on the morphology of electrospun nanofibers and textiles.'

- Polymer* 42 (2001): 261–272; (b) Garg, K.; Bowlin, G. L. ‘Electrospinning jets and nanofibrous structures.’ *Biomicrofluidics* 5 (2011): 013403.
- 16. Brown, T. D.; Dalton, P. D.; Hutmacher, D. W. ‘Direct writing by way of melt electrospinning.’ *Adv. Mater.* 23 (2011): 5651–5657.
  - 17. Liang, D.; Hsiao, B. S.; Chu, B. ‘Functional electrospun nanofibrous scaffolds for biomedical applications.’ *Adv. Drug Deliv. Rev.* 59 (2007): 1392–1412.
  - 18. Lopez, F. L.; Shearman, G. C.; Gaisford, S.; Williams, G. R. ‘Amorphous formulations of indomethacin and griseofulvin prepared by electrospinning.’ *Mol. Pharm.* 11 (2014): 4327–4338.
  - 19. (a) Chaudhary, H.; Gauri, S.; Rathee, P.; Kumar, V. ‘Development and optimization of fast dissolving oro-dispersible films of granisetron HCl using Box–Behnken statistical design.’ *Bull. Fac. Pharm. Cairo Univ.* 51 (2013): 193–201; (b) Hoffmann, E. M.; Breitenbach, A.; Breitkreutz, J. ‘Advances in orodispersible films for drug delivery.’ *Expert Op. Drug Del.* 8 (2011): 299–316; (c) Illangakoon, U. E.; Gill, H.; Shearman, G. C.; Parhizkar, M.; Mahalingam, S.; Chatterton, N. P.; Williams, G. R. ‘Fast dissolving paracetamol/caffeine nanofibers prepared by electrospinning.’ *Int. J. Pharm.* 477 (2014): 369–379.
  - 20. Seager, H. ‘Drug-delivery products and the Zydis fast-dissolving dosage form.’ *J. Pharm. Pharmacol.* 50 (1998): 375–382.
  - 21. Yu, D. G.; Shen, X. X.; Branford-White, C.; White, K.; Zhu, L. M.; Bligh, S. W. ‘Oral fast-dissolving drug delivery membranes prepared from electrospun polyvinylpyrrolidone ultrafine fibers.’ *Nanotechnology* 20 (2009): 055104.
  - 22. Adeli, E. ‘Irbesartan-loaded electrospun nanofibers-based PVP K90 for the drug dissolution improvement: Fabrication, *in vitro* performance assessment, and *in vivo* evaluation.’ *J. Appl. Polym. Sci.* 132 (2015): 42212.
  - 23. Yu, D.-G.; Branford-White, C.; Shen, X.-X.; Zhang, X.-F.; Zhu, L.-M. ‘Solid dispersions of ketoprofen in drug-loaded electrospun nanofibers.’ *J. Dispersion Sci. Technol.* 31 (2010): 902–908.
  - 24. Illangakoon, U. E.; Nazir, T.; Williams, G. R.; Chatterton, N. P. ‘Mebeverine-loaded electrospun nanofibers: Physicochemical characterization and dissolution studies.’ *J. Pharm. Sci.* 103 (2014): 283–292.
  - 25. Li, X.; Lin, L.; Zhu, Y.; Liu, W.; Yu, T.; Ge, M. ‘Preparation of ultrafine fast-dissolving cholecalciferol-loaded poly(vinyl pyrrolidone) fiber mats via electrospinning.’ *Polym. Composite.* 34 (2013): 282–287.
  - 26. Chen, J.; Wang, X.; Zhang, W.; Yu, S.; Fan, J.; Cheng, B.; Yang, X.; Pan, W. ‘A novel application of electrospinning technique in sublingual membrane: Characterization, permeation and *in vivo* study.’ *Drug. Dev. Ind. Pharm.* 42 (2016): 1365–1374.
  - 27. Li, X.-Y.; Wang, X.; Yu, D.-G.; Ye, S.; Kuang, Q.-K.; Yi, Q.-W.; Yao, X.-Z. ‘Electrospun borneol–PVP nanocomposites.’ *J. Nanomater.* 2012 (2012): 731382.
  - 28. Wang, C.; Ma, C.; Wu, Z.; Liang, H.; Yan, P.; Song, J.; Ma, N.; Zhao, Q. ‘Enhanced bioavailability and anticancer effect of curcumin-loaded electrospun nanofiber: *In vitro* and *in vivo* study.’ *Nanoscale Res. Lett.* 10 (2015): 439.

29. Li, X.; Kanjwal, M. A.; Lin, L.; Chronakis, I. S. 'Electrospun polyvinyl-alcohol nanofibers as oral fast-dissolving delivery system of caffeine and riboflavin.' *Colloids Surf. B* 103 (2013): 182–188.
30. Krstic, M.; Radojevic, M.; Stojanovic, D.; Radojevic, V.; Uskokovic, P.; Ibric, S. 'Formulation and characterization of nanofibers and films with carvedilol prepared by electrospinning and solution casting method.' *Eur. J. Pharm. Sci.* 101 (2017): 160–166.
31. Taepaiboon, P.; Rungsardthong, U.; Supaphol, P. 'Drug-loaded electrospun mats of poly(vinyl alcohol) fibres and their release characteristics of four model drugs.' *Nanotechnol.* 17 (2006): 2317–2329.
32. Rasekh, M.; Karavasili, C.; Soong, Y. L.; Bouropoulos, N.; Morris, M.; Armitage, D.; Li, X.; Fatouros, D. G.; Ahmad, Z. 'Electrospun PVP-indomethacin constituents for transdermal dressings and drug delivery devices.' *Int. J. Pharm.* 473 (2014): 95–104.
33. Baskakova, A.; Awwad, S.; Jimenez, J. Q.; Gill, H.; Novikov, O.; Khaw, P. T.; Brocchini, S.; Zhilyakova, E.; Williams, G. R. 'Electrospun formulations of acyclovir, ciprofloxacin and cyanocobalamin for ocular drug delivery.' *Int. J. Pharm.* 502 (2016): 208–218.
34. Samprasit, W.; Akkaramongkolporn, P.; Ngawhirunpat, T.; Rojanarata, T.; Kaomongkolgit, R.; Opanasopit, P. 'Fast releasing oral electrospun PVP/CD nanofiber mats of taste-masked meloxicam.' *Int. J. Pharm.* 487 (2015): 213–222.
35. Vrbata, P.; Berka, P.; Stranska, D.; Dolezal, P.; Musilova, M.; Cizinska, L. 'Electrospun drug loaded membranes for sublingual administration of sumatriptan and naproxen.' *Int. J. Pharm.* 457 (2013): 168–176.
36. Yu, D.-G.; Yang, J.-M.; Branford-White, C.; Lu, P.; Zhang, L.; Zhu, L.-M. 'Third generation solid dispersions of ferulic acid in electrospun composite nanofibers.' *Int. J. Pharm.* 400 (2010): 158–164.
37. Brettmann, B. K.; Myerson, A. S.; Trout, B. L. 'Solid-state nuclear magnetic resonance study of the physical stability of electrospun drug and polymer solid solutions.' *J. Pharm. Sci.* 101 (2012): 2185–2193.
38. Demuth, B.; Farkas, A.; Pataki, H.; Balogh, A.; Szabo, B.; Borbas, E.; Soti, P. L.; Vigh, T.; Kiserdei, E.; Farkas, B.; Mensch, J.; Verreck, G.; Van Assche, I.; Marosi, G.; Nagy, Z. K. 'Detailed stability investigation of amorphous solid dispersions prepared by single-needle and high speed electrospinning.' *Int. J. Pharm.* 498 (2016): 234–244.
39. Tiwari, S. B.; Rajabi-Siahboomi, A. R., 'Extended-release oral drug delivery technologies: Monolithic matrix systems.' In *Drug Delivery Systems*, edited by Jain, K. K., 217–243. Totowa: Humana Press, 2008.
40. Bhardwaj, N.; Kundu, S. C. 'Electrospinning: A fascinating fiber fabrication technique.' *Biotechnol. Adv.* 28 (2010): 325–347.
41. (a) Piskin, E.; Bölgön, N.; Egri, S.; Isoglu, I. A. 'Electrospun matrices made of poly( $\alpha$ -hydroxy acids) for medical use.' *Nanomedicine* 2 (2007): 441–457; (b) Schiffman, J. D.; Schauer, C. L. 'A review: Electrospinning of biopolymer nanofibers and their applications.' *Polym. Rev.* 48 (2008): 317–352.

42. Xie, J.; Wang, C. H. 'Electrospun micro- and nanofibers for sustained delivery of paclitaxel to treat C6 glioma *in vitro*.' *Pharm. Res.* 23 (2006): 1817–1826.
43. Xie, J.; Tan, R. S.; Wang, C. H. 'Biodegradable microparticles and fiber fabrics for sustained delivery of cisplatin to treat C6 glioma *in vitro*.' *J. Biomed. Mater. Res. A* 85 (2008): 897–908.
44. Park, Y.; Kang, E.; Kwon, O. J.; Hwang, T.; Park, H.; Lee, J. M.; Kim, J. H.; Yun, C. O. 'Ionically crosslinked Ad/chitosan nanocomplexes processed by electrospinning for targeted cancer gene therapy.' *J. Control. Release* 148 (2010): 75–82.
45. Puppi, D.; Piras, A. M.; Detta, N.; Dinucci, D.; Chiellini, F. 'Poly(lactic-co-glycolic acid) electrospun fibrous meshes for the controlled release of retinoic acid.' *Acta Biomater.* 6 (2010): 1258–1268.
46. Luong-Van, E.; Grondahl, L.; Chua, K. N.; Leong, K. W.; Nurcombe, V.; Cool, S. M. 'Controlled release of heparin from poly(epsilon-caprolactone) electrospun fibers.' *Biomaterials* 27 (2006): 2042–2050.
47. Hall Barrientos, I. J.; Paladino, E.; Brozio, S.; Passarelli, M. K.; Moug, S.; Black, R. A.; Wilson, C. G.; Lamprou, D. A. 'Fabrication and characterisation of drug-loaded electrospun polymeric nanofibers for controlled release in hernia repair.' *Int. J. Pharm.* 517 (2017): 329–337.
48. Zhang, Z.; Tang, J.; Wang, H.; Xia, Q.; Xu, S.; Han, C. C. 'Controlled antibiotics release system through simple blended electrospun fibers for sustained antibacterial effects.' *ACS Appl. Mater. Interfaces* 7 (2015): 26400–26404.
49. Jiang, J.; Chen, G.; Shuler, F. D.; Wang, C. H.; Xie, J. 'Local sustained delivery of 25-hydroxyvitamin D3 for production of antimicrobial peptides.' *Pharm. Res.* 32 (2015): 2851–2862.
50. He, T.; Wang, J.; Huang, P.; Zeng, B.; Li, H.; Cao, Q.; Zhang, S.; Luo, Z.; Deng, D. Y.; Zhang, H.; Zhou, W. 'Electrospinning polyvinylidene fluoride fibrous membranes containing anti-bacterial drugs used as wound dressing.' *Colloids Surf. B* 130 (2015): 278–286.
51. Zamani, M.; Morshed, M.; Varshosaz, J.; Jannesari, M. 'Controlled release of metronidazole benzoate from poly epsilon-caprolactone electrospun nanofibers for periodontal diseases.' *Eur. J. Pharm. Biopharm.* 75 (2010): 179–185.
52. Yu, D.-G.; Branford-White, C.; Li, L.; Wu, X.-M.; Zhu, L.-M. 'The compatibility of acyclovir with polyacrylonitrile in the electrospun drug-loaded nanofibers.' *J. Appl. Polym. Sci.* (2010): 1509–1515.
53. Lu, H.; Wang, Q.; Li, G.; Qiu, Y.; Wei, Q. 'Electrospun water-stable zein/ethyl cellulose composite nanofiber and its drug release properties.' *Mater. Sci. Eng. C* 74 (2017): 86–93.
54. Jalvandi, J.; White, M.; Gao, Y.; Truong, Y. B.; Padhye, R.; Kyriatzi, I. L. 'Polyvinyl alcohol composite nanofibres containing conjugated levofloxacin-chitosan for controlled drug release.' *Mater. Sci. Eng. C* 73 (2017): 440–446.

55. Natu, M. V.; de Sousa, H. C.; Gil, M. H. 'Effects of drug solubility, state and loading on controlled release in bicomponent electrospun fibers.' *Int. J. Pharm.* 397 (2010): 50–58.
56. Verreck, G.; Chun, I.; Peeters, J.; Rosenblatt, J.; Brewster, M. E. 'Preparation and characterization of nanofibers containing amorphous drug dispersions generated by electrostatic spinning.' *Pharm. Res.* 20 (2003): 810–817.
57. Xie, Z.; Buschle-Diller, G. 'Electrospun poly(D,L-lactide) fibers for drug delivery: The influence of cosolvent and the mechanism of drug release.' *J. Appl. Polym. Sci.* 115 (2010): 1–8.
58. Zamani, M.; Prabhakaran, M. P.; Ramakrishna, S. 'Advances in drug delivery via electrospun and electrosprayed nanomaterials.' *Int. J. Nanomedicine* 8 (2013): 2997–3017.
59. Wang, M.; Wang, L.; Huang, Y. 'Electrospun hydroxypropyl methyl cellulose phthalate (HPMCP)/erythromycin fibers for targeted release in intestine.' *J. Appl. Polym. Sci.* 106 (2007): 2177–2184.
60. Shen, X.; Yu, D.; Zhu, L.; Branford-White, C.; White, K.; Chatterton, N. P. 'Electrospun diclofenac sodium loaded Eudragit L100–55 nanofibers for colon-targeted drug delivery.' *Int. J. Pharm.* 408 (2011): 200–207.
61. Akhgari, A.; Heshmati, Z.; Afrasiabi Garekani, H.; Sadeghi, F.; Sabbagh, A.; Sharif Makhmalzadeh, B.; Nokhodchi, A. 'Indomethacin electrospun nanofibers for colonic drug delivery: *In vitro* dissolution studies.' *Colloids Surf. B* 152 (2017): 29–35.
62. Karthikeyan, K.; Guhathakarta, S.; Rajaram, R.; Korrapati, P. S. 'Electrospun zein/eudragit nanofibers based dual drug delivery system for the simultaneous delivery of aceclofenac and pantoprazole.' *Int. J. Pharm.* 438 (2012): 117–122.
63. Zhao, J.; Jiang, S.; Zheng, R.; Zhao, X.; Chen, X.; Fan, C.; Cui, W. 'Smart electrospun fibrous scaffolds inhibit tumor cells and promote normal cell proliferation.' *RSC Adv.* 4 (2014): 51696–51702.
64. Zhao, J. W.; Cui, W. G. 'Fabrication of acid-responsive electrospun fibers via doping sodium bicarbonate for quick releasing drug.' *Nanosci. Nanotechnol. Lett.* 6 (2014): 339–345.
65. Salehi, R.; Irani, M.; Eskandani, M.; Nowruzi, K.; Davaran, S.; Haririan, I. 'Interaction, controlled release, and antitumor activity of doxorubicin hydrochloride from pH-sensitive p(NIPAAm-MAA-VP) nanofibrous scaffolds prepared by green electrospinning.' *Int. J. Polym. Mater.* 63 (2014): 609–619.
66. Kaassis, A. Y. A.; Young, N.; Sano, N.; Merchant, H. A.; Yu, D.-G.; Chatterton, N. P.; Williams, G. R. 'Pulsatile drug release from electrospun poly(ethylene oxide)-sodium alginate blend nanofibres.' *J. Mater. Chem. B* 2 (2014): 1400–1407.
67. Alhusein, N.; Blagbrough, I. S.; De Bank, P. A. 'Electrospun matrices for localised controlled drug delivery: Release of tetracycline hydrochloride from layers of polycaprolactone and poly(ethylene-co-vinyl acetate).' *Drug Deliv. Trans. Res.* 2 (2012): 477–488.

68. Alhusein, N.; De Bank, P. A.; Blagbrough, I. S.; Bolhuis, A. ‘Killing bacteria within biofilms by sustained release of tetracycline from triple-layered electrospun micro/nanofibre matrices of polycaprolactone and poly(ethylene-co-vinyl acetate).’ *Drug Del. Trans. Res.* 3 (2013): 531–541.
69. Alhusein, N.; Blagbrough, I. S.; De Bank, P. A. ‘Zein/polycaprolactone electrospun matrices for localised controlled delivery of tetracycline.’ *Drug Del. Trans. Res.* 3 (2013): 542–550.
70. Alhusein, N.; Blagbrough, I. S.; Beeton, M. L.; Bolhuis, A.; De Bank, P. A. ‘Electrospun zein/PCL fibrous matrices release tetracycline in a controlled manner, killing *Staphylococcus aureus* both in biofilms and *ex vivo* on pig skin, and are compatible with human skin cells.’ *Pharm. Res.* 33 (2016): 237–246.
71. Huang, L. Y.; Branford-White, C.; Shen, X. X.; Yu, D. G.; Zhu, L. M. ‘Time-engineering biphasic drug release by electrospun nanofiber meshes.’ *Int. J. Pharm.* 436 (2012): 88–96.
72. Rockwood, D. N.; Chase, D. B.; Akins, R. E.; Rabolt, J. F. ‘Characterization of electrospun poly(*N*-isopropyl acrylamide) fibers.’ *Polymer* 49 (2008): 4025–4032.
73. (a) Wang, N.; Zhao, Y.; Jiang, L. ‘Low-cost, thermoresponsive wettability of surfaces: Poly(*N*-isopropylacrylamide)/polystyrene composite films prepared by electrospinning.’ *Macromol. Rapid Commun.* 29 (2008): 485–489; (b) Gu, S.-Y.; Wang, Z.-M.; Li, J.-B.; Ren, J. ‘Switchable wettability of thermo-responsive biocompatible nanofibrous films created by electrospinning.’ *Macromol. Mater. Eng.* 295 (2010): 32–36.
74. Lin, X.; Tang, D.; Cui, W.; Cheng, Y. ‘Controllable drug release of electrospun thermoresponsive poly(*N*-isopropylacrylamide)/poly(2-acrylamido-2-methylpropanesulfonic acid) nanofibers.’ *J. Biomed. Mater. Res. A* 100 (2012): 1839–1845.
75. Song, F.; Wang, X. L.; Wang, Y. Z. ‘Poly (*N*-isopropylacrylamide)/poly (ethylene oxide) blend nanofibrous scaffolds: Thermo-responsive carrier for controlled drug release.’ *Colloids Surf. B* 88 (2011): 749–754.
76. Hu, J.; Li, H.-Y.; Williams, G. R.; Yang, H.-H.; Tao, L.; Zhu, L.-M. ‘Electrospun poly(*N*-isopropylacrylamide)/ethyl cellulose nanofibers as thermoresponsive drug delivery systems.’ *J. Pharm. Sci.* 105 (2016).
77. Li, H.; Williams, G. R.; Wu, J.; Wang, H.; Sun, X.; Zhu, L. M. ‘Poly(*N*-isopropylacrylamide)/poly(*L*-lactic acid-co-caprolactone) fibers loaded with ciprofloxacin as wound dressing materials.’ *Mater. Sci. Eng. C* 79 (2017): 245–254.
78. Li, H.; Williams, G. R.; Wu, J.; Lv, Y.; Sun, X.; Wu, H.; Zhu, L. M. ‘Thermosensitive nanofibers loaded with ciprofloxacin as antibacterial wound dressing materials.’ *Int. J. Pharm.* 517 (2017): 135–147.
79. Xu, X.; Chen, X.; Ma, P.; Wang, X.; Jing, X. ‘The release behavior of doxorubicin hydrochloride from medicated fibers prepared by emulsion-electrospinning.’ *Eur. J. Pharm. Biopharm.* 70 (2008): 165–170.

80. Wang, C.; Wang, M. ‘Formation of core–shell structures in emulsion electrospun fibres: A comparative study.’ *Aust. J. Chem.* 67 (2014): 1403–1413.
81. Hu, J.; Wei, J.; Liu, W.; Chen, Y. ‘Preparation and characterization of electrospun PLGA/gelatin nanofibers as a drug delivery system by emulsion electrospinning.’ *J. Biomater. Sci. Polym. Ed.* 24 (2013): 972–985.
82. Sy, J. C.; Klemm, A. S.; Shastri, V. P. ‘Emulsion as a means of controlling electrospinning of polymers.’ *Adv. Mater.* 21 (2009): 1814–1819.
83. (a) Yang, Y.; Li, X.; He, S.; Cheng, L.; Chen, F.; Zhou, S.; Weng, J. ‘Biodegradable ultrafine fibers with core–sheath structures for protein delivery and its optimization.’ *Polym. Adv. Technol.* 22 (2011): 1842–1850; (b) Briggs, T.; Arinze, T. L. ‘Examining the formulation of emulsion electrospinning for improving the release of bioactive proteins from electrospun fibers.’ *J. Biomed. Mater. Res. A* 102 (2014): 674–684.
84. Yang, Y.; Li, X.; Cui, W.; Zhou, S.; Tan, R.; Wang, C. ‘Structural stability and release profiles of proteins from core–shell poly (DL-lactide) ultrafine fibers prepared by emulsion electrospinning.’ *J. Biomed. Mater. Res. A* 86 (2008): 374–385.
85. Tian, L.; Prabhakaran, M. P.; Ding, X.; Kai, D.; Ramakrishna, S. ‘Emulsion electrospun vascular endothelial growth factor encapsulated poly(L-lactic acid-co-ε-caprolactone) nanofibers for sustained release in cardiac tissue engineering.’ *J. Mater. Sci.* 47 (2011): 3272–3281.
86. (a) Tian, L.; Prabhakaran, M. P.; Ding, X.; Ramakrishna, S. ‘Biocompatibility evaluation of emulsion electrospun nanofibers using osteoblasts for bone tissue engineering.’ *J. Biomater. Sci. Polym. Ed.* 24 (2013): 1952–1968; (b) Spano, F.; Quarta, A.; Martelli, C.; Ottobrini, L.; Rossi, R. M.; Gigli, G.; Blasi, L. ‘Fibrous scaffolds fabricated by emulsion electrospinning: From hosting capacity to *in vivo* biocompatibility.’ *Nanoscale* 8 (2016): 9293–9303.
87. Gordon, V.; Marom, G.; Magdassi, S. ‘Formation of hydrophilic nanofibers from nanoemulsions through electrospinning.’ *Int. J. Pharm.* 478 (2015): 172–179.
88. Hu, J.; Prabhakaran, M. P.; Tian, L.; Ding, X.; Ramakrishna, S. ‘Drug-loaded emulsion electrospun nanofibers: Characterization, drug release and *in vitro* biocompatibility.’ *RSC Adv.* 5 (2015): 100256–100267.
89. Zhao, X.; Lui, Y.; Toh, P.; Loo, S. ‘Sustained release of hydrophilic L-ascorbic acid 2-phosphate magnesium from electrospun polycaprolactone scaffold – a study across blend, coaxial, and emulsion electrospinning techniques.’ *Materials* 7 (2014): 7398–7408.
90. Ma, J.; Meng, J.; Simonet, M.; Stingelin, N.; Peijs, T.; Sukhorukov, G. B. ‘Biodegradable fibre scaffolds incorporating water-soluble drugs and proteins.’ *J. Mater. Sci. Mater. Med.* 26 (2015): 205.
91. Li, K.; Sun, H.; Sui, H.; Zhang, Y.; Liang, H.; Wu, X.; Zhao, Q. ‘Composite mesoporous silica nanoparticle/chitosan nanofibers for bone tissue engineering.’ *RSC Adv.* 5 (2015): 17541–17549.

92. (a) Burke, L.; Mortimer, C. J.; Curtis, D. J.; Lewis, A. R.; Williams, R.; Hawkins, K.; Maffeis, T. G.; Wright, C. J. 'In-situ synthesis of magnetic iron-oxide nanoparticle–nanofibre composites using electrospinning.' *Mater. Sci. Eng. C* 70 (2017): 512–519; (b) Zhang, H.; Xia, J.; Pang, X.; Zhao, M.; Wang, B.; Yang, L.; Wan, H.; Wu, J.; Fu, S. 'Magnetic nanoparticle-loaded electrospun polymeric nanofibers for tissue engineering.' *Mater. Sci. Eng. C* 73 (2017): 537–543.
93. Wang, L.; Wang, M.; Topham, P. D.; Huang, Y. 'Fabrication of magnetic drug-loaded polymeric composite nanofibres and their drug release characteristics.' *RSC Adv.* 2 (2012): 2433.
94. Kim, H. W.; Lee, H. H.; Knowles, J. C. 'Electrospinning biomedical nanocomposite fibers of hydroxyapatite/poly(lactic acid) for bone regeneration.' *J. Biomed. Mater. Res. A* 79 (2006): 643–649.
95. Lee, I. W.; Li, J.; Chen, X.; Park, H. J. 'Electrospun poly(vinyl alcohol) composite nanofibers with halloysite nanotubes for the sustained release of sodium-pantothenate.' *J. Appl. Polym. Sci.* 133 (2016): 42900.
96. Valarezo, E.; Tammaro, L.; González, S.; Malagón, O.; Vittoria, V. 'Fabrication and sustained release properties of poly( $\epsilon$ -caprolactone) electrospun fibers loaded with layered double hydroxide nanoparticles intercalated with amoxicillin.' *Appl. Clay Sci.* 72 (2013): 104–109.
97. Fazli, Y.; Shariatinia, Z. 'Controlled release of cefazolin sodium antibiotic drug from electrospun chitosan–polyethylene oxide nanofibrous mats.' *Mater. Sci. Eng. C* 71 (2017): 641–652.
98. Jalvandi, J.; White, M.; Gao, Y.; Truong, Y. B.; Padhye, R.; Kyratzis, I. L. 'Slow release of levofloxacin conjugated on silica nanoparticles from poly ( $\epsilon$ caprolactone) nanofibers.' *Int. J. Polym. Mater.* 66 (2017): 507–513.
99. Li, L.; Zhou, G.; Wang, Y.; Yang, G.; Ding, S.; Zhou, S. 'Controlled dual delivery of BMP-2 and dexamethasone by nanoparticle-embedded electrospun nanofibers for the efficient repair of critical-sized rat calvarial defect.' *Biomaterials* 37 (2015): 218–229.
100. Brettmann, B. K.; Cheng, K.; Myerson, A. S.; Trout, B. L. 'Electrospun formulations containing crystalline active pharmaceutical ingredients.' *Pharm. Res.* 30 (2013): 238–246.
101. (a) Haidar, M. K.; Eroglu, H. 'Nanofibers: New insights for drug delivery and tissue engineering.' *Curr. Top. Med. Chem.* 17 (2016): 1564–1579; (b) Kitsara, M.; Agbulut, O.; Kontziamasis, D.; Chen, Y.; Menasche, P. 'Fibers for hearts: A critical review on electrospinning for cardiac tissue engineering.' *Acta Biomater.* 48 (2017): 20–40; (c) Asghari, F.; Samiei, M.; Adibkia, K.; Akbarzadeh, A.; Davaran, S. 'Biodegradable and biocompatible polymers for tissue engineering application: A review.' *Artif. Cells Nanomed. Biotechnol.* 45 (2017): 185–192; (d) Rezvani, Z.; Venugopal, J. R.; Urbanska, A. M.; Mills, D. K.; Ramakrishna, S.; Mozafari, M. 'A bird's eye view on the use of electrospun nanofibrous scaffolds for bone tissue engineering: Current state-of-the-art, emerging directions and future trends.'

- Nanomedicine* 12 (2016): 2181–2200; (e) O'Connor, R. A.; McGuinness, G. B. ‘Electrospun nanofibre bundles and yarns for tissue engineering applications: A review.’ *Proc. Inst. Mech. Eng. H* 230 (2016): 987–998; (f) Kong, B.; Mi, S. ‘Electrospun scaffolds for corneal tissue engineering: A review.’ *Materials* 9 (2016): 614; (g) Khorshidi, S.; Solouk, A.; Mirzadeh, H.; Mazinani, S.; Lagaron, J. M.; Sharifi, S.; Ramakrishna, S. ‘A review of key challenges of electrospun scaffolds for tissue-engineering applications.’ *J. Tissue Eng. Regen. Med.* 10 (2016): 715–738.
102. Yu, D.-G.; Branford-White, C.; Williams, G. R.; Bligh, S. W. A.; White, K.; Zhu, L.-M.; Chatterton, N. P. ‘Self-assembled liposomes from amphiphilic electrospun nanofibers.’ *Soft Matter* 7 (2011): 8239–8247.
103. Song, H.-H.; Gong, X.; Williams, G. R.; Quan, J.; Nie, H.-L.; Zhu, L.-M.; Nan, E.-L.; Shao, M. ‘Self-assembled magnetic liposomes from electrospun fibers.’ *Mater. Res. Bull.* 53 (2014): 280–289.