

# Antibacterial Electrospun Nanofibres



Wazed Ali, Rahul Gadkari, Sanchi Arora, Viraj Somkuwar, and Anupam Chowdhury

**Abstract** Several methods are available for the production of nanofibres, such as rotary jet spinning, melt fibrillation, electrospinning, sol-gel method, phase separation, gas-jet technique, self-assembly and template synthesis. However, electrospinning surpasses all of them in terms of efficiency and effectiveness. Electrospinning enables the production of continuous nanofibres from a wide range of materials. It also facilitates precise control over several fibre/membrane properties like diameter, morphology, composition, porosity, etc., that too by using simple equipment and even simpler procedure. Nanofibres produced from electrospinning technique exhibit diverse features, such as high porosity, high surface area-to-volume ratio, small pore size, low weight and good mechanical properties. Considering these favourable characteristics, electrospun membranes are extensively used to fabricate bioactive products for application in various areas like healthcare, energy harvesting and storage, biomedical, environmental engineering, defence and security. This chapter focuses on the potential use of various natural, synthetic, functionalised, encapsulated and composite electrospun nanofibrous membranes for antibacterial bioactive applications.

**Keywords** Nanofibres · Electrospinning · Antibacterial · Plant extract · Composite nanofibres

## 1 Introduction

Nanotechnology is an interesting interdisciplinary area with potential applications in almost all fields of science and technology: healthcare, material science, energy studies, environmental studies, mechanics, optics, electronics, plastics, aerospace

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W. Ali (✉) · R. Gadkari · S. Arora · V. Somkuwar · A. Chowdhury  
Department of Textile and Fibre Engineering, Indian Institute of Technology Delhi, New Delhi,  
India  
e-mail: [wazed@textile.iitd.ac.in](mailto:wazed@textile.iitd.ac.in)

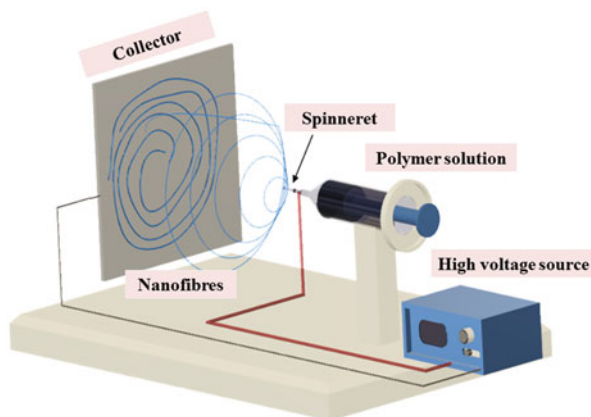
and so on. Some typical morphologies of nanomaterials that have been researched upon extensively are nanofibres, nanowires, nanorods, nanotubes, nanoparticles, etc. Among these, nanofibres have recently gained substantial interest of the research community owing to their large surface area-to-volume ratio, feasibility of generating diverse surface functionalities and superior mechanical properties that render them suitable for use in myriad applications. Out of the numerous existing techniques for spinning nanofibres, electrospinning has emerged as most potent of producing continuous nanofibres from a variety of natural as well as synthetic polymers (Panthi et al. 2015; Joshi et al. 2008).

Over the years, bioactive and antibacterial/antimicrobial materials are gaining rising importance not only in healthcare sector but also in other applications like, sportswear, defence clothing, protective and industrial textiles, geomembranes, etc. These are important for creating a healthy and hygienic atmosphere at home, workplace and in general environment. In order to tackle bacterial infections, composite nanofibres, i.e. nanofibres integrated with powerful antibacterial agents such as silver, copper, copper oxide (CuO), zinc oxide (ZnO) in the form of nanoparticles are being developed (Ditaranto et al. 2018; Gadkari et al. 2017a; Saquing et al. 2009). In general, fabrication of antibacterial nanofibres via electrospinning technique involves integration of biocides in fibres. This can be achieved by various methods, such as using a homogenous blend of antibacterial agent in the polymer solution as feed for electrospinning, adding nanostructures of antibacterial substance(s) in feed solution for electrospinning fibres embedded with nanostructures, enclosing an antibacterial agent in the core of nanocomposite fibre by a sheath of other polymer, treating the electrospun nanofibres with antibacterial agent(s), etc. (Xue et al. 2017). However, the rising concern related to the adverse effects of synthetic antibacterial agents on the health of the human population and on the environment has driven the research on development of electrospun antibacterial fibres towards usage of natural plant extracts and essential oils having bactericidal property. The present chapter presents an overview of some significant studies focused upon the electrospinning of such antibacterial nanofibres or nanofibrous membranes.

## 2 Electrospinning Process

Electrospinning is the most popular and vastly used technology for making nano-scale fibres from both natural and synthetic polymers. Traditionally, synthetic fibres are obtained via melt spinning, which results in fibre diameter ranging between 5  $\mu\text{m}$  to 200  $\mu\text{m}$ , whereas the diameter of electrospun fibres falls within a range of several nanometres (Brown et al. 2016; Bhardwaj and Kundu 2010; Stankus et al. 2006). Moreover, production of polymeric fibres via melt spinning uses mechanical force to extract the melt polymer through spinnerets, whereas electrospinning is an electrohydrodynamic process that uses electric potential to draw polymer solution into fibres. The electrospinning phenomenon was first observed way back in 1600

**Fig. 1** Schematic depiction of a horizontal electrospinning set-up



century by W. Gilbert, an English Physicist. He observed a water droplet placed over a dry surface reshaping in a conical form on being brought into proximity of a piece of rubbed amber (Brown et al. 2016). In 1749, Nollet demonstrated separation of water drops from charged water jet falling into a collector set at different electric potential. Later, Rayleigh (1897), Zeleny (1914) and Formhals (1934) also observed electro spraying effect (Bhardwaj and Kundu 2010). All of these studies became the basis for Antonin Formhals in 1934 to fabricate the first system for producing electrospun yarn using a voltage of 57 kV, whereafter a thorough research on electrospinning gained momentum.

Typically, an electrospinning set-up consists of three basic units: a variable high DC voltage supply, a metallic spinneret (syringe) with a very fine orifice and a collector plate with some conductive element (Fig. 1). The granules of polymer to be electrospun are generally dissolved in some solvent to prepare a solution with requisite viscosity. A very high voltage is applied between the drop of polymer (held inside the spinneret tip by its surface tension) and a collector plate or target electrode placed at certain distance from the spinneret tip. Application of high voltage results in development of an electrostatic force on the polymer drop, which deforms the polymer drop into a conical shape called Taylor cone. When this electrostatic force reaches a critical value, it overcomes the surface tension, resulting in ejection of a charged jet from Taylor cone towards the collector plate. The solvent evaporates leaving behind the solid polymer in fibrous form, which gets deposited onto the collector forming nonwoven fibrous webs or mats (Persano et al. 2013; Balamurugan et al. 2011; Zhang et al. 2005; Zaikov 2016; Baker et al. 2006; Tamura and Kawakami 2010). Besides, during the stretching of polymer stream and simultaneous evaporation of solvent, the size of polymeric strand can be drastically decreased (up to six orders of magnitude), resulting in nano-sized fibres (Tamura and Kawakami 2010). The continuous deposition of fibres generates 2D nanofibrous web of very high porosity (even 90%), thus making them suitable for use in high efficiency filters, scaffolds for tissue engineering, artificial implants, wound dressings, masks, etc. Other than these applications, electrospun nanofibres are also used

in energy harvesting and storage devices for smart textiles, solar cells, fuel cells, chemical and biological protection sensors, etc. (Ramakrishna et al. 2006; Riboldi et al. 2005; Reneker and Chun 1996; Reneker et al. 2000; Rho et al. 2006).

Conventional electrospinning set-ups are horizontal or vertical (Bhardwaj and Kundu 2010; Persano et al. 2013; Balamurugan et al. 2011), though some researchers have also reported designing typical systems for the production of complex nanofibrous structures to suit specific requirements and applications. Balamurugan et al. (2011) reported a system combining electrospinning and electro spraying techniques for the fabrication of nanocomposite membranes. Stankus et al. (2006) also combined electro spraying with electrospinning to integrate smooth muscle cells into biodegradable fibre matrix. Baker et al. (2006) used a circular mandrel to produce 3D electrospun polystyrene scaffolds which could be used for cell growth and cell to cell interaction. Numerous other studies about modification of electrospinning set-ups like development of coaxial electrospinning system for highly concentrated solutions (Yu et al. 2011), combination of melt electrospinning and solution electrospinning for hybrid electrospinning of scaffolds (Il Yoon et al. 2013), using differently profiled collectors for creating different alignment of fibres (Stocco et al. 2017; Levitt et al. 2017), etc. are also available.

### 3 Bioactive Antibacterial Electrospun Nanofibres

For any antibacterial material to function efficiently, the bacteria should essentially have good contact with it. Thus, the high surface area-to-volume ratio of electrospun nanofibres enhances the bactericidal effect of materials developed from them (Li et al. 2015). Moreover, the fibrous mats developed by electrospinning can be easily tailored for different applications during or after the generation of fibres. In addition, various strategies such as doping of antimicrobial agent(s) in precursor solution, surface functionalisation (Su et al. 2012; Schiffman and Elimelech 2011), solution blending (Charernsriwilaiwat et al. 2012), coaxial spinning (Khalf and Madihally 2017; Liu et al. 2011), coating of antibacterial/antimicrobial agents on nanofibrous mat (Rieger et al. 2016; Kang et al. 2009) can also be easily adopted as per required applications.

#### 3.1 *Natural Polymer Based Nanofibres*

Natural polymer based electrospun nanofibres have been extensively used for various medical applications, like wound healing, artificial tissues, medical implants and drug delivery. Natural polymers are chosen over synthetic ones due to better biocompatibility, biodegradability, poor immunogenicity and enhanced cell proliferation (Bhattarai et al. 2005; Wang et al. 2016). However, it is difficult to spin natural polymers in their pure form. For instance, in case of pure alginate, fibre

formation is not possible for less concentrated solutions due to occurrence of gelation. On the other hand, at higher concentration, the solution becomes too viscous to be extruded through spinneret (Bhattarai et al. 2006). Similarly, electrospinning of pure chitosan solution poses problems due to its polycationicity, rigid chemical structure and typical inter- and intra-molecular interactions (Pakravan et al. 2011). Hence, natural polymers are often either blended with synthetic copolymers or chemically modified to be electrospun into nanofibres.

### 3.1.1 Chitosan Based Nanofibres

Chitosan is a deacetylated derivative of chitin, which is a natural polysaccharide found in crustaceans, insects and some fungi exoskeletons. Chitosan derivatives, nanoparticles and nanofibres exhibit antibacterial activity against several fungi, viruses and bacteria (Gadkari et al. 2017b). However, as already mentioned, it is difficult to electrospin it. Many researchers have reported combining chitosan with other polymers to enhance its spinnability. Jung et al. (2007) dissolved poly(ethylene terephthalate) (PET) and chitosan in trifluoroacetic acid and electrospun this polymeric blend solution to make PET/chitosan nanofibrous mats having fibres with 500–800 nm diameter. Homayoni et al. (2009) suggested that the difficulty associated with electrospinning of pure chitosan due to its high viscosity can be resolved using alkaline treatment for hydrolysing its polymer chains, and thus decreasing its molecular weight. They demonstrated successful production of chitosan nanofibres of appropriate quality and processing stability by electrospinning a solution of alkaline treated chitosan in aqueous acetic acid. They also reported an increase in the mean diameter of the chitosan nanofibres upon decreasing the acetic acid concentration in the solvent.

Nguyen et al. (2011) manufactured core/shell structured poly(lactic acid)(PLA)/chitosan fibres through coaxial electrospinning process at different core feed rates of 1  $\mu\text{L}/\text{min}$ , 2  $\mu\text{L}/\text{min}$  and 4  $\mu\text{L}/\text{min}$ . The antibacterial performance of the nanofibrous mats thus produced against *E. coli* bacterium was found to be 99–100% for a bacterial concentration of  $10^3$  CFU/mL. Ahmed et al. (2018) fabricated electrospun fibrous mats from polyvinyl alcohol (PVA)/chitosan blend and PVA/chitosan/ZnO nanoparticles blend. On assessing them for their antibacterial activity against *E. coli*, *P. aeruginosa*, *B. subtilis* and *S. aureus* by disc diffusion method, they were found to create zone of inhibitions with average diameter of  $14.1 \pm 0.8$  mm,  $15.8 \pm 1.0$  mm,  $13.0 \pm 0.7$  mm and  $5.4 \pm 0.5$  mm, respectively, in case of PVA/chitosan mats and  $20.2 \pm 1.0$  mm,  $21.8 \pm 1.5$  mm,  $15.5 \pm 0.8$  mm and  $21.5 \pm 0.5$  mm, respectively, in case of PVA/chitosan/ZnO mats.

### 3.1.2 Protein Based Nanofibres

A protein is a linear polymer of amino acids and is known to exhibit miscellaneous functions. Over the years, researchers have been increasingly exploiting the

structural and functional qualities of fibrous proteins in order to enhance the efficiency of synthetic biomaterials. A variety of proteins and their blends with other polymer(s) have been electrospun by several researchers. This section highlights the use of various protein fibres for application in antibacterial materials.

Lin et al. (2012) prepared electrospun fibrous mats from pure collagen, pure zein and their blend using 40% (w/v) solutions of these polymeric polymers in 70% (v/v) aqueous acetic acid. Though they could not process pure collagen into micro- or nanofibres, pure zein and collagen/zein blend could be efficiently electrospun into bead-free fibrous mats. Besides, they also observed an increase in average fibre diameter from 423 nm to 910 nm on increasing zein weight fraction in collagen/zein blend from 0.33 to 0.67. In order to impart an inherent bactericidal effect, the authors also incorporated berberine drug in the feed solution while electrospinning collagen/zein membrane. In a similar study, Wongkanya et al. (2017) electrospun sodium alginate (SA) and soy protein isolated (SPI) blend fibres with poly(ethylene oxide) (PEO) polymer using different weight percentages of SA, SPI and PEO. They also added vancomycin (Van) drug (0.1 wt.% of polymer), which resists Gram-positive type bacteria, to the feed solution. The zone of inhibition created by Van-loaded SA/PEO/SPI fibres against *S. aureus* bacteria was found to be 21–22.8 mm.

Zhou et al. (2017) used 10:1 (v/v) mixture of 8% collagen solution in hexafluoroisopropanol and bioactive glass (BG) precursor solution to electrospin collagen/BG nanofibres with  $494 \pm 153$  nm average diameter, good thermal stability and hydrophilicity. These fibres were also found to inhibit adhesion and proliferation of *S. aureus* bacterium due to release of Ca, P and Si ions. Khajavi et al. (2016) explored the feasibility of electrospinning nanofibrous scaffolds from different blend compositions of keratin (extracted from quail feather wastes), PVA and silver nanoparticles (Ag-NPs) at 20 kV voltage, 15 cm tip to collector distance and 1 mL/h feed rate. Increase in process efficiency was reflected in formation of uniform nanofibres with fewer beads upon increase in proteinaceous or keratin content. Besides, these mats showed 93–98% antibacterial activity against *S. aureus* (higher for higher keratin content), and almost 100% against *E. coli*.

### 3.1.3 Cellulose Based Nanofibres

Cellulose is a biocompatible and biodegradable polysaccharide consisting of linear chains of  $\beta(1 \rightarrow 4)$  linked d-glucose monomer units. Cellulose based electrospun nanostructures and their derivatives find enormous applications in pharmaceutical industry. This section presents examples of studies conducted on production of antibacterial nanofibres from different derivatives of cellulose.

Carboxymethyl cellulose (CMC) is a classic derivative of cellulose and is non-toxic, biodegradable, biocompatible, water soluble, and has reported usage in variety of biomedical applications, food, detergents, etc. However, alike other natural polymers, it is not suitable to electrospin it in its pure form and needs blending with synthetic copolymers. Shi et al. (2016) developed CMC/PEO membrane by electrospinning an aqueous solution of CMC and PEO in equal amount, at

22 kV with a solution feed rate of 2 mL/h. Subsequent to this, they immersed the membrane in AgNO<sub>3</sub> solution for 2 h, followed by irradiation under ultraviolet (UV) lamp for deposition of silver nanoparticles (Ag-NPs) on fibres. The scanning electron micrographs of the membranes thus developed using AgNO<sub>3</sub> solutions of different concentrations showed presence of several Ag-NPs on their surface as well as in between the adjacent nanofibres. Treatment with 0.10 mol/L AgNO<sub>3</sub> solution was observed to facilitate uniform growth of Ag-NPs on membrane surface, as well as preserved the integrity of 3D membranous structure unlike AgNO<sub>3</sub> solutions with concentration lower or higher than this. Besides, this membrane was also found to exhibit 100% activity against both *S. aureus* and *E. coli* bacteria.

Cellulose acetate (CA) is the only derivative of cellulose that can be processed in an electrospinning set-up in its pure form itself. Several studies are available demonstrating increasing interest in electrospinning CA based antibacterial nanofibres using various antibacterial compounds. In one such study, Sultana et al. (2016) electrospun CA nanofibrous from differently concentrated solutions of CA in acetic acid/acetone. Lower concentration of CA (10% w/v) yielded nanofibres with many beads, whereas bead-free nanofibres were obtained when concentration of CA was increased to 14% (w/v). In case of feed solution loaded with 2% (w/v) tetracycline hydrochloride drug, clear area of inhibition was observed against *B. cereus* and *E. coli* bacteria, unlike pure CA membrane that exhibited no inhibition zone.

Cyclodextrin (CD), another derivative of cellulose, has attracted attention of the research community for the development of antimicrobial nanofibres. Celebioglu et al. (2014) electrospun nanofibres from extremely concentrated (160%) aqueous suspensions of cyclodextrin inclusion complexes (CD-IC) loaded with an antibacterial agent triclosan, using two forms of chemically modified CD, namely hydroxypropyl-beta-cyclodextrin (HPβCD) and hydroxypropyl-gamma-cyclodextrin (HPγCD). Bead-free membranes were obtained with average fibre diameter as 520 ± 250 nm and 1100 ± 660 nm, corresponding to former and latter forms of CD, which also showed very good antibacterial effect against both *E. coli* and *S. aureus*.

### **3.2 Nanofibres Encapsulating Bioactive Plant Extract**

Plants are sources of numerous chemical compounds that display antibacterial activity against a range of bacteria. For thousands of years, plant oils and extracts have been in use for a wide range of purposes (Hammer et al. 1999), antimicrobial functionality being the most popular. Additionally, the over-rising concerns about the hazardous side effects of conventionally used synthetic compounds in medication and nourishment industry, as well as the escalating resistance of pathogens to antibiotics have led to a growing interest among the research community to integrate natural extracts with polymers to electrospin nanofibres for various applications like wound dressings, scaffolds for tissue engineering, drug delivery and active food packaging systems (Sridhar et al. 2015; Khan and Shi Xiangyang 2018; Zhang et al. 2017a). Crude extracts can be easily obtained from fresh plants or from milled dried

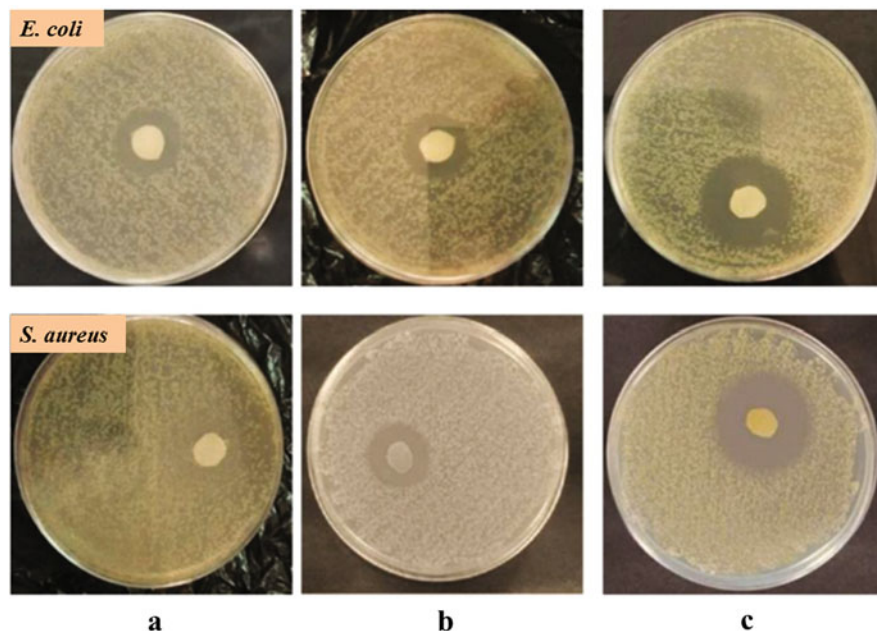
plants via organic solvent extraction. Numerous extracts of crude plants, for example *Garcinia mangostana*, *grewia mollis*, *aloe vera*, *centella asiatica*, *tecomella undulata*, *baicalein*, *chamomile*, *memecylon edule*, *Indigofera aspalathoides* and *Azadirachta indica*, have been reported to be successfully encapsulated in electrospun fibres (Motealleh et al. 2014; Suganya et al. 2011; Chan et al. 2017; Charernsriwilaiwat et al. 2013; Agnes Mary and Giri Dev 2015; Al-Youssef et al. 2013).

Ganesan et al. (Ganesan and Pradeepa 2017) electrospun 108–519 nm diameter fibres from a blend of 80% PVA and 20% tridax daisy (or *Tridax procumbens*) leaves extract. The developed nanofibrous mat created 45 mm and 36 mm zone of inhibition for *S. aureus* and *E. coli*, respectively, demonstrating strong resistivity against both bacteria. Yao et al. (2017) prepared an electrospun membrane from a mixture of 17% gelatin solution (in formic acid) and aqueous PVA, also containing gotu kola or *centella asiatica* extract and demonstrated it to be biodegradable, facilitate dermal wound healing and exhibit antibacterial activity against *S. aureus*, *E. coli* and *P. aeruginosa* with minimum inhibitory concentration (MIC) of 6.25 mg/mL for *S. aureus* and 25 mg/mL for both *E. coli* and *P. aeruginosa*. Yousefi et al. (2017) fabricated 90/10 chitosan/PEO nanofibrous mat loaded with Henna (*Lawsonia inermis*) leaves extract. Though Henna extract did not affect the electrospinnability of the precursor blend solution, but the fibre diameter was affected by its concentration. It was also observed that on increasing Henna extract loading from 1 to 2 wt%, the zone of inhibition against *E. coli* increased from 16 mm to 25 mm and from 14 mm to 18 mm for *S. aureus*, as seen in Fig. 2.

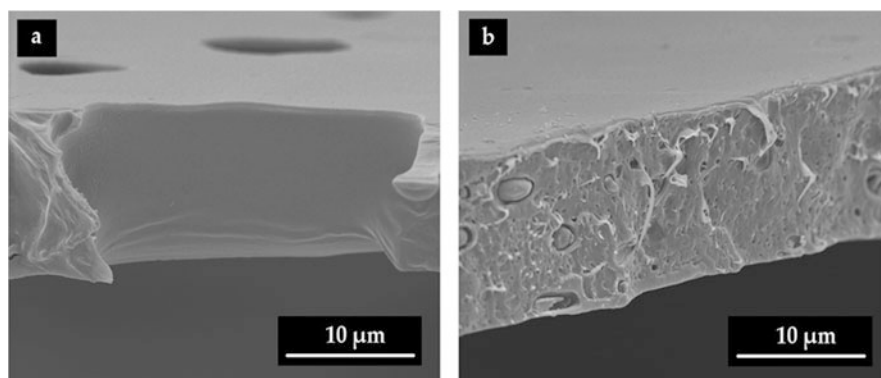
Radusin et al. (2019) electrospun films from pure PLA solution and PLA solution loaded with 10 wt.% wild garlic or *Allium ursinum* L (AU) extract at 2000  $\mu\text{L/h}$  flow rate and 14 kV applied voltage. After annealing the films under hydraulic press at 135 °C, without pressure for  $5 \pm 1$  s, followed by air-cooling at room temperature, PVA and AU were observed to exhibit ‘island-and-sea’ morphology, indicating successful encapsulation of AU droplets of size  $2.3 \pm 0.5$   $\mu\text{m}$  in the PLA matrix (Fig. 3). The PVA/AU film was found to exhibit high antibacterial activity against *E. coli*, but just reasonable against *S. aureus*. Zeyohanness and Zulkifli (2018) successfully electrospun bead-free nanofibres from 10% PVA solution loaded with rose myrtle or *Rhodomyrtus tomentosa* extract (RTE) in different proportions (0.25%, 0.5%, 1.5% and 2.5%). The concentration of RTE was observed to affect the average fibre diameter as well as the antibacterial activity of the fibres against *B. subtilis*, *E. coli*, *P. aeruginosa* and *E. faecalis* bacteria.

Essential oils (EOs) are usually derived from aromatic plants and are mixtures of various chemical compounds like linalool, pinene, eugenol and cymene, etc. They can be biosynthesised from various plant organs, including flowers, herbs, buds, leaves, fruits, bark, seeds, wood and roots (El Asbahani et al. 2015). EOs, for example *cinnamon*, *lemongrass*, *candeia*, *tea tree*, *lavender* and *thyme* have been extensively explored for integration of antibacterial property in electrospun fibrous mats. Zhang et al. (2017b) electrospun PLA solution loaded with different concentrations of tea tree and manuka oils at a flow rate of 2 mL/h, applied voltage of 18.5 kV and needle to collector distance of 15 cm to form antibacterial nanofibres.





**Fig. 2** Antibacterial activity of electrospun nonwoven mats of chitosan/PEO/Henna extract: (a) chitosan/PEO (0 wt.% Henna extract), (b) 1 wt.% Henna extract loading and (c) 2 wt.% Henna extract loading (Yousefi et al. 2017)



**Fig. 3** Scanning electron micrographs of cross-section of electrospun films of (a) neat PLA, (b) PLA/AU (Radusin et al. 2019)

The used EOs not only improved the antibacterial activity of PLA fibres, but also their mechanical properties, i.e. elongation at break and tensile strength. The bactericidal action of EOs was attributed to the partition of hydrocarbons into the bacterial membrane, further causing damage to the cytoplasmic membranes, thus disrupting their functions and ultimately causing cell lysis. Jung et al. (2020) developed

**Table 1** Antibacterial effect of nanofibrous membranes containing cinnamon oil (Jung et al. 2020)

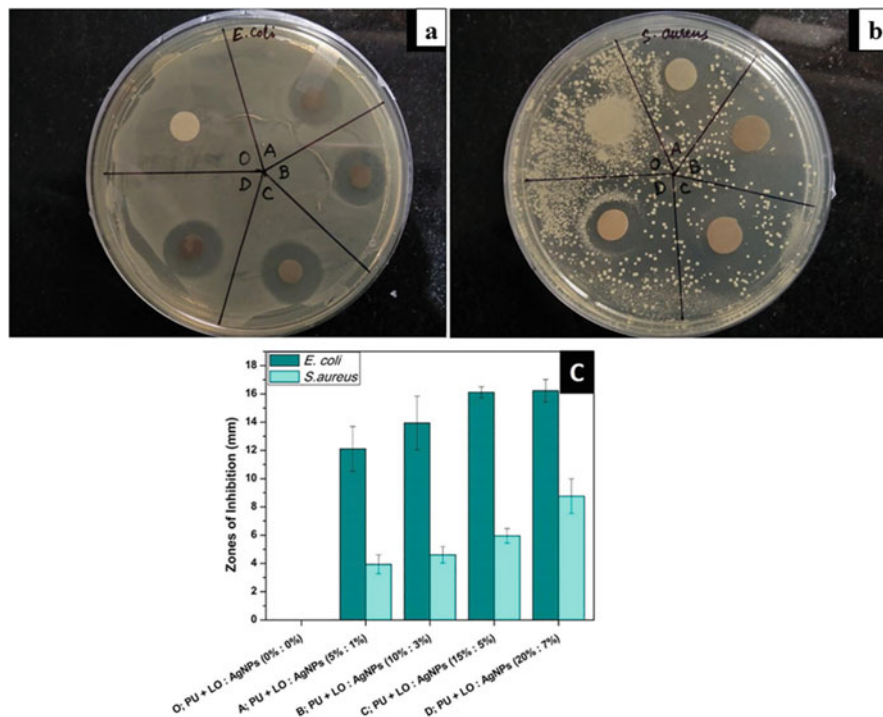
Bacteria	Contact time (h)	Number of bacterial colonies (CFU/mL)		Bacterial reduction (%)
		Control	Nanofibrous membranes containing cinnamon oil	
<i>S. aureus</i>	0	$2.5 \times 10^5$	$2.5 \times 10^5$	–
	24	$1.3 \times 10^5$	$9.0 \times 10^5$	99.9
<i>K. pneumoniae</i>	0	$2.1 \times 10^5$	$2.1 \times 10^5$	–
	24	$1.7 \times 10^5$	$3.0 \times 10^5$	0

membranes of core/sheath nanofibres of cinnamon oil (4.9 wt.%) /PVA by emulsion electrospinning. Upon evaluation of their antibacterial activity against *S. aureus* under dynamic contact condition, a reduction of 99.9% in the number of bacterial colonies was observed in comparison with the inoculated buffer solution (Table 1). However, the same nanofibrous membranes did not exhibit any inhibitory effect against *K. pneumoniae* because of more complex cell wall structure of this bacteria. Unalan et al. (2019) manufactured peppermint essential oil (PEP) loaded poly( $\epsilon$ -caprolactone) (PCL) electrospun fibrous mats with a smooth, uniform and bead-free morphology; and with average fibre diameter reducing from 1.6  $\mu\text{m}$  to 0.9  $\mu\text{m}$  with increase in concentration of PEP from 1.5 to 6% (v/v). Other than bringing reduction in fibre diameter, higher PEP concentrations also led to reduction in bacterial viability for *S. aureus* and *E. coli* strains. However, as *E. coli* consists of a double membrane with the outer membrane having a layer of lipopolysaccharide that prevents the penetration of certain antibacterial compounds (Burt and Reinders 2003), the PEP loaded fibrous mats were less effective against *E. coli*.

Sofi et al. (2019) fabricated composite nanofibres consisting of polyurethane (PU) loaded with different concentrations of lavender oil (0%, 5%, 10%, 15% and 20%) and Ag-NPs (0%, 1%, 2%, 5% and 7%). As seen in Fig. 4, the growth of *E. coli* and *S. aureus* strains was not inhibited by pure PU fibrous mats. However, all the mats containing fibres loaded with Ag-NPs and lavender oil were effective in suppressing the bacterial growth.

### 3.3 Composite Nanofibres

Numerous polymers have been electrospun into nanofibres over the past few decades. However, these single-component electrospun nanofibres typically have limited properties and cannot perform multiple functions (Gao et al. 2019; Sahay et al. 2012; Yang et al. 2014). Of late, many researchers have focused their work around inclusion of other nanoscaled structures in the nanofibres to create composite nanofibres with added functionalities. This section comprises studies related to functionalisation of electrospun nanofibres by incorporation of nanoparticles to improve their antibacterial activity.

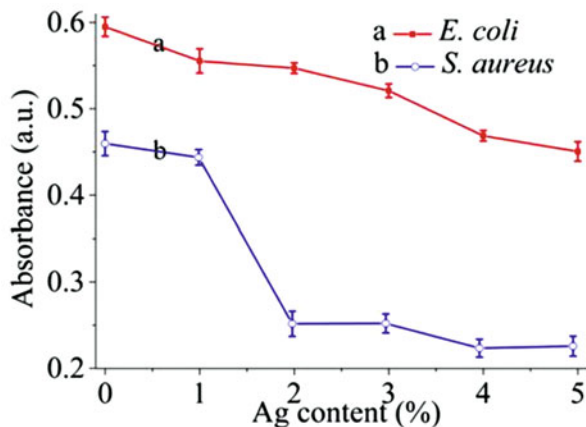


**Fig. 4** Zone of inhibition of pure PU fibre and PU/lavender oil/Ag-NPs composite fibre mats against *E. coli* and *S. aureus*; (a, b) Photos of agar plates from antibacterial testing; (c) average diameter of zone of inhibition (Sofi et al. 2019)

Augustine et al. (2014) added ZnO nanoparticles of size  $\sim 60$  nm in polycaprolactone (PCL) solution, in varying proportions, to electrospin PCL/ZnO nanocomposite membrane. For higher concentration of ZnO nanoparticles, fibre diameter was less than that of pure PCL fibres (2500 nm), and roughening of fibre surface was observed due to their agglomeration. The antibacterial activity assessment of these membrane revealed that membranes with less than 5 wt.% ZnO nanoparticles showed no activity against both bacteria. Notably, the bactericidal effect of ZnO nanoparticles activates only when they come in direct contact with the walls of the bacterial cells. At lower concentrations, most of the nanoparticles were trapped inside the polymer only, with very few superficial nanoparticles being able to make direct contact with bacterial cell wall.

Zhang et al. (2016) electrospun PVA/Ag-NPs composite nanofibrous membranes with varying concentration of embedded Ag-NPs (1–5%). The antibacterial activity of these nanofibres against *S. aureus* and *E. coli*, as evaluated via UV absorption method, has been depicted in Fig. 5 through the variation curves between UV-vis absorbance intensity and PVA/Ag-NPs nanofibres with different Ag-NPs content.

**Fig. 5** Variation curves between UV-vis absorbance and PVA/Ag-NPs nanofibres with different Ag-NPs content against (a) *E. coli* and (b) and *S. aureus* (Zhang et al. 2016)



The solution of bacterial suspension without any antibacterial agent in the broth medium was observed to be very turbid after incubation, supporting high UV-vis absorption. Besides, the UV-vis absorbance of PVA/Ag-NPs nanofibres was found to decrease with increase in concentration of Ag-NPs, for both bacteria. It was also observed that antibacterial activity of these fibres was better against *S. aureus* than against *E. coli*.

Tijing et al. (2012) electrospun mats from pure PU nanofibres and composite nanofibres of PU incorporated with tourmaline nanoparticles (TM-NPs) in varying proportion. Pure PU nanofibrous mat showed no zone of inhibition, i.e. absence of any antibacterial activity, against both *E. coli* and *Streptococci* bacterial strains. On the other hand, PU/TM-NPs composite mats showed distinct inhibition zone, with increasing average diameter corresponding to increasing TM-NPs content. It is important to note here that TM-NPs decrease the membrane fluidity of *E. coli*, leading to increase in cell membrane permeability, and subsequent cell death. Moreover, TM-NPs possess piezoelectric and pyroelectric effects, which facilitate in killing Gram-positive *Streptococci* bacterium.

## 4 Summary

The key attributes of electrospun nanofibres, such as high surface area-to-volume ratio, tuneable mechanical and physical properties, excellent porosity make them a popular choice for various applications. The last decade has seen an increased number of publications involving electrospinning of natural and synthetic nanofibres with bactericidal property. This number is, in fact, expected to rise even further in the upcoming years to fulfil the growing need for improvement in materials for wound dressings, dressings for dermal bacterial infections, artificial skin, implants and scaffolds for tissue engineering, membranes for air/water purification, protective

masks, sportswear, food packaging material, etc. There is still a tremendous scope for quality enhancement by exploring new antibacterial materials and finding ways to make their electrospinning feasible. Besides, a lot can be done to upscale the electrospinning set-ups for mass production of nanofibrous webs, as well as to make the process cost-effective and the final products affordable.

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