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Review Article

Emerging Applications of Electrospun Nanofibers Using Various Fabrication Techniques

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ABSTRACT

Nanofibers are fibers having nanometric diameters, made from a variety of polymeric materials, generally developed using electrospinning methods. It can incorporate multiple drugs, such as proteins, peptides, antibodies, and small molecules, either loaded inside or adhered to the surface. They offer enormous potential in the biomedical field, including drug delivery, regenerative medicine, stent coating, implants, and controlled drug release. The application of nanofibers in tissue regeneration have attracted a lot of interest due to their distinctive composition and structural characteristics. The application of nanofiber technology has also gained attention for wound healing, considering most conventional wound dressings have problems and drawbacks include infection, irritation, pain, and weak adhesive properties. This review highlights the major properties of nanofibers, their fabrication techniques, novel delivery approaches using electro spun technology for controlled drug release applications, and various other applications in the medical and healthcare field. The importance of Quality by Design (QbD) principles in the production of nanofibers, recent clinical trials conducted, patents filed in the field of nanofiber technology and various product developed by electrospinning technology were also discussed.

Keywords: Electrospun nanofiber, Self-assembly, Template synthesis, Tissue engineering, Scaffold.

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INTRODUCTION

One of the newest nanotechnologies, nanofibers, have numerous uses in delivering sophisticated systems to human bodies to combat various illnesses. The dimensions are 500–1000 nm (diameter). Drug delivery to a particular region in the body is made easier by the distinctive physical characteristics of nanofibers. The ratio of volume to surface area, tunable porosity, tiny pore size, low in density, and many highly automated qualities are important characteristics of nanofibers that can be adjusted for desired functions. Nanofibers are good materials for a variety of applications because of these characteristics. In addition to biomedical engineering, examining the potential functional and structural uses of nanofibers in areas including energy generation and storage, environmental treatment, water and healthcare has been a priority.[1]

Nanofibers are produced successfully using various materials like polycaprolactone, polymethacrylate, gelatin, polyacrylonitrile, polyethylene terephthalate, poly-L-lactide, polyglycolic acid, polybenzimidazole, nylon-6, polyvinyl alcohol, polyurethane, polystyrene, chitosan, polycarbonate, and others. Research on the Electromagnetic Wave (EMW) absorption characteristics of nanomaterials, particularly nanofibers, has attracted much attention and offers a huge number of potential applications in the future.[2]

The process of electrospinning is one of the most famous methods to make nanofibers since it is effortless to use and suitable for a variety of ceramics, polymers, and metals. Melting, flash spinning, force rotating, bicomponent spinning, drawing, phase separation, are other conventional old methods for nanofabrication. The majority of these techniques contain fibers combined into nanowebs, which are nonwoven random fiber mats made with fibers ranging between a few to hundreds of nanometers.[3,4] With significant potential for the future, particularly in the search for cancer treatment, nanotechnology in medicine has received much attention.[5]

Nanofiber mats offer a framework with strong interconnectivity, excellent absorption, controlled moisture and porosity, and gas permeability, which produces a setting that safeguards the wound from external infection. Nanofiber retains more moisture in its structure and keeps the area around the wound moist while it

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heals. As a result, the nanofibers cannot stick to the surface of the wound.[6]

The complexity of regulating the efficacy and safety of medications utilizing nanoengineered materials has significantly increased in recent years. A wide range of health issues are addressed by the use of nanotechnology and nanomaterials in the production of pharmaceuticals, including improved bioavailability, changed biodistribution, greater pharmacological action, stability of biodegradable drugs, and targeted drug administration. It is far more challenging to analyse the potential risks of nonbiological complicated drugs, and there is an urgent need for an integrated approach to address this problem. The United States Food and Drug Administration (US FDA) proposed a risk-based approach to drug engineering in 2000, which was later called as Quality-by-Design by the International Council of Harmonisation of Technical Requirements for Pharmaceuticals for Human Use. This approach aims to identify, analyze, and control all factors that could affect a new drug's quality and safety.[7]

This review mainly introduces the fabrication technology of nanofibers, focusing more on the electrospinning method, the current novel drug delivery systems based on modernized electrospinning fibers that hold promising potential for future application, a special focus on quality by design principles, essential parameters affecting nanofiber technology, and the application of nanofibers in various streams of science, medicine, and engineering.

NANOFIBER TECHNOLOGY

The creation of nanofibers utilizing various polymer types is possible using a variety of approaches. The design of porous nanofibers involves both conventional and cutting-edge techniques, which include electrospinning, phase separation, template synthesis, self-assembly, and many other methods. The most popular method of fabrication is electrospinning.

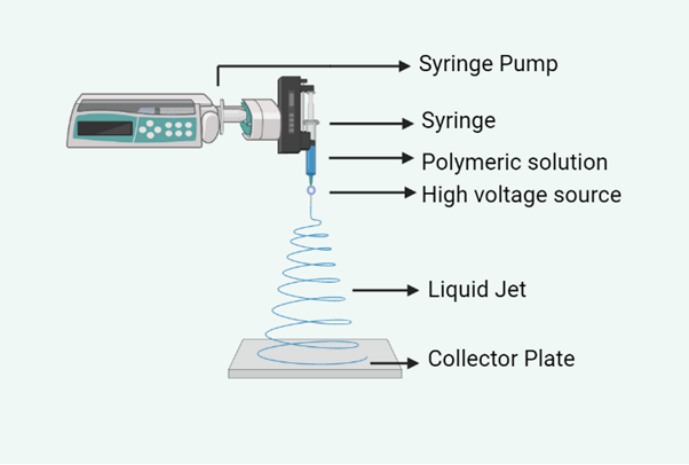

Figure 1: Electrospinning method.

Electrospinning method

The electrohydrodynamic phenomenon known as "electrospinning" is used to extrude and stretch tiny strands of polymer melts or solutions.[8] The necessary parts of the electrospinning device are a high-voltage generator, a supply of the system, and a collector, shown in Figure 1. An electrospinning device's supply system, collector, and high-voltage generator are necessary parts. The supply system part for solution electrospinning usually consists of a needle that is attached to a syringe containing a melted polymer or a solvent-based polymer solution (melt electrospinning). Electric charges are produced on the surface of the polymeric droplet at the tip of the needle in both cases when a voltage is applied between the grounded metallic collector and the needle ranging from 5 to 65 kV.[9] The outcome is that the droplet warps and takes on the form of a cone (Taylor cone). Can be spun electrically. This indicates that by modifying the viscosity, operating parameters as well as conductivity of the material, the size and shape of the nanofibers may be altered (voltage, needle-collector distance, humid conditions, and temperature). The polymer molecular weight and solution concentration is another factor essential for the development of fibers during electrospinning as that affects the entanglement of the polymer chains. When the liquid jet cannot be stabilized by chain entanglement, beads rather than homogeneous fibers grow. However, sufficient chain entanglement encourages the creation of fibrous structures as well as a continuous jet upon solvent evaporation.[10]

Self-assembly technique

This method has a large production capability and is also simple to carry out. The resulting nanofibers have a consistent shape and structure. The procedure for producing high-quality fiber is continuous. It still takes time and is only applicable to a certain group of polymers. Porosity maintenance and inadequate drug loading present significant difficulties. With this technique, the parts self-assemble into a nanomesh with the desired shape and functionality.[11]

Phase separation technique

The extraction of porous nanofibers using solvents with specified volumes is a phase separation technique. The intended product is produced after this phase under the circumstances of freezing and drying. Thermoresponsive polymers are best candidates for this technique. The dynamic behaviour of this type of nanofibers can be analysed by methods like dye-photosensitized T-jump technique which involves indirect heating by using 532 nm laser pulses and direct heating T-jump technique, which uses 1.2 μm laser pulses. Both methods provide similar results related to the sol-gel behaviour of nanofibers. There are serious stability problems with this method.[12]

Template synthesis methods

In this method, a fibril solid or hollow-shaped tubule was used to transfer the polymer solution into a solidification solution. This method enables the fabrication of many different basic materials, including metals, semiconductors, and carbon. This method allows for the fabrication of different types of nanofibers without the assistance of a specialist and allows for modification, but it is also a time-consuming process. This method cannot produce continuous nanofibers one at a time while utilizing numerous processes.[13]

QUALITY BY DESIGN FUNDAMENTALS AND PARAMETERS FOR THE PRODUCTION OF NANOFIBERS

Safety, efficacy and quality are the goals of the medication development process. End-product testing is the conventional method for evaluating product quality, which limits knowledge of the process and its crucial elements. This method of maintaining quality has many drawbacks; hence, QbD is implemented to increase product quality by giving little room for process variation. Following the release of multiple International Conference on Harmonization guidelines Q8, Q9, Q10, and Q11, regulatory bodies (the FDA, and the European Medicines Agency, or EMA) have approved QbD. According to the ICH Q8 guideline, QbD is a technique for establishing the process's initial goals and making them simpler to attain, comprehend, and control, as well as risk management for the quality of the finished product.[14]

It is a drawn-out process that calls for an expert's engagement in product and manufacturing process expertise, an assessment of variable risk, and statistical analysis. It enhances the effectiveness of the drug development process and the manufacturing process.[15] It offers a cutting-edge method for precisely and consistently producing goods.

It determines how crucial process variables can be changed to continuously make a therapeutic product with the required features. For this, the relation of product attributes, formulation, and factors in the making process (such as medicine components, excipient qualities, and process parameters) are established, and causes of variability are recognized. Using these data, a flexible and dependable manufacturing process is subsequently developed. This method allows for ongoing adjustments and the production of consistent products. As a result, some QbD components could define the product quality profile you want to achieve, identify important quality traits and process factors, identify sources of variability, and manage production procedures to ensure constant quality across time.

Creating the Quality Target Product Profile (QTPP) comes first. For nano-based goods, QTPP includes quality requirements that must be fulfilled to ensure a good quality of the finished product while also taking product efficacy and safety into account. In the second stage of QbD-based development, Critical Quality Attributes (CQAs) are identified.[15] CQAs are QTPP-derived product quality characteristics that affect the final product's quality; as a result, they need to be scrutinized. To achieve the intended quality, such parameters listed in Table 1 must be maintained throughout the development and manufacturing process. Any kind of change in process or formulation variables may pose a risk to CQAs.[16] The effect of these parameters directly compromises the quality of the product that can be seen by Ishikawa diagram in Figure 2. The production parameters like distance, diameter of tip, applied voltage, flow rate which directly affects the manufacturing of nanofibers. The solution parameters like molecular weight, concentration, conductivity, surface tension, and viscosity affect the thickness and texture of nanofibers. The stability parameters such as temperature and humidity also play huge role in smooth manufacturing of nanofibers. The characterization of such nanofibers is then analysed through surface: volume ratio, density, tunable porosity, pore size, and mechanical properties. Critical Process Parameters (CPPs) and Critical Material Attributes (CMAs) for the product in question, risk assessment by ICH Q9 to identify Design of Experiments (DoEs), design space to identify critical material attributes (CMAs),[17] and establishing a controlled strategy with continuous innovation and experimentation throughout the life cycle of the product.[18]

NOVEL DRUG DELIVERY APPLICATIONS

Numerous medication delivery systems have been created using various electrospun technologies. The triaxial electrospun technique enables the loading of different medications into a particular carrier with a complicated structure using various approaches. This has more benefits than traditional electrospinning approaches.[19] Drugs can be released under control and in the correct order of kinetics using rationally developed nanofibers.

Solubility enhancement for poorly soluble drug

Research has been performed on electrospun core-shell fibers for medications with limited solubility. Hydrophilic filament and polymeric matrix-containing acyclovir model drug have been described.[20] When utilized as a polymer to create a shell, cellulose acetate gave the medicine a sustained release pattern with a manageable rate. The inner fluid was a thick phospholipid solution with diclofenac sodium in ethanol solvent, but the outer eudragit coat was spinnable with a solvent N, N-dimethylacetamide and ethanol. Eudragit's solubility at higher pH values caused the core to dissociate into tiny phospholipid solution particles. Due to its smaller size after dissociating from phospholipid solution became more soluble. In their work, Yang *et al.* demonstrated how to enhance the permeability and solubility for biopharmaceutical classification system class-II and IV drugs, respectively, by

Table 1: Effect of different parameters on the quality of electrospun nanofibers.

Parameters	Effect on the quality of electrospun nanofibers
Voltage	When the voltage is increased above a critical amount, the diameter reduces at first, then grows after a point. Increased repulsion forces cause the initial decrease in diameter.
Distance of the electrode from the collector	There is insufficient time for solvent evaporation at a short distance. As a result, nanofibers develop flattened shapes.
Flow rate	The volume of the polymer solution in the Tylor cone increases as the flow rate increases, reducing the diameter at first. Beads form as the flow rate increases.
Surface tension	Proper jet initiation is aided by lower surface tension.
conductivity	The charge carrying capacity of high conductivity solutions is large, resulting in a higher applied voltage. When the voltage is increased, the diameter shrinks.
Viscosity	On their route from the needle to the collection, more viscous liquids tend to produce droplets. Beads are formed from these drops.
Solution concentration	As the concentration rises, the viscosity rises, forming droplets that dry as they approach the collection.
Temperature	As the temperature rises, the solution's concentration rises as well, enhancing viscosity and the beaded morphology is reduced as the viscosity rises.
Relative humidity	The pearl effect is amplified when the relative humidity rises.

electrospinning a variety of delicate solutions with spinnable fluid in Figure 3.[21]

Controlled release of drug

For the abatement of *Helicobacter pylori*, a number of mucoadhesive drug delivery systems, including cholestyramine nanocapsules, chitosan nanomeshes, and carboxy vinyl mucoadhesive nanospheres, have been proposed. Because pharmacological dosage forms with large specific surfaces have significant potential for interaction with biological surfaces, the use of nanofibers for bioadhesion is of interest.[22] This unique approach can be adjusted as needed in addition to aiding the zero-order release profiles, according to a contrasting release profile of this form of technology with drug depot and the delivery aligned inside drug-free liners. The drug release pathways are depicted in Figure 4, where the first potential mechanism involved was drug diffusion across a swellable membrane. Drug release through the end of a nanofiber was the second conceivable mechanism, however it may not be the primary one given the enormous length of nanofibers in comparison to fibre diameter. When the drug that was on the surface of the shell diffused out, it created some pores. These pores allowed media to reach the inner core, where it produced a diffusion route. When the release profiles of this type of drug delivery system were compared to depot systems embedded within drug-free shells, it became clear that the release profiles of the new system could be altered in addition to help preserve zero order release profiles. Therefore, while maintaining zero-order kinetics, electrospun nanofibers with functionalized drugs could provide prolonged drug release. Triaxial electrospinning offers a very creative method for creating new nanostructures, allowing for the expansion of drug delivery applications and obtaining the appropriate functional capabilities. Future research should concentrate on developing electrospun adaptive systems providing drugs for the therapy of various acute and chronic conditions like diabetes, cancer, and ophthalmic disorders.

According to reports, ferulic acid works as an antioxidant against radicals such as hydroxyl, superoxide, and nitric oxide. Therefore, it has enormous potential to cure cancer of the tongue, breast, prostate, lung, colon, and stomach.[23] Although the drug itself did not form a filament, it was thought that this technique could be incorporated into nanofibers in the form of a drug depot as seen in Figure 5. The triaxial procedure was mostly used with unspinnable medicinal solutions. Numerous investigations, including X-ray Diffraction (XRD) spectroscopy, Fourier Transform Infrared (FTIR), and *in vitro* release experiments, revealed the presence of crystalline ferulic acid peaks, supporting the drug's dispersion within the matrix system. Drugs were released at a zero-order rate due to the core-sheath system's persistent action. The disciplines of biomaterials and pharmaceutics would greatly benefit from new ways of producing this type of nanostructure, especially "top-down" techniques that can be easily scaled up.[24]

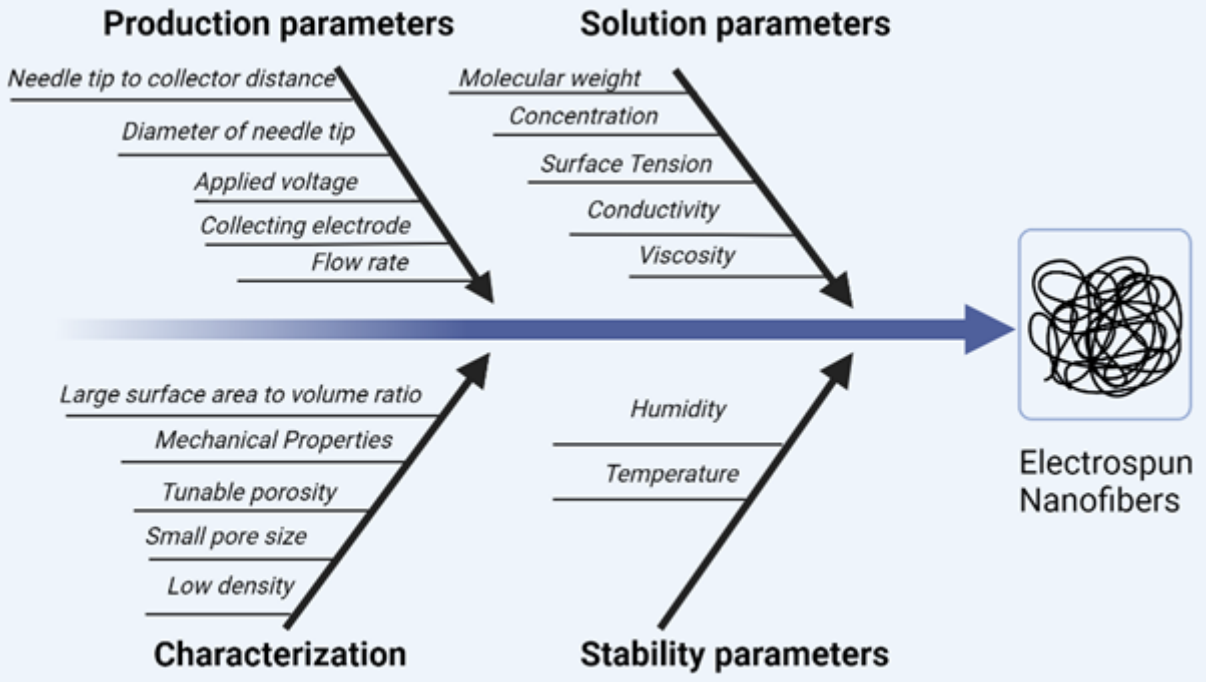

Figure 2: Ishikawa diagram–formulation development parameters for electrospun nanofibers.

Dual drug delivery system

Han *et al.* attempted to create a triaxial electrospinning-based dual-drug delivery device. Different colored dyes were utilized in this instance as a model medication. A solution of keyacid uranine (model drug) and polyvinylpyrrolidone, was used as the inner core layer. Poly (-caprolactone) solutions in a mixture of chloroform solvent and trifluoroethanol and poly (-caprolactone) with key acid blue in the solvent of trifluoroethanol were utilized as the central and outermost layers, respectively. Voltages ranging from 18 to 22 kV were used throughout the electrospinning process. The outcome of an *in vitro* release investigation showed 80% release of the outer layer dye in just one hour from the nanofiber sheath. This significant release of dye led to the creation of water channels, which made the external sheath absorptive. This improved absorptive property of the external sheath could cause the core to release drug in bursts as well.[25] According to this study, it may be possible to induce the release of pharmaceuticals from the core using stimuli-responsive polymers depending on changes in temperature or pH. It is anticipated that the hybrid nanofibrous mat will transport both hydrophilic and hydrophobic medications and proteins. It shows promising results for pharmaceutical products or biomedical applications, such as tissue engineering scaffolds.[26]

APPLICATIONS

Diagnostic applications of nanofibers

Biosensors can provide dependable information for diabetes prevention and diagnosis. The development of effective, stable, and affordable glucose sensors for the continuous monitoring of blood glucose levels has attracted considerable attention. Titanium dioxide/Copper nanofibers electrodes are a viable option as glucose sensors. A nanofiber electrode was created to electrochemically detect glucose in 0.1 M sodium hydroxide. Nanofiber-based glucose sensors mostly use optical and electrochemical detection to sense glucose levels. The nanofiber electrode tests with amperometric current-time values can yield results with high selectivity and strong stability.[27]

Pathological stimuli-responsive self-assembly peptide nanofibers accumulate different imaging agents as cargos in the targeted stimuli-rich regions. It enhances imaging signals, tumor/disease accessibility and retention, and biocompatibility, thereby providing sensitive tumor/disease imaging in preclinical as well as clinical studies.[28]

Self-assembled nanofibers are also used for cancer theranostics owing to their spatiotemporal responsiveness, *in situ* assembly, and diverse bioactivity. Recent advances in self-assembled peptide nanofibers focus on the dynamic process of capturing cancer cells from the outside in. The *in situ* self-assembly could be in response to pathological or physiological changes or dependent on various processes at different locations of tumors, such as forming a thrombus in tumor vasculature, barrier formation on the cancer cell membrane, and disrupting the cancer cell organelles. These nanofibers could form a drug depot *in situ* for the sustained release of chemotherapeutic drugs.[29]

Tumor complexity makes the development of highly sensitive tumor imaging probes an arduous task. A peptide-based near-infrared probe responsive to fibroblast activation protein-α specifically forms nanofibers on the surface of cancer-associated fibroblasts *in situ*. The assembly/aggregation-induced retention

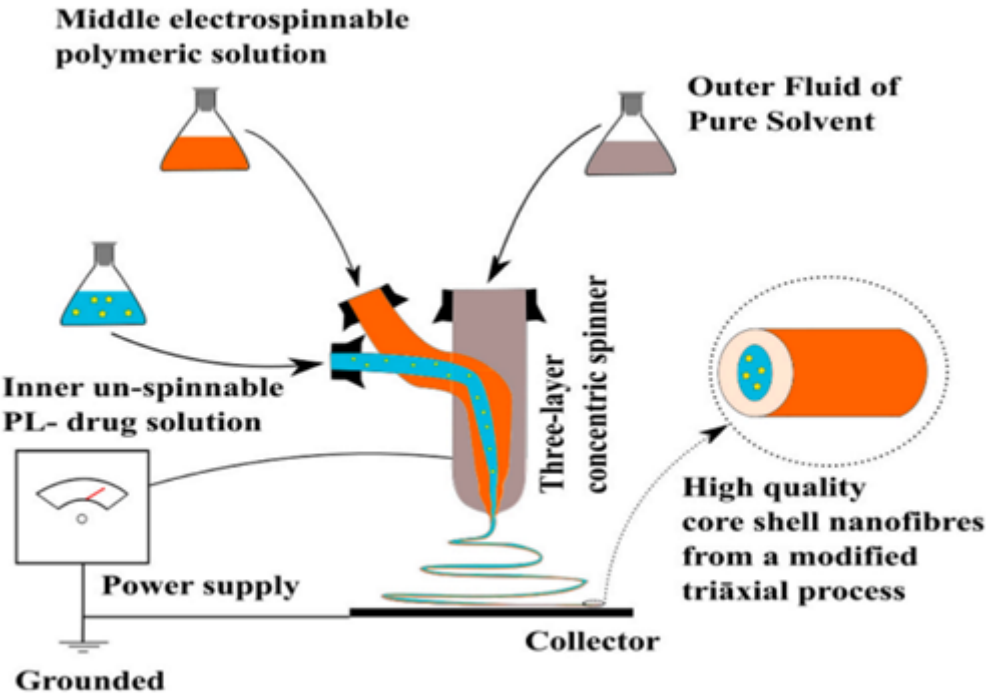

Figure 3: Triaxial electrospinning process used for poorly soluble drugs. Reprinted from with permission from Elsevier.[19] Copyright ® 2021.

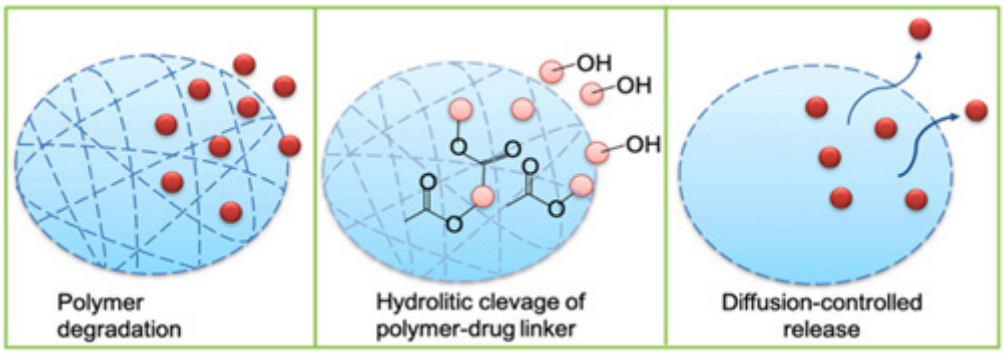

Figure 4: Drug release mechanisms. Reprinted from Ghosal *et al*. (2021) with permission from Elsevier.[19] Copyright ® 2021.

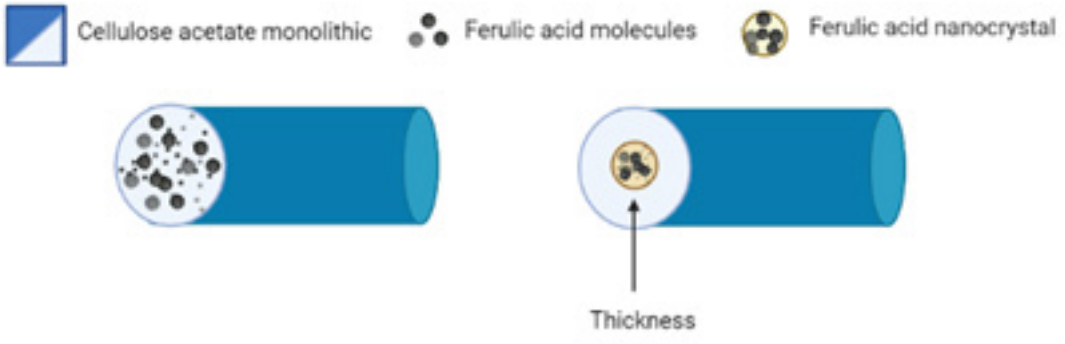

Figure 5: Nanodepot system of ferulic acid.

effect results in enhanced accumulation and retention of the probe around the tumor, resulting in a 5.5-fold signal enhancement in the tumor, and also provides a longer detectable window which helps in visualizing even small tumors.[30]

Therapeutic applications of nanofibers

The chrysin-curcumin nanofibers show anti-inflammatory characteristics at different stages of the healing process. A Poly(caprolactone)-Poly(ethylene glycol)-curcumin-chrysin solution was electrospun, and the process resulted in the creation of a yellow electrospun mat with little to no chrysin build-up on the surface. Chrysin-loaded nanofibers, curcumin-loaded nanofibers, and chrysin-curcumin-loaded nanofibers all have some dose-dependent effects on the wound-healing process.[31]

Shalumon Xing *et al.* investigated the antibacterial activity of Zinc nanoparticles electrospun onto sodium alginate polyvinyl alcohol fibers. Eighteen hours of incubation resulted in a 99.9% reduction in the number of bacteria. The nanofibers containing silver demonstrated strong antibacterial efficacy and efficiency. They evaluated the antibacterial activity of these structures against *Klebsiella pneumoniae* (*K. pneumoniae*) and *Staphylococcus aureus* (*S. aureus*) by *in vitro* studies using microplate proliferation tests. In order to investigate the *in vitro* cell compatibility of the scaffolds, they also examined the properties of osteoblast development and fibroblast adhesion, viability, and proliferation in NIH 3T3 fibroblasts. The authors came to the conclusion that cobalt is relatively more economical with reduced toxicity with the study results, hence it can be used to manufacture more antimicrobial electrospun nanofibers at lower costs.[32]

A brand-new class of bioactive nanofibers are made from collagen hydrolysate and essential oils (such as thyme or oregano). Collagen was successfully electrospun with the addition of the essential oils, producing porous nanofibers having a diameter that ranges from 471 nm to 580 nanometers. Microbiological experiments against *Staphylococcus aureus, Escherichia coli, Candida albicans*, and *Pseudomonas aeruginosa* revealed that the addition of essential oils to the novel collagen nanofibers enhanced their antibacterial characteristics. By cultivating a specific clone 929 fibroblastic cell line *in vitro* and measuring cell survival, the biocompatibility of collagen and that of collagen with essential oils was determined.[33] The electrospun collagen nanofibres with thyme essential oil have a slight and moderate cytotoxic effect only at 1000 $\mu g \cdot mL^{-1}$ concentration, while the electrospun collagen nanofibres with oregano essential oil have a slight and moderate cytotoxic effect at 500 $\mu g \cdot mL^{-1}$ concentration. In this case, the cytotoxicity limits of collagen nanofibres loaded with essential oils are significantly high in comparison to earlier reported results about the cytotoxicity concentrations for thyme oil. The adjustment of electrospun parameters has resulted in the creation of new collagen-based nanofiber mats with an addition of essential oils. These mats could be used as protective garments, tissue engineering materials, or wound treatments.[34]

The use of cell-filled scaffolds for the treatment of osteoarthritis are constrained by the morbidity of the donor site, high prices, and low performance.[35] To overcome these drawbacks, cell-free fibrous hyaluronic acid nano-scaffold was made that supplies two growth factors that are Stromal cell-Derived Factor (SDF)-1 and Transforming Growth Factor (TGF)-3. They have shown to improve cartilage repair and regeneration which promotes the invasion and recruitment of mesenchymal stem cells, and SDF-1 (SDF-1; SDF), migrate, and TGF promotes matrix synthesis.[36] However, scaffolds that released SDF *in vivo* caused a worse cartilage repair response (worse mechanics). When compared with nano-scaffolds releasing TGF alone, SDF and TGF both improved cell scores, demonstrating the bioactivity of both drugs *in vitro*.[37]

The low specificity of the anti-cancer drugs is a major obstacle for targeted therapy. After surgery, an appropriate concentration of the anticancer drug should be maintained in the local area to reduce this toxicity to normal cells. The anticancer drug-loaded nanofibers can provide sustained drug release at such local sites. As a result, nanofibers are one of the most effective methods for minimizing the risk of local recurrence of cancer following surgery and can be implanted directly into solid tumor cells for treatment.[38]

Drug delivery applications of nanofibers

Nanofibers are used as vehicles for the delivery of many bioactive compounds like drugs, gene sequences, and growth factors as they have high surface-to-volume ratio. Natural polymers are more prone to mimic an extracellular matrix whereas synthetic polymers are commonly used as reinforcements and sensors.[39]

In a study, the bacteria *Lactobacillus acidophilus* was encapsulated into nanofibers made of polyvinyl alcohol and polyvinylpyrrolidone with two different molar masses. This gave the solid dosage form a longer shelf life and made it easier for patients to administer it. Additionally, viability testing demonstrated that, when kept at (or below) 7°C, the nanofibers can maintain their long-term stability for large numbers of living bacteria. Furthermore, the developed biohybrid nanowebs can offer novel potential treatments for bacterial vaginosis because all of the nanowebs created in this study disintegrated immediately upon contact with water.[40]

Adhesion prevention during ventral operations may get benefit from the use of nanofiber-based barriers in conjunction with anti-adhesion medications. Nanofibers improve the antiadhesion capability of such barriers. An ibuprofen-loaded electrospun sandwich scaffold was created in a study which served as a physical barrier and as an efficient mediator delivering the drug to the wounded site. In comparison to other scaffolds, the Sandwich

Table 2: Clinical trials.

Sl. No.	Status of trial	Study	Condition	Interventions	Location
1	Ongoing	Using a nanofiber scaffold to be used in patients above 55 years for Rotator Cuff Healing.	Rotator Cuff Tears.	-	American Health Network Avon, Indiana, US Baptist Health System Lexington in Kentucky, United Status. Associated Orthopedists of Detroit Saint Clair Shores in Michigan, United Status.
2	Completed	Mouth-to-mouth Ventilation process efficiency through breathable Self-Sterilizing Respirator during BLS in COVID-19 Pandemic.	Ventilation during resuscitation process.	Procedure: Mouth-to-mouth ventilation Procedure: Quantitative analysis.	Faculty of Medicine, Masaryk University Brno in Czechia.
3	Completed	Spinner Device evaluation for the Application of Wound Dressing for various areas: for treating Split Skin Graft Donor Sites.	Skin wound.	Device: A Spinner Device: A Jelonet.	Burn Unit in Sheba Medical CenterRamat Gan in Israel. Kaplan Hospital Rehovot, Israel. Sourasky Medical Center Tel Aviv in Israel.
4	Unknown	Modified Antibiotic Nanofibers used as antimicrobial agent for Regenerative Endodontics Procedures.	Necrosis and Pulp.	Procedure: electrospun TAP nanofibers. Procedure: modified TAP paste.	

scaffold greatly reduced the first burst of drug release within 60 min and delayed the administration of ibuprofen over 14 days, in which long-term anti-adhesion properties are anticipated. Ibuprofen's ability to efficiently suppress fibroblast adhesion and proliferation was demonstrated in an *in vitro* investigation, with the produced sandwich maintaining the least amount of L-929 adherence (20%) after five days of continuous culture showing greater anti-inflammation activity.[41]

Surface Deacetylated Chitin Nanofibers (SDACNFs) and chitosan have been studied by Koizumi *et al.* for their preventative benefits against 5-fluorouracil (5-FU)-induced intestinal mucositis, the most well-known adverse effect of 5-FU chemotherapy. Chitosan and SDACNFs decreased intestinal crypt cell hyperproliferation and death and eliminated intestinal mucositis-related histological abnormalities. As a result, SDACNFs and chitosan may be effective agents for preventing mucositis caused by anticancer medications.[42]

Drug release applications of nanofibers

In order to achieve sustained ciprofloxacin release, various hydrophobic polymers alone in monolithic nanofibers or in combination with hydrophilic polymers in blended nanofibers were tested in a study. Up to 40 days, poly(methyl methacrylate) nanofibers released only 1.5% of ciprofloxacin. The release profile of ciprofloxacin was completely altered when hydrophilic polymers like polyvinyl alcohol, poly(ethylene oxide), and chitosan were added. The combination of poly(methyl methacrylate) and poly(ethylene oxide) produced burst drug release, the combination of polyvinyl alcohol and poly(ethylene oxide) produced a mixture of burst and sustained drug release, and the combination of 10% chitosan and ciprofloxacin produced sustained drug release. As a result, nanofiber drug release can be altered by blending it with various polymers.[43]

The spinal tracts get permanently disrupted by Spinal Cord Injuries (SCI), which ultimately result in functional impairment.

In order to replicate the mechanical properties of the spinal cord, hydrogels and self-assembling peptide nanofibers are frequently utilized. In a study, a biodegradable, three-dimensionally aligned nanofibers-hydrogel scaffold for nerve injury treatment provided sustained non-viral drug/gene delivery and contact guidance. The scaffold is made up of electrospun nanofibers made of aligned poly (-caprolactone-co-ethyl ethylene phosphate) that were distributed in a three-dimensional pattern within a collagen hydrogel. The controlled release of neurotrophin-3 for spinal cord injury treatment demonstrated by encapsulating it in such nanofibers increased nerve generation in the targeted area.[44]

PATENTS AND CLINICAL TRIALS

Recently, Iain Cameron Hosie and Bhuvaneswari Kannan created multilayer nanofibers with bioactive components for managing wound healing.[45] A matrix made of multifunctional nanofibers that are stacked together and each layer includes at least one or more functional bioactives that can be used alone or in combination to manage the entire wound cycle. Every nanofiber and its coat composition dissolve at a desirable, regulated release rate to release the bioactive agent when exposed to moisture or on wet skin, or a non-dissolving layer may serve as a sacrificial layer of the skin. Another innovation relates to a brand-new electrostatic spinning machine supply coating method for nanofibers.

A spinning motor, a coating device, electrostatically spun threads, metering pumps, and a liquid supply pipe make up the unique supply feeding mechanism. When a coating procedure is carried out on a polymer solution, the coating of the polymer solution on the electrostatically spun threads is more uniform, so that the applications of a polymer solvent are saved. The utmost advantage of the new supply feeding mechanism is that it is reasonable in design and simple in structure, and the issues of repeated coating in a short period and no coating for a long period are successfully overcome.[46] The complexity of electrospun nanofibers utilizing diverse processes has received significant attention from numerous other unique technologies. The various trials conducted over diverse applications of electrospun nanofibers are discussed below.

Rotator Cuff healing using a nanofiber scaffold

Following rotator cuff surgery, tendon failure to recover is common. Chronic pain, poor outcomes, and a substantial financial cost to society can all result from non-healing. A recently FDA-approved scaffold called a rotium nanofiber has been demonstrated in animal experiments to enhance tendon repair to the bone. Rotium is a nonwoven, microfiber matrix with FDA approval for rotator cuff restoration that is made of Polyglycolide (PGL) and Poly L-lactide-co-Caprolactone (PLCL) and the first prospective randomized clinical study in people to examine differences in healing and strength will be this one. The outcome to be determined is whether employing a nanofiber scaffold lowers the chance of postoperative failure for patients over the age of 55 undergoing Rotator Cuff Surgery (RCR).[47]

A breathable self-sterilizing respiratory system with mouth-to-mouth ventilation efficiency during Corona virus disease-2019 (COVID-19) Pandemic

The major goal of this trial is to evaluate the effectiveness of mouth-to-mouth breathing using breathable, self-sterilizing nanosized fiber respirators during the COVID-19 pandemic. On 3 different mannequins, including the Professional Adult Medium Skin Cardiopulmonary resuscitation monitoring, training Manequin with Cardiopulmonary resuscitation monitor (Prestan), hundred volunteers (medical students as trainers and others as volunteers) will perform a two-minute cycle in accordance with European Resuscitation Council (ERC) guidelines 2021. (Laerdal). The dummy will be used in randomized order. The effectiveness of mouth-to-mouth breaths will be assessed using "visible breath" and "not visible breath" metrics. The spectator might see the chest rise on the first mannequin. Rescuers are expected to be able to successfully provide mouth-to-mouth rescue breaths to the dummy during BLS simulation training while wearing permeable nanofiber respirators with layers of activated copper in compliance with safety rules. The outcomes were effectiveness of mouth-to-mouth ventilation procedure and analysis of the ventilation procedure.[47]

Spinner device evaluation for wound dressing application for the treatment of split skin graft donor sites

For external burns and wounds, the device is a convenient, transportable electrospinning device that makes customized *in situ* nanofiber therapies. This dressing's characteristics include nonadherence, superior absorbency, bacterial prevention, quick and painless peeling, high conformability, and wound coverage. The aim of the trial is to measure the efficacy and safety of the Spinner device and wound dressing in treating donor-site wounds with sizes ranging from 10 to 200 cm^2. The outcome observed were changes in the value of dermal safety (Draize) score and changes in wound healing.[47]

Modernized antibiotic nanofibers for regenerative endodontics as an antimicrobial product

Apexification or regenerative endodontic therapies using plasma rich in platelets, injectable platelet-rich fibrin, and platelet-rich fibrin, which are used in combined form or alone, can be utilized to treat young teeth with pulp necrosis. Compared to apexification, revascularization therapy has additional benefits, such as encouraging root-end development and strengthening.[48] This study seeks to investigate the antimicrobial efficacy of presynthesized new antibiotic-loaded pun fibers employed in patients having immature necrotic teeth since apical healing will not occur when inflammatory and infected tissue exists in the

oral region. The outcome measures observed were antibacterial impact (2-3 weeks) after taking intracanal medications on the number of bacteria that are colony-forming units and nanofiber morphology (1 week) under the Scanning Electron Microscope (SEM), surface morphology will be examined.[49] Table 2 summarizes these clinical trials with their status, condition and intervention.

CONCLUSION

With the ability to use biodegradable or nonbiodegradable materials that nanofibers offer, it is possible to achieve remarkable attributes such as finer control over drug delivery application and release kinetics. Growth hormones, antibiotics, antimicrobial peptides, and enzymes can all be delivered into the body by immobilizing them on nanofibers. Nanofibers with the ability to be loaded with bioactive substances provide the perfect environment for treating infections, developing bacterial biofilms, prolonging drug release, and accelerating the healing process. Irrespective of the method of synthesis, nanofibers have produced scaffolds with a large surface area and improved porosity. These characteristics have been proven to significantly influence the adhesion and other properties of nanofibers. As a result, these matrices are being investigated as scaffolds for many applications and for the controlled delivery of drugs, proteins, and DNA. All these findings largely confirm the idea that nanofiber-based scaffolds have great potential for use in a range of biomedical applications.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

QBD: Quality by design; **EMW:** Electromagnetic wave; **US FDA:** United States Food and Drug Administration; **XRD:** X-ray diffraction; **FTIR:** Fourier Transform infrared; **EMA:** European Medicines Agency; **QTTP:** Quality target product profile; **CQAs:** Critical quality attributes; **CPPs:** Critical process parameters; **CMAs:** Critical material attributes; **DOEs:** Design of experiments; **SDACNFs:** Surface deacetylated chitin nanofibers; **5-FU:** 5-fluorouracil; **HA:** Hyaluronic acid; **TGF:** Transforming growth factor; **SDF:** Stromal cell-derived factor; **Fe_2O_3:** Ferric oxide; **CNF:** Carbon nanofiber; **PGL:** Polyglycolide; **PLCL:** Poly L-lactide-co-caprolactone; **RCR:** Rotator cuff surgery; **COVID-19:** Corona virus disease-2019; **ERC:** European Resuscitation Council; **SEM:** Scanning electron microscope.

SUMMARY

Nanofibers are manufactured from a range of polymeric materials and have nanometric diameters. They are typically created using electrospinning techniques. It can include a variety of medications, including proteins, peptides, antibodies, and tiny compounds, either loaded internally or externally. They offer a great number of possibilities in the biomedical industry, including controlled medication release, stent coating, regenerative medicine, and drug delivery. Due to their unique composition and structural traits, nanofibers have found extensive use in the field of tissue regeneration. Given that most standard wound dressings have issues and drawbacks such as infection, irritability, pain, and inadequate adhesive characteristics, the use of nanofiber technology for wound healing has also attracted attention. This review focuses on the key characteristics of nanofibers, their manufacturing processes, novel drug delivery strategies utilizing electrospinning technology, as well as a number of other applications in the medical and healthcare industries. Recent clinical trials, patent applications in the field of nanofiber technology, the significance of Quality by Design (QbD) principles in the production of nanofibers, and numerous products created using electrospinning technology were also covered.

REFERENCES

1. Teck C. Progress in Polymer Science nanofiber technology: Current status and emerging developments. Prog Polym Sci. 2017;70:1-17.
2. Yuan X, Sha A, Huang S, Zhao H, Chen X, Zhang Y, *et al*. Boron nitride nanosheets decorated N-doped carbon nanofibers as a wide-band and lightweight electromagnetic wave absorber. J Alloys Compd. 2022;890:161903.
3. Ondarçuhu T, Joachim C. Drawing a single nanofibre over hundreds of microns. Europhys Lett. 1998;42(2):215-20.
4. Nayak R, Padhye R, Kyratzis IL, Truong YB, Arnold L. Recent advances in nanofibre fabrication techniques. Text Res J. 2012;82(2):129-47.
5. Rawal M, Singh A, Amiji MM. Quality-by-design concepts to improve nanotechnology-based drug development. Pharm Res. 2019;36(11):153.
6. Bastogne T. Quality-by-design of nanopharmaceuticals–a state of the art. Nanomedicine. 2017;13(7):2151-7.
7. Rasouli R, Barhoum A, Bechelany M, Dufresne A. Nanofibers for biomedical and healthcare applications. Macromol Biosci. 2019;19(2):e1800256.
8. Ibrahim YS, Hussein EA, Zagho MM, Abdo GG, Elzatahry AA. Melt electrospinning designs for nanofiber fabrication for different applications. Int J Mol Sci. 2019;20(10):2455.
9. Mele E, Bayer IS, Nanni G, Heredia-Guerrero JA, Ruffilli R, Ayadi F, *et al*. Biomimetic approach for liquid encapsulation with nanofibrillar cloaks. Langmuir. 2014;30(10):2896-902.
10. Zhang W, Ronca S, Mele E. Electrospun nanofibres containing antimicrobial plant extracts. Nanomaterials. 2017;7(2):42.
11. Endres T, Zheng M, Beck-Broichsitter M, Samsonova O, Debus H, Kissel T. Optimising the self-assembly of siRNA loaded PEG-PCL-lPEI nano-carriers employing different preparation techniques. J Control Release. 2012;160(3):583-91.
12. Tsuboi Y, Yoshida Y, Okada K, Kitamura N. Phase separation dynamics of aqueous solutions of thermoresponsive polymers studied by a laser T-jump technique. J Phys Chem B. 2008;112(9):2562-5.
13. Qi A, Chan P, Ho J, Rajapaksa A, Friend J, Yeo L. Template-free synthesis and encapsulation technique for layer-by-layer polymer nanocarrier fabrication. ACS Nano. 2011;5(12):9583-91.
14. Cunha S, Costa CP, Moreira JN, Sousa Lobo JM, Silva AC. Using the Quality by Design (QbD) approach to optimize formulations of lipid nanoparticles and nanoemulsions: A review. Nanomedicine. 2020;28:102206.
15. Cook J, Cruañes MT, Gupta M, Riley S, Crison J. Quality-by-Design: Are we there yet? AAPS Pharm Sci Tech. 2014;15(1):140-8.

16. Simões A, Veiga F, Figueiras A, Vitorino C. A practical framework for implementing Quality by Design to the development of topical drug products: Nanosystem-based dosage forms. Int J Pharm. 2018;548(1):385-99.
17. Martins S, Tho I, Souto E, Ferreira D, Brandl M. Multivariate design for the evaluation of lipid and surfactant composition effect for optimisation of lipid nanoparticles. Eur J Pharm Sci. 2012;45(5):613-23.
18. Gupta MS, Kumar TP, Gowda DV, Rosenholm JM. Orodispersible films: Conception to quality by design. Adv Drug Deliv Rev. 2021;178:113983.
19. Ghosal K, Augustine R, Zaszczynska A, Barman M, Jain A, Hasan A, *et al*. Novel drug delivery systems based on triaxial electrospinning based nanofibers. React Funct Polym. 2021;163:104895.
20. Liu ZP, Cui L, Yu DG, Zhao ZX, Chen L. Electrosprayed core-shell solid dispersions of acyclovir fabricated using an epoxy-coated concentric spray head. Int J Nanomedicine. 2014;9(1):1967-77.
21. Yang C, Yu DG, Pan D, Liu XK, Wang X, Bligh SWA, *et al*. Electrospun pH-sensitive core-shell polymer nanocomposites fabricated using a tri-axial process. Acta Biomater. 2016;35:77-86.
22. Umamaheshwari RB, Ramteke S, Jain NK. Anti-Helicobacter pylori effect of mucoadhesive nanoparticles bearing amoxicillin in experimental gerbils model. AAPS Pharm Sci Tech. 2004;5(2):60-8.
23. Hassanzadeh P, Atyabi F, Dinarvand R. Nanoencapsulation: A promising strategy for biomedical applications of ferulic acid. Biomed Rev. 2017;28:22-30.
24. Yang GZ, Li JJ, Yu DG, He MF, Yang JH, Williams GR. Nanosized sustained-release drug depots fabricated using modified tri-axial electrospinning. Acta Biomater. 2017;53:233-41.
25. Han D, Steckl AJ. Triaxial electrospun nanofiber membranes for controlled dual release of functional molecules. ACS Appl Mater Interfaces. 2013;5(16):8241-5.
26. Yan S, Xiaoqiang L, Shuiping L, Xiumei M, Ramakrishna S. Controlled release of dual drugs from emulsion electrospun nanofibrous mats. Colloids Surf B Biointerfaces. 2009;73(2):376-81.
27. Tian L, Hu W, Zhong X, Liu B. Glucose sensing characterisations of TiO2/CuO nanofibres synthesised by electrospinning. Mater Res Innov. 2015;19(3):160-5.
28. Jiang Q, Liu X, Liang G, Sun X. Self-assembly of peptide nanofibers for imaging applications. Nanoscale. 2021;13(36):15142-50.
29. Liu N, Zhu L, Li Z, Liu W, Sun M, Zhou Z. *In situ* self-assembled peptide nanofibers for cancer theranostics. Biomater Sci. 2021;9(16):5427-36.
30. Zhao XX, Li LL, Zhao Y, An HW, Cai Q, Lang JY, *et al. In situ* Self-Assembled Nanofibers Precisely Target Cancer-Associated Fibroblasts for Improved Tumor Imaging. Angew Chem Int Ed Engl. 2019;58(43):15287-94.
31. Mohammadi Z, Sharif Zak M, Majdi H, Mostafavi E, Barati M, Lotfimehr H, *et al*. The effect of chrysin–curcumin-loaded nanofibres on the wound-healing process in male rats. Artif Cells Nanomed Biotechnol. 2019;47(1):1642-52.
32. Maleki DS, Sharifi S, Jahangiri A. Electrospun nanofibers as versatile platform in antimicrobial delivery: Current state and perspectives. Pharm Dev Technol. 2019;24(10):1187-99.
33. Costa MF, Durço AO, Rabelo TK, De Barreto RSS, Guimarães AG. Effects of carvacrol, thymol and essential oils containing such monoterpenes on wound healing: A systematic review. J Pharm Pharmacol. 2019;71(2):141-55.
34. Berechet MD, Gaidau C, Miletic A, Pilic B, Râpă M, Stanca M, *et al*. Bioactive properties of nanofibres based on concentrated collagen hydrolysate loaded with thyme and oregano essential oils. Materials. 2020;13(7):1618.
35. Baker BM, Gee AO, Metter RB, Nathan AS, Marklein RA, Burdick JA, *et al*. The potential to improve cell infiltration in composite fiber-aligned electrospun scaffolds by the selective removal of sacrificial fibers. Biomaterials. 2008;29(15):2348-58.
36. Baker BM, Shah RP, Silverstein AM, Esterhai JL, Burdick JA, Mauck RL. Sacrificial nanofibrous composites provide instruction without impediment and enable functional tissue formation. Proc Natl Acad Sci U S A. 2012;109(35):14176-81.
37. Martin AR, Patel JM, Locke RC, Eby MR, Saleh KS, Davidson MD, *et al*. Nanofibrous hyaluronic acid scaffolds delivering TGF-β3 and SDF-1α for articular cartilage repair in a large animal model. Acta Biomater. 2021;126:170-82.
38. Luo X, Xie C, Wang H, Liu C, Yan S, Li X. Antitumor activities of emulsion electrospun fibers with core loading of hydroxycamptothecin via intratumoral implantation. Int J Pharm. 2012;425(1-2):19-28.
39. De Nascimento LTR, De Amoêdo CVMM, Silva CF, Costa Cruz SBS, Gondim BLC, Mondelli RFL, *et al*. Current applications of biopolymer-based scaffolds and nanofibers as drug delivery systems. Curr Pharm Des. 2019;25(37):3997-4012.
40. Nagy ZK, Wagner I, Suhajda A, Tobak T, Harasztos AH, Vigh T, *et al*. Nanofibrous solid dosage form of living bacteria prepared by electrospinning. Express Polym Lett. 2014;8(5):352-61.
41. Mao Y, Chen M, Guidoin R, Li Y, Wang F, Brochu G, *et al*. Potential of a facile sandwiched electrospun scaffold loaded with ibuprofen as an anti-adhesion barrier. Mater Sci Eng C Mater Biol Appl. 2021;118:111451.
42. AL-Jbour ND, Beg MD, Gimbun J, Alam AKMM. An overview of chitosan nanofibers and their applications in the drug delivery process. Curr Drug Deliv. 2019;16(4):272-94.
43. Zupančič Š, Sinha-Ray S, Sinha-Ray S, Kristl J, Yarin AL. Long-term sustained ciprofloxacin release from PMMA and hydrophilic polymer blended nanofibers. Mol Pharm. 2016;13(1):295-305.
44. Nguyen LH, Gao M, Lin J, Wu W, Wang J, Chew SY. Three-dimensional aligned nanofibers-hydrogel scaffold for controlled non-viral drug/gene delivery to direct axon regeneration in spinal cord injury treatment. Sci Rep. 2017;7(1):42212.
45. Nejaddehbashi F, Hashemitabar M, Bayati V, Abbaspour M, Moghimipour E, Orazizadeh M. Application of polycaprolactone, chitosan, and collagen composite as a nanofibrous mat loaded with silver sulfadiazine and growth factors for wound dressing. Artif Organs. 2019;43(4):413-23.
46. Danyang Co. Ltd., Novel supply coating mechanism of nanofiber electrostatic spinning machine.
47. Trials NC [cited 6/2/2023]. Available from: https://clinicaltrials.gov/ct2/show/NCT04867265.
48. Mallishery S, Shah T. Regenerative endodontics–looking inward. J adv Med Med res. 2020;32(7):83-98.
49. Elgamal SG, El-Shafei JM, Azzazy HME, Dwedar RA, El-Khodary SA. Evaluation of the Antimicrobial Effect of Pre-Synthesized Novel Antibiotic Electrospun Nanofibers as an Intracanal Delivery Strategy for Regenerative Endodontics: A Randomized Clinical Trial. Open Access Maced J Med Sci. 2022;10(D):303-10.

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