Study on the Application of MNSs/PLGA Nanocomposites in Biomedicine

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Abstract. The ways and effects of metal-based nanostructure (MNSs) to improve the physical or biomedical properties of PLGA polymers were studied. Firstly, the preparation methods of MNSs/PLGA nanocomposites are introduced, including solvent method, non-solvent method, MNSs direct modification method and single emulsion-solvent evaporation method. Then the applications of MNSs/PLGA nanocomposites in biomedical fields such as drug delivery, biological imaging and tissue regeneration are reviewed. Finally, the potential advantages and development trends of MNSs/PLGA nanocomposites in the treatment of diseases were analyzed on the basis of in vitro / vivo studies.

1. Introduction
Biodegradable synthetic polymers, such as polylactic acid (PLA) [1-2], polyglycolic acid (PGA) [3] and poly(lactic acid-glycolic acid) (PLGA) [4-5], have been proverbially used for biomedical science. Among above-mentioned biodegradable synthetic polymers, PLGA is extensively used in food, biomedical engineering and biomedical field [6-7]. However, the poor mechanical properties and poor biological activity of pure PLGA are the main bottlenecks of its application. In order to get over these drawbacks, metal-based nanostructure has been added to PLGA, to synthesize metal-based polymer materials with excellent biodegradability. Commonly used metal nanomaterials include pure metals (such as silver, platinum), metal oxides (such as iron oxide, titanium dioxide) and metal alloys (such as magnesium-tin Alloy).

Compared with the traditional composites, the combination of MNSs and PLGA matrix greatly changes the physical, chemical and biological properties of the composites. MNSs/PLGA nanocomposites have a variety of excellent electrical, mechanical, antibacterial and anticancer properties. In this paper, firstly, the preparation route of MNSs/PLGA composites is introduced. Then the applications of these composites in biomedical fields such as drug delivery, biological imaging and tissue regeneration are reviewed.

2. Preparation of MNSs/PLGA nanocomposites
Generally speaking, MNSs/PLGA nanocomposites can be prepared by the following four different techniques. The first is the solvent method (Figure 1a). Dissolve PLGA in a solvent such as CHCl₃,
and then mix MNS into the above solution in the magnetic stirrer [8]. The MNSs/PLGA composite membrane can be obtained in this way. In evidence, the biggest odds of the above-mentioned means is the simple preparation route and the high solubility of the polymer in volatile solvents.

The second is physical and chemical solvent/non-solvent method (Figure 1b). This way offers a high-speed, repeatable and adjustable ideas used to produce spongy-like connectable structures which can be used as a scaffold for drug delivery, tissue regeneration and any other application. Specifically, dissolve PLGA in a solvent such as acetone at 25°C. Then metal-based nanoparticles (such as Ag) aqueous solution is added to above solution. To precipitate (ethanol, for example) in a non-solvent solution at last[9], that is, MNSs is combined into the PLGA copolymer mold. The main barrier of the second technique is the change in acetone concentration. If the concentration of acetone is greater, smaller molecular chains will be released, blocking the holes and causing scaffolds to collapse, freeze-drying will occur.

In addition to the above two materials, MNSs can be mixed with circular PLGA nanoscale particles in two distinct patterns: the first is MNSs modified PLGA, but only if the exterior of PLGA nanoscale particles is multi-functional. The second kind is MNSs inserted PLGA, in which MNSs is inserted in circular PLGA nanoscale particles. Most of these two methods are made by emulsion system[10].

The third is MNSs direct modification method(Figure 1c). This method supplies amplified interaction between MNSs and PLGA interface and gets better physical, chemical and mechanical performance. The process is one of the most valid ways to prevent MNSs assembling and preserve its characteristics[11]. By this means, PLGA nanoscale particles are scattered in H2O and incorporated into the silver nitrate solution containing metal ions [12]. Subsequently, the obtained mixture is blended with NaBH₄, a type of reducing agent, to bring about the ultimate outcome. The primary restriction of such means is that the contaminants are wrapped in polymeric substance in the process of the mixture process.

The fourth is MNSs encapsulated PLGA (Figure 1d). This synthesis technique consists of two stages. At the beginning, PLGA is dissolved and MNS is dispersed in a similar low boiling point solvent such as CH₂Cl₂ or CHCl₃. Oil-in-water emulsions are formed by put in H₂O and stabilizers, as formed of polyvinyl alcohol, to the oil phase of the PLGA solution. The following procedure is to vaporize low-boiling volatile solvent through uninterrupted mixing at 25°C or under decompression conditions. The organic solvent is evaporated and precipitated, and the polymer obtained is nanospheres. The advantage of this method is that there is no need to supplement surfactants to generate emulsions. The principal drawback of above-mentioned method is that it limits the ability of disperse hydrophilic polymer molecule in oil-in-water emulsions[13]. In addition, it is difficult to collect or remove old oil from emulsion as external phase.

In addition to the above methods, electrospinning (Figure 1e) is also used to prepare nanofiber composites, which is widely used in biomedicine[14].

Figure 1. Five methods for construction of MNS/PLGA nanocomposites[8-9,12-14].
3. Application of MNSs/PLGA nanocomposites in biomedicine

3.1 Sustained drug release

Sustained drug release means that anti-cancer drugs or gene drugs are first delivered to the lesion site through passive targeting, and then the drug is released. So far, PLGA and its composites have been used as impressive transporters for sustained drug release in the form of thin films or nanoparticles. Neat and functionalized PLGA particles can improve the location of anti-cancer drugs or gene drugs at the mark site and contribute to the continuous emancipate of encapsulated drugs. PLGA nanoscale particles release anti-cancer drugs or gene drugs through brief spreading in the process of spontaneous deesterification of PLGA matrix. The molecular release rate depends on the concentration of the drug, the model of force among drugs and PLGA, and the molar ratio of monomers in PLGA. Drug release can be accelerated by improving the ratio of GA to LA monomer, resulting in stronger hydrophilicity of PLGA [15]. It can also be adjusted by introducing three-level components, that is, the components are placed and floated in the PLGA carrier[16-17].

Compared with the polymer matrix, due to the existence of nanoscale metals in the compound structure, this fresh type of nanocomposites has more ideal makings properties. The combination of MNSs into PLGA has been used to adjust its release rate and perform additional performance, for instance photothermal therapy or biological imaging. The role of different types of MNS/PLGA in medical science are able to realize distinct effect. For instance, magnetic NPs has been utilized to regulate the transfer of biomolecules by producing extrinsic magnetic fields that are not the same with the physical and chemical conditions of mammalian bodies [18]. PLGA-PEG-PLGA triblock copolymer contains Fe3O4 NPs. Sulfamethoxazole was loaded into the above Fe3O4/PLGA compound and evaluate its drug release effect in the appearance or nonappearance of an outside magnetic field. Compared with no magnetic field, the nanocomposites can maintain the magnetism in the predetermined position or lesion location. Under opposite conditions, Fe2O3 NPs will cause carrier contraction, resulting in faster speed. In another study, Fe3O4-packaged PLGANPs was prepared to transport an anti-cancer drug paclitaxel. The drug release in vitro of magnetic NPs depends on acidity useful for the treatment of cancer cells with low pH value. Compared with free paclitaxel, these iron(III) oxide nanocomposites were more poisonous to MG-63 cells[19].

3.2 Biological imaging and phototherapy

Biological imaging includes the method of real-time non-invasive visualization of biological processes. Biological imaging can observe subcellular structures, for example Mitochondria, Lysosomes, Golgi bodies, DNA and Ribosomes, Cells tissues, and Multicellular organisms. Within cell biology, the technology is employed to follow cellular processes, calculate levels of ions or metabolites, and define the interplay of living elements.

Nanomaterials have important application value in biological imaging owing to their dimension, shape, ingredient, catalysis, plasma, magnetism or luminescence. The nanomaterials used in biological imaging are designed according to the target location and disease type. Ligand-functionalized NPs can improve the targeting efficiency and improve the imaging results.

For example, its surface was modified with folic acid to prepare both fluorescent and magnetic PLGA-based nanoscale particles (Figure 2). NPs containing emission conjugated polymer and lipid coated Iron Oxides can easily internalize MCF-7 breast cancer cells and has no apparent toxicity to normal cells [20].

Figure 2. Schematic diagram of both fluorescent and magnetic PLGA-based nanoscale particles [20].
3.3 Tissue engineering

Tissue engineering pushes forwards self-healing of injured tissue by using cells with the ability to proliferate and differentiate. In the above process, a scaffold is needed to establish an unnatural surrounding to sustain the cells, which are predestinated to regenerate damaged tissues and growth factors that stimulate cell differentiation. PLGA is a kind of synthetic copolymer with biocompatibility and biodegradability. It has been used in tissue engineering not only with adjustable degradation rate, but also easily decomposed into non-toxic carbon dioxide. The application of PLGA NPs in tissue engineering generally concentrates on bone regeneration and damage rehabilitation. Give an example, TiO2/PLGA nanocomposite biofilm has been used as unnatural wound bandage in vivo [21]. As mentioned earlier, metal-based NPs have favorable antibacterial performance. In MNSs, Mg nanostructure is regularly used in the area of tissue engineering. Studies have demonstrated that Mg nanostructure can significantly improve the adhesion of human osteocytes, improve the adhesion of osteoblasts, improve bone healing and accelerate bone regeneration and differentiation of osteoblasts has biological activity, can speed up bone regeneration [22], and their effectiveness is associated with the concentration of Mg2+ released [23]. Mg/PLGA biodegradable microspheres were produced by S/O/W emulsion method with MgO and MgCO3 nanostructures co-embedded PLGA microspheres. In vivo studies displayed that Mg/PLGA NPs released Mg2+, efficaciously regenerated mouse skull faults [24].

![Figure 3. Schematic diagram of the preparation of PLGA microspheres with Mg2+][24].

Other metal nanomaterials, such as gold nanoparticles (GNPs), are also applied for biocompatible nanomaterials [25]. GNPs enhances cell proliferation with the attachment of biomolecules. In addition, GNPs binds to mercaptan groups to form conjugates with a large number of materials. On this account, PLGA nanofibers containing mercaptan group (-SH) were synthesized and used to combine with GNPs. Owing to the strong covalent bond of Au-S covalent bond, it can be used for the self-assembly of PLGA-SH on the Au surface, attaching the PLGA-SH sheet to the GNPs. PLGA/GNPs have strong osteogenic potential [26].

![Figure 4. Schematic of GNPs/PLGA nanofiber production][25].
4. Conclusions and perspectives
This article reviews the preparation methods of MNSs/PLGA nanocomposites and their applications in biomedical fields such as drug delivery, bioimaging, and tissue regeneration. The combination of MNSs and PLGA improves the mechanical performances of PLGA, which is beneficial to the combination of PLGA and biological tissue. In addition, nano-metals or their oxides bring new performances for PLGA. In chemical PTT, X-ray and PA imaging fields, nanoparticles or capsules of PLGA are modified or embedded in nano-metals, e.g. GNPs, as a contrast agent for disease diagnosis and treatment. In connection with tissue engineering, the prevalence of multidrug-resistant microorganisms has given rise to new attentiveness in the use of antibacterial MNSs. In the group of the nano-fillers, Ag-Nps is the most commonly used commercial nanocomposites. These nanoparticles have been integrated into the PLGA substrate. As an effective antibacterial nanocomposite, MNSs/PLGA-based scaffold has the advantage of promoting bone regeneration in hard tissue engineering. In addition to the above devices, metals and metal oxides and magnetic nanoparticles are also introduced into PLGA for drug/biomolecule delivery. These nanocomposites show faster drug release under external magnetic field.

Although extensive experimental studies have been carried out on MNSs/PLGA composites, due to the existence of MNSs, its application in clinical field is still limited by safety. In this aspect, biosynthetic MNSs with lower cytotoxicity may build the path for safer drug applications. Over and above that, the following study ought to alleviate these problems through making use of bioactive molecules to participate in functionalized MNSs. The size and structure of metal-based nanostructures should be balanced. For example, although smaller Ag-NPs have better antibacterial activity, they have substantial cytotoxicity to host cells. Therefore, the expected biological activity and cytotoxicity of the nano-metals used should be balanced. In order to form a completely safe profile, it is necessary to conduct clinical trials of the final product before it is commercialized.

References
[1] Zagho M, Hussein E A, Elzatahry T, et al. A Recent overview in functional polymer composites for biomedical applications[J]. Polymers, 2018, 10(7): 739.
[2] Leite J, Mano F. Biomedical applications of natural-based polymers combined with bioactive glass nanoparticles[J]. Journal of materials chemistry B, 2017, 5(24): 4555-4568.
[3] Lee J Y, Kim S E. Young-Pil. Osteogenesis and new bone formation of alendronate-immobilized porous PLGA microspheres in a rat calvarial defect model[J]. Journal of industrial and engineering chemistry, 2017, 52: 277-286.
[4] Kamalakannan R, Mani G, Muthusamy P, et al. Caffeine-loaded gold nanoparticles conjugated with PLA-PEG-PLA copolymer for in vitro cytotoxicity and anti-inflammatory activity[J]. Journal of Industrial and Engineering Chemistry, 2017, 51: 113-121.
[5] Shim Y B, Jung H, Woong J, et al. Fabrication of hollow porous PLGA microspheres using sucrose for controlled dual delivery of dexamethasone and BMP2[J]. Journal of Industrial and Engineering Chemistry, 2016, 37: 101-106.
[6] Mohanty S, Panda S, Purohit D, et al. A comprehensive review on PLGA - based nanoparticles used for rheumatoid arthritis[J]. Research Journal of Pharmacy and Technology, 2019, 12(3): 1481-1488.
[7] Martins C, Sousa F, Araujo F, et al. Functionalizing PLGA and PLGA Derivatives for Drug Delivery and Tissue Regeneration Applications[J]. Advanced Healthcare Materials, 2018, 7(1): 1701035.
[8] Irfan H L, Jeenat A, Nagi R E R, et al. Transition Metal Oxides: a rare interaction of ferroelectricity and magnetism. [J]. Nanoscale research letters, 2019, 14(1): 142-146.
[9] Pramila K, Ravi K Y, Deepak K S, et al. Biogenesis of metal nanoparticles and their pharmacological applications: present status and application prospects[J]. Journal of Nanostructure in Chemistry, 2018, 1-38.
[10] Ping Y, Zhang J, Xing T, et al. Green synthesis of silver nanoparticles using grape seed extract
and their application for reductive catalysis of Direct Orange 26 [J]. Journal of Industrial & Engineering Chemistry, 2018, 58: 74-79.

[11] Nazarzadeh Z, Ehsan M, Franklin R. Recent progress in the industrial and biomedical applications of tragacanth gum: A review[J]. Carbohydrate Polymers, 2019, 212: 450-467.

[12] Makvandi P, Ali G W, Della S F, et al. Biosynthesis and characterization of antibacterial thermosensitive hydrogels based on corn silk extract, hyaluronic acid and nanosilver for potential wound healing[J]. Carbohydrate Polymers, 2019, 223-235.

[13] Makvandi P, Ali G W, Della S F, et al. Hyaluronic acid/corn silk extract based injectable nanocomposite: A biomimetic antibacterial scaffold for bone tissue regeneration. [J]. Materials Science & Engineering C, 2020, 107-110.

[14] Nazarzadeh Z, Ehsan M, Pooyan B, et al. Antimicrobial gum bio-based nanocomposites and their industrial and biomedical applications. [J]. Chemical Communications, 2019, 55(99): 14871-14885.

[15] Giliopoulos, D, Zamboulis P. Polymer/Metal Organic Framework (MOF) Nanocomposites for Biomedical Applications.[J]. Molecules, 2020, 25(1): 185.

[16] Kouli M, Banis G, Savvidou M G, et al. A Study on Magnetic Removal of Hexavalent Chromium from Aqueous Solutions Using Magnetic/Zeolite-X Composite Particles as Adsorbing Material[J]. International Journal of Molecular Sciences, 2020, 21(8): 2707.

[17] Ceren A D, Nuray Y, Ayşe K, et al. Synthesis and characterization of Fe-mptms-PLGA nanocomposites for anticancer drug loading and release studies[J]. Artificial Cells, Nanomedicine, and Biotechnology, 2017, 45(7): 1408-1414.

[18] Ehsan N Z, Rezvan J, Parvaneh N, et al. Metal-Based Nanostructures/PLGA Nanocomposites: Antimicrobial Activity, Cytotoxicity, and Their Biomedical Applications. ACS Applied Materials & Interfaces, 2020, 12 (3): 3279-3300.

[19] Giri T. Prospects of pharmaceuticals and biopharmaceuticals loaded microparticles prepared by double emulsion technique for controlled delivery[J]. Saudi Pharmaceutical Journal, 2013, 21(2): 125-141.

[20] Ana S, Meltem S, Marina M, et al. PLGA/Nano-ZnO Composite Particles for Use in Biomedical Applications: Preparation, Characterization, and Antimicrobial Activity[J]. Journal of Nanomaterials, 2016, 21(8): 2707.

[21] Corinne N R, Feini Q, Dong H K, et al. Electrospun PLGA Nanofiber Scaffolds Release Ibuprofen Faster and Degrade Slower After In Vivo Implantation[J]. Annals of biomedical engineering, 2017, 45(10): 2348-2359.

[22] Dheda A, Keertan D, Gumbo T, et al. The epidemiology, pathogenesis, transmission, diagnosis, and management of multidrug-resistant, extensively drug-resistant, and incurable tuberculosis[J]. The Lancet Respiratory Medicine, 2017, 5(4): 291-360.

[23] Kirsi S, Ilkka M, Tuula A N, et al. Growth mode and physiological state of cells prior to biofilm formation affect immune evasion and persistence of staphylococcus aureus[J]. Microorganisms, 2020, 8(1):106.

[24] Lin Z, Wu J, Qiao W, et al. Precisely Controlled Delivery of Magnesium Ions Through Sponge-Like Monodisperse PLGA/Nano-MgO-Alginate Core-Shell Microsphere Device to Enable In-situ Bone Regeneration. Biomaterials, 2018, 174, 1-16.

[25] Xue Y R, Li X, Li H B, et al. Quantifying Thiol-Gold Interactions Towards the Efficient Strength Control. Nat. Commun. 2014, 5, 1-8.

[26] Lee D H, Lee S L, Heo M, et al. Poly(Lactide-Co-Glycolide) Nanofibrous Scaffolds Chemically Coated with Gold-Nanoparticles as Osteoinductive Agents for Osteogenesis[J]. Appl. Surf. Sci. 2018, 432, 303-307.