compuscript

Acta Materia Medica

Review Article

Advances in therapeutic nanodrug delivery systems for infectious lung diseases: a review

Gang Sheng[a,b,1], **Na Tian**[a,b,1], **Huijuan Duan**[a,b], **Zhaogang Sun**[a,b,*], **Hongqian Chu**[a,b,*]

[a]Translational Medicine Center, Beijing Chest Hospital, Capital Medical University, 101149, China
[b]Beijing Tuberculosis and Thoracic Tumor Research Institute, Beijing 101149, China

[1]These authors contributed equally to this work.
***Correspondence:** sunzhaogang@bjxkyy.cn (Z. Sun); chuhongqian@bjxkyy.cn (H. Chu)
Tel.: +86-10-89509566

Received: 19 June 2022; Revised: 11 August 2022; Accepted: 20 August 2022

Published online: 30 August 2022

DOI 10.15212/AMM-2022-0019

ABSTRACT

Infectious lung diseases are inflammatory diseases of the lungs caused by infectious agents such as bacteria, viruses or fungi. Oral or intravenous administration of antibiotics is the most common method of treatment, but some drugs have poor release stability, high systemic toxicity and susceptibility to drug resistance. Nanodrug delivery systems are promising alternatives for the treatment of infectious lung diseases, because they provide the advantages of enhancing the stability and solubility of delivered drugs, increasing pulmonary accumulation, decreasing systemic toxicity and ameliorating drug resistance. This review provides a brief overview of recent advances in approaches and ideas in pulmonary drug delivery methods. We believe that nano-based therapeutic strategies offer great potential to broaden the scope of treatment of infectious lung diseases and enhance therapeutic efficacy.

Keywords: infectious lung diseases, nanodrug delivery, treatment

1. INTRODUCTION

Infectious lung diseases have become a global public-health problem. These diseases are usually caused by bacteria, viruses or fungi, and result in inflammation and structural changes in the lung parenchyma or interstitial disease. Lung injury is also an important cause of secondary lung infections, often in association with primary disease, thus leading to adverse clinical outcomes [1]. The main current treatments for infectious lung diseases include antibiotics and vaccines [2, 3]. The primary mode of drug delivery remains oral or intravenous administration. Inhalation has gained increasing attention as an important local administration route. However, each of these modes has distinct advantages and limitations in the treatment of lung diseases. Oral drugs have high patient compliance and are easy to use, but the drugs must be absorbed through the gastrointestinal mucosa; thus, the mucosal penetration and stability of the drug pose challenges, particularly for poorly soluble drugs [4, 5]. Many new drugs have remained undeveloped because of their low solubility [4]. Intravenous administration, a systemic drug administration route, avoids crossing the gastrointestinal barrier but requires high-dose administration to achieve favorable therapeutic effects, thus inevitably affecting normal organs (e.g., the spleen, liver or kidneys). In addition, poor patient compliance and high treatment costs are associated with intravenous drug administration. Long-term intravenous drug injection often causes adverse reactions, such as phlebitis and bacterial drug resistance. Inhalation is a local drug administration route for the management of lung diseases. Through special atomization devices, such as pressurized metered-dose inhalers or dry powder inhalers (DPIs), drug particles can be directly inhaled into the lungs, thus decreasing the adverse effects of systemic drug delivery, allowing for lower dosages and increasing drug bioavailability [6]. However, some drugs can also be deposited in the trachea, oral cavity or other non-infected sites, thus potentially damaging normal tissues and showing low therapeutic efficacy [7].

To overcome the limitations of the above administration routes, scientists have incorporated traditional drugs directly into nano-scale drug particles or combined appropriate carrier materials with Active Pharmaceutical Ingredient (APIs) to form nano-scale (approximately 1–1000 nm) particles to improve various characteristics of the drugs, which are called nanodrugs. Nanodrugs have incomparable advantages over traditional drugs [8]. Nanomaterials can significantly

improve the solubility, stability and tissue penetration of drugs. Surface modification of nanocarriers can be used to enable targeting to sites of disease occurrence. A variety of drugs with different functions and mechanisms can also be simultaneously encapsulated in nanocarriers to enhance their synergistic therapeutic effects. In addition, nanodrugs have great potential to decrease the rate of drug resistance after the widespread use of antibiotics in infectious diseases.

In this review, we summarize recently used strategies for the treatment of infectious lung diseases based on nanodrug delivery systems, with a focus on the following carrier materials: (1) polymers, (2) liposomes, (3) solid liposomes, (4) inorganic nanoparticles, (5) nanomicelles and (6) combinational nanosystems.

2. ADVANGATES OF THE PULMONARY NANODRUG DELIVERY SYSTEM

2.1 Increasing drug stability and solubility to achieve controlled release

Some drugs require sustained release during clinical application to maintain therapeutic effects. However, ordinary drug particles are generally exposed directly to the internal environment, and the drug rapidly disintegrates and disappears after diffusion. In nanomedicines, a core-shell structure is used to wrap the drug inside the cavity or sandwich it in a protective layer; the drug then diffuses outward at a constant rate across the nano-shell, thus resulting in controlled drug release and a prolonged half-life. The nanocarrier protects the drug from direct contact with plasma proteins, digestive enzymes and other machineries involved in drug degradation and removal, thus enhancing the stability of the drug. Nanocarriers also enhance drug solubility. Most drugs used in the lungs are lipid soluble, and can easily penetrate and enter cells. However, some antibiotics, such as aminoglycosides and β-lactams, have poor lipid solubility. If the drug structure is directly altered or the drug is modified, the therapeutic efficacy is decreased, and the potential biotoxicity is increased. Nanomaterials with good bio-penetration and modifiability are excellent choices for drug delivery. The use of nanocarriers can alter the lipid solubility of these drugs, increase their solubility in target organs and improve their therapeutic effects [9]. Some hydrophobic drugs can be physically modified by polymeric carriers to increase their solubility and delivery efficiency [10].

2.2 Weakened first-pass elimination effects and enhanced pulmonary accumulation of drugs

Drugs used to treat infectious lung diseases are usually administered orally or intravenously. Oral medications have the best patient compliance and are easy to administer. However, drugs often must cross the gastrointestinal mucosal barrier or pass through the bloodstream to reach the body, and most drugs are not effectively deposited in the lungs [11, 12]. To achieve therapeutic concentrations of drugs in the lungs, patients often must take large doses of drugs, thus inevitably resulting in severe systemic toxicity. Nanocarriers differ from prodrugs in that they can be arbitrarily modified through the surface attachment of, or encapsulation in, molecules with lung-targeting properties—such as chemical groups, nucleic acid fragments or peptides—thus enabling targeted deposition of drugs and increasing drug concentrations in the lungs to achieve better therapeutic effects [13, 14]. In addition, cell-membrane biomimetic nanoparticles are a new type of drug delivery system (DDS) in which cell membranes with various biological activities are wrapped by nanoparticles, thus causing organisms to recognize them as "self-components" and avoiding their removal. The "nanoghost" model has become a new breakthrough in the field of DDS [15].

2.3 Combating drug resistance

Drug resistance is an adaptive process of natural selection that occurs mainly in patients who require long-term antibiotic use. Patients with drug-resistant infectious lung diseases often continue to receive low doses and irregular application of antibiotics or immunosuppressive drugs [16, 17]. When drug resistance occurs, clinicians must change the antibiotics or increase the dose, thus posing a heavy financial burden on patients and potentially leading to more severe drug toxicity [18]. The mechanisms of bacterial drug resistance have been widely explored and are generally considered to be associated with the following mechanisms: (1) catabolic inactivation of antibiotics; (2) alteration of the drug's target of action; (3) altered permeability or other properties of bacterial cell membranes; (4) specific drug efflux pump systems; and (5) formation of bio-permeable membranes. Nanodrug carriers "camouflage" drugs, thus enabling evasion of drug recognition by pathogens and avoiding expulsion of drugs outside bacteriophages [19, 20]. In vitro studies have shown that nanodrugs, particularly liposomal drugs, can enter bacterial or host cells, release drugs in large quantities and continuously exert powerful antibacterial effects. Biofilms are biological colonies formed by aggregated and closely connected microorganisms on the surfaces of tissues; conventional antibiotics with relatively large particle sizes have difficulty in crossing the tight bacterial interstices, thus leading to the development of drug resistance [21]. Nanodrug carriers, which usually have a particle size of only 1–1000 nm, can pass through the gaps between bacteria in biofilms and inhibit the biofilm formation process, thus overcoming biofilm-induced drug resistance [19].

3. NANODRUG DELIVERY SYSTEMS FOR THE TRATMENT OF LUNG DISEASES

3.1 Polymeric microparticles and nanoparticles

The polymers currently used in the medical field can be divided into natural polymers and synthetic polymers. Most natural polymers are of animal, plant or microbial origin, such as chitosan, alginate, gelatin or albumin.

Natural polymers are considered available carriers for drug delivery because of their biodegradability, biocompatibility and low toxicity [22]. Given the demand for higher drug delivery performance and more stable delivery, synthetic polymers have been widely studied. Compared with natural polymers, synthetic polymers have predictable properties and functions, owing to their advantages such as adjustable degradation kinetics and mechanical properties. We next introduce two promising polymers developed in recent years: chitosan (CS) and poly(lactide-co-glycolide) (PLGA).

3.1.1 Chitosan. CS is a naturally occurring linear polysaccharide found in the exoskeletons of crustaceans such as shrimp, lobsters and crabs, as well as in some microorganisms, such as fungi and yeast. CS is the most widely used natural polymer in DDSs. The structure of CS is similar to that of cellulose [23] and differs only in the group attached to the C2 position. Consequently, CS has favorable fibrillogenic and moisturizing properties. CS is the product of partial acetylation of the natural polysaccharide chitin, which has favorable cell affinity and biodegradability, and low cytotoxicity [24]. Several studies have shown that CS has immune enhancement, anti-tumor, anti-inflammatory, anti-bacterial and anti-pulmonary fibrosis effects in the lungs [25-27]. CS particles increase the bioavailability and decrease the adverse effects of drugs by prolonging the residence time of locally administered drugs or by opening the tight junctions between epithelial cells [28]. These features not only enable proper delivery of the carriers but also avoid being uptaked by macrophages [29]. CS contains many free amino groups with cations, which can be cross-linked with multivalent anions. Therefore, CS can be easily combined with positively charged substances such as drugs, other polymers or modifying chemical groups, via electrostatic action. CS has low solubility in water, but better solubility in hydrochloric and acetic acids. In addition, inhalable CS drug formulations should contain aerodynamic sizes ranging from 1 to 5 mm (dry) to successfully reach the lungs, swell after deposition in moist lungs and provide sustained controlled drug release through a polymeric matrix. Researchers have prepared CS derivatives such as hydroxypropyl CS, carboxymethyl CS and quaternized CS to increase water solubility and tissue penetration [27]. Si et al. [30] have developed a cationic antimicrobial polymer named 2,6-diamino CS (2,6-DAC) with enhanced biodegradability and biocompatibility; CS membrane permeability; and proton sponge effects of CS, which has excellent antibacterial activity. In some studies, CS nanoparticles (CSNPs) have been coupled with bioactive components such as functionally modified peptides [31], cell membranes, cell receptors [32] and metal elements [29, 33] to endow them with targeted killing or strong protective abilities. Ding et al. [34] have prepared red-blood-cell-hitchhiking nanoparticles (RBC-MPSS-CSNP) by using an ionotropic gelation technique to load methylprednisolone sodium succinate into CSNP and mixed it with red blood cells from the rat abdominal aorta (**Figure 1a**). RBC-MPSS-CSNPs have a long duration of action in vivo and preferentially accumulate in the lungs, thus facilitating sustained release of drugs to the lungs and overcoming the drawback of the short duration of action of methylprednisolone sodium succinate in vivo (**Figure 1b**).

CS has many amino groups and consequently can interact with mucous membranes with acidic groups, thus enhancing the deposition of drugs in mucous membranes. Therefore, CS modification has good potential for application in local mucosal drug delivery. Scolari et al. [35] have reported functionalized alginate nanoparticles loaded with rifampicin (RIF) and the antioxidant ascorbic acid (ASC) with CS and the nonionic surfactant T80, thus providing a hydrophilic protective layer through the "sugar cluster effect" (**Figure 2a**). Experimental data have shown that nanoparticles loaded with RIF/ASC are mucus inert and can penetrate the mucus layer of the respiratory tract (**Figure 2b**) and become internalized. The nanoparticles have no influence on cell metabolism during this process.

Researchers have attempted to treat chronic lung diseases with CS nanoparticles, such as pulmonary cystic fibrosis (CF), tuberculosis (TB) and chronic obstructive pulmonary disease (COPD) [36]. Mutation of the CTFR gene leads to pulmonary CF and subsequently to various pulmonary infections, particularly *Pseudomonas aeruginosa* infection. Gene therapy is a feasible approach for the treatment of CF. Viruses are usually transfected with target genes, thus potentially resulting in dangerous induction of an immune response. Therefore, safe and effective delivery of target genes to target cells remains a challenge [37]. Fernández et al. [38] have designed a non-viral gene transfection system (CS-pEGFP-siRNA complex) based on CS nanoparticles and used it to simultaneously deliver pEGFP and siRNA. Successful transfection efficiency was achieved when cells were either transfected with CS-pEGFP or co-transfected with CS-pEGFP-siRNA at an N/P charge ratio of 12. In another study, CS nanoparticles were used to deliver wild-type CTFR-mRNA (wtCTFR-mRNA), and the sodium-channel permeability of capsaicin-free epithelial cells was used to increase transfection efficiency, thus successfully restoring CTFR gene activity in transfected cells [39]. Beyond gene therapy, CS nanoparticles play a role in treating CF-induced *P. aeruginosa* infection. To overcome biofilm formation, researchers have used DNase-1 to functionalize CS nanoparticles, thus effectively inhibiting biofilm formation. DNase-I functionalization of ciprofloxacin-loaded CS nanoparticles can overcome biofilm-mediated resistance of *P. aeruginosa*. The functional combination of CS nanoparticles with secretory leukocyte protease inhibitor inhibits not only the reproduction of *P. aeruginosa* but also the inflammation caused by the infection [40]. In a study examining the antibacterial activity of CS-coated antibiotic nanoparticles against *P. aeruginosa*, ten antibiotics

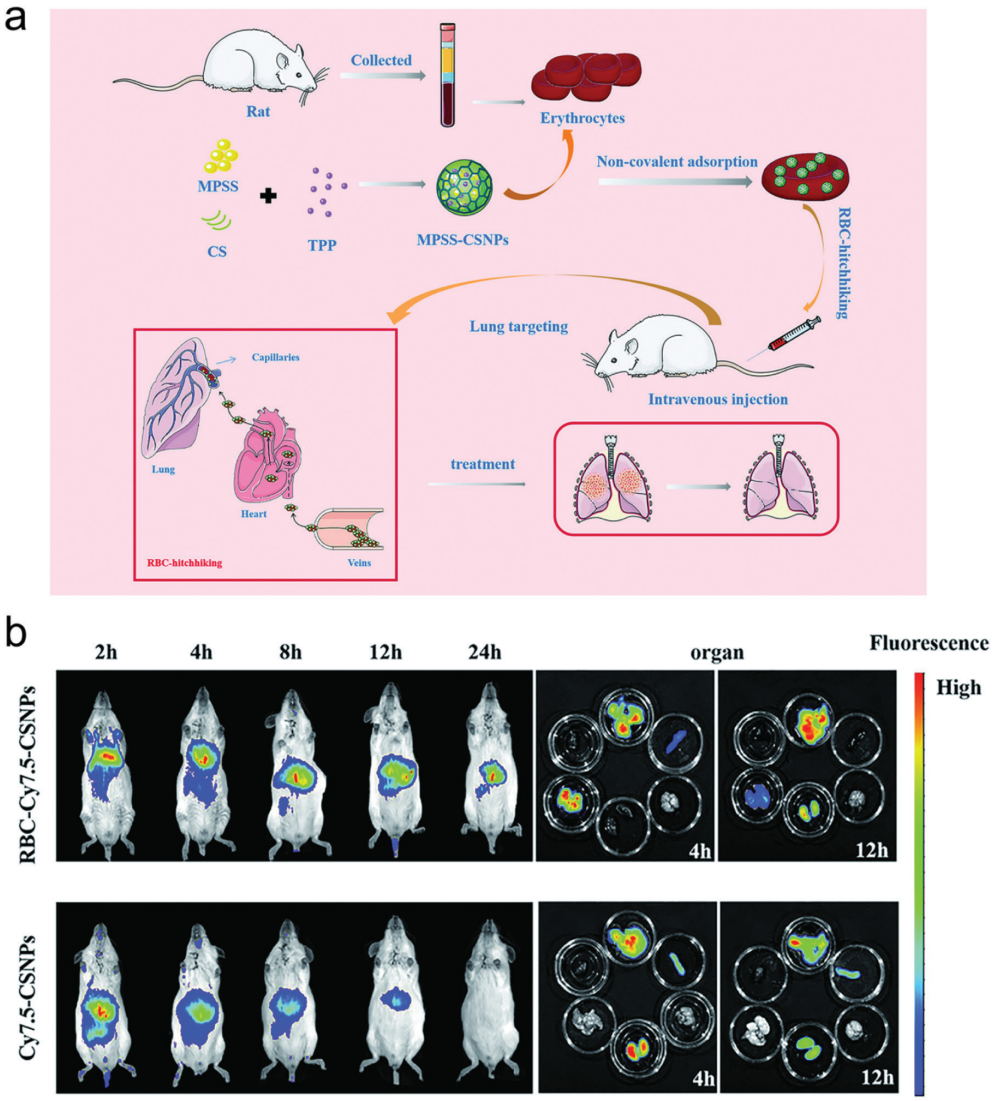

Figure 1 | (a) The fundamental principle of RBC hitchhiking: RBC-hitchhiking nanoparticles enter the lungs and weaken drug clearance from the liver. (b) Cy7.5-CSNPs and RBC-Cy7.5-CSNPs were injected intravenously into rats, and in vivo imaging and post-dissection organ imaging were performed at specific times. Copyright 2022, Science Direct [34].

were mixed with CS and CS oligosaccharide with various degrees of deacetylation. All CS showed synergistic effects with sulfamethoxazole, a sulfonamide antimicrobial drug [41]. Raafat et al. [42] have demonstrated that CS has a bactericidal effect, although it may not act on a single classical target. An important aspect of its bactericidal effect is the combination of CS with phytic acid, and the extraction of potential membrane lipids (mainly lipophytic acid). In addition to applications in infections caused by pulmonary fibrosis, CS has been applied in TB. The effect of monotherapy in the treatment of TB is limited and tends to lead to drug-resistant TB. Therefore, CS is usually combined with drugs for the treatment of TB. Nanocarriers are used for the simultaneous delivery of multiple drugs with potential for the treatment of TB. Cunha et al. [43] have prepared CS microparticles (aerodynamic diameter of 4 μm) loaded with isoniazid and rifabutin with enhanced drug loading rate (isoniazid: 93%; rifabutin: 99%), macrophage activation ability and antibacterial ability (96%). CS has also been studied in COVID-19. Wang et al. [44] have used CS to encapsulate bovine-serum-albumin-integrated silymarin/curcumin. The presence of CS effectively improved the dispersion and deposition of drugs in the lungs. After the treatment, serum CPR and IL-6 levels significantly decreased in a COVID-19 mouse model, and the pathological changes in the lungs of mice were ameliorated.

3.1.2 Poly(lactide-co-glycolide) and its derivatives. PLGA has been widely used as a synthetic polymer in

Acta Materia Medica

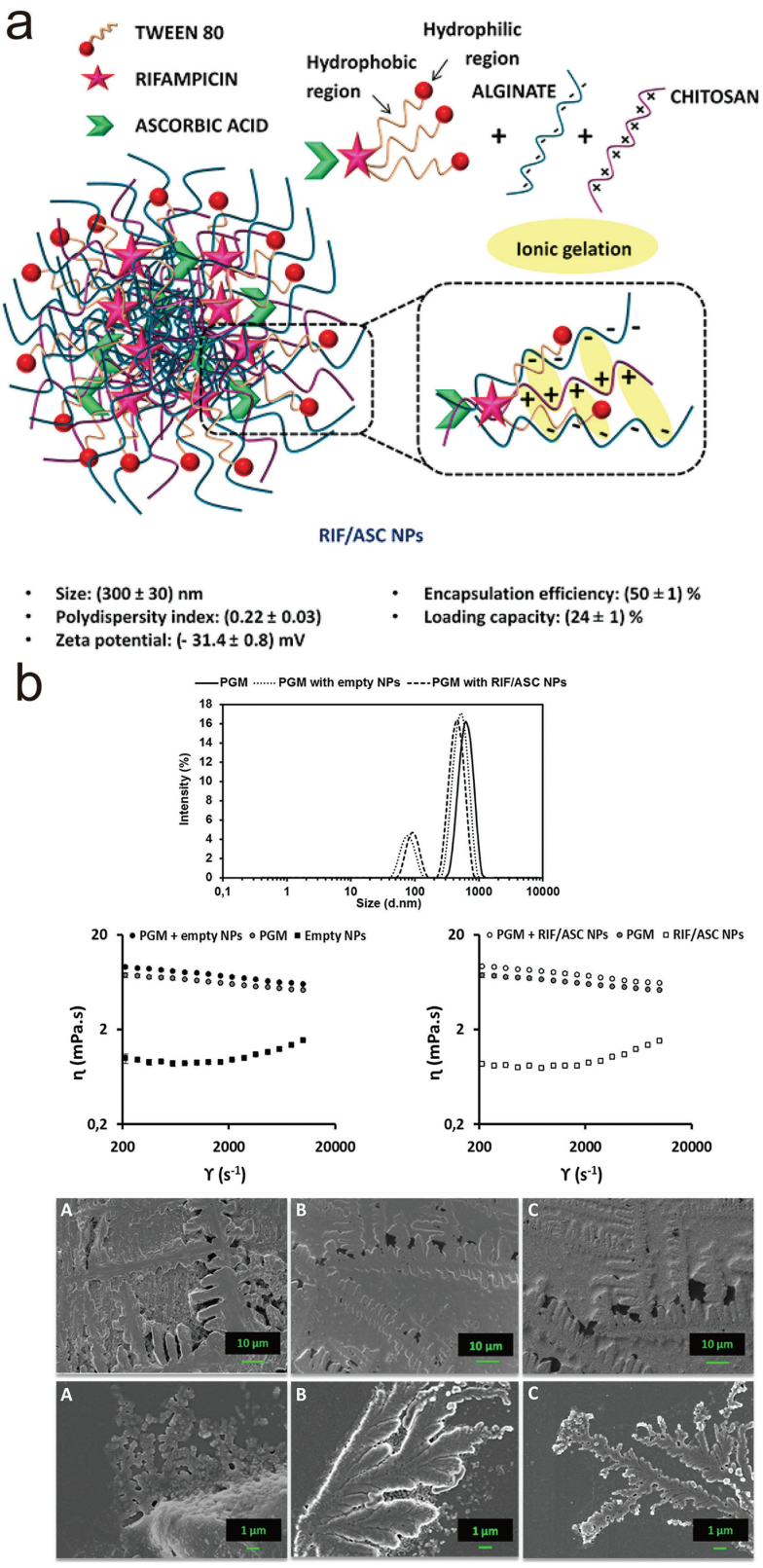

Figure 2 | (a) General schematic description and main characteristics of RIF/ASC nanoparticles. (b) Porcine gastric mucin (PGM) as an in vitro model to clarify the respiratory mucus adsorption of nanoparticles loaded with RIF/ASC. Copyright 2021, American Chemical Society [35].

slow-release DDSs because of its excellent biodegradability and biocompatibility [45]. PLGA can be randomly polymerized from lactic and glycolic acids, and various aspects, including molecular weight, degradability and size, can be enhanced through mixture of different proportions of the two monomers [46, 47]. PLGA, through its hydrophobic core, increases the stability of hydrophobic drugs [48]. Coupling it with PEG increases the water solubility of the drug, thus enhancing its mucosal adhesion. PEG-modified PLGA (PEG-PLGA) particles have progressed in pulmonary drug delivery [49]. Li et al. [50] have explored the in vitro drug release, mucus penetration and macrophage uptake of PEG-PLGA at different PEG ratios and molar masses, and examined the activity of microspheres in vivo in rats. The PEG2000-DSPE/PLGA1:1 group showed higher mucus permeability, lower macrophage uptake and a longer lung retention time than the 0.25:1 group. PLGA has free end groups that can be modified into hydroxyl end groups (PLGA-OH), carboxyl end groups (PLGA-COOH) or ester end groups (PLGA-COOR); the encapsulation efficiency and sustained drug release characteristics of PLGA derivatives change with the presence of different modified end groups [51].

In recent years, the research focus on PLGA-based DDSs has gradually shifted from complex modifications to the core drugs in the system. In addition to traditional anti-infective and anti-tumor chemical drugs, some bioactive drugs, such as polypeptide drugs, are gradually attracting clinical attention. Because of the special properties (appropriate molecular weight, poor mucosal permeability and easy degradation in vivo) of peptides compared with traditional drugs [52], polypeptide drug delivery carriers are in urgent need of exploration. Laura. et al. [53] have reported a nanostructure in which CSE4 (a polypeptide drug that enhances the telomerase activity of cells) is encapsulated in PLGA-PEI. PLGA-PEI nanoparticles loaded with CSE4 have been found to protect ProSP-C cells against bleomycin-induced damage by decreasing DNA damage and promoting alveolar structural repair, thus exerting therapeutic effects in patients with idiopathic pulmonary fibrosis. Fluorinated fragments of fluorocarbons with hydrophobic and lipophobic properties have been incorporated into peptide drugs. The fluorination modification prevents binding of the peptide drug to mucin glycoproteins, thus resulting in an approximately 240-fold increase in the mucus penetration of the peptide drug [54]. Wang et al. [55] have encapsulated calcitonin gene-associated peptide (CGRP) in PLGA-PEG nanoparticles to increase the duration of action and stability of CGRP. The experimental group of PLGA-PEG-coated CGRP had a thinner pulmonary vascular wall and less inflammatory cell infiltration than the free CGRP group, thus showing the potential of PLGA-PEG nanocarriers to increase the therapeutic effects of CGRP treatment of inflammatory lung damage. Jannuzzi et al. [56] have loaded a single-chain variable fragment (scFv) of an antibody against *Pichia pastoris* into PLGA nanoparticles and evaluated its anti-paracoccidioidomycosis effect on animals. ScFv was released at a controlled rate in vivo, and the fungal load in experimental animals significantly decreased. The combination of traditional nanomaterials with new drugs may enrich their properties and provide new hope for the treatment of lung diseases. Recent studies have shown that PLGA can carry multiple drugs and enhance their efficacy in exerting potent antibacterial and anti-inflammatory effects. PLGA can also serve as a carrier for several other drugs, such as sparfloxacin [57], thymoquinone [58], levofloxacin [59] and luteolin [60], thus improving their therapeutic efficacy and decreasing their systemic toxicity. PLGA-based DDSs enable controlled release of drugs, thus showing a promising future in the treatment of pulmonary mycoses.

Beyond CS and PLGA, other natural polymers, such as hyaluronic, alginate and synthetic polymers, are used in the treatment of infectious lung diseases. Hyaluronic acid modification promotes the entry of CS nanoparticles loaded with a model plasmid (pCMV-β-Gal) into alveolar epithelial cells and increases the transfection efficiency of the plasmid [61]. Polymer-lipid nanoparticles formed by conjugation of poly(β-amino esters) with poly(ethylene glycol)-lipids can be administered in the whole body, thus facilitating mRNA transport to the lungs, with little interception in the liver [62]. Furthermore, polymer micelles (PCD/PPC/PPE) constructed from a polycaprolactone-polyethylene glycol carrier (PCL-PEG-COOH, PPC), polycaprolactone-polyethylmethacrylate cationic carrier (PCL-PDMAEMA, PCD) and polycaprolactone-polyethylene glycol carrier connected with high-affinity targeting peptide (Esbp) by a solvent evaporation method targeting inflammatory endothelial cells (PCL-PEG-Esbp, PPE) have shown good biocompatibility and lung targeting ability in animal models of acute lung injury, thus effectively enhancing the effect of dexamethasone against this injury [63].

3.2 Liposomes

Liposomes are phospholipid-containing carriers composed of naturally occurring phospholipids, such as phosphatidylcholine, phosphatidylserine, soybean lecithin or egg yolk lecithin, which are sometimes complemented with other lipids and are easily prepared [64]. In addition, the surface charge, particle size and drug encapsulation efficiency of liposomes can be easily adjusted [65]. Hydrophilic drugs are encapsulated within the capsule lumen formed by liposomes, and lipophilic drugs are entrapped in the lipid bilayer [66]. Liposomes are widely known for their versatility as DDSs for a variety of diseases, including cancer and infections [67]. Liposomal drugs are currently the only nanodrugs primarily used in clinical settings, because of their excellent biocompatibility and slow-release properties. Because liposomes are similar to cell membranes, liposome-based DDSs can easily penetrate the blood barrier and reach target organs

[68]. Most currently applied liposomal drugs are administered intravenously. Liposomes have been a focus of development as potential drug carriers over the past few decades, partly because of their biocompatibility as naturally derived lipids, and their strong non-targeted tissue penetration and tissue adhesion [69]. Another advantage of liposomes for drug delivery is their ability to slow the release of antibiotics and decrease organ burden in patients [70].

However, conventional liposomes, such as Lipofectamine 2000, have high efficacy, and the lack of on-demand release of their contents limits their therapeutic utility [71]. Therefore, in recent years, researchers have focused on liposomal nanodrug delivery systems for organ targeting, and controlled and sustained drug release. Surface modification or coupling of different functional groups, such as enzymes, aptamers, nucleic acid molecules, cell membranes, small molecules and chemical groups, can achieve such functions. Acute respiratory distress syndrome is a serious complication of SARS-CoV-2 viral infection. Pooladanda et al. [72] have prepared liposomes in which BRD4 and siRNA were complexed with cationic lipid (BRD4-siRNA-LP) through the thin-film hydration method; the BRD4-siRNA-LP markedly suppressed LPS-induced pulmonary inflammation. Weng et al. [73] have reported a liposome-based DDS for delivering methylprednisolone (MPS) to the lungs (MPS-NSSLs-SPANb) as a treatment for idiopathic pulmonary fibrosis. Alveolar surfactant protein A (SP-A) is highly expressed in human type II alveolar epithelial cells but scarcely expressed in other organs, and thus is an ideal target for lung tissue. The DDS can be targeted to lung tissue via SP-A nanobodies (SPANbs). Immunohistochemistry indicated that MPS-NSSLs-SPANb specifically bound lung tissue, but not liver and kidney tissue. Animal imaging also indicated a high accumulation of MPS-NSSLs-SPANb from 15 min to 6 h (**Figure 3**).

Angiotensin-converting enzyme 2 (ACE2) protects lung epithelial cells but also serves as a functional receptor for SARS-CoV-2, a virus that can cause lethal damage to the lungs. The presence of excess soluble ACE2 in the lungs may neutralize some viruses, thus resisting pulmonary attack. On the basis of this idea, Arisoy et al. [74] have prepared ACE2-loaded decoy liposomes that limit SARS-CoV-2 infection, thus resulting in 4 log fold Vero E6 cell death. Gupta et al. [75] have found that a combination of bacteriophage lysozyme and levofloxacin liposomes (LVX liposomes) is effective against the biofilm formed by *Staphylococcus aureus*. A possible mechanism is that lysozyme disrupts the bacterial cell wall, and LVX plays a bactericidal role. This combination of enzymes and liposomal drugs provides a new pathway for the treatment of invasive *S. aureus* lung disease.

In addition to the loaded drugs and targeted receptors affecting drug therapeutic efficacy, the influence of the surface charges of nanomaterials on DDSs is gradually receiving more attention. A relationship exists between the charge properties of liposomes and the biological barrier in the lungs. Studies have shown that liposomes with neutral and negative charges have better permeability, retention time and stability in bronchoalveolar lavage fluid containing alveolar surfactant [76]. The use of LVX liposomes for the treatment of CF-induced *P. aeruginosa* infection has been reported, and the anionic liposomes have good encapsulation rates, slow-release properties and low epithelial cytotoxicity. However, cationic liposomes are unable to encapsulate LVX and therefore may not be suitable as nanocarriers for LVX [77]. A similar study has prepared neutral and negatively charged liposomes by loading different proportions of cholesterol and gentamicin. The minimum inhibitory concentration and minimum bactericidal concentration of neutral liposomes against planktonic cells of *P. aeruginosa* and *Klebsiella oxytoca* were more favorable than those of free gentamicin, whereas those of the negative liposomes were equivalent to those of free gentamicin. The negatively charged formulation exhibited the same bacteriostatic concentration as that of free gentamicin. However, the drug encapsulation efficiency of negatively charged formulations was more favorable than that of neutral liposomes, owing to the stronger binding effect between negatively charged liposomes and positively charged gentamicin [78].

In 2018, an ARIKAYCE® KIT (Amikacin Liposome Inhalation Suspension: ALIS) from Insmed Company was approved for marketing in the United States. It is the only drug specifically used to treat non-tuberculous mycobacterial (NTM) lung disease caused by Mycobacterium avium complex infection [79]. Amikacin is an aminoglycoside drug that achieves antibacterial effects by interfering with the synthesis of bacterial proteins. It penetrates the biofilm of Mycobacterium avium complex and exerts strong antibacterial activity in vitro [80, 81]. ALIS has been prepared by mixture of 70 mg/mL of amikacin dissolved in water with lipids (dipalmitoylphosphatidylcholine and cholesterol) dissolved in ethanol at a specific flow-rate ratio. Compared with the prototype drug amikacin, ALIS administered through inhalation resulted in significantly greater drug exposure in lung tissue and alveolar macrophage uptake, thus effectively prolonging the drug retention time in the lungs [82, 83]. However, ALIS application causes some adverse events, including (1) respiratory toxicity (dyspnea, cough, hemoptysis and dysphonia) and (2) ototoxicity (mainly hearing loss). ALSI has low nephrotoxicity, thus making it a viable option for patients with renal insufficiency [83, 84]. A cohort study has indicated that the therapeutic effects of ALSI are superior to those of tobramycin inhalation in patients with CF with chronic *P. aeruginosa* infection [85]. Beyond ALIS, researchers have evaluated the efficacy of liposomal ciprofloxacin for NTM caused by *Mycobacterium avium* subsp. *hominissuis* and *Mycobacterium abscessus* sp. Ciprofloxacin liposomes have enhanced biofilm penetration and

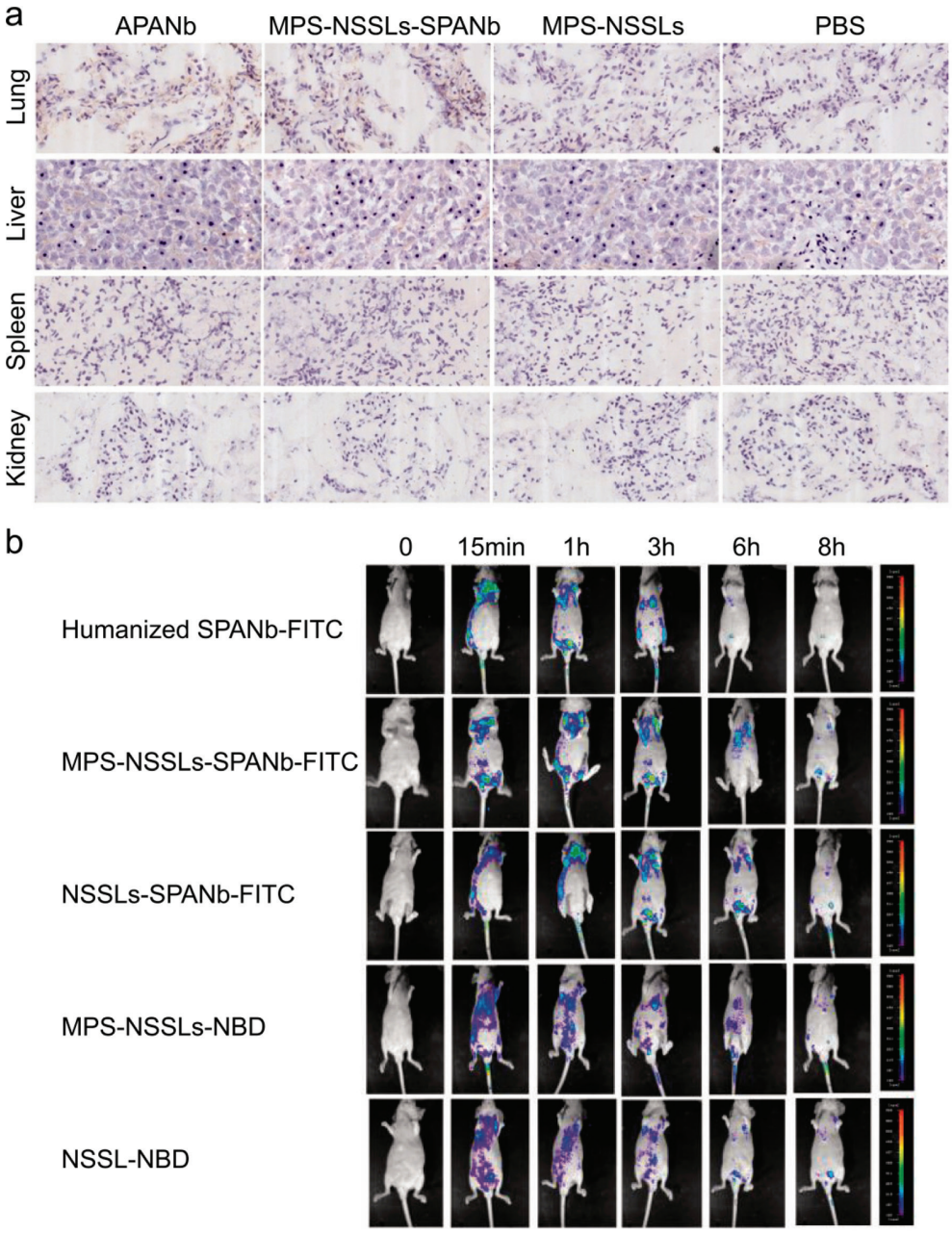

Figure 3 | Images of immunohistochemical staining of human tissue specimens and MPS-NSSLs-SPANb binding to SPA antigen in vitro.

(a) The magnification is ×20. Arrows indicate positive staining. (b) Real-time in vivo imaging of nude mice injected with the indicated reagents. Copyright 2021, Taylor & Francis Online [73].

macrophage uptake, thus significantly decreasing the bacterial load in patients with NTM [86].

Liposomal antibiotic nanoparticles have unique potential in treating pulmonary infections caused by *P. aeruginosa*. The use of liposomal nanoparticles in combination with ciprofloxacin and mucin significantly enhance the adhesion ability of lipid drugs to lung epithelial cell monolayers and prolong the retention time in the lungs [87]. Another study has investigated the dissolution characteristics of ciprofloxacin liposomes in artificial bronchodilator sputum medium. Ciprofloxacin liposomes have slow-release properties and release antibiotics at concentrations higher than the minimum inhibitory concentration over time. They

can penetrate the mucus barrier and kill *P. aeruginosa* [88]. Liposomal amphotericin B (LAmB) is another commercially available intravenous liposomal drug used primarily for the treatment of fungal infectious diseases of the lung. LAmB has been shown in animal and clinical trials to be effective in decreasing fungal loads. In addition to its therapeutic effects, LAmB has a preventive effect on pulmonary fungal diseases [89].

In addition to delivering antibiotics, liposomes have shown promise in the development of vaccines for infectious diseases. 2′,3′-Cyclic guanosine monophosphate-adenosine monophosphate (cGAMP) is a natural agonist of stimulator of interferon genes (STING). Wang et al. [90] have developed a potential "universal" mucosal adjuvant (PS-GAMP) for influenza vaccines by loading cGAMP into prepared liposomes based on lung surface active ingredients, which enter alveolar macrophages (AMs) via SP-A and SP-D-mediated endocytosis; subsequently, cGAMP passage through cellular gaps into alveolar epithelial cells further activates the STING pathway, thus inducing the production of immune type I immune mediators and promoting the recruitment and differentiation of $CD11b^+$ dendritic cells (DCs) and $CD8^+$ T cells, as well as humoral immunity (**Figure 4a**). Owing to its compositional similarity to alveolar surface-active substances, PS-GAMP can enter AMs without damaging the cells. AMs exhibit high uptake of PS-GAMP (**Figure 4b**). Bernasconi et al. [91] have evaluated a novel influenza virus vaccine platform that achieves enhanced vaccine immunogenicity and host protection by combining liposomal nanoparticles (LNPs) with the vaccine adjuvant CTA1-DD. The constructed liposomal influenza vaccine, CTA1-3M2e-DD (FPM2e), remains extremely immunogenic, and lung tissue-specific responses have been observed after immunization of mice. Unexpectedly, no significant pathological damage has been observed in the mouse lungs.

3.3 Solid lipid nanoparticles

Solid lipid nanoparticles (SLNs) are made up of a solid lipid core, a surfactant shell and water [92]. Since the early 1990s, SLNs have been investigated as an alternative to polymeric nanoparticles [93]. Compared with particles made from PLA or PLGA, SLNs have several advantages, such as greater physical stability, controlled release, stronger drug targeting effects, faster degradation, lower cytotoxicity, higher tolerability and ease of scale-up production [92, 94-96]. Chattopadhyay et al. [97] have prepared various solid lipid nanoparticle suspensions containing ketoprofen and indomethacin as model compounds by using supercritical fluid extraction of emulsions. The feasibility of delivering different SLN nanosuspensions as aerosols for inhalation has been investigated by nebulizing the prepared nanosuspension formulations with Aradigm's AERx® system. Nanoparticles with an average volume diameter of <50 nm exhibit excellent suitability for deep lung deposition. In addition, the drugs remain in a steady state within the SLN with drug loading as high as 10% and 20% w/w, well above the saturation limits in these lipids. Jaafar-Maalej et al. [98] have developed lipid nanoparticles loaded with beclomethasone dipropionate through a high-shear homogenization method and nebulized them in the form of aerosols. The lipid nanoparticles displayed high entrapment efficiency values reaching 99% and in vitro diffusion-controlled release of the lipid matrix. Moreover, the nebulized SLN and NLC have been demonstrated to have an aerodynamic behavior suitable for deep lung delivery. LN can reasonably be considered a promising drug carrier system that paves the way to lipophilic drug-targeting strategies via nebulization. Wang et al. [99] have prepared SLNs consisting of lecithin, cholesterol and a lipid-polyethylene glycol conjugate by solvent evaporation. Dry powders of SLNs have been prepared by thin-film freeze-drying (TFFD), spray drying or conventional shelf freeze-drying methods. The dry powder prepared by TFFD has been found to exhibit more preferable aerosol properties than that prepared by spray drying. In addition, SLNs encapsulated with siRNA can be successfully converted into aerosolized dry powder by TFFD without negative effects on the function of the siRNA. The resultant dry powder vaccines are expected to be delivered through non-invasive routes, such as intranasal or pulmonary administration. Ma et al. [100] have designed a mannose-modified macrophage-targeting SLN, MAN-IC-SLN, loaded with a prodrug of isoniazid, with the aim of treating latent TB infection. Cellular uptake of modified SLNs has been found to be higher in macrophages (97.2%) than that of unmodified SLNs (42.4%). In addition, pH-sensitive antibiotic release and targeted drug release have been achieved in macrophage endosomes, thus enhancing drug retention and efficacy. In an in vitro latent TB infection model and in vivo antibiotic efficacy tests, MAN-IC-SLNs have shown significantly greater antibiotic effectiveness than free drug solution. Thus, macrophage-targeted and pH-sensitive SLNs may serve as a promising platform for treating latent TB infection. Li et al. [101] have prepared curcumin solid lipid nanoparticles (Cur-SLNs) by using a solvent emulsification diffusion–low temperature curing method. Through freeze-drying technology, they micronized the Cur-SLN micropowder and combined it with flower-shaped lactose (FL) to create a Cur-SLN-FL powder mist agent, which increased the drug loading of curcumin and substantially enhanced the performance of its drug release in artificial lung fluid. Furthermore, the in vitro cytotoxic effect of Cur-SLN-FL powder was investigated in mouse fibroblast (L929) cells and human normal lung epithelial cells (BEAS-2B), thus confirming that this powder has a favorable safety profile in lung cells and is expected to be a safe and efficient intrapulmonary drug delivery method by inhalation. Mehrabani et al. [102] have developed DPIs comprising amphotericin B-loaded solid lipid nanoparticles (AMB-SLNs) through lyophilization and the use of lactose as an inhalation carrier to

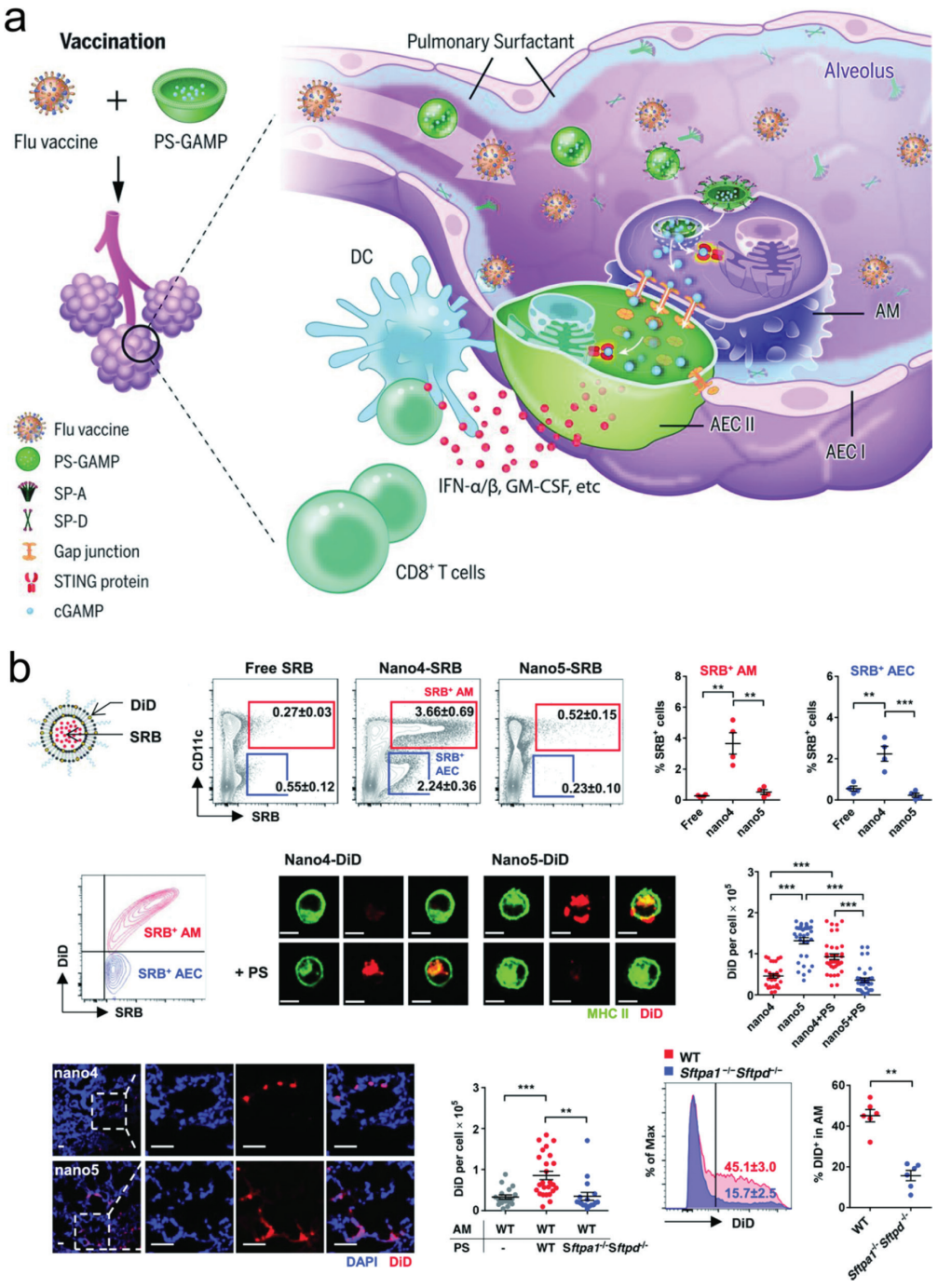

Figure 4 | (a) Schematic diagram of the principle of PS-GAMP-mediated adjuvant. (b) Uptake of PS-GAMP by AMs. Copyright 2020, HHS Public Access [90].

prevent invasive pulmonary aspergillosis. Statistically, the optimized nanoparticles had the smallest size and the lowest polydispersity index. Morphological studies of polydispersity index formulations have indicated the formation of non-aggregated spherical particles with uniform sizes and smooth surfaces. Moreover, the highest fine particle fraction was obtained with 10% lactose, thus demonstrating the aerodynamic properties suitable for lung administration. Consequently, the formulation was considered effective in delivering drugs to the peripheral respiratory tract. Chokshi et al. [103] have prepared mannose-appended rifampicin containing solid lipid nanoparticles (Mn-RIF-SLNs) for the management of pulmonary TB. Cytotoxicity studies using

Acta Materia Medica

the J774A.1-cell line have shown that the developed Mn-RIF-SLNs, as compared with free drugs, are non-toxic and biocompatible. Fluorescence microscopy and fluorescence-activated cell-sorting analysis have demonstrated significantly higher intracellular uptake of coumarin-6-loaded mannosylated SLNs (Mn-C6-SLNs) than Un-C6-SLNs. An oral pharmacokinetic study in adult Sprague Dawley rats has indicated markedly greater RIF bioavailability in Mn-RIF-SLNs than RIF solution. Biodistribution studies have shown higher drug levels in the lungs with Mn-RIF-SLNs than Un-RIF-SLNs. Therefore, the developed Mn-RIF-SLNs can be used as a potential tool for the delivery of anti-tubercular drugs to the lungs for the treatment of TB.

Active targeting of AMs may increase the efficacy of "old" drugs currently used clinically to treat pulmonary TB. Previous studies have shown that respirable solid lipid nanoparticle assemblies (SLNas) loaded with RIF and surface decorated with a mannose-based surfactant can effectively target AMs through a mannose-receptor-mediated mechanism. Truzzi et al. [104] have shown the in vivo biodistribution of these mannosylated SLNas and compared their behavior with that of non-functionalized SLNas and bare RIF. The biodistribution of SLNas in mice after intratracheal instillation has been assessed by whole-body real-time fluorescence imaging in living animals, and RIF has been quantified in excised organs and plasma. In addition, the uptake of SLNas has been determined with fluorescence microscopy on AMs and alveolar epithelium. The results have indicated that RIF-loaded nanocarriers are suitable for effective targeting of AMs via a mannose-receptor-mediated pathway, and may provide a potentially more efficient inhaled therapy for the treatment of pulmonary TB. Costa et al. [105] have reported a similar mannose-based targeting study showing substantial in vitro uptake of mannose-containing nanoparticles by macrophages, thus suggesting that these functionalized nanocarriers may provide a promising platform to deliver anti-infective drugs for the treatment of infectious lung diseases. Rodenak-Kladniew et al. [106] have developed SLN/CS/eugenol by incorporating CS (a cationic biopolymer) and eugenol (an essential oil) into a lipid matrix. The nanoparticles provided sustained release of encapsulated ofloxacin for 24 h and exhibited strong bactericidal activity against *P. aeruginosa* and *S. aureus*. Moreover, the nasal administration of hybrid SLN to mice reached therapeutic ofloxacin levels in the lungs, thus demonstrating its potential promise as a system for pulmonary inhalation therapy. Gaspar et al. [107] have prepared SLNs containing rifabutin (RFB) for pulmonary delivery of drugs to treat TB, then evaluated their antimycobacterial activity in a mouse model infected with H37Rv. RFB-SLN resulted in enhanced activity of RFB against *Mycobacterium tuberculosis* infection compared with that in untreated animals, thus suggesting that RFB-SLN microencapsulation may be a promising treatment method for TB. In another study, Pastor et al. [108] have manufactured lipid nanoparticles loaded with colistimethate sodium to enhance antimicrobial therapy against *P. aeruginosa* in patients with CF. The obtained nanoparticles exhibited antimicrobial activity against clinically isolated *P. aeruginosa* and appeared to be less toxic than free colistimethate sodium in an in vitro study.

Lipid nanoparticles may be a promising carrier system for the pulmonary delivery of drugs [109-111]. Nevertheless, the selection of compounds and carrier systems for pulmonary delivery must be adjusted through practical therapeutic approaches to ensure the stability and pulmonary selectivity of the formulation in vivo.

3.4 Inorganic nanoparticles

Many inorganic materials including silica, gold, iron oxide and alumina are used for the synthesis of nanoparticles. Inorganic nanoparticle carriers offer high biocompatibility, high stability, high delivery efficiency and resistance to microbial degradation. Inorganic substances, because of their plasmonic and magnetic properties, can be used for the treatment of pulmonary diseases. For example, mesoporous silica nanoparticles (MSNs) are multifunctional nanocarriers that can provide multiple functions to improve the treatment of lung diseases. The mesoporous scaffold protects the drug until it reaches the target site. In addition, the MSN surface is easily functionalized to provide desired nanoparticle biocompatibility, targeting fractions and controlled drug release characteristics. In the past few years, substantial progress and successful results have been achieved in the use of MSNs for lung delivery. Although few studies have used MSNs for the treatment of COPD, acute respiratory distress syndrome and some infectious lung diseases, studies have shown that these nanoparticles have high potential for the treatment of these respiratory diseases. Moreover, the inherent characteristics of MSNs may offer distinct benefits in the treatment of inflammation and lung infections. Higher doses of particularly insoluble drugs can be delivered topically by using porous carrier matrices to minimize their adverse effects and improve their biodistribution. Gulin-Sarfraz et al. [112] have manufactured two sizes of mesoporous silica particles (MSPs) loaded with dexamethasone and investigated their feasibility as delivery carriers for the treatment of airway inflammation in mice. Two mouse models of airway inflammation were administered drug-loaded aerosol particles through inhalation. Aerosolized dexamethasone-loaded MSPs successfully treated airway inflammation induced by melphalan, thus indicating that MSPs are effective pulmonary delivery carriers of corticosteroids for the treatment of chemical lung injury. This study emphasizes the potential of MSPs as drug carriers for the treatment of respiratory diseases. To address the problems of antimicrobial resistance and off-target toxicity caused by overuse of antibiotics, Rathnayake et al. [113] have prepared a targeted drug delivery nano-assembly containing liposome-coated colistin (Col)-loaded porous silica particles in which the liposomal coating is attached to

a *P. aeruginosa*-targeting peptide LL-37 (Col@MSN@LL-(LL-37)), which can deliver antibiotics to extracellular and intracellular bacteria. Col@MSN@LL-(LL-37) efficiently targeted the clinical *P. aeruginosa* strain PA14 and effectively inhibited the growth of intracellular PA14 in lung epithelial cells. Furthermore, no significant cytotoxic effects of Col@MSN@LL-(LL-37) were observed in mammalian cells. Thus, this lipid-encapsulated targeted nano-assembly provides an antibiotic delivery platform for the treatment of a wide range of intracellular infections. Tenland et al. [114] have developed MSNs incorporating the antimicrobial peptide NZX, which inhibits *M. tuberculosis*. Most of the NZX is adsorbed in the pores of MSNs by electrostatic interactions in this carrier. NZX-MSPs can be effectively taken up by cells, particularly primary macrophages. In addition, compared with the free peptide, NZX-MSPs exhibited significantly better *M. tuberculosis* eradication in primary macrophages infected with *M. tuberculosis* H37Rv and in a murine TB model. These MSPs are nontoxic to mammalian cells at therapeutic concentrations. In another study, Xu et al. [115] have synthesized silica (SiO_2) nanoparticles through a laser ablation method. The inhibitory effect of SiO_2 nanoparticles on carbonic anhydrase protects alveolar epithelial cells against H_2O_2-induced oxidative stress and acts as an antibacterial agent against *Klebsiella pneumoniae*. This protective effect of SiO_2 nanoparticles has been observed in alveolar epithelial cells (A549) through measurements of MTT, ROS levels, CAT and SOD activity, and GSH content. Finally, the antibacterial activity of nanoparticles against *K. pneumoniae* has been confirmed and attributed mainly to the interaction of nanoparticles with the cell wall and the disruption of the bacterial membrane. In conclusion, these data may provide useful information for the development of pneumonia treatment and management. After pneumonia treatment, Clemens et al. [116] evaluated the efficacy of moxifloxacin (MXF) via disulfide snap-top redox-operated MSN (MSN-SS-MXF) delivery through various administration routes in a mouse model of tularemia. *Francisella tularensis* Live Vaccine Strain was administered intranasally to BALB/c mice, and 1 day later, the mice were treated with free MXF or MSN-SS-MXF via intravenous (i.v.), intramuscular (i.m.) or subcutaneous (s.c.) routes, administered in three doses every 48 h. MSN-SS-MXF was significantly more effective than the free drug in decreasing bacterial load in the lungs, regardless of the route of administration. Interestingly, biodistribution analysis of MSN silica indicated considerable amounts in the lungs, liver and spleen in i.v.-treated animals, but lower levels in i.m.- or s.c.-treated animals. These results are consistent with those of flow cytometry studies. In summary, the results show the potential of MSN-SS-MXF in optimizing the therapeutic efficacy of infectious lung diseases. Iron oxide nanoparticles are also frequently used as drug delivery carriers. Tewes et al. [117] have formulated porous microparticles loaded with superparamagnetic iron oxide nanoparticles in combination with a target-directed magnetic gradient field to achieve targeted delivery of aerosols to specific regions of the lung. The microparticles were characterized by various physicochemical methods. The results indicated that, when loaded with drugs, these particles may be useful in treating localized lung diseases, such as tumor nodules or bacterial infectious lesions. Miranda et al. [118] have prepared a magnetically responsive microparticulate system for pulmonary delivery of an anti-TB drug candidate (P3). The microparticles were developed through a casting method, and were found to be suitable for P3 entry into the lower airways and phagocytosis by alveolar macrophages. These microparticles may increase TB treatment efficiency and patient compliance, because a predefined amount of drug could be released at a predetermined time and at a desired frequency, according to treatment requirements.

In summary, inorganic nanoparticles offer several advantages, such as excellent intrinsic physical properties and versatile surfaces, thus showing great potential for the construction of multifunctional nanoprobes for in vivo therapy. However, studies on their in vivo behavior, particularly their pharmacokinetics, remain in early stages, and further research is needed.

3.5 Nanomicelles

Nanomicelles are self-assembling nanosized colloidal dispersions with a hydrophobic core and hydrophilic shell, usually in the particle size range of 10 to 100 nm [119]. They are currently used as drug delivery vehicles by encapsulating hydrophobic drugs in a mixed micellar hydrophobic core to solubilize them [120]. In addition, nanomicelles are a valuable emerging delivery technique for preventing or minimizing drug degradation, decreasing adverse effects and enhancing the biocompatibility of poorly soluble drugs [121]. Gilani et al. [122] have prepared CS-stearic acid conjugated nanomicelles for pulmonary delivery of amphotericin B (AmB) to prevent invasive pulmonary fungal infections. Favorable drug encapsulation efficiency was obtained for different ratios of AmB and polymers. A drug encapsulation efficiency as high as 97% was obtained with AmB loaded in DC-SA micelles. The in vitro deposition profiles showed that DC-SA nanomicelles maintained their stability during the nebulization process, with a nebulization efficiency as high as 56%. In addition, the solubility of AmB in DC-SA micellar solution was greatly increased compared to AmB alone in water, thus enhancing the antifungal activity of the drug in some cases such as criptococcus neoformans. The results suggest that DC-SA micelles have the potential to be used as a suitable vehicle for spray drug delivery. Farhangi et al. [123] have prepared polymeric CS-lipid nanomicelles containing high ciprofloxacin loading capacity by using a thin-film hydration method. The formulation had an optimal size for slow release of ciprofloxacin for approximately 8 h. Moreover, the encapsulation

of ciprofloxacin in nanomicelles resulted in an 8-fold increase in antibacterial activity against *P. aeruginosa* for the treatment of lower respiratory tract infections. In another study, Farhangi et al. [124] prepared DPI formulations by spray drying CS nanomicelles containing ciprofloxacin. A two-level full factorial design was used in the study. The optimal nanoaggregate was prepared and examined for its antibacterial inhibitory effects against *P. aeruginosa*, *K. pneumoniae* and *Streptococcus pneumoniae*. This formulation significantly increased the antibacterial effect of ciprofloxacin against *P. aeruginosa* and may be used as an effective DPI for the treatment of pulmonary infections. Ren et al. [125] have fabricated pH-responsive zwitterionic polyurethane nanomicelles. This material can spontaneously self-assemble into nanomicelles in water and is stable in the physiological environment. The system has been found to increase the efficiency of reactive oxygen species production under white-light irradiation, enhance the antibacterial efficiency at acidic infection sites and significantly increase the biocompatibility of the material during in vivo circulation. Pourhajibagher et al. [126] have investigated the antimicrobial effects of nanomicellar curcumin (N-MCur)-mediated antimicrobial photodynamic therapy in combination with sonodynamic antimicrobial chemotherapy, known as photo-sonodynamic antimicrobial chemotherapy (PSACT), on virulence gene expression patterns of *Acinetobacter baumannii*. Changes in gene expression involved in *A. baumannii* treated with PSACT were determined by quantitative real-time-PCR. N-MCur-mediated PSACT regulated the expression of pathogenicity-associated genes in *A. baumannii.* Therefore, PSACT can be considered a promising application for the treatment of *A. baumannii* as an alternative to the systemic application of antibiotics. Furthermore, de Barros AODS et al. [127] have synthesized two hydroxychloroquine- and azithromycin-based nanosystems (polymeric nanomicelles and nanoparticles) and evaluated their efficacy in the in vitro treatment of COVID-19. The anti-SARS-CoV-2 effects of nanomicelles co-loaded with hydroxychloroquine and azithromycin were superior to those of nanoparticles. Direct pulmonary application, indicated that both nanoparticles reached the interior portions of the lungs. The nanosystems did not show cytotoxicity in vitro, thus corroborating their safety profile. Therefore, these nanoparticles may provide a safe therapeutic option for COVID-19 treatment.

3.6 Combinational nanosystems

In recent years, a growing number of combination therapy studies have increased the efficacy of treatments against infectious diseases. By triggering several mechanisms of death, combination therapy will provide new opportunities for effective infectious disease treatment. Jasim et al. [128] have investigated the in vitro synergistic activity of polymyxin B in combination with 2 nm silver nanoparticles by using a checkerboard assay against *P. aeruginosa* isolated from the lungs of patients with CF. Clinically relevant concentrations of polymyxin B (2 mg/L) in combination with silver nanoparticles (4 mg/L) successfully inhibited the growth of polymyxin-resistant *P. aeruginosa* isolates. Given the relative non-toxicity of silver nanoparticles and polymyxins in lung tissues, the combination of silver nanoparticles with polymyxins and the development of new nebulizer formulations for CF may have considerable clinical applications. Pérez-Cantero et al. [129] have studied the in vivo efficacy of oral AmB encapsulated in modified CS nanoparticles (Nanomerics' Molecular Envelope Technology) supplemented with a standardized extract of cultured *Lentinula edodes* mycelia (AHCC®) as a prophylactic therapy in a murine model of invasive pulmonary aspergillosis. The inclusion of the AHCC® supplement considerably increased the effectiveness of the AmB therapy in terms of load reduction of aspergillosis and survival, thereby enhancing the immunogenicity of the vaccine. Bernasconi et al. [91] have explored whether the combination of the self-adjuvanted vaccine CTA1-3M2e-DD (FPM2e) and LNPs might be beneficial for vaccine efficacy. They have evaluated the immunogenicity and protective potential of the combined vectors in an influenza mouse model. The nanoparticle formulation strongly enhanced pulmonary protection against a highly virulent PR8 strain of influenza viral infection, and the lung tissue did not exhibit gross pathological changes after recovery from infection, thus demonstrating that the combination of an enzymatically active adjuvant and LNPs dramatically enhances immunogenicity and protection against infection. Mehdizadeh et al. [130] have prepared a nanofluid containing antibiotics and f-MWCNTs through an ultrasonic method and examined the antibacterial effects of the f-MWCNT plus ciprofloxacin combination against *K. pneumoniae* by evaluating the expression of virulence genes. The combination of f-MWCNT and antibiotics increased the efficacy of antibiotics at lower doses and the drug delivery efficiency of ciprofloxacin. Kingstad-Bakke et al. [131] have investigated whether PLGA-based cationic pathogen-like particles loaded with TLR4 (glucopyranosyl lipid adjuvant) or TLR9 (CpG) agonists mixed with influenza virus nucleoproteins might induce antigen-specific CD4 and CD8 T cells with or without a nanoemulsion adjuvant. This vaccine formulation, when administered intranasally to mice, elicited a strong mucosal imprinted T-cell memory in the lungs and airways, and conferred long-term protection against pathogenic influenza viral infection. These findings have important implications for the development of vaccines against respiratory pathogens, including influenza virus and SARS-CoV-2.

4. CONCLUSION

This review provides a brief overview of advances in approaches and ideas for representative pulmonary

Table 1 | DDSs for the treatment of infectious lung diseases.

Carrier	Active pharmaceutical ingredients	Couplers	Indication	Reference
Polymeric				
Chitosan	Itraconazole	Peptide ligand	Cryptococcal pneumonia	[31]
Chitosan	Hesperidin	PLGA-PEG	Acute lung injury	[132]
Chitosan	Methylprednisolone sodium succinate	RBC	COVID-19	[34]
Chitosan	Chemically modified messenger RNA	PLGA	Cystic fibrosis	[133]
Chitosan	Isoniazid (and rifampin)	Genipin	TB	[134]
Chitosan	rifampicin		TB	[30]
Chitosan	Gadofullerene	α-Fe_2O_3	Pneumonia	[135]
Chitosan	Cp-1 phage endolysin		Pneumonia	[136]
Chitosan	Ciprofloxacin		Pneumonia	[137]
PLGA	Tobramycin	PEG	Pneumonia	[138]
PLGA	Lopinavir		COVID-19	[139]
PLGA	Sparfloxacin and tacrolimus	γ3-peptide (NNQKIVNLKEKV AQLEA)	Pneumonia	[57]
Polymethacrylic acid	Amphotericin B		Pulmonary aspergillosis	[140]
α-Cyclodextrin	Moxifloxacin	4-(hydroxymethyl) phenylboronic acid pina-col ester	Pneumonia	[141]
Poly-β-cyclodextrins	ETH and booster, etc.		TB	[142]
N,N,N-trimethyl chitosan chloride, TMC	Ag		TB	[33]
Alginate	Rifampicin and the antioxidant ascorbic acid	Chitosan and Tween 80	Lung intracellular infections	[35]
Liposome				
Liposome	Amikacin		Mycobacterium avium complex lung disease	[83, 143-145]
Liposome	Amikacin		Cystic fibrosis	[85, 146]
Liposome	Amphotericin B		Invasive pulmonary aspergillosis	[147, 148]
Liposome	Gentamicin		Pneumonia	[149]
Liposome	Everolimus	Hyaluronic acid	Pulmonary fibrosis	[150]
Liposome	Remdesivir		COVID-19	[151]

Table 1 | Continued

Carrier	Active pharmaceutical ingredients	Couplers	Indication	Reference
Solid liposome				
Solid liposome	Berberine	Chitosan	COPD	[36]
Solid liposome	Dimethyl fumarate		Pneumonia	[152]
Solid liposome	Carvacrol		Pneumonia	[153]
Solid liposome	Icariin		Pulmonary fibrosis	[154]
Inorganic nanoparticles				
TiO_2	Oligonucleotides		Influenza	[155]
Mesoporous silica	Dexamethasone		Acute lung injury	[156]
Mesoporous silica	N-acetylcysteine		Acute lung injury	[157]
Nanomicelles				
Nanomicelles	Curcumin		COVID-19	[158]
Poly(ethylene glycol)-poly(lactide) micelles	Luteolin		Pneumonia	[60]
Amphiphilic phosphorus dendron (C11G3) nanomicelles	Curcumin	Tyramine bearing two dimethylphosphonate sodium salts	Pneumonia	[159]
Polyvinyl caprolactam–polyvinylacetate–PEG 6000	Rifampicin and curcumin	Mannose	TB	[160]

Acta Materia Medica

drug delivery methods in recent years. New therapeutic strategies using polymers, liposomes, solid liposomes, inorganic nanoparticles and nanomicelles as drug delivery carriers were discussed, and several DDSs are summarized (**Table 1**). In general, surface modification of nanomaterials and targeted drug delivery remain current research hotspots. Pulmonary nanodrug delivery systems are highly promising therapeutic tools for the treatment of infectious lung diseases. Nanomaterials have advantages in delivering drugs for treating infectious lung diseases, such as targeted delivery, increased solubility of drugs, diminished toxicity and synergistic therapeutic effects. However, with the exception of some liposomal antibiotics, these delivery systems remain far from clinical application, because of their unevaluated potential for toxicity. In addition, some studies have been performed only in vitro, in cellular assays; therefore, more systematic and sophisticated animal studies are needed to assess their efficacy and toxicity, most nanotoxicologists and toxicologists have not assessed systematic toxicity, such as acute toxicity, chronic toxicity, inflammatory toxicity, or reproductive and developmental toxicity. Such toxicity evaluations are time-consuming and expensive, thus greatly hindering the clinical transformation of nanomedicines. Whether a particular property of a drug delivery system is favorable or unfavorable is difficult to ascertain, and depends on the given therapeutic goals and protocols.

ACKNOWLEGEDMENTS

This work was supported by the Natural Science Foundation of China (No. 82001946) and Beijing Municipal Natural Science Foundation (No. 7214300).

DECLARATION OF COMPETING INTERESTS

The authors declare no conflicts of interest.

REFERENCES

[1] Trottein F, Alcorn JF: Editorial: Secondary Respiratory Infections in the Context of Acute and Chronic Pulmonary Diseases. *Frontiers in Immunology* 2019, 10:2764.

[2] Mottais A, Le Gall T, Sibiril Y, Ravel J, Laurent V, d'Arbonneau F, et al.: Enhancement of Lung Gene Delivery After Aerosol: A New Strategy Using Non-viral Complexes with Antibacterial Properties. *Bioscience Reports* 2017, 37:BSR20160618.

[3] Wang L, Li Y, Zhang X, Li H: Characteristics of Nosocomial Infection and Its Effects on the Survival of Chemotherapy Patients with Advanced Non-Small Cell Lung Cancer. *Oncology Letters* 2017, 14:7379–7383.

[4] Bremmell KE, Prestidge CA: Enhancing Oral Bioavailability of Poorly Soluble Drugs with Mesoporous Silica based Systems: Opportunities and Challenges. *Drug Development and Industrial Pharmacy* 2019, 45:349–358.

[5] Ling J, Mangal S, Park H, Wang S, Cavallaro A, Zhou QT: Simultaneous Particle Size Reduction and Homogeneous Mixing to Produce Combinational Powder Formulations for Inhalation by the Single-Step Co-Jet Milling. *Journal of Pharmaceutical Sciences* 2019, 108:3146–3151.

[6] Khairnar SV, Jain DD, Tambe SM, Chavan YR, Amin PD: Nebulizer Systems: A New Frontier for Therapeutics and Targeted Delivery. *Therapeutic Delivery* 2022, 13:31–49.

[7] Islam MS, Saha SC, Gemci T, Yang IA, Sauret E, Ristovski Z, et al.: Euler-Lagrange Prediction of Diesel-Exhaust Polydisperse Particle Transport and Deposition in Lung: Anatomy and Turbulence Effects. *Science Report* 2019, 9:12423.

[8] Hallan SS, Sguizzato M, Esposito E, Cortesi R: Challenges in the Physical Characterization of Lipid Nanoparticles. *Pharmaceutics* 2021, 13:549.

[9] O'Driscoll CM, Griffin BT: Biopharmaceutical Challenges Associated with Drugs with Low Aqueous Solubility--the Potential Impact of Lipid-Based Formulations. *Advanced Drug Delivery Reviews* 2008, 60:617–624.

[10] Davoodi P, Lee LY, Xu Q, Sunil V, Sun Y, Soh S, et al.: Drug Delivery Systems for Programmed and On-Demand Release. *Advanced Drug Delivery Reviews* 2018, 132:104–138.

[11] Gomes P, Vale N, Moreira R: Cyclization-Activated Prodrugs. *Molecules* 2007, 12:2484–2506.

[12] Lennernäs H. Intestinal Permeability and Its Relevance for Absorption and Elimination. *Xenobiotica* 2007, 37:1015–1051.

[13] El-Helw AR, Fahmy UA: Improvement of Fluvastatin Bioavailability by Loading on Nanostructured Lipid Carriers. *International Journal of Nanomedicine* 2015, 10:5797–5804.

[14] He F, Wen N, Xiao D, Yan J, Xiong H, Cai S, et al.: Aptamer-Based Targeted Drug Delivery Systems: Current Potential and Challenges. *Current Medical Chemistry* 2020, 27:2189–2219.

[15] Wang H, Liu Y, He R, Xu D, Zang J, Weeranoppanant N, et al.: Cell Membrane Biomimetic Nanoparticles for Inflammation and Cancer Targeting in Drug Delivery. *Biomaterials Science* 2020, 8:552–568.

[16] Spampinato C, Leonardi D: Candida Infections, Causes, Targets, and Resistance Mechanisms: Traditional and Alternative Antifungal Agents. *Biomed Research International* 2013, 2013:204237.

[17] Yoshikawa TT: Antimicrobial Resistance and Aging: Beginning of the End of the Antibiotic Era? *Journal of the American Geriatrics Society* 2002, 50:S226–S229.

[18] Whitty CJM: Antimicrobial Resistance: Learning Lessons from Antiparasitic, Antibacterial and Antimycobacterial Drug Resistance in Low-Income Settings. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 2019, 113:105–107.

[19] Tewabe A, Marew T, Birhanu G: The Contribution of Nano-Based Strategies in Overcoming Ceftriaxone Resistance: A Literature Review. *Pharmacology Research & Perspectives* 2021, 9:e00849.

[20] Baptista PV, McCusker MP, Carvalho A, Ferreira DA, Mohan NM, Martins M, et al.: Nano-Strategies to Fight Multidrug Resistant Bacteria-"A Battle of the Titans". *Frontiers in Microbiology* 2018, 9:1441.

[21] Azam MW, Khan AU: Updates on the Pathogenicity Status of Pseudomonas Aeruginosa. *Drug Discovery Today* 2019, 24:350–359.

[22] George A, Shah PA, Shrivastav PS: Natural Biodegradable Polymers Based Nano-Formulations for Drug Delivery: A Review. *International Journal of Pharmaceutics* 2019, 561:244–264.

[23] Elgadir MA, Uddin MS, Ferdosh S, Adam A, Chowdhury AJK, Sarker MZI: Impact of Chitosan Composites and Chitosan Nanoparticle Composites on Various Drug Delivery Systems: A Review. *Journal of Food Drug Analysis* 2015, 23:619–629.
[24] Kravanja G, Primožič M, Knez Ž, Leitgeb M: Chitosan-Based (Nano)Materials for Novel Biomedical Applications. *Molecules* 2019, 24:1960.
[25] Kim YS, Li Q, Youn HY, Kim DY: Oral Administration of Chitosan Attenuates Bleomycin-Induced Pulmonary Fibrosis in Rats. *In Vivo* 2019, 33:1455–1461.
[26] Muralidharan A, Russell MS, Larocque L, Gravel C, Sauvé S, Chen Z, et al.: Chitosan Alters Inactivated Respiratory Syncytial Virus Vaccine Elicited Immune Responses without Affecting Lung Histopathology in Mice. *Vaccine* 2019, 37:4031–4039.
[27] Gulati N, Dua K, Dureja H: Role of Chitosan Based Nanomedicines in the Treatment of Chronic Respiratory Diseases. *International Journal of Biological Macromolecules* 2021, 185:20–30.
[28] Sezer AD, Cevher E: Topical Drug Delivery Using Chitosan Nano- and Microparticles. *Expert Opinion on Drug Delivery* 2012, 9:1129–1146.
[29] Silva AS, Sousa AM, Cabral RP, Silva MC, Costa C, Miguel SP, et al.: Aerosolizable Gold Nano-in-Micro Dry Powder Formulations For Theragnosis and Lung Delivery. *International Journal of Pharmaceutics* 2017, 519:240–249.
[30] Si Z, Hou Z, Vikhe YS, Thappeta KRV, Marimuthu K, De PP, et al.: Antimicrobial Effect of A Novel Chitosan Derivative and Its Synergistic Effect with Antibiotics. *ACS Applied Materials & Interfaces* 2021, 13:3237–3245.
[31] Tang Y, Wu S, Lin J, Cheng L, Zhou J, Xie J, et al.: Nanoparticles Targeted Against Cryptococcal Pneumonia by Interactions Between Chitosan and Its Peptide Ligand. *Nano Letters* 2018, 18:6207–6213.
[32] Zhu X, Yu Z, Feng L, Deng L, Fang Z, Liu Z, et al.: Chitosan-Based Nanoparticle Co-Delivery of Docetaxel and Curcumin Ameliorates Anti-Tumor Chemoimmunotherapy in Lung Cancer. *Carbohydrate Polymers* 2021, 268:118237.
[33] Abdel-Aziz MM, Elella MHA, Mohamed RR: Green Synthesis of Quaternized Chitosan/Silver Nanocomposites for Targeting Mycobacterium Tuberculosis and Lung Carcinoma Cells (A-549). *International Journal of Biological Macromolecules* 2020, 142:244–253.
[34] Ding Y, Lv B, Zheng J, Lu C, Liu J, Lei Y, et al.: RBC-Hitchhiking Chitosan Nanoparticles Loading Methylprednisolone for Lung-Targeting Delivery. *Journal of Controlled Release* 2022, 341:702–715.
[35] Scolari IR, Volpini X, Fanani ML, De La Cruz-Thea B, Natali L, Musri MM, et al.: Exploring the Toxicity, Lung Distribution, and Cellular Uptake of Rifampicin and Ascorbic Acid-Loaded Alginate Nanoparticles as Therapeutic Treatment of Lung Intracellular Infections. *Molecular Pharmacology* 2021, 18:807–821.
[36] Liu H, Li Y, Zhang X, Shi M, Li D, Wang Y: Chitosan-Coated Solid Lipid Nano-Encapsulation Improves the Therapeutic Antiairway Inflammation Effect of Berberine against COPD in Cigarette Smoke-Exposed Rats. *Canadian Respiratory Journal* 2022, 2022:8509396.
[37] Shirley JL, de Jong YP, Terhorst C, Herzog RW: Immune Responses to Viral Gene Therapy Vectors. *Molecular Therapy* 2020, 28:709–722.
[38] Fernández Fernández E, Santos-Carballal B, Weber WM, Goycoolea FM: Chitosan as A Non-Viral Co-Transfection System in A Cystic Fibrosis Cell Line. *International Journal of Pharmaceutics* 2016, 502:1–9.
[39] Kolonko AK, Efing J, González-Espinosa Y, Bangel-Ruland N, van Driessche W, Goycoolea FM, et al.: Capsaicin-Loaded Chitosan Nanocapsules for wtCFTR-mRNA Delivery to a Cystic Fibrosis Cell Line. *Biomedicines* 2020, 8:364.
[40] Hill M, Twigg M, Sheridan EA, Hardy JG, Elborn JS, Taggart CC, et al.: Alginate/Chitosan Particle-Based Drug Delivery Systems for Pulmonary Applications. *Pharmaceutics* 2019, 11:379.
[41] Tin S, Sakharkar KR, Lim CS, Sakharkar MK: Activity of Chitosans in Combination with Antibiotics in Pseudomonas Aeruginosa. *International Journal of Biological Sciences* 2009, 5:153–160.
[42] Raafat D, von Bargen K, Haas A, Sahl HG: Insights into the Mode of Action of Chitosan as An Antibacterial Compound. *Applied and Environmental Microbiology* 2008, 74:3764–3773.
[43] Cunha L, Rodrigues S, Rosa da Costa AM, Faleiro L, Buttini F, Grenha A: Inhalable Chitosan Microparticles for Simultaneous Delivery of Isoniazid and Rifabutin in Lung Tuberculosis Treatment. *Drug Development and Industrial Pharmacy* 2019, 45:1313–1320.
[44] Hanafy NAN, El-Kemary MA: Silymarin/Curcumin Loaded Albumin Nanoparticles Coated by Chitosan as Muco-Inhalable Delivery System Observing Anti-Inflammatory and Anti COVID-19 Characterizations in Oleic Acid Triggered Lung Injury and in Vitro COVID-19 Experiment. *International Journal of Biological Macromolecules* 2022, 198:101–110.
[45] Le Reste PJ, Pineau R, Voutetakis K, Samal J, Jégou G, Lhomond S, et al.: Local Intracerebral Inhibition of IRE1 by MKC8866 Sensitizes Glioblastoma to Irradiation/Chemotherapy in Vivo. *Cancer Letters* 2020, 494:73–83.
[46] Su Y, Zhang B, Sun R, Liu W, Zhu Q, Zhang X, et al.: PLGA-Based Biodegradable Microspheres in Drug Delivery: Recent Advances in Research and Application. *Drug Delivery* 2021, 28:1397–1418.
[47] Ahmad N, Ahmad R, Alrasheed RA, Almatar HMA, Al-Ramadan AS, Amir M, et al.: Quantification and Evaluations of Catechin Hydrate Polymeric Nanoparticles Used in Brain Targeting for the Treatment of Epilepsy. *Pharmaceutics* 2020, 12:203.
[48] Xie Y, Tan X, Huang J, Huang H, Zou P, Hu J: Atorvastatin-Loaded Micelles with Bone-Targeted Ligand for the Treatment of Osteoporosis. *Drug Delivery* 2017, 24:1067–1076.
[49] Jin F, Liu D, Yu H, Qi J, You Y, Xu X, et al.: Sialic Acid-Functionalized PEG-PLGA Microspheres Loading Mitochondrial-Targeting-Modified Curcumin for Acute Lung Injury Therapy. *Molecular Pharmacology* 2019, 16:71–85.
[50] Li J, Zheng H, Xu EY, Moehwald M, Chen L, Zhang X, et al.: Inhalable PLGA Microspheres: Tunable Lung Retention and Systemic Exposure via Polyethylene Glycol Modification. *Acta Biomaterialia* 2021, 123:325–334.
[51] Li X, Wei Y, Wen K, Han Q, Ogino K, Ma G: Novel Insights on the Encapsulation Mechanism of PLGA Terminal Groups on Ropivacaine. *European Journal of Pharmaceutics and Biopharmaceutics* 2021, 160:143–151.
[52] Fröhlich E, Salar-Behzadi S. Oral Inhalation for Delivery of Proteins and Peptides to the Lungs. *European Journal of Pharmaceutics and Biopharmaceutics* 2021, 163:198–211.

[53] Pintado-Berninches L, Montes-Worboys A, Manguan-García C, Arias-Salgado EG, Serrano A, Fernandez-Varas B, et al.: GSE4-Loaded Nanoparticles a Potential Therapy for Lung Fibrosis that Enhances Pneumocyte Growth, Reduces Apoptosis and DNA Damage. *Faseb Journal* 2021, 35:e21422.

[54] Ge C, Yang J, Duan S, Liu Y, Meng F, Yin L: Fluorinated α-Helical Polypeptides Synchronize Mucus Permeation and Cell Penetration toward Highly Efficient Pulmonary siRNA Delivery against Acute Lung Injury. *Nano Letters* 2020, 20:1738–1746.

[55] Wang F, Deng Y, Wang J, Yu L, Ding F, Lian W, et al.: The PLGA Nanoparticles for Sustainable Release of CGRP to Ameliorate the Inflammatory and Vascular Disorders in the Lung of CGRP-Deficient Rats. *Drug Delivery* 2021, 28:865–872.

[56] Jannuzzi GP, Souza NA, Françoso KS, Pereira RH, Santos RP, Kaihami GH, et al.: Therapeutic Treatment with ScFv-PLGA Nanoparticles Decreases Pulmonary Fungal Load in A Murine Model of Paracoccidioidomycosis. *Microbes and Infection* 2018, 20:48–56.

[57] Yang Y, Ding Y, Fan B, Wang Y, Mao Z, Wang W, et al.: Inflammation-Targeting Polymeric Nanoparticles Deliver Sparfloxacin and Tacrolimus for Combating Acute Lung Sepsis. *Journal of Controlled Release* 2020, 321:463–474.

[58] Saghir SAM, Al-Gabri NA, Ali AA, Al-Attar AR, Al-Sobarry M, Al-Shargi OYA, et al.: Ameliorative Effect of Thymoquinone-Loaded PLGA Nanoparticles on Chronic Lung Injury Induced by Repetitive Intratracheal Instillation of Lipopolysaccharide in Rats. *Oxidative Medicine and Cellular Longevity* 2021, 2021:5511523.

[59] Agnoletti M, Rodríguez-Rodríguez C, Kłodzińska SN, Esposito TVF, Saatchi K, Mørck Nielsen H, et al.: Monosized Polymeric Microspheres Designed for Passive Lung Targeting: Biodistribution and Pharmacokinetics After Intravenous Administration. *ACS Nano* 2020, 14:6693–6706.

[60] Miao J, Lin F, Huang N, Teng Y: Improving Anti-Inflammatory Effect of Luteolin with Nano-Micelles in the Bacteria-Induced Lung Infection. *Journal of Biomedical Nanotechnology* 2021, 17:1229–1241.

[61] Fernández-Paz E, Feijoo-Siota L, Gaspar MM, Csaba N, Remuñán-López C: Microencapsulated Chitosan-Based Nanocapsules: A New Platform for Pulmonary Gene Delivery. *Pharmaceutics* 2021, 13:1377.

[62] Kaczmarek JC, Kauffman KJ, Fenton OS, Sadtler K, Patel AK, Heartlein MW, et al.: Optimization of a Degradable Polymer-Lipid Nanoparticle for Potent Systemic Delivery of mRNA to the Lung Endothelium and Immune Cells. *Nano Letters* 2018, 18:6449–6454.

[63] Zhang J, Su M, Yin Z: Construction of Inflammatory Directed Polymer Micelles and Its Application in Acute Lung Injury. *AAPS Pharmaceutical Science and Technology* 2020, 21:217.

[64] Wang DY, van der Mei HC, Ren Y, Busscher HJ, Shi L: Lipid-Based Antimicrobial Delivery-Systems for the Treatment of Bacterial Infections. *Frontiers in Chemistry* 2019, 7:872.

[65] Joseph RR, Tan DWN, Ramon MRM, Natarajan JV, Agrawal R, Wong TT, et al.: Characterization of Liposomal Carriers for the Trans-scleral Transport of Ranibizumab. *Science Report* 2017, 7:16803.

[66] Ponkshe P, Feng S, Tan C: Inhalable Liposomes for Treating Lung Diseases: *Clinical Development and Challenges*. *Biomedical Materials* 2021, 16:054101.

[67] Guan J, Shen Q, Zhang Z, Jiang Z, Yang Y, Lou M, et al.: Enhanced Immunocompatibility of Ligand-Targeted Liposomes by Attenuating Natural IgM Absorption. *Nature Communications* 2018, 9:2982.

[68] Ye H, Zhou L, Jin H, Chen Y, Cheng D, Jiang Y: Sorafenib-Loaded Long-Circulating Nanoliposomes for Liver Cancer Therapy. *Biomed Research International* 2020, 2020:1351046.

[69] Discher DE, Ortiz V, Srinivas G, Klein ML, Kim Y, Christian D, et al.: Emerging Applications of Polymersomes in Delivery: from Molecular Dynamics to Shrinkage of Tumors. *Progress in Polymer Science* 2007, 32:838–857.

[70] Bassetti M, Vena A, Russo A, Peghin M: Inhaled Liposomal Antimicrobial Delivery in Lung Infections. *Drugs* 2020, 80:1309–1318.

[71] Deng W, Chen W, Clement S, Guller A, Zhao Z, Engel A, et al.: Controlled Gene and Drug Release from A Liposomal Delivery Platform Triggered by X-ray Radiation. *Nature Communications* 2018, 9:2713.

[72] Pooladanda V, Thatikonda S, Muvvala SP, Devabattula G, Godugu C: BRD4 Targeting Nanotherapy Prevents Lipopolysaccharide Induced Acute Respiratory Distress Syndrome. *International Journal of Pharmaceutics* 2021, 601:120536.

[73] Weng D, Yin ZF, Chen SS, He X, Li N, Chen T, et al.: Development and Assessment of the Efficacy and Safety of Human Lung-Targeting Liposomal Methylprednisolone Crosslinked with Nanobody. *Drug Delivery* 2021, 28:1419–1431.

[74] Arisoy S, Kocas M, Comoglu T, Guderer I, Banerjee S: Development of ACE2 Loaded Decoy Liposomes and Their Effect on SARS-CoV-2 for Covid-19 Treatment. *Pharmaceutics Development and Technology* 2022, 27:290–300.

[75] Gupta PV, Nirwane AM, Nagarsenker MS: Inhalable Levofloxacin Liposomes Complemented with Lysozyme for Treatment of Pulmonary Infection in Rats: *Effective Antimicrobial and Antibiofilm Strategy*. *AAPS Pharmaceutical Science and Technology* 2018, 19:1454–1467.

[76] Zhao J, Qin L, Song R, Su J, Yuan Y, Zhang X, et al.: Elucidating Inhaled Liposome Surface Charge on Its Interaction with Biological Barriers in the Lung. *European Journal of Pharmaceutics and Biopharmaceutics* 2022, 172:101–111.

[77] Derbali RM, Aoun V, Moussa G, Frei G, Tehrani SF, Del'Orto JC, et al.: Tailored Nanocarriers for the Pulmonary Delivery of Levofloxacin Against Pseudomonas Aeruginosa: *A Comparative Study. Molecular Pharmacology* 2019, 16:1906–1916.

[78] Alhariri M, Majrashi MA, Bahkali AH, Almajed FS, Azghani AO, Khiyami MA, et al.: Efficacy of Neutral and Negatively Charged Liposome-Loaded Gentamicin on Planktonic Bacteria and Biofilm Communities. *International Journal of Nanomedicine* 2017, 12:6949–6961.

[79] Parvathaneni V, Kulkarni NS, Shukla SK, Farrales PT, Kunda NK, Muth A, et al.: Systematic Development and Optimization of Inhalable Pirfenidone Liposomes for Non-Small Cell Lung Cancer Treatment. *Pharmaceutics* 2020, 12:206.

[80] Uchiya KI, Asahi S, Futamura K, Hamaura H, Nakagawa T, Nikai T, et al.: Antibiotic Susceptibility and Genotyping of Mycobacterium avium Strains That Cause Pulmonary and Disseminated Infection. *Antimicrobial Agents and Chemotherapy* 2018, 62: e02035–17.

[81] Hoy SM: Amikacin Liposome Inhalation Suspension in Refractory Mycobacterium avium Complex Lung Disease: A Profile of Its Use. *Clinical Drug Investigation* 2021, 41:405–412.
[82] Zhang J, Leifer F, Rose S, Chun DY, Thaisz J, Herr T, et al.: Amikacin Liposome Inhalation Suspension (ALIS) Penetrates Non-Tuberculous Mycobacterial Biofilms and Enhances Amikacin Uptake into Macrophages. *Frontiers in Microbiology* 2018, 9:915.
[83] Golia A, Mahmood BR, Fundora Y, Thornby KA, Chahine EB: Amikacin Liposome Inhalation Suspension for Mycobacterium avium Complex Lung Disease. *The Senior Care Pharmacist* 2020, 35:162–170.
[84] Griffith DE, Eagle G, Thomson R, Aksamit TR, Hasegawa N, Morimoto K, et al.: Amikacin Liposome Inhalation Suspension for Treatment-Refractory Lung Disease Caused by Mycobacterium avium Complex (CONVERT). A Prospective, Open-Label, Randomized Study. *American Journal of Respiratory and Critical Care Medicine* 2018, 198:1559–1569.
[85] Bilton D, Pressler T, Fajac I, Clancy JP, Sands D, Minic P, et al.: Amikacin Liposome Inhalation Suspension for Chronic Pseudomonas aeruginosa Infection in Cystic Fibrosis. *Journal of Cystic Fibrosis* 2020, 19:284–291.
[86] Blanchard JD, Elias V, Cipolla D, Gonda I, Bermudez LE: Effective Treatment of Mycobacterium Avium Subsp. Hominissuis and Mycobacterium Abscessus Species Infections in Macrophages, Biofilm, and Mice by Using Liposomal Ciprofloxacin. *Antimicrobial Agents and Chemotherapy* 2018, 62:e00440-18.
[87] Chai G, Park H, Yu S, Zhou F, Li J, Xu Q, et al.: Evaluation of Co-delivery of Colistin and Ciprofloxacin in Liposomes Using an in Vitro Human Lung Epithelial Cell Model. *International Journal of Pharmaceutics* 2019, 569:118616.
[88] Tran TT, Yu H, Vidaillac C, Lim AYH, Abisheganaden JA, Chotirmall SH, et al.: An evaluation of Inhaled Antibiotic Liposome versus Antibiotic Nanoplex in Controlling Infection in Bronchiectasis. *International Journal of Pharmaceutics* 2019, 559:382–392.
[89] Adler-Moore J, Lewis RE, Brüggemann RJM, Rijnders BJA, Groll AH, Walsh TJ: Preclinical Safety, Tolerability, Pharmacokinetics, Pharmacodynamics, and Antifungal Activity of Liposomal Amphotericin B. *Clinical Infectious Diseases* 2019, 68:S244–S259.
[90] Wang J, Li P, Yu Y, Fu Y, Jiang H, Lu M, et al.: Pulmonary Surfactant-Biomimetic Nanoparticles Potentiate Heterosubtypic Influenza Immunity. *Science* 2020, 367:eaau0810.
[91] Bernasconi V, Norling K, Gribonika I, Ong LC, Burazerovic S, Parveen N, et al.: A Vaccine Combination of Lipid Nanoparticles and A Cholera Toxin Adjuvant Derivative Greatly Improves Lung Protection Against Influenza Virus Infection. *Mucosal Immunology* 2021, 14:523–536.
[92] Mehnert W, Mäder K: Solid Lipid Nanoparticles: Production, Characterization and Applications. *Advanced Drug Delivery Reviews* 2001, 47:165–196.
[93] Müller RH, Mäder K, Gohla S: Solid Lipid Nanoparticles (SLN) for Controlled Drug Delivery - A Review of the State of the Art. *European Journal of Pharmaceutics and Biopharmaceutics* 2000, 50:161–177.
[94] Hu FQ, Zhang Y, Du YZ, Yuan H: Nimodipine Loaded Lipid Nanospheres Prepared by Solvent Diffusion Method in a Drug Saturated Aqueous System. *International Journal of Pharmaceutics* 2008, 348:146–152.
[95] Yoon G, Park JW, Yoon I-S: Solid Lipid Nanoparticles (SLNs) and Nanostructured Lipid Carriers (NLCs): Recent Advances in Drug Delivery. *Journal of Pharmaceutical Investigation* 2013, 43:353–362.
[96] Yu W, Liu C, Liu Y, Zhang N, Xu W: Mannan-Modified Solid Lipid Nanoparticles for Targeted Gene Delivery to Alveolar Macrophages. *Pharmaceutical Research* 2010, 27:1584–1596.
[97] Chattopadhyay P, Shekunov BY, Yim D, Cipolla D, Boyd B, Farr S: Production of Solid Lipid Nanoparticle Suspensions Using Supercritical Fluid Extraction of Emulsions (SFEE) for Pulmonary Delivery Using the AERx System. *Advanced Drug Delivery Reviews* 2007, 59:444–453.
[98] Jaafar-Maalej C, Andrieu V, Elaissari A, Fessi H: Beclomethasone-Loaded Lipidic Nanocarriers for Pulmonary Drug Delivery: Preparation, Characterization and in Vitro Drug Release. *Journal of Nanoscience and Nanotechnology* 2011, 11:1841–1851.
[99] Wang JL, Hanafy MS, Xu H, Leal J, Zhai Y, Ghosh D, et al.: Aerosolizable SiRNA-Encapsulated Solid Lipid Nanoparticles Prepared by Thin-Film Freeze-Drying for Potential Pulmonary Delivery. *International Journal of Pharmaceutics* 2021, 596:120215.
[100] Ma C, Wu M, Ye W, Huang Z, Ma X, Wang W, et al.: Inhalable Solid Lipid Nanoparticles for Intracellular Tuberculosis Infection Therapy: Macrophage-Targeting and pH-Sensitive Properties. *Drug Delivery and Translational Research* 2021, 11:1218–1235.
[101] Li N, Li X, Cheng P, Yang P, Shi P, Kong L, et al.: Preparation of Curcumin Solid Lipid Nanoparticles Loaded with Flower-Shaped Lactose for Lung Inhalation and Preliminary Evaluation of Cytotoxicity In Vitro. *Evidence-Based Complementary and Alternative Medicine* 2021, 2021:4828169.
[102] Mehrabani Yeganeh E, Bagheri H, Mahjub R: Preparation, Statistical Optimization and In-vitro Characterization of a Dry Powder Inhaler (DPI) Containing Solid Lipid Nanoparticles Encapsulating Amphotericin B: Ion Paired Complexes with Distearoyl Phosphatidylglycerol. *Iranian Journal of Pharmaceutical Research* 2020, 19:45–62.
[103] Chokshi NV, Rawal S, Solanki D, Gajjar S, Bora V, Patel BM, et al.: Fabrication and Characterization of Surface Engineered Rifampicin Loaded Lipid Nanoparticulate Systems for the Potential Treatment of Tuberculosis: An in Vitro and in Vivo Evaluation. *Journal of Pharmaceutical Sciences* 2021, 110:2221–2232.
[104] Truzzi E, Nascimento TL, Iannuccelli V, Costantino L, Lima EM, Leo E, et al.: In Vivo Biodistribution of Respirable Solid Lipid Nanoparticles Surface-Decorated With a Mannose-Based Surfactant: A Promising Tool for Pulmonary Tuberculosis Treatment? *Nanomaterials (Basel)* 2020, 10:568.
[105] Costa A, Sarmento B, Seabra V: Mannose-Functionalized Solid Lipid Nanoparticles Are Effective in Targeting Alveolar Mmacrophages. *European Journal of Pharmaceutical Sciences* 2018, 114:103–113.
[106] Rodenak-Kladniew B, Scioli Montoto S, Sbaraglini ML, Di Ianni M, Ruiz ME, Talevi A, et al.: Hybrid Ofloxacin/eugenol Co-loaded Solid Lipid Nanoparticles with Enhanced and Targetable Antimicrobial Properties. *International Journal of Pharmaceutics* 2019, 569:118575.
[107] Gaspar DP, Gaspar MM, Eleutério CV, Grenha A, Blanco M, Gonçalves LMD, et al.: Microencapsulated Solid

Lipid Nanoparticles as a Hybrid Platform for Pulmonary Antibiotic Delivery. *Molecular Pharmaceutics* 2017, 14:2977–2990.

[108] Pastor M, Moreno-Sastre M, Esquisabel A, Sans E, Viñas M, Bachiller D, et al.: Sodium Colistimethate Loaded Lipid Nanocarriers for the Treatment of Pseudomonas Aeruginosa Infections Associated with Cystic Fibrosis. *International Journal of Pharmaceutics* 2014, 477:485–494.

[109] Mirchandani Y, Patravale VB, SB: Solid Lipid Nanoparticles for Hydrophilic Drugs. *Journal of Controlled Release* 2021, 335:457–464.

[110] Zhao Y, Chang YX, Hu X, Liu CY, Quan LH, Liao YH: Solid Lipid Nanoparticles for Sustained Pulmonary Delivery of Yuxingcao Essential Oil: Preparation, Characterization and in Vivo Evaluation. *International Journal of Pharmaceutics* 2017, 516:364–371.

[111] Liparulo A, Esposito R, Santonocito D, Muñoz-Ramírez A, Spaziano G, Bruno F, et al.: Formulation and Characterization of Solid Lipid Nanoparticles Loading RF22-c, a Potent and Selective 5-LO Inhibitor, in a Monocrotaline-Induced Model of Pulmonary Hypertension. *Frontiers in Pharmacology* 2020, 11:83.

[112] Gulin-Sarfraz T, Jonasson S, Wigenstam E, von Haartman E, Bucht A, Rosenholm JM: Feasibility Study of Mesoporous Silica Particles for Pulmonary Drug Delivery: Therapeutic Treatment with Dexamethasone in a Mouse Model of Airway Inflammation. *Pharmaceutics* 2019, 11:149.

[113] Rathnayake K, Patel U, Pham C, McAlpin A, Budisalich T, Jayawardena SN: Targeted Delivery of Antibiotic Therapy to Inhibit Pseudomonas aeruginosa Using Lipid-Coated Mesoporous Silica Core-Shell Nanoassembly. *ACS Applied Bio Materials* 2020, 3:6708–6721.

[114] Tenland E, Pochert A, Krishnan N, Umashankar Rao K, Kalsum S, Braun K, et al.: Effective Delivery of the Anti-Mycobacterial Peptide NZX in Mesoporous Silica Nanoparticles. *PLoS One* 2019, 14:e0212858.

[115] Xu N, Lu W, Meng L, Feng X, Xuan J, Liu F, et al.: Carbonic Anhydrase Inhibition, Antioxidant Activity against Alveolar Epithelial Cells and Antibacterial Effect against Klebsiella Pneumoniae Enabled by Synthesized Silica Nanoparticles through Laser Ablation Technique. *Life Sciences* 2021, 278:119032.

[116] Clemens DL, Lee BY, Plamthottam S, Tullius MV, Wang R, Yu CJ, et al.: Nanoparticle Formulation of Moxifloxacin and Intramuscular Route of Delivery Improve Antibiotic Pharmacokinetics and Treatment of Pneumonic Tularemia in a Mouse Model. *ACS Infectious Diseases* 2019, 5:281–291.

[117] Tewes F, Ehrhardt C, Healy AM: Superparamagnetic Iron Oxide Nanoparticles (SPIONs)-Loaded Trojan Microparticles for Targeted Aerosol Delivery to the Lung. *European Journal of Pharmaceutics and Biopharmaceutics* 2014, 86:98–104.

[118] Miranda MS, Rodrigues MT, Domingues RMA, Costa RR, Paz E, Rodríguez-Abreu C, et al.: Development of Inhalable Superparamagnetic Iron Oxide Nanoparticles (SPIONs) in Microparticulate System for Antituberculosis Drug Delivery. *Advanced Healthcare Materials* 2018, 7:e1800124.

[119] Trivedi R, Kompella UB: Nanomicellar Formulations for Sustained Drug Delivery: Strategies and Underlying Principles. *Nanomedicine (Lond)* 2010, 5:485–505.

[120] Bose A, Roy Burman D, Sikdar B, Patra P: Nanomicelles: Types, Properties and Applications in Drug Delivery. *IET Nanobiotechnology* 2021, 15:19–27.

[121] Cholkar K, Patel A, Vadlapudi AD, Mitra AK: Novel Nanomicellar Formulation Approaches for Anterior and Posterior Segment Ocular Drug Delivery. *Recent Patents on Nanotechnology* 2012, 2:82–95.

[122] Gilani K, Moazeni E, Ramezanli T, Amini M, Fazeli MR, Jamalifar H: Development of Respirable Nanomicelle Carriers for Delivery of Amphotericin B by Jet Nebulization. *Journal of Pharmaceutical Sciences*. 2011, 100:252–259.

[123] Farhangi M, Kobarfard F, Mahboubi A, Vatanara A, Mortazavi SA: Preparation of an Optimized Ciprofloxacin-loaded Chitosan Nanomicelle with Enhanced Antibacterial Activity. *Drug Development and Industrial Pharmacy* 2018, 44:1273–1284.

[124] Farhangi M, Mahboubi A, Kobarfard F, Vatanara A, Mortazavi SA: Optimization of a Dry Powder Inhaler of Ciprofloxacin-loaded Polymeric Nanomicelles by Spray Drying Process. *Pharmaceutical Development and Technology* 2019, 24:584–592.

[125] Ren B, Li K, Liu Z, Liu G, Wang H: White Light-triggered Zwitterionic Polymer Nanoparticles Based on an AIE-active Photosensitizer for Photodynamic Antimicrobial Therapy. *Journal of Materials Chemistry B* 2020, 8:10754–10763.

[126] Pourhajibagher M, Talaei N, Bahador A: Evaluation of Antimicrobial Effects of Photo-sonodynamic Antimicrobial Chemotherapy Based on Nano-micelle Curcumin on Virulence Gene Expression Patterns in Acinetobacter baumannii. *Infectious Disorder Drug Targets* 2022, 22:e201221199163.

[127] de Barros AODS, Pinto SR, Dos Reis SRR, Ricci-Junior E, Alencar LMR, Bellei NCJ, et al.: Polymeric Nanoparticles and Nanomicelles of Hydroxychloroquine Co-loaded with Azithromycin Potentiate Anti-SARS-CoV-2 Effect. *Journal of Nanostructure Chemistry* 2022, 26:1–19.

[128] Jasim R, Schneider EK, Han M, Azad MAK, Hussein M, Nowell C, et al.: A Fresh Shine on Cystic Fibrosis Inhalation Therapy: Antimicrobial Synergy of Polymyxin B in Combination with Silver Nanoparticles. *Journal of Biomedical Nanotechnology* 2017, 13:447–457.

[129] Pérez-Cantero A, Serrano DR, Navarro-Rodríguez P, Schätzlein AG, Uchegbu IF, Torrado JJ, et al.: Increased Efficacy of Oral Fixed-Dose Combination of Amphotericin B and AHCC® Natural Adjuvant against Aspergillosis. *Pharmaceutics* 2019, 11:456.

[130] Mehdizadeh M, Sheikhpour M, Salahshourifar I, Siadat SD, Saffarian P: An in Vitro Study of Molecular Effects of a Combination Treatment with Antibiotics and Nanofluid Containing Carbon Nano-tubes on Klebsiella pneumoniae. *Iranian Journal of Public Health* 2021, 50:2292–2301.

[131] Kingstad-Bakke B, Toy R, Lee W, Pradhan P, Vogel G, Marinaik CB, et al.: Polymeric Pathogen-Like Particles-Based Combination Adjuvants Elicit Potent Mucosal T Cell Immunity to Influenza A Virus. *Frontiers in Immunology* 2021, 11:559382.

[132] Jin H, Zhao Z, Lan Q, Zhou H, Mai Z, Wang Y, et al.: Nasal Delivery of Hesperidin/Chitosan Nanoparticles Suppresses Cytokine Storm Syndrome in a Mouse Model of Acute Lung Injury. *Front Pharmacology* 2020, 11:592238.

[133] Haque A, Dewerth A, Antony JS, Riethmüller J, Schweizer GR, Weinmann P, et al.: Chemically Modified hCFTR mRNAs Recuperate Lung Function in a Mouse Model of Cystic Fibrosis. *Science Report* 2018, 8:16776.

[134] Wu T, Liao W, Wang W, Zhou J, Tan W, Xiang W, et al.: Genipin-crosslinked Carboxymethyl Chitosan Nanogel for Lung-targeted Delivery of Isoniazid and Rifampin. *Carbohydrate Polymers* 2018, 197:403–413.

[135] Huang J, Guo J, Zou X, Zhu J, Wu S, Zhang T: Bioinspired Heteromultivalent Chitosan-α-Fe_2O_3/Gadofullerene Hybrid Composite for Enhanced Antibiotic-Resistant Bacterial Pneumonia. *Journal of Biomedical Nanotechnology* 2021, 17:1217–1228.

[136] Gondil VS, Dube T, Panda JJ, Yennamalli RM, Harjai K, Chhibber S: Comprehensive evaluation of chitosan nanoparticle based phage lysin delivery system; a novel approach to counter S. pneumoniae infections. *International Journal of Pharmaceutics* 2020, 573:118850.

[137] Arauzo B, Lobera MP, Monzon A, Santamaria J: Dry Powder Formulation for pulmonary infections: Ciprofloxacin loaded in chitosan sub-micron particles generated by electrospray. *Carbohydrate Polymers* 2021, 273:118543.

[138] Ernst J, Klinger-Strobel M, Arnold K, Thamm J, Hartung A, Pletz MW, et al.: Polyester-based Particles to Overcome the Obstacles of Mucus and Biofilms in the Lung for Tobramycin Application under Static and Dynamic Fluidic Conditions. *European Journal of Pharmaceutics and Biopharmaceutics* 2018, 131:120–129.

[139] Tan Q, He L, Meng X, Wang W, Pan H, Yin W, et al.: Macrophage Biomimetic Nanocarriers for Anti-inflammation and Targeted Antiviral Treatment in COVID-19. *Journal of Nanobiotechnology* 2021, 19:173.

[140] Shirkhani K, Teo I, Armstrong-James D, Shaunak S: Nebulised Amphotericin B-polymethacrylic Acid Nanoparticle Prophylaxis Prevents Invasive Aspergillosis. *Nanomedicine* 2015, 11:1217–1226.

[141] Wang Y, Yuan Q, Feng W, Pu W, Ding J, Zhang H, et al.: Targeted Delivery of Antibiotics to the Infected Pulmonary Tissues Using ROS-responsive Nanoparticles. *Journal of Nanobiotechnology* 2019, 17:103.

[142] Machelart A, Salzano G, Li X, Demars A, Debrie AS, Menendez-Miranda M, et al.: Intrinsic Antibacterial Activity of Nanoparticles Made of β-Cyclodextrins Potentiates Their Effect as Drug Nanocarriers against Tuberculosis. *ACS Nano* 2019, 13:3992–4007.

[143] Griffith DE, Thomson R, Flume PA, Aksamit TR, Field SK, Addrizzo-Harris DJ, et al.: Amikacin Liposome Inhalation Suspension for Refractory Mycobacterium avium Complex Lung Disease: Sustainability and Durability of Culture Conversion and Safety of Long-term Exposure. *Chest* 2021, 160:831–842.

[144] Winthrop KL, Flume PA, Thomson R, Mange KC, Yuen DW, Ciesielska M, et al.: Amikacin Liposome Inhalation Suspension for Mycobacterium avium Complex Lung Disease: A 12-Month Open-Label Extension Clinical Trial. *Annals of the American Thoracis Society* 2021, 18:1147–1157.

[145] Henriette Zweijpfenning SM, Chiron R, Essink S, Schildkraut J, Akkerman OW, Aliberti S, et al.: Safety and Outcomes of Amikacin Liposome Inhalation Suspension for Mycobacterium Abscessus Pulmonary Disease: A NTM-NET study. *Chest* 2022, 162:76–81.

[146] Bilton D, Fajac I, Pressler T, Clancy JP, Sands D, Minic P, et al.: Long-term Amikacin Liposome Inhalation Suspension in Cystic Fibrosis Patients with Chronic P. Aeruginosa Infection. *Journal of Cystic Fibrosis* 2021, 20:1010–1017.

[147] Ledoux MP, Guffroy B, Nivoix Y, Simand C, Herbrecht R: Invasive Pulmonary Aspergillosis. *Seminars in Respiratory and Critical Care Medicine* 2020, 41:80–98.

[148] Ambati S, Ellis EC, Pham T, Lewis ZA, Lin X, Meagher RB: Antifungal Liposomes Directed by Dectin-2 Offer a Promising Therapeutic Option for Pulmonary Aspergillosis. *mBio* 2021, 12:e00030-21.

[149] Burton AJ, Giguère S, Arnold RD: Pharmacokinetics, Pulmonary Disposition and Tolerability of Liposomal Gentamicin and Free Gentamicin in Foals. *Equine Veterinary Journal* 2015, 47:467–472.

[150] Pandolfi L, Marengo A, Japiassu KB, Frangipane V, Tsapis N, Bincoletto V, et al.: Liposomes Loaded with Everolimus and Coated with Hyaluronic Acid: A Promising Approach for Lung Fibrosis. *International Journal of Molecular Sciences* 2021, 22:7743.

[151] Li J, Zhang K, Wu D, Ren L, Chu X, Qin C, et al.: Liposomal Remdesivir Inhalation Solution for Targeted Lung Delivery as a Novel Therapeutic Approach for COVID-19. *Asian Journal of Pharmaceutical Sciences* 2021, 16:772–783.

[152] Pinto BF, Ribeiro LNB, da Silva G, Freitas CS, Kraemer L, Oliveira FMS, et al.: Inhalation of Dimethyl Fumarate-encapsulated Solid Lipid Nanoparticles Attenuate Clinical Signs of Experimental Autoimmune Encephalomyelitis and Pulmonary Inflammatory Dysfunction in Mice. *Clinical Science (Lond)* 2022, 136:81–101.

[153] Carvalho FO, Silva ÉR, Nunes PS, Felipe FA, Ramos KPP, Ferreira LAS, et al.: Effects of the Solid Lipid Nanoparticle of Carvacrol on Rodents with Lung Injury from Smoke Inhalation. *Naunyn Schmiedebergs Archives of Pharmacology* 2020, 393:445–455.

[154] Wang J, Sun Y, Tian X: The Inhibitory Effect of Icariin Nanoparticles on Angiogenesis in Pulmonary Fibrosis. *Journal of Nanoscience and Nanotechnology* 2021, 21:5429–5435.

[155] Levina A, Repkova M, Shikina N, Ismagilov Z, Kupryushkin M, Pavlova A, et al.: Pronounced Therapeutic Potential of Oligonucleotides Fixed on Inorganic Nanoparticles Against Highly Pathogenic H5N1 Influenza A Virus in vivo. *European Journal of Pharmaceutics and Biopharmceutics* 2021, 162:92–98.

[156] García-Fernández A, Sancho M, Bisbal V, Amorós P, Marcos MD, Orzáez M, et al.: Targeted-Lung Delivery of Dexamethasone Using Gated Mesoporous Silica Nanoparticles. A New Therapeutic Approach for Acute Lung Injury Treatment. *Journal of Controlled Release* 2021, 337:14–26.

[157] Kim S, Kim SY, Rho SJ, Kim SH, Song SH, Kim CH, et al.: Biocompatible N-acetyl-nanoconstruct Alleviates Lipopolysaccharide-induced Acute Lung Injury in vivo. *Science Report* 2021, 11:22662.

[158] Hassaniazad M, Inchehsablagh BR, Kamali H, Tousi A, Eftekhar E, Jaafari MR, et al.: The Clinical Effect of Nano Micelles Containing Curcumin as a Therapeutic Supplement in Patients with COVID-19 and the Immune Responses Balance Changes Following Treatment: A Structured Summary of a Study Protocol for a Randomised Controlled Trial. *Trials* 2020, 21:876.

[159] Li J, Chen L, Li C, Fan Y, Zhan M, Sun H, et al.: Phosphorus Dendron Nanomicelles as a Platform for Combination

Anti-inflammatory and Antioxidative Therapy of Acute Lung Injury. *Theranostics* 2022, 12:3407–3419.

[160] Galdopórpora JM, Martinena C, Bernabeu E, Riedel J, Palmas L, Castangia I, et al.: Inhalable Mannosylated Rifampicin-Curcumin Co-Loaded Nanomicelles with Enhanced In Vitro Antimicrobial Efficacy for an Optimized Pulmonary Tuberculosis Therapy. *Pharmaceutics* 2022, 14:959.