# PN Drugs Against Intracellular Infections

Subjects: Nanoscience & Nanotechnology

Contributor: Arturo Sánchez , Susana P. Mejía , Jahir Orozco

Polymeric nanocarriers (PNs) are a promising alternative for delivering intracellularly antimicrobials of high toxicity, low solubility and low bioavailability to reduce dose and side effects and improve their therapeutic efficacy. They may prevent unwanted drug interactions and degradation thus decreasing the development of resistance in microorganisms.

polymeric nanocarrier intracellular infection antimicrobial drug biodistribution pharmacokinetic

Polymeric nanocarriers (PNs) have demonstrated to be a promising alternative to treat intracellular infections. They have outstanding performance in delivering antimicrobials intracellularly to reach an adequate dose level and improve their therapeutic efficacy. PNs offer opportunities for preventing unwanted drug interactions and degradation before reaching the target cell of tissue and thus decreasing the development of resistance in microorganisms. The use of PNs has the potential to reduce the dose and adverse side effects, providing better efficiency and effectiveness of therapeutic regimens, especially in drugs having high toxicity, low solubility in the physiological environment and low bioavailability.

## 1. Introduction

In the frontier of different areas, nanochemistry uses a variety of methods to assemble materials at a nanometer-scale size, with new unique features in respect to the bulk material counterparts, including electronic, magnetic, optical, chemical, and mechanical properties. The interdisciplinarity of nanochemistry involves scientific and technical knowledge from diverse fields such as natural- and material-sciences, engineering, medicine, and pharmacy, among others. Although it is not an easy task, it is aimed at searching for new or existing (bio)materials, understanding their interactions and providing new functionalities towards new products and unexpected applications; these perspectives are encouraging and highly promising, up to the point of being able to generate a revolutionary knowledge of the world at the nanoscale. Within the health care field, nanochemistry offers outstanding opportunities for the design of diagnostic and therapeutic tools; for example, nanoconjugates and nanoplatforms assembled for the controlled and site-specific delivery of active principles with enhanced pharmacological properties against intracellular microorganisms.

Intracellular infectious diseases represent a major challenge in health care due to the low specificity of available treatments and the appearance of co-infections and drug-resistant pathogens, which limits the existing therapies [1][2][3][4][5][6]. Recent advances in the field of nanotechnology offer alternatives to improve the biological activity of

existing drugs, which is of great potential to help to overcome the drawbacks inherent in the treatment of intracellular infectious diseases. Encapsulation of drugs into nanoparticles (NP) represents a valuable option to improve drug solubility and biodistribution, prevent undesirable interactions and drug degradation before reaching the target tissues and cells and non-specific accumulation of drugs in other tissues. In this context, nanoencapsulated drugs hold the potential to increase the efficiency and effectiveness of therapeutic regimens, particularly in drugs with low solubility, short half-life, variable absorption, and undesirable interactions [7][8][9][10][11][12]. Such improvement comes from the reduction of dose and drug frequencies in patients, which dramatically impact on decreasing both toxic and side effects that drugs usually have intrinsically.

In the last years, the encapsulation of drugs into synthetic and natural polymeric nanocarriers (PNs) has been one of the most explored systems for drug delivery. The resultant nanosystems enjoy high biocompatibility and biodegradability, being very reproducible and amenable for mass production. PNs are usually designed to be stable, have high drug loading capacity and the ability to transport one or more active ingredients (with similar or different physicochemical properties), or a combination of therapeutic agents in the same formulation. Such features are very appealing for delivering drugs by different routes and multipurpose clinical approaches [13][14][15]. Size, surface charge and morphology of PNs can be tailored on demand to produce solid capsules/nanoparticles, amphiphilic structures (micelles), and hyperbranched structures (dendrimers), and vesicles [16][17][18][19][20][21][22], which sizes can usually be smaller than 100 nm but eventually reach hundreds of nm, depending upon the active ingredient or drug, the type and length of the polymer and the preparation method [23][24][25]. Besides, the polymers can be chemically modified to encapsulate, adsorb, disperse, or bound the drug [26][27][28].

Along with modulation of size, form and physicochemical properties of the polymer, to functionalize PNs with specific ligands may actively drive drugs to site-specific for the efficient drug uptaking, thereby readily reaching therapeutic intracellular levels [29][30][31]. Moreover, the PNs are extremely versatile in controlling the drug release profile. For instance, PNs can be tuned for the immediate or sustained delivery of drugs in a localized place either by natural diffusion of the drug or by osmotic, erosion or degradation processes. However, such mechanisms have evolved towards more refined ones based on physical, chemical, or biochemical external stimuli. In this context, triggered local changes in pH and temperature, in the number of reactive oxygen species (ROS), the reductive or oxidative states, and conformational changes in the polymer have been extensively used [32].

In recent years, a large number of drug delivery systems have been described in many medical areas, including intracellular infections [30][33]. Indeed, the global infectious disease therapeutics market size was valued at $46.88 billion in 2018 and is estimated to reach $64.5 in 2023 [34]. Yet, many challenges are still needed to face before many antimicrobial-based nanoformulations hit the market. They are related to the fact that pathogenic microorganisms are established mainly in phagocytic cells. Besides, nanocarriers injected intravenously are mostly recognized and cleaned either by phagocytic cells from the endothelial reticulum system (RES) or by the mononuclear phagocyte system (MPS) going “passively” to the macrophages, the reservoir of most intracellular pathogens [35]. Another challenge of nanocarriers is the necessity to increase their drug loading capacity to administrate less amount of material but sufficient to reach a therapeutic concentration of drug at the site of infection that avoids toxicity and side effects [36][37][38][39]. Other imminent necessities are related to the increase of

stability and better control of the drug release profile. Therefore, it is of paramount importance to design antimicrobial-based nanocarriers, not only with high drug loading capacity and well-controlled release but site-directed to infected cells.

This review provides an overview of the use of different organic nanoparticle precursors and the main assembly methods to produce nanoplatforms of tuned physicochemical and morphological properties and surface chemistry for controlled release of antimicrobials in a target cell or tissue, in a physiological environment, depending on the active principle nature and administration route. It points out the remarkable versatility of these nanosystems and details the facing challenges, as well as the opportunities of the nanoplatforms to deliver antimicrobial drugs to efficiently and effectively treat intracellular infections. It finally mentions some nanotoxicology aspects essential to be considered and prospects in the topic that expect to stimulate advances and new opportunities in this promising field.

## 2. Polymeric Nanocarriers against Intracellular Infections: General Aspects

The efficient treatment of intracellular infections by antimicrobials is highly challenging. Challenges are related to the evasion of intracellular infectious agents by host phagocytic killing mechanisms, the establishment of intracellular survival machinery and the worldwide misuse of antibiotics, which are rising multidrug resistance of pathogens [40][41]. Besides, many conventional antimicrobial-based treatments possess low cellular penetration and therefore, drug distribution at the subcellular level is not uniform; thus, the site of infection may remain without treatment. Once inside the cell, antimicrobials activity can be influenced by enzymatic inactivation, and changes in pH and chemical environment, as discussed in the next sections. It may result in a low intracellular concentration of antimicrobials, thus limiting the efficacy and efficiency of the therapy [42][43].

PNs are an alternative approach extensively studied to overcome some limitations of antimicrobial therapies by encapsulating drugs to improve their ability to enter the cells and release the cargo intracellularly [44][45][46]. As detailed in the following sections, cellular internalization of PNs includes phagocytotic-mediated-, clathrin-mediated-, caveolae-mediated- and receptor-mediated-endocytosis or a mixture of them [47][48]. After cellular uptake, it engineers the escape of endocytic vesicles formed to avoid lysosomal degradation for cytosolic delivery as well as the possible intracellular trafficking of PNs targeting subcellular compartments where determined intracellular pathogens reside. PNs protect antimicrobials of degradation, increase their solubility and bioavailability for their controlled and targeted release.

They may reduce drug dose and adverse side effects and provide better efficiency and efficacy of therapeutic regimens. By modulating properties and functionality of PNs they can specifically address the target tissue or cell depending on the route of administration (oral, parenteral, intranasal, topical, and intravenous, among others).

PNs are commonly built by self-assembly of biocompatible and biodegradable polymeric materials to minimize non-specific cytotoxicity on healthy tissues with convenient degradation kinetic profiles and complete metabolization of

degradation products [49][50]. A myriad of nanoplatforms with specific morphological characteristics (nanorods, nanoworms, nanodiscs, among others), size and surface chemistry, can be produced depending on a diversity of precursors, fabrication methods and antimicrobials' chemical nature. Among them, nanocapsules and nanospheres are the most common. While nanocapsules of vesicular structure generally have the drug immersed in a liquid core surrounded by a solidified polymeric shell, nanospheres are a solid/mass polymeric matrix in which drug is encapsulated inside or over the structure surface. Active principles can be associated with nanostructures by physical encapsulation, covalent conjugation, adsorption, electrostatic- and van der Waal-interactions. Modulation of cellular uptake, extracellular transport and intracellular drug delivery is achieved by tuning size, shape, surface chemistry, antimicrobials' intrinsic properties and microenvironments that PNs need to overstep. Targeting can be passive, activated by ligands attached to the outermost nanoparticle surface and drug release by natural ways or triggered by an external or internal stimulus [51][52].

In nanotherapeutics based on passive targeting, nanocarriers must reach the site of action by physiological or physicochemical changes that occur naturally in the body. They include differences in the pH among tissues (tumor microenvironment), defective vasculature enhanced permeability and retention effect (EPR), capture by the mononuclear phagocytic system and differences in the redox properties of the systems. In active targeting, the surface of nanocarriers is modified to generate affinity with bioreceptors or cellular biomarkers, tissues or organs through ligand-receptor interactions [53]. Ligand-nanocarrier conjugation uses different coupling methodologies, including the formation of disulfide bonds, cross-linking, covalent coupling, ionic interactions, layer-by-layer assembly, among other strategies [54]. Coating the nanocarriers with targeting ligands such as peptides, antibodies, lectins, sugars, folate- and mannose-receptors, among others, drive them to the site of action, thereby increasing the concentration of the active principle in the place and avoiding non-specific accumulations and the concomitant adverse effects [55][56][57].

Drugs are commonly released from nanocapsules rapidly or in a sustained fashion. It depends on the formulation. Drug release can be by diffusion through the porous or polymeric chains of the PNs or by osmotic-, erosion- or degradation-processes. However, natural release mechanisms are migrating towards more sophisticated ones based on physical, chemical or biochemical external stimuli. For example, the response of PNs can be triggered by stimulation with radiation, ultrasound, magnetic fields and temperature. The response can be modulated by changes in the pH and ionic strength, the cellular environment, or by enzymes [58]. Among stimuli-responsive PNs, nanogel-based PNs establish three-dimensional polymeric networks at nanoscale level with high capacity to water uptake and cross-linking. While they change their volume by absorbing water, the surrounding environment influences their behavior, thus generating stimuli-responsive systems (pH, ionic strength, electric field or temperature). Nanogels are prepared by natural or synthetic polymers, including chitosan, methylcellulose, ethylcellulose, dextran, polysaccharide-based polymers, dextrin, poly(oleic acid-Y-N-isopropyl acrylamide), polyvinyl alcohol, alginate, hyaluronic acid, poly(N-isopropyl acrylamide), among others. Nanoencapsulation of active principles of different natures can be achieved either by nanohydrogels or by nano-organo-gels, having higher loading capacity, controlled release and biocompatibility regarding other types of nanoparticles.

Nanohydrogels encapsulate hydrophobic and hydrophilic compounds, where the cross-linked network functions as a matrix holding the absorbed liquid medium, which modulates the diffusion of the active principles [59]. In contrast, nano-organo-gels have a micelle-like structure having hydrophobic regions (that hold oily compounds) attached to the hydrophilic regions at the center of the nanostructure. Nanogels performance can be modulated by changing their size and surface charge, or by incorporating targeting ligands, changes in cross-linking density or PEGylation strategies. Encapsulated active principles are released by hydrolytic degradation of the gel network. Recent studies have demonstrated the disruption of nanogels by using cross-linking agents sensible to temperature, light, differences in pH, use of disulfide bond linkages and cleavage by glutathione (GSH) enzyme. Therefore, the active principle can reach intracellular targets after nanogels are endocytosed, promoting endolysosome escape and improving intracellular therapy [59][60][61].

## 3. Future Outlooks

Attempts to fight intracellular infectious agents have given us important lessons, bringing out how urgently needs to be the efforts to develop alternative antimicrobial therapies that manage to increase antimicrobial efficacy and decrease microbial resistance as compared to conventional antimicrobial treatments. In this context, PNs have emerged as a nanoplatform having broad prospects for the development of highly promising site-directed antimicrobial therapies for the management of intracellular infections whose products on the market expect to be established shortly, as judged by the number of research publications and patents currently granted in the field.

Recent advances in nanoparticulate formulations, including hybrid strategies, nanoencapsulation of natural products, targeting ligand-based formulations and smart materials, have shown to allow tuning the bio-physicochemical properties of PNs for enhanced antimicrobial efficacy. However, there are still many challenges to face to improve PN-based antimicrobial technology towards scaling-up at the industrial level and reach the market. For example, PNs captured by the immune system hinders the site-specific drug delivery facilitated by highly selective targeting ligands leading to ineffective internalization. Nanosystems toxicity and nonspecific biodistribution limit in vivo practical applicability. Furthermore, proper standardization is not trivial both in vitro and in vivo, which limit systematic comparative studies. Therefore, it is necessary to continue researching to advance the limited understanding of the fundamental processes of PNs in and out of the human body, given the multiple interactions of antimicrobial nanotherapeutics among them, with the protein corona and with the vast diversity of organs, tissues and cells on their journey from administration to therapeutic targets.

A higher number of standardized production methods, validated studies of toxicity, bioequivalence, clinical studies, and the establishment of reference materials may impact on gaining a better knowledge of the antimicrobial nanotherapeutic systems and their associated pro and contra, in the way to creating products that ensure quality, safety and efficiency. The final balance of this process defines the scope of PN-encapsulated drugs, to quickly reach high acceptance in the market, thereby offering added value as compared to conventional antimicrobial therapies.

## References


1. Dodds Ashley, E.; Lewis, R.; Lewis, J.; Martin, C.; Andes, D. Pharmacology of Systemic Antifungal Agents. Clin. Infect. Dis. 2006, 43, doi:10.1086/504492.
2. Jayaraman, R. Antibiotic resistance: An overview of mechanisms and a paradigm shift. Curr. Sci. 2009, 96,1475–1484.
3. Ladaviere, C.; Gref, R. Toward an optimized treatment of intracellular bacterial infections: Input of nanoparticulate drug delivery systems. Nanomedicine (Lond) 2015, 10, 3033–3055, doi:10.2217/nnm.15.128.
4. Zazo, H.; Colino, C.I.; Lanao, J.M. Current applications of nanoparticles in infectious diseases. J. Control. Release 2016, 224, 86–102, doi:10.1016/j.jconrel.2016.01.008.
5. Costa-Gouveia, J.; Aínsa, J.A.; Brodin, P.; Lucía, A. How can nanoparticles contribute to antituberculosis therapy? Drug Discov. Today 2017, 22, 600–607, doi:10.1016/j.drudis.2017.01.011.
6. Lee, N.-Y.; Ko, W.-C.; Hsueh, P.-R. Nanoparticles in the Treatment of Infections Caused by Multidrug-Resistant Organisms. Front. Pharmacol. 2019, 10, 1153, doi:10.3389/fphar.2019.01153.
7. Tukulula, M.; Gouveia, L.; Paixao, P.; Hayeshi, R.; Naicker, B.; Dube, A. Functionalization of PLGA Nanoparticles with 1,3-beta-glucan Enhances the Intracellular Pharmacokinetics of Rifampicin in Macrophages. Pharm. Res. 2018, 35, 111, doi:10.1007/s11095-018-2391-8.
8. Jahagirdar, P.S.; Gupta, P.K.; Kulkarni, S.P.; Devarajan, P. V Polymeric curcumin nanoparticles by a facile in situ method for macrophage targeted delivery. Bioeng. Transl. Med. 2019, 4, 141–151, doi:10.1002/btm2.10112.
9. Singh, L.; Kruger, H.G.; Maguire, G.E.M.; Govender, T.; Parboosing, R. The role of nanotechnology in the treatment of viral infections. Ther. Adv. Infect. Dis. 2017, 4, 105–131, doi:10.1177/2049936117713593.
10. Biswaro, L.S.; Garcia, M.P.; da Silva, J.R.; Neira Fuentes, L.F.; Vera, A.; Escobar, P.; Azevedo, R.B. Itraconazole encapsulated PLGA-nanoparticles covered with mannose as potential candidates against leishmaniasis. J. Biomed. Mater. Res. B. Appl. Biomater. 2019, 107, 680–687, doi:10.1002/jbm.b.34161.
11. Hu, J.; Wei, P.; Seeberger, P.H.; Yin, J. Mannose-Functionalized Nanoscaffolds for Targeted Delivery in Biomedical Applications. Chem. Asian J. 2018, 13, 3448–3459, doi:10.1002/asia.201801088.
12. Gao, W.; Chen, Y.; Zhang, Y.; Zhang, Q.; Zhang, L. Nanoparticle-based local antimicrobial drug delivery. Adv. Drug Deliv. Rev. 2018, 127, 46–57, doi:10.1016/j.addr.2017.09.015.

13. Mir, M.; Ahmed, N.; ur Rehman, A. Recent applications of PLGA based nanostructures in drug delivery. Colloids Surf. B Biointerfaces 2017, 159, 217–231, doi:10.1016/j.colsurfb.2017.07.038.
14. Xu, Y.; Kim, C.-S.; Saylor, D.M.; Koo, D. Polymer degradation and drug delivery in PLGA-based drug-polymer applications: A review of experiments and theories. J. Biomed. Mater. Res. B. Appl. Biomater. 2017, 105, 1692–1716, doi:10.1002/jbm.b.33648.
15. Kamaly, N.; Yameen, B.; Wu, J.; Farokhzad, O.C. Degradable controlled-release polymers and polymeric nanoparticles: Mechanisms of controlling drug release. Chem. Rev. 2016, 116, 2602–2663, doi:10.1021/acs.chemrev.5b00346.
16. Bentz, K.C.; Savin, D.A. Hollow polymer nanocapsules: Synthesis, properties, and applications. Polym. Chem. 2018, 9, 2059–2081, doi:10.1039/C8PY00142A.
17. Gao, M.; Yang, Y.; Bergfel, A.; Huang, L.; Zheng, L.; Bowden, T.M. Self-assembly of cholesterol end-capped polymer micelles for controlled drug delivery. J. Nanobiotechnol. 2020, 18, 13, doi:10.1186/s12951-020-0575-y.
18. Ganda, I.S.; Zhong, Q.; Hali, M.; Albuquerque, R.L.C.; Padilha, F.F.; da Rocha, S.R.P.; Whittum-Hudson, J.A. Dendrimer-conjugated peptide vaccine enhances clearance of Chlamydia trachomatis genital infection. Int. J. Pharm. 2017, 527, 79–91, doi:10.1016/j.ijpharm.2017.05.045.
19. Zhu, Y.; Yang, B.; Chen, S.; Du, J. Polymer vesicles: Mechanism, preparation, application, and responsive behavior. Prog. Polym. Sci. 2017, 64, 1–22, doi:10.1016/j.progpolymsci.2015.05.001.
20. Zhao, Y.; Li, X.; Zhao, X.; Yang, Y.; Li, H.; Zhou, X.; Yuan, W. Asymmetrical Polymer Vesicles for Drug delivery and Other Applications. Front. Pharmacol. 2017, 8, 374, doi:10.3389/fphar.2017.00374.
21. Letchford, K.; Burt, H. A review of the formation and classification of amphiphilic block copolymer nanoparticulate structures: Micelles, nanospheres, nanocapsules and polymersomes. Eur. J. Pharm. Biopharm. 2007, 65, 259–269, doi:10.1016/j.ejpb.2006.11.009.
22. Colorado, D.; Fernandez, M.; Orozco, J.; Lopera, Y.; Muñoz, D.L.; Acín, S.; Balcazar, N. Metabolic Activity of Anthocyanin Extracts Loaded into Non-ionic Niosomes in Diet-Induced Obese Mice. Pharm. Res. 2020, 37, 152, doi:10.1007/s11095-020-02883-z.
23. Hickey, J.W.; Santos, J.L.; Williford, J.-M.; Mao, H.-Q. Control of polymeric nanoparticle size to improve therapeutic delivery. J. Control. Release 2015, 219, 536–547, doi:10.1016/j.jconrel.2015.10.006.
24. Rezvantalab, S.; Drude, N.I.; Moraveji, M.K.; Guvener, N.; Koons, E.K.; Shi, Y.; Lammers, T.; Kiessling, F. PLGA-Based Nanoparticles in Cancer Treatment. Front. Pharmacol. 2018, 9, 1260, doi:10.3389/fphar.2018.01260.

25. Cowen, T.; Karim, K.; Piletsky, S.A. Solubility and size of polymer nanoparticles. Polym. Chem. 2018, 9, 4566–4573, doi:10.1039/C8PY00829A.
26. Ekladious, I.; Colson, Y.L.; Grinstaff, M.W. Polymer–drug conjugate therapeutics: Advances, insights and prospects. Nat. Rev. Drug Discov. 2019, 18, 273–294, doi:10.1038/s41573-018-0005-0.
27. Girase, M.L.; Patil, P.G.; Ige, P.P. Polymer-drug conjugates as nanomedicine: A review. Int. J. Polym. Mater. Polym. Biomater. 2019, 1–25, doi:10.1080/00914037.2019.1655745.
28. Shirure, V.S.; George, S.C. Design considerations to minimize the impact of drug absorption in polymer-based organ-on-a-chip platforms. Lab Chip 2017, 17, 681–690, doi:10.1039/c6lc01401a.
29. Sun, Y.; Chen, D.; Pan, Y.; Qu, W.; Hao, H.; Wang, X.; Liu, Z.; Xie, S. Nanoparticles for antiparasitic drug delivery. Drug Deliv. 2019, 26, 1206–1221, doi:10.1080/10717544.2019.1692968.
30. Batalha, I.L.; Bernut, A.; Schiebler, M.; Ouberai, M.M.; Passemar, C.; Klapholz, C.; Kinna, S.; Michel, S.; Sader, K.; Castro-Hartmann, P.; et al. Polymeric nanobiotics as a novel treatment for mycobacterial infections. J. Control. Release 2019, 314, 116–124, doi:10.1016/j.jconrel.2019.10.009.
31. Reynolds, N.; Dearnley, M.; Hinton, T.M. Polymers in the Delivery of siRNA for the Treatment of Virus Infections. Top. Curr. Chem. 2017, 375, 38, doi:10.1007/s41061-017-0127-6.
32. Mena-Giraldo, P.; Pérez-Buitrago, S.; Londoño-Berrío, M.; Ortiz-Trujillo, I.C.; Hoyos-Palacio, L.M.; Orozco, J. Photosensitive nanocarriers for specific delivery of cargo into cells. Sci. Rep. 2020, 10, 2110, doi:10.1038/s41598-020-58865-z.
33. Trousil, J.; Syrova, Z.; Dal, N.-J.K.; Rak, D.; Konefal, R.; Pavlova, E.; Matejkova, J.; Cmarko, D.; Kubickova, P.; Pavlis, O.; et al. Rifampicin Nanoformulation Enhances Treatment of Tuberculosis in Zebrafish. Biomacromolecules 2019, 20, 1798–1815, doi:10.1021/acs.biomac.9b00214.
34. Tropical Diseases 3rd International Conference on Tropical and Infectious Diseases. Available online: https://tropicaldiseases.infectiousconferences.com/ (accessed on: 18 July 2020).
35. Briones, E.; Colino, C.I.; Lanao, J.M. Delivery systems to increase the selectivity of antibiotics in phagocytic cells. J. Control. Release 2008, 125, 210–227, doi:10.1016/j.jconrel.2007.10.027.
36. Zaioncz, S.; Khalil, N.M.; Mainardes, R.M. Exploring the Role of Nanoparticles in Amphotericin B Delivery. Curr. Pharm. Des. 2017, 23, 509–521, doi:10.2174/1381612822666161027103640.
37. Hakkimane, S.S.; Shenoy, V.P.; Gaonkar, S.L.; Bairy, I.; Guru, B.R. Antimycobacterial susceptibility evaluation of rifampicin and isoniazid benz-hydrazone in biodegradable polymeric nanoparticles against Mycobacterium tuberculosis H37Rv strain. Int. J. Nanomed. 2018, 13, 4303–4318, doi:10.2147/IJN.S163925.

38. Patra, J.K.; Das, G.; Fraceto, L.F.; Campos, E.V.R.; Rodriguez-Torres, M.D.P.; Acosta-Torres, L.S.; Diaz-Torres, L.A.; Grillo, R.; Swamy, M.K.; Sharma, S.; et al. Nano based drug delivery systems: Recent developments and future prospects. J. Nanobiotechnol.2018, 16, 71, doi:10.1186/s12951-018-0392-8.

39. Cunha-Azevedo, E.P.; Py-Daniel, K.R.; Siqueira-Moura, M.P.; Bocca, A.L.; Felipe, M.S.S.; Tedesco, A.C.; Pires Junior, O.R.; Lucci, C.M.; Azevedo, R.B. In vivo evaluation of the efficacy, toxicity and biodistribution of PLGA-DMSA nanoparticles loaded with itraconazole for treatment of paracoccidioidomycosis. J. Drug Deliv. Sci. Technol. 2018, 45, 135–141, doi:10.1016/j.jddst.2018.02.014.

40. World Health Organization Antibiotic Resistance. Available online: https://www.who.int/news-room/fact-sheets/detail/antibiotic-resistance (accessed on 29 April 2020).

41. Thakur, A.; Mikkelsen, H.; Jungersen, G. Intracellular pathogens: Host immunity and microbial persistence strategies. J. Immunol. Res. 2019, 2019, doi:10.1155/2019/1356540.

42. Maurin, M.; Raoult, D. Use of aminoglycosides in treatment of infections due to intracellular bacteria. Antimicrob. Agents Chemother. 2001, 45, 2977–2986.

43. Xie, S.; Tao, Y.; Pan, Y.; Qu, W.; Cheng, G.; Huang, L.; Chen, D.; Wang, X.; Liu, Z.; Yuan, Z. Biodegradable nanoparticles for intracellular delivery of antimicrobial agents. J. Control. Release 2014, 187, 101–117.

44. Ficai, A.; Grumezescu, A.M. Nanostructures for Antimicrobial Therapy; Elsevier: Amsterdam, The Netherlands, 2017; ISBN 0323461514.

45. Aderibigbe, B.A. Polymeric therapeutic delivery systems for the treatment of infectious diseases. Ther. Deliv. 2017, 8, 557–576.

46. Abed, N.; Couvreur, P. Nanocarriers for antibiotics: A promising solution to treat intracellular bacterial infections. Int. J. Antimicrob. Agents 2014, 43, 485–496.

47. Khalid, M.; El-Sawy, H.S. Polymeric nanoparticles: Promising platform for drug delivery. Int. J. Pharm. 2017, 528, 675–691.

48. Donahue, N.D.; Acar, H.; Wilhelm, S. Concepts of nanoparticle cellular uptake, intracellular trafficking, and kinetics in nanomedicine. Adv. Drug Deliv. Rev. 2019, 143, 68–96.

49. Kamaly, N.; He, J.C.; Ausiello, D.A.; Farokhzad, O.C. Nanomedicines for renal disease: Current status and future applications. Nat. Rev. Nephrol. 2016, 12, 738.

50. Devarajan, P. V.; Jain, S. Targeted Drug Delivery: Concepts and Design; Springer: Berlin/Heidelberg, Germany, 2016; ISBN 331937625X.

51. Lakkireddy, H.R.; Bazile, D. Building the design, translation and development principles of polymeric nanomedicines using the case of clinically advanced poly (lactide (glycolide))–poly

(ethylene glycol) nanotechnology as a model: An industrial viewpoint. Adv. Drug Deliv. Rev. 2016, 107, 289–332.

52. Zhang, Y.; Chan, H.F.; Leong, K.W. Advanced materials and processing for drug delivery: The past and the future. Adv. Drug Deliv. Rev. 2013, 65, 104–120.
53. Farokhzad, O.C.; Langer, R. Impact of Nanotechnology on Drug Delivery. ACS Nano 2009, 3, 16–20, doi:10.1021/nn900002m.
54. Amgoth, C.; Phan, C.; Banavoth, M.; Rompivalasa, S.; Tang, G. Polymer Properties: Functionalization and Surface Modified Nanoparticles. In Role of Novel Drug Delivery Vehicles in Nanobiomedicine; IntechOpen: London, UK, 2019.
55. Koo, O.M.; Rubinstein, I.; Onyuksel, H. Role of nanotechnology in targeted drug delivery and imaging: A concise review. Nanomed. Nanotechnol. Biol. Med. 2005, 1, 193–212.
56. Binnemars-Postma, K.; Storm, G.; Prakash, J. Nanomedicine strategies to target tumor-associated macrophages. Int. J. Mol. Sci. 2017, 18, 979.
57. Dinarvand, R.; Sepehri, N.; Manoochehri, S.; Rouhani, H.; Atyabi, F. Polylactide-co-glycolide nanoparticles for controlled delivery of anticancer agents. Int. J. Nanomed. 2011, 6, 877.
58. Canaparo, R.; Foglietta, F.; Giuntini, F.; Della Pepa, C.; Dosio, F.; Serpe, L. Recent developments in antibacterial therapy: Focus on stimuli-responsive drug-delivery systems and therapeutic nanoparticles. Molecules 2019, 24, 1991.
59. Suhail, M.; Rosenholm, J.M.; Minhas, M.U.; Badshah, S.F.; Naeem, A.; Khan, K.U.; Fahad, M. Nanogels as drug-delivery systems: A comprehensive overview. Ther. Deliv. 2019, 10, 697–717.
60. Li, D.; van Nostrum, C.F.; Mastrobattista, E.; Vermonden, T.; Hennink, W.E. Nanogels for intracellular delivery of biotherapeutics. J. Control. Release 2017, 259, 16–28.
61. Ye, Y.; Yu, J.; Gu, Z. Versatile protein nanogels prepared by in situ polymerization. Macromol. Chem. Phys. 2016, 217, 333–343.