The Thymoquinone Inhalable Nano-drug Delivery System with COVID-19 Therapeutic Potential

Chengwei Jiang

School of Pharmacy, University College London, London, UK
Email: chengwei.jiang@ucl.ac.uk

***Abstract*—The COVID-19 pandemic, stemming from a novel coronavirus outbreak in 2019, has resulted in significant loss of life and widespread disruption to societal and economic functions globally. Thymoquinone (TQ), a natural compound sourced from Nigella sativa, has garnered attention for its potential in mitigating and treating COVID-19. This paper provides an overview of inhaled nano-formulations designed for the localized pulmonary delivery of TQ. These formulations hold promise for enhancing the efficacy of TQ in combating respiratory infections, including COVID-19. However, challenges persist in translating TQ-loaded inhaled nano-formulations into clinical applications. Factors such as stability, bioavailability, and safety must be carefully addressed to ensure the feasibility and effectiveness of these formulations in clinical settings. This review highlights the current advancements and hurdles in utilizing TQ-loaded inhaled nano-formulations for combating COVID-19 and underscores the need for further research and development in this promising area.**

***Keywords*—COVID-19, nano-drug, delivery system, thymoquinone, SARS-CoV-2**

I. Introduction

A. Alternative Therapies for COVID-19

The coronavirus illness (COVID-19), outbreaking in the end of 2019, has sparked a worldwide epidemic. Clinical studies have tested a variety of therapeutic modalities, and new medication classes have also been created. The primary active ingredients in these medications were those with antiviral, anti-inflammatory, immunomodulatory, antioxidant, and other similar properties [1, 2]. Glucocorticoids protected tissue and organ damage by reducing cytokine storms, while Remdesivir, Lopinavir/ritonavir, and Chloroquine conducted anti-SARS-CoV-2 effects by altering the life cycle and the reproduction of the virus [3]. In addition, a few COVID-19 vaccinations are available and have been shown to be successful in controlling case expansion [4]. Additionally, it has been suggested that several substances produced from consumable natural products may have antiviral properties and the potential to become COVID-19 nutraceuticals [5]. These items could provide new opportunities to shield the entire world's population from the ongoing COVID-19 epidemic.

B. Natural Products with anti-SARS-CoV-2 Activity

In the past, pharmaceutical compounds made from natural products were proved to have anti-SARS effects. On SARS-CoV-2, a comparable inhibition was also detectable. According to Khan *et al.* [6], 10 natural substances, including spermidine, resveratrol, phytoestrogen, trehalose, baicalin, curcumin, quercetin, coumarin, and epigallocatechin 3-gallate (EGCG), are employed in treating COVID-19. Such substances primarily interfere with the infection and multiplication of SARS-CoV-2 by inhibiting endo-lysosomes and mTOR signaling pathways. Additionally, newer in vitro and in vivo research have shown that additional natural substances have anti-SARS-CoV-2 effects (Table I). Natural products have been identified as a unique approach to COVID-19 treatment and prevention. One of the therapeutic plants with a long history of usage in folklore traditional medicine, even before the arrival of modern medicine, was Nigella sativa (NS), or black cumin seed (Fig. 1) [7]. NS belongs to the Ranunculaceae family and is native to the Mediterranean area and western Asia, including India, Pakistan, and Afghanistan [8].

C. Thymoquinone

According to reports, the primary bioactive component extracted from NS is Thymoquinone (TQ). Fig. 2 shows that TQ (2-isopropyl-5-methylbenzo-1,4-quinone) makes for around 30–48% of the total ingredient recovered from black seeds [8].

Studies conducted in vitro and in vivo have documented and confirmed the numerous bioactivities of TQ. They displayed strong biological effects against oxidative [9], anti-inflammatory [10], chemoprotective, and chemo-curative [11]. Furthermore, Seadawy *et al.* [12] reported that TQ had a similar binding ability to the active site of SARS-CoV-2 M^{pro}, compared with standard compounds. This indicates that TQ may be able to combat SARS-CoV-2 by preventing the major viral protease (M^{pro}) from working. Thus, TQ may provide essential additional assistance when treating COVID-19 under circumstances when core basic requirements are unknown.

The absorption and disposal of TQ have, however, only been the subject of a few research that have been published in the past. Its strong hydrophobicity, limited solubility,

Manuscript received December 3, 2023; revised February 1, 2024; accepted March 13, 2024; published August 20, 2024.

and stability in biological fluids may be to blame for this [13]. An effective solution to TQ's poor biopharmaceutical qualities in the oral administration method may be a pulmonary drug delivery system based on nanotechnology. In this paper, we summarised the potential nanocarriers that might be used to deliver TQ to the lungs, potentially resolving the issue of poor solubility and bioavailability in the oral delivery system, and we also showed the advantages of TQ-loaded nanoparticles for the prevention and treatment of COVID-19.

TABLE I. ANTI-COVID 19 EFFECTS OF NATURAL PRODUCTS

Plant	Compound	EC50 value	SI	IC50	Reported anti SARS and SARS-CoV-2 method	Reference
Nigella sativa	Thymoquinone	No data	No data	3.9 μmol/L	Inhibits SARS-CoV-2 Mpro	[12]
Thymus vulgaris	Thymol	No data	No data	617 μmol/L	Inhibits SARS-CoV-2 Mpro	
Origanum bulgare	Carvacrol	No data	No data	464 μmol/L	Inhibits SARS-CoV-2 Mpro	
Citrus	Hesperidine	No data	No data	5.58 μmol/L	Inhibits SARS-CoV-2 Mpro	
Curcuma longa	Curcumin	7.9 μg/mL	NOT Calculable	No data	Inhibits SARS-CoV-2 entry	[14]
Scutellaria baicalensis	Baicalin	10.27 μmol/L	>19	No data	Inhibits SARS-CoV-2 3CLpro	[15]
	Baicalein	1.69 μmol/L	>118	No data		
Various organism	Trehalose	No data	No data	No data	Inhibits cellular organelles associated with SARS-CoV-2 replication	[16]
Wheat germ	Spermidine and Spermine	No data	No data	149 μmol/L	Induces autophagy infected by SARS-CoV-2 and inhibits mTOR signaling pathway	[17]
Citrus sinensis	Naringenin	No data	No data	No data	Inhibits SARS-CoV-2 Mpro	[18]
Grape	Resveratrol	66 μmol/L	NOT Calculable	No data	Inhibits SARS-CoV-2 replication	[19]
Blueberry	Pterostilbene	19 μmol/L	NOT Calculable	No data	Inhibits SARS-CoV-2 replication	[19]
Quercetum	Quercetin	No data	No data	42.79 μmol/L	Inhibits SARS-CoV 3CLpro	[20]
		10.6 μmol/L	NOT Calculable	No data	Tetra-O-galloyl-B-D-glucose (TGG) and luteolin, block the entry of SARS-CoV into host cells	[21]
		No data	No data	No data	Potential target against SARS-CoV-2	[22]
Licorice root	Glycyrrhizin	No data	No data	No data	Binds to ACE2 and block it and inhibit the entry of SARS-CoV-2 into the cells	[23]
		0.44 mg/mL	NOT Calculable	No data	Inhibits SARS-CoV-2 Mpro	[24]
Panax ginseng	Ginsenoside-Rb1	No data	No data	100 μmol/L	Inhibits SARS-CoV 3CLpro	[25]
Stephania japonica	Cepharanthine	0.98 μmol/L	NOT Calculable	No data	Inhibits SARS-CoV 2 GX_P2V infection	[26]
Black tea	Theaflavin	No data	No data	8.44 μg/mL	Inhibits SARS-CoV-2 3CLpro	[27]
Green tea	Epigallocatechin-3-gallate	No data	No data	7.58 μg/mL	Inhibits SARS-CoV-2 3CLpro	[27]

Fig. 1. Nigella Sativa (NS) (black cumin seed).

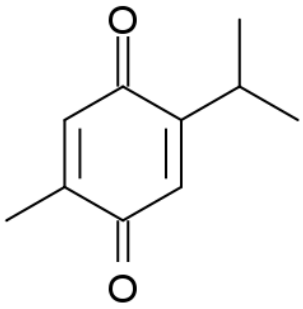

Fig. 2. Thymoquinone.

II. RESULTS AND DISCUSSION

A. Thymoquinone Nanoparticles

Due to its hydrophobicity and poor solubility, TQ's limited systemic bioavailability may restrict its usage in clinics' oral administration systems, necessitating greater dosages or concentrations to be effective [28]. Additionally, due to its limited chemical stability and rapid first-pass metabolism, it degrades and is eliminated from the physiological environment [29]. To get over these issues, several nanocarriers have been used to the pulmonary administration of natural compounds. Nano-drug delivery via the pulmonary route is particularly notable for its wide surface area, high vascularization, and weak blood-alveolar barrier, as well as its local targeting, avoidance of first-pass hepatic metabolism, and quick beginning of action [30].

B. Polymeric Inhaled Nanoparticles Loaded TQ

The FDA has authorised the use of poly(lactic-co-glycolic acid) (PLGA), a biocompatible and noncytotoxic polymer, as a carrier and excipient in drug delivery to produce sustained release of the medicine. Furthermore, the problems of poor transport across mucosal barriers and the negative surface potential of the negatively charged PLGA nanoparticles have been

overcome by blending PLGA nanoparticle formulations with alginate, chitosan, pectin, and poly (propylene fumarate), Polyvinyl Alcohol (PVA) and poly(orthoester). Tomoda *et al.* [31] stated that nanoparticles-in-microparticles dry powder may be administered to deep lungs and conducted activity based on the development of polymeric nanotechnology. This PLGA nanoparticle was created and spray-dried with trehalose as excipients, with an average size diameter of around 200 nm. Significantly, rat inhalation of spray-dried drug-loaded PLGA nanoparticles showed greater drug retention in the lung (about 13 times higher) than that of medicines administered by intravenous injection. The nanoprecipitation method involved adding an acetone-dissolved polymer, with or without an emulsifier or stabiliser, dropwise into the continuously rotating aqueous phase, followed by a vacuum to evaporate the organic solvent. Similar formulations have been developed on the TQ using both of these methods. The emulsification solvent evaporation technique, in contrast, included adding polymers that had been dissolved in volatile organic solvent to the constantly spinning aqueous phase, either with or without the addition of an emulsifier, and sonicating them [32]. Because it was simpler to scale up and produced high entrapment effectiveness, the nanoprecipitation approach was the most popular method for creating PLGA nanoparticles [32]. According to Ganea *et al.* [33], they created the TQ-loaded PLGA nanoparticles by evaporating the solvent from an emulsion and using anionic molecular micelles as emulsifiers. All of the molecular micelles they looked at had excellent emulsifying properties, which led to the highest possible TQ entrapment efficiency and monodispersed particle sizes under 200 nm. Dialysis was used to examine the release of TQ from molecularly modified nanoparticles, and the levels were lower than those of the free drug. As well, thymoquinone-loaded PLGA-PVA nanoparticles have been developed by Saghir *et al.* and they assessed TQ-PLGA-PVA nanoparticles' potential impact on pulmonary fibrosis. The outcome shown that TQ-PLGA-PVA nanoparticles might reduce pulmonary fibrosis brought on by bleomycin by reducing lung inflammation and bleomycin-induced oxidative stress.

A nontoxic, FDA-approved polymer with water solubility, Polyethylene Glycol (PEG) has been used extensively to encapsulate various medications, and the majority of the polymeric nanoparticles made using PEG polymers have entered clinical trials. PEG became a strong contender for the encapsulation of TQ as a result of this. Bhattacharya *et al.* [34] prepared the PEG200-TQ nanoparticles and PEG4000-TQ nanoparticles, and they found that the particle size was lower than 50 nm (PEG200-TQ-NPs, 38 ± 4.9 nm; PEG4000-TQ-NPs, 16 ± 3.2 nm) and their encapsulation efficiency were 97.5% and 90% respectively. It means that PEG has potential as a nanocarrier for TQ. PEG has also been employed as a new formulation in medication delivery systems for the lungs. For instance, PEG was used as a carrier in a mouse model to carry the drug Paclitaxel (PTX) to the lungs, and the results revealed that PEG-PTX improved the drug's effectiveness and decreased pulmonary toxicity [35]. PEGylation suggested a viable delivery method for inhalation, according to their study.

A biodegradable Polyester Called Polycaprolactone (PCL) has ester bonds that may break down under physiological circumstances [32]. The FDA has approved PCL as a biomaterial that may be utilised for medication delivery and tissue engineering. Some other polymer materials used in the delivery of Thymoquinone could also be found such as PHA-mPEG, PHV-mPEG, β-cyclodextrin [36], Ethylcellulose (EC) and Polycaprolactone (PCL) [37]. They all have the potential to be used in the pulmonary drug delivery system.

C. Chitosan-based TQ Nanoparticle

Chitosan (CS) was a good natural polymer material and showed good biocompatible propertiesComplexes of CS nanoparticles exhibited a variety of properties, including tiny size and selective adsorption. The majority of drugs delivered by CS nanoparticles have extended blood circulation times, regulated or sustained drug release, and lower toxicity to healthy tissues [38]. Thymoquinone loaded chitosan-lecithin micelles have been prepared [13] and they reported that they were able to create stable polymeric micelles of thymoquinone with excellent entrapment efficiency by employing the self-assembly approach (98.77%). Additionally, this nanocarrier may regulate the TQ release rate. As an inhaler formulation, this nanocarrier has also been employed effectively. According to Wang *et al.* [39], they created a specific drug-loaded chitosan nanoparticles dry powder inhaler formulation and discovered that it was capable of both delivering the medication to the lung and controlling its release. Similar to this, Debnath *et al.* [40] created a dry powder inhalation formulation based on chitosan nanoparticles. According to their findings, the aerodynamic particle size was 1.76 μm, indicating that it could be administered to the lungs effectively.

D. Lipid-based TQ Nanoparticles

Lipid-based nanoparticles were created as the first nano-drug delivery method by combining surfactants, oil, and water [41]. These particles may serve a number of purposes, including enhancing medication bioavailability, stability, and penetrating power [42]. Additionally, the drug release could be sustained by lipidic nanocarriers, which helped to reduce the peak drug concentration in systemic circulation and prevent adverse effects [43]. Targeting certain cells or organs might potentially be done using the drug-loaded lipid-based nanocarriers [36]. Several lipid-based nanoparticles loaded TQ reported have been summarized below.

1) TQ-loaded Nanostructured Lipid Carriers (NLCs)

The lipid matrix of NLC was made up of a mixture of liquid and solid lipids (oils). TQ-loaded NLCs helped to improve drug bioavailability and pharmacokinetics, regulate drug release, shield TQ from intense first-pass metabolism, and prevent P-gp efflux of the drug transporters [44]. There have been several reports of NLCs with thymoquinone loads. Based on the Compritol 888

ATO Pluronic F127 system, certain thymoquinone-loaded NLCs were created by Elmowafy *et al.* [45]. They claimed that these TQ-loaded NLCs had higher encapsulation efficiency (EE%), lying between 84.6 ± 5% and 96.2 ± 1.6%. besides, the bioavailability of TQ-loaded NLCs was 2.03- and 3.97-fold higher than free thymoquinone. Similarly, Abdelwanhb *et al.* prepared TQ-NLCs and evaluated the particle size, zeta potential, and *in vitro* toxicity. They found that the particle size was about 75 nm with negative zeta potential (−31 ± 0.1 mV) and they claimed the pharmacokinetic profile of TQ-NLCs in rats and rabbits [46]. In addition, Garbuzenko *et al.* [47] also developed a multifunctional inhalation NLC-based delivery system. The *In vitro* and *in vivo* results proved that its delivery system could increase the efficiency of treating lung disease.

2) TQ-loaded Solid Lipid Nanoparticles (SLNs)

SLNs were initially reported in the 1990s. The nanoparticles utilised for controlled drug release at the time were created by combining one or more solid lipids [48]. Solid Lipid Nanoparticles (SLNs), which might be utilised to safeguard drugs from chemical deterioration and regulate drug release, were formerly a revolutionary colloidal drug delivery technology. Additionally, this carrier was physically stable, well-tolerated, biodegradable, and effective in encapsulating lipophilic medications within its lipid framework [49]. Many lipophilic compounds, including thymoquinone, had the potential to be delivered as drugs through SLNs.

Some scientists used the solvent injection method to prepare TQ-SLNs. In a pharmacokinetic study, researchers found that the bioavailability of TQ-SLNs was 5-fold stronger than that of the pure TQ suspension [50]. Besides, solid lipid nanoparticles have been evaluated in the pulmonary drug delivery system. Wang *et al.* [38] used Aggregation-Caused Quenching (ACQ) probe, P4, to clarify the effect of particle size on SLN cellular uptake in cells line A549 and RAW 264.7. This research supported the development of SLN as a nanocarrier employed in inhaled pulmonary medication delivery systems in addition to reporting the mechanisms between particle size and cell uptake behaviours. According to the study, greater particle size SLN produced stronger fluorescence intensity and thus increased cellular absorption in the range of 120 to 480 nm. Finally, they asserted that following internalisation, they were mostly found in the endosomes. For the construction of thymoquinone-loaded SLN with acceptable particle size employed for pulmonary drug administration, this study offered an opportunity and important information.

3) Liposomes

One of the effective nano-drug delivery technologies, liposomes have been employed extensively and received FDA marketing approval. It was simple to make liposomes. They were self-assembled carriers with a hydrophilic core and an exterior coating of hydrophobic lipids [51]. A wide variety of liposome particle sizes between 50 nm and 1000 nm might be created depending on the phospholipid and cholesterol molecules employed. Because they include phospholipid layers, liposomes are a perfect vehicle for encapsulating naturally occurring substances with low solubility, including thymoquinone.

By using a thin-film hydration approach, Fahmy [52] created DPPC TQ-loaded liposomes and assessed their entrapment efficiency (99%) and particle size (722.9 ± 12 nm). In addition, he noted that after 72 hours of incubation with lung cancer cell line A549, TQ-loaded liposomes led to dose-dependent cell death. The half-maximal effective concentration of TQ-loaded liposome was 335 μg/ml. This means that liposome was a good carrier for thymoquinone, and it showed the potential in the treatment of lung cancer. The knowledge gained from these experiments will be helpful in creating the inhaled TQ-loaded liposome that will treat lung diseases.

III. Conclusion

The symptoms of COVID-19 are now a major public health concern, and many scientists are working to create efficient treatments and medications. This article highlighted various well-known thymoquinone-loaded nanotechnologies that have been created as a new inhalation delivery method. These provide a fresh opportunity to cure COVID-19. Thymoquinone-loaded nanoparticles give thymoquinone additional beneficial properties by boosting its solubility, stability, and bioavailability. Inhaled administration provides additional benefits, such as lowering hepatic metabolism, especially for pulmonary diseases like COVID-19, and local inhalation delivery may be achieved. However, the majority of research largely focused on oral delivery and system distribution. This paper provided some useful information to develop a TQ-loaded inhaled nano-formulation targeting SARS-CoV-2.

Conflict of Interest

The author declares no conflict of interest.

References

[1] C. Stasi, S. Fallani, F. Voller, *et al.*, "Treatment for COVID-19: An overview," *European Journal of Pharmacology*, 173644, 2020.

[2] S. S. Jean and P. R. Hsueh, "Old and re-purposed drugs for the treatment of COVID-19," *Expert Review of Anti-infective Therapy*, vol. 18, no. 9, pp. 843–847, 2020.

[3] R. Wu, L. Wang, H. C. D. Kuo, *et al.*, "An update on current therapeutic drugs treating COVID-19," *Current Pharmacology Reports*, vol. 6, no. 3, pp. 56–70, 2020.

[4] S. J. Thomas, E. D. Moreira Jr, N. Kitchin, *et al.*, "Safety and efficacy of the BNT162b2 mRNA COVID-19 vaccine through 6 months," *New England Journal of Medicine*, 2021.

[5] B. Patel, S. Sharma, N. Nair, *et al.*, "Therapeutic opportunities of edible antiviral plants for COVID-19," *Molecular and Cellular Biochemistry*, pp. 1–20, 2021.

[6] N. Khan, X. Chen, and J. D. Geiger, "Possible therapeutic use of natural compounds against COVID-19," *Journal of Cellular Signaling*, vol. 2, no. 1, p. 63, 2021.

[7] S. Dabeer, M. A. Rather, S. Rasool, *et al.*, "History and traditional uses of black seeds (Nigella sativa)," in *Black Seeds (Nigella Sativa)*, Elsevier, 2022, pp. 1–28.

[8] P. Negi, C. Rathore, G. Sharma, *et al.*, "Thymoquinone a potential therapeutic molecule from the plant Nigella sativa: Role of colloidal carriers in its effective delivery," *Recent Patents on Drug Delivery & Formulation*, vol. 12, no. 1, pp. 3–22, 2018.

[9] K. Staniek and L. Gille, "November. Is thymoquinone an antioxidant?" in *BMC Pharmacology*, BioMed Central, vol. 10, no. 1, p. 1, 2010.

[10] M. E. Gazzar, R. E. Mezayen, J. C. Marecki, *et al.*, "Anti-inflammatory effect of thymoquinone in a mouse model of allergic lung inflammation," *International Immunopharmacology*, vol. 6, no. 7, pp. 1135–1142, 2006.

[11] A. S. Haron, S. S. S. Alwi, L. S. Yazan, *et al.*, "Cytotoxic effect of Thymoquinone-loaded Nanostructured Lipid Carrier (TQ-NLC) on liver cancer cell integrated with hepatitis B genome, Hep3B," *Evidence-Based Complementary and Alternative Medicine*, 2018.

[12] M. G. Seadawy, A. F. Gad, M. Shamel, *et al.*, "In vitro: Natural compounds (Thymol, Carvacrol, Hesperidine, and Thymoquinone) against Sars-Cov2 strain isolated from Egyptian patients," *Biomedical Journal of Scientific & Technical Research*, vol. 34, no. 3, pp. 26750–26757, 2020.

[13] P. Negi, G. Sharma, C. Verma, *et al.*, "Novel thymoquinone loaded chitosan-lecithin micelles for effective wound healing: Development, characterization, and preclinical evaluation," *Carbohydrate Polymers*, vol. 230, 115659, 2020.

[14] M. Bormann, M. Alt, L. Schipper, *et al.*, "Turmeric root and its bioactive ingredient curcumin effectively neutralize SARS-CoV-2 in vitro," *Viruses*, vol. 13, no. 10, 1914, 2021.

[15] H. Su, S. Yao, W. Zhao, *et al.*, "Discovery of baicalin and baicalein as novel, natural product inhibitors of SARS-CoV-2 3CL protease in vitro," BioRxiv, 2020.

[16] D. Martinon, V. F. Borges, A. C. Gomez, *et al.*, "Potential fast COVID-19 containment with trehalose," *Frontiers in Immunology*, vol. 11, 1623, 2020.

[17] N. C. Gassen, J. Papies, T. Bajaj, *et al.*, "Analysis of SARS-CoV-2-controlled autophagy reveals spermidine, MK-2206, and niclosamide as putative antiviral therapeutics," BioRxiv, 2020.

[18] S. Khaerunnisa, H. Kurniawan, R. Awaluddin, *et al.*, "Potential inhibitor of COVID-19 main protease (Mpro) from several medicinal plant compounds by molecular docking study," preprints, 2020030226, 2020.

[19] B. M. T. Ellen, N. D. Kumar, E. M. Bouma, *et al.*, "Resveratrol and pterostilbene potently inhibit SARS-CoV-2 replication in vitro," BioRxiv, 2020-09, 2021.

[20] L. Chen, J. Li, C. Luo, *et al.*, "Binding interaction of quercetin-3-β-galactoside and its synthetic derivatives with SARS-CoV 3CLpro: Structure-activity relationship studies reveal salient pharmacophore features," *Bioorganic & Medicinal Chemistry*, vol. 14, no. 24, pp. 8295–8306, 2006.

[21] L. Yi, Z. Li, K. Yuan, *et al.*, "Small molecules blocking the entry of severe acute respiratory syndrome coronavirus into host cells," *Journal of Virology*, vol. 78, no. 20, pp. 11334–11339, 2004.

[22] P. K. Agrawal, C. Agrawal, and G. Blunden, "Quercetin: Antiviral significance and possible COVID-19 integrative considerations," *Natural Product Communications*, vol. 15, no. 12, 1934578X20976293, 2020.

[23] Y. Wan, J. Shang, R. Graham, *et al.*, "Receptor recognition by the novel coronavirus from Wuhan: An analysis based on decade-long structural studies of SARS coronavirus," *Journal of Virology*, vol. 94, no. 7, e00127-20, 2020.

[24] L. V. D. Sand, M. Bormann, M. Alt, *et al.*, "Glycyrrhizin effectively neutralizes SARS-CoV-2 in vitro by inhibiting the viral main protease," BioRxiv, 2020.

[25] C. Y. Wu, J. T. Jan, S. H. Ma, *et al.*, "Small molecules targeting severe acute respiratory syndrome human coronavirus," *Proceedings of the National Academy of Sciences*, vol. 101, no. 27, pp. 10012–10017, 2004.

[26] H. H. Fan, L. Q. Wang, W. L. Liu, *et al.*, "Repurposing of clinically approved drugs for treatment of coronavirus disease 2019 in a 2019-novel coronavirus-related coronavirus model," *Chinese Medical Journal*, vol. 133, no. 9, 1051, 2020.

[27] M. Jang, Y. I. Park, Y. E. Cha, *et al.*, "Tea polyphenols EGCG and theaflavin inhibit the activity of SARS-CoV-2 3CL-protease in vitro," *Evidence-Based Complementary and Alternative Medicine*, 2020.

[28] M. A. Khan, Y. H. Aldebasi, S. A. Alsuhaibani, *et al.*, "Therapeutic potential of thymoquinone liposomes against the systemic infection of Candida albicans in diabetic mice," *PLoS One*, vol. 13, no. 12, e0208951, 2018.

[29] S. Alam, Z. I. Khan, G. Mustafa, *et al.*, "Development and evaluation of thymoquinone-encapsulated chitosan nanoparticles for nose-to-brain targeting: A pharmacoscintigraphic study," *International Journal of Nanomedicine*, vol. 7, 5705, 2012.

[30] Z. Liang, R. Ni, J. Zhou, *et al.*, "Recent advances in controlled pulmonary drug delivery," *Drug Discovery Today*, vol. 20, no. 3, pp. 380–389, 2015.

[31] K. Tomoda, T. Ohkoshi, K. Hirota, *et al.*, "Preparation and properties of inhalable nanocomposite particles for treatment of lung cancer," *Colloids and Surfaces B: Biointerfaces*, vol. 71, no. 2, pp. 177–182, 2009.

[32] S. Kumari, M. G. Swetha, and S. Mayor, "Endocytosis unplugged: Multiple ways to enter the cell," *Cell Research*, vol. 20, no. 3, pp. 256–275, 2010.

[33] G. M. Ganea, S. O. Fakayode, J. N. Losso, *et al.*, "Delivery of phytochemical thymoquinone using molecular micelle modified poly (D, L lactide-co-glycolide) (PLGA) nanoparticles," *Nanotechnology*, vol. 21, no. 28, 285104, 2010.

[34] S. Bhattacharya, M. Ahir, M. Patra, *et al.*, "PEGylated-thymoquinone-nanoparticle mediated retardation of breast cancer cell migration by deregulation of cytoskeletal actin polymerization through miR-34a," *Biomaterials*, vol. 51, pp. 91–107, 2015.

[35] T. Luo, C. Loira-Pastoriza, H. P. Patil, *et al.*, "PEGylation of paclitaxel largely improves its safety and anti-tumor efficacy following pulmonary delivery in a mouse model of lung carcinoma," *Journal of Controlled Release*, vol. 239, pp. 62–71, 2016.

[36] M. Teymouri, M. Mashreghi, E. Saburi, *et al.*, "The trip of a drug inside the body: From a lipid-based nanocarrier to a target cell," *Journal of Controlled Release*, vol. 309, pp. 59–71, 2019.

[37] M. A. Mona and A. Mottaleb, "Biodegradable thymoquinone nanoparticles for higher therapeutic efficiency in murine colorectal cancer," *Ijppr. Human*, vol. 7, pp. 436–450, 2016.

[38] W. Wang, B. Yang, Z. Huang, *et al.*, "Investigating the effect of particle size on cellular uptake by aggregation-caused quenching probe–encapsulating solid lipid nanoparticles, Inhaled," *Journal of Pharmaceutical Innovation*, pp. 1–7, 2021.

[39] H. Wang, J. Holgate, S. Bartlett, *et al.*, "Assessment of nicotine release from nicotine-loaded chitosan nanoparticles dry powder inhaler formulations via locomotor activity of C57BL/6 mice," *European Journal of Pharmaceutics and Biopharmaceutics*, vol. 154, pp. 175–185, 2020.

[40] S. K. Debnath, S. Saisivam, M. Debanth, *et al.*, "Development and evaluation of Chitosan nanoparticles based dry powder inhalation formulations of Prothionamide," *PloS One*, vol. 13, no. 1, e0190976, 2018.

[41] L. Sercombe, T. Veerati, F. Moheimani, *et al.*, "Advances and challenges of liposome assisted drug delivery," *Frontiers in Pharmacology*, vol. 6, p. 286, 2015.

[42] G. Bozzuto and A. Molinari, "Liposomes as nanomedical devices," *International Journal of Nanomedicine*, vol. 10, p. 975, 2015.

[43] S. B. Lim, A. Banerjee, and H. Önyüksel, "Improvement of drug safety by the use of lipid-based nanocarriers," *Journal of Controlled Release*, vol. 163, no. 1, pp. 34–45, 2012.

[44] S. Roohinejad, R. Greiner, I. Oey, *et al.*, *Emulsion-Based Systems for Delivery of Food Active Compounds: Formation, Application, Health and Safety*, John Wiley & Sons, 2018.

[45] M. Elmowafy, A. Samy, M. A. Raslan, *et al.*, "Enhancement of bioavailability and pharmacodynamic effects of thymoquinone via Nanostructured Lipid Carrier (NLC) formulation," *Aaps Pharmscitech*, vol. 17, no. 3, pp. 663–672, 2016.

[46] S. Makled, N. Boraie, and N. Nafee, "Nanoparticle-mediated macrophage targeting—A new inhalation therapy tackling tuberculosis," *Drug Delivery and Translational Research*, vol. 11, no. 3, pp. 1037–1055, 2021.

[47] O. B. Garbuzenko, A. Kuzmov, O. Taratula, *et al.*, "Strategy to enhance lung cancer treatment by five essential elements: inhalation delivery, nanotechnology, tumor-receptor targeting, chemo-and gene therapy," *Theranostics*, vol. 9, no. 26, 8362, 2019.

[48] C. Rathore, M. J. Rathbone, D. K. Chellappan, *et al.*, "Nanocarriers: More than tour de force for thymoquinone," *Expert Opinion on Drug Delivery*, vol. 17, no. 4, pp. 479–494, 2020.

[49] V. Mishra, K. K. Bansal, A. Verma, *et al.*, "Solid lipid nanoparticles: Emerging colloidal nano drug delivery systems," *Pharmaceutics*, vol. 10, no. 4, p. 191, 2018.

[50] S. Ramachandran and S. Thangarajan, "A novel therapeutic application of solid lipid nanoparticles encapsulated thymoquinone (TQ-SLNs) on 3-nitroproponic acid induced Huntington's disease-like symptoms in Wistar rats," *Chemico-Biological Interactions*, vol. 256, pp. 25–36, 2016.

[51] W. H. Lee, C. Y. Loo, D. Traini, *et al.*, "Inhalation of nanoparticle-based drug for lung cancer treatment: advantages and challenges," *Asian Journal of Pharmaceutical Sciences*, vol. 10, no. 6, pp. 481–489, 2015.

[52] H. M. Fahmy, "In vitro study of the cytotoxicity of thymoquinone/ curcumin fluorescent liposomes," *Naunyn-Schmiedeberg's Archives of Pharmacology*, vol. 392, no. 11, pp. 1465–1476, 2019.