UTU Publishing Group

Bulletin of Biomedical Science

Journal Homepage: www.utu.com.sg

REVIEW

Application of Lipid-based Nanoparticles in the Diagnosis and Treatment of Hepatocellular Carcinoma

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ARTICLE INFO

Received 28 December 2023
Accepted 26 January 2024
Online 16 February 2024

KEY WORDS:
Hepatocellular carcinoma;
Lipid-based nanoparticles;
Liposomes;
Solid lipid nanoparticles;
Nanostructured lipid carriers

Abstract

Hepatocellular carcinoma (HCC) is one of the diseases with high morbidity and mortality in the world. Due to the difficulty of early diagnosis, it is often found to have reached the middle and late stages, and there are shortcomings in the treatment, so the diagnosis and treatment of HCC need new ideas and methods. In recent years, nanoparticles have emerged in the diagnosis and treatment of HCC, in which lipid-based nanoparticles have advantages that other types of nanoparticles could not surpass. It has many types, such as liposomes, solid lipid nanoparticles and nanostructured lipid carriers. Because of its superior biocompatibility, low toxicity and high bioavailability, it provides a new idea and means for the diagnosis and treatment of HCC. This review introduces several common types of lipid-based nanoparticles and summarizes their applications in the diagnosis and treatment of HCC.

Introduction

The global incidence of liver cancer ranks fifth, and primary liver cancer is the sixth most commonly diagnosed cancer in the world in 2020, and also the third cause of cancer-related death in the world (Sung et al., 2021). Currently, the highest incidence of primary liver cancer in the world is in Asia and Africa (McGlynn et al., 2021). Studies have shown that the incidence of liver cancer in men is higher than that in women (Chidambaranathan-Reghupaty et al., 2021). Primary liver cancer is mainly divided into two types, one is hepatocellular carcinoma (HCC), which originates from the main parenchymal cells of the liver, and the other is intrahepatic cholangiocarcinoma (ICC), which originates from the bile duct

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(Chidambaranathan-Reghupaty et al., 2021). HCC accounts for about 80-90% of all cases of primary liver cancer and is the most common type of primary liver cancer (Sohn et al., 2021). At present, the most important exogenous risk factors for primary liver cancer are HBV and HCV infection, in addition to type 2 diabetes mellitus, obesity and non-alcoholic fatty liver disease (McGlynn et al., 2021). Smoking and ingestion of aflatoxin-contaminated food are also exogenous risk factors (Chuang et al., 2009).

The early symptoms of HCC are not obvious and difficult to detect, and the diagnosis often reaches the late stage of HCC, which is difficult to cure completely. At present, the conventional treatments for HCC include surgery, transcatheter arterial chemoembolization (TACE), ablation, immunotherapy and so on, but due to drug resistance and recurrence of the disease, all kinds of treatments have shortcomings. Treatment outcomes need to be further improved. Therefore, new and more effective methods for early diagnosis and treatment of HCC are needed. Nanoparticles, as a new field, have come into people's vision in recent years, and play a role in cancer diagnosis and treatment. Nanoparticles are defined as particles with a size between 1-100 nm in one dimension (Najahi-Missaoui et al., 2020). Nanoparticles can be administered orally, inhaled, intravenously, and through skin penetration. Nanoparticles are usually composed of three layers: the core, the shell and the surface. The core is the main material of the nanoparticles; the shell is the middle layer; and the surface is the outermost layer, which can be functionalized with other particles (Khan et al., 2019). Nanoparticles include polymer-based nanoparticles, solid nanoparticles, carbon-based nanoparticles and lipid-based nanoparticles (Najahi-Missaoui et al., 2020). Nanoparticles are able to improve the solubility and stability of encapsulated drugs, improving safety and drug effectiveness (Kou et al., 2018).

Among them, the most common group of nanomedicines approved by the Food and Drug Administration (FDA) are lipid-based nanoparticles (Anselmo and Mitragotri, 2019). Lipid-based nanoparticles have a variety of advantages, such as biocompatibility, high bioavailability, large payload capacity and simple formulation, as well as a series of physical and chemical properties to regulate their biological properties (Sercombe et al., 2015). The nanoparticles also have the lowest systemic toxicity compared to other nanodelivery systems (Puri et al., 2009). And it can maintain high solubility in the aqueous phase compared to polymer nanoparticles and inorganic nanoparticles (Mitchell et al., 2021). The material mentioned above can either bind to endogenous lipoproteins that are taken up after binding to specific cellular receptors, or interact with lipids in the cell membrane to enhance cellular uptake, contributing to endosomal escape (Zelphati and Szoka, 1996; Akinc et al., 2010; Schroeder et al., 2010). This review systematically describes several types of lipid-based nanoparticles and their applications in the diagnosis and treatment of HCC, providing a more effective new idea for the current diagnosis and treatment of HCC.

Diagnosis and Treatment of HCC and Its Deficiencies

1. Diagnosis of HCC and Its Shortcoming

Current common diagnostic techniques, such as imaging and histological techniques, are not effective enough in the early diagnosis of HCC (Gao et al., 2018). In recent years, the poor therapeutic effect of HCC is related to the difficulties in early diagnosis of HCC, so more effective diagnostic methods for HCC are very important. The early diagnosis of tumors is convenient for patients and doctors to seek more suitable and effective treatment methods to improve the therapeutic effect, alleviate the pain of patients and prolong patients' the survival time. At present, the early diagnosis of HCC is mostly based on imaging examination and serological examination (Luo et al., 2020). Computed tomography (CT) and magnetic resonance imaging (MRI) have made significant progress in recent years, greatly improving the diagnosis of HCC, but they are too expensive to be used for extensive screening (Villanueva et al., 2010). At present, the main imaging screening method is ultrasound, which is non-invasive and non-radiation, but it is not easy to distinguish benign and malignant nodules in small cirrhosis, and the diagnostic results are affected by the operator's level of operation (Li et al.,2015).

Common serological protein biomarkers include alpha-fetoprotein (AFP), cancer embryonic antigen (CEA) and Golgi protein 73 (GP73) (Gao et al., 2018). In serological testing, the most widely used HCC biomarker worldwide is AFP, which was first introduced as an HCC serological marker in the 1960s (Johnson et al., 1999). However, about 30% of patients with early HCC cannot be detected by AFP, and serum AFP remains normal in 15-30% of patients with advanced HCC (Luo et al., 2020). Serum AFP is also seen to be elevated in other conditions, such as acute and chronic hepatitis, intrahepatic cholangiocarcinoma, and embryonal tumors, leading to a decrease in specificity (Piñero et al., 2020). AFP is no longer recommended by the American Association for the Study of Liver Diseases for the early diagnosis of HCC (Luo et al., 2020). Other tumor markers such as osteopontin (OPN), vascular endothelial growth factor (VEGF) and angiopoietin-2 (ANG-2) have been proposed in recent years, but their accuracy is still uncertain (Piñero et al., 2020). In addition, the diagnostic accuracy of AFP combined with abnormal prothrombin was improved compared with that of AFP alone (Ji

et al., 2016). However, there are others who believe that when these biomarkers are used in combination, their specificity is reduced (Durazo et al.,2008).

In addition, liquid biopsy is also a promising detection tool, which refers to all non-solid biomaterials used in HCC diagnosis and monitoring, such as circulating tumor cells (CTCs), circulating RNA, circulating tumor DNA (ctDNA), and extracellular vesicles (EVs) (Maravelia et al., 2021). However, it also has some limitations, such as the low level of early detection of ctDNA and the lack of standardized procedures for sample preparation and data analysis (Mocan et al., 2020); early detection of CTCs is also very challenging due to the lack of specific markers. Therefore, we seek more effective diagnostic methods to diagnose HCC more timely and accurately (Chen et al.,2020).

2. Treatment of HCC and Its Shortcomings

There are many methods to treat HCC, such as surgical resection, liver transplantation, transcatheter arterial chemoembolization (TACE), ablation therapy, and systemic therapy (Kong et al., 2019). When the tumor has not spread outside the liver, the choice of treatment depends on the location and burden of the tumor (Galle et al., 2018). The prognosis of liver cancer is relatively poor. In the early stage, the most effective treatments are surgical resection, liver transplantation, or percutaneous local ablation (Giraud et al., 2021). However, at this early stage, the 5-year recurrence rate of HCC after surgical resection is as high as 70% (Bruix et al., 2016). Surgical resection is only suitable for early patients with low postoperative mortality (Zou et al., 2018), but the portion of suitable patients merely accounts for 5-15% (Anwanwan et al., 2020). More than 80% of patients cannot tolerate surgery because of comorbidities or multicentric lesions (Cardarelli-Leite et al., 2020). Extensive liver resection also greatly increases the risk of postoperative liver failure (Kong et al., 2019). Compared with hepatectomy, liver transplantation has better therapeutic effect, but it also faces the problems of organ shortage and high price (Wang et al., 2021).

Most HCCs are diagnosed in the intermediate or advanced stages, at which time the five-year survival rate is only 16% (Giraud et al., 2021). TACE is one of the main methods for the treatment of advanced HCC, but the differences between different individuals make the therapeutic effect of TACE quite different (Kong et al., 2021). Moreover, the treatment of TACE needs to be carried out for many times to achieve the therapeutic effect, so it is very easy to lead to liver failure after treatment, and the incomplete necrosis of tumors is also very easy to recur (Liu et al., 2022). Ablation therapy is also considered to be an effective means for the treatment of HCC, of which the most common and widely used is radiofrequency ablation (RFA), but its therapeutic effect is affected by the size of tumor (Izzo et al., 2019). Although its efficacy and safety are widely recognized, its recurrence rate is high, especially in patients with tumor diameter > 3 cm after treatment (Izzo et al., 2019). Systemic anti-tumor therapy includes immunotherapy, molecular targeted drug therapy, chemotherapy, etc (He et al., 2023). However, these treatments also have the problem of toxicity due to long-term use of drugs, and the wide application of immunotherapy is prone to immune-related adverse events (Liu et al., 2021). At the same time, immune checkpoint inhibitors, which have attracted much attention in immunotherapy, also have similar problems, such as insufficient efficacy when used alone and toxicity when used for a long time (Jácome et al., 2021; Xia et al., 2022). At the same time, the therapeutic effect of immunotherapy is also different because of the diversity and heterogeneity of tumor types (Meng et al., 2021). Therefore, we need to seek more effective ways and means to make up for the shortcomings of these treatmentmethods.

Common Lipid-based Nanoparticles and Their Characteristics

There are many types of nanoparticles used in nano-drug delivery systems, and lipid-based nanoparticles have advantage that other inorganic nanoparticles and polymer nanoparticles cannot surpass. For example, lipid-based nanoparticles have the least systemic toxicity and high solubility in the aqueous phase (Mitchell et al., 2021). There are many forms of lipid-based nanoparticles, such as nanoassemblies, solid lipid nanoparticles, liposomes, nanostructured lipid carriers, nanoemulsions, microemulsions, lipid-coated nanoparticles and so on (Mahmoud et al., 2022). Several types of lipid-based nanoparticles are briefly described below.

1. Liposomes and Their Characteristics

Liposomes were first discovered by Bangham and his collaborators in the United Kingdom and the first report was published in 1964 (Bangham and Horne, 1964). Liposomes are the most abundant subpopulation of lipid-based nanoparticles (Sarfraz et al., 2018). It is mainly composed of glycerophospholipid and sphingomyelin, and thus is amphiphilic, i.e., hydrophilic and hydrophobic (Wang et al., 2023). Amphiphilic phospholipids are assembled to form the fat-soluble end of cyclic lipid bilayer, which enables the lipophilic drug to be adsorbed on the surface of liposome or wrapped by the phospholipid bilayer; at the same time, the water-soluble head of phospholipid can form an outer surface and a water-containing center, which enables the hydrophilic drug to be wrapped in the aqueous phase of vesicle (Sheoran et al., 2022; Wang et al., 2023). Phospholipids are able to

mimic native cell membranes, promote interactions between liposomes and mammalian cell membranes, and improve cellular uptake (He et al., 2019). The chemical properties of phospholipids have an impact on the clearance, permeability and drug distribution of liposome preparations (Nsairat et al., 2022). Liposomes are spherical vesicles ranging in size from about 20 nm to 2.5 μm, consisting of one or more concentric or non-concentric membranes. The size of vesicles and the number of membranes will affect the drug encapsulation rate and clearance rate (Wang et al., 2023).

Liposomeshavethecharacteristicsofbiocompatibility,non-toxicityandnon-immunogenicity, biodegradability and structural versatility (Mathiyazhakan et al., 2018), so they are used as an excellent drug delivery system and have been successfully applied in cancer treatment and pain treatment (Nsairat et al., 2022). Its biocompatibility, biodegradability and other characteristics can improve the low drug availability caused by water insolubility, low bioavailability and high toxicity (Nsairat et al., 2022). Traditional liposomes are easily recognized by the host immune system such as mononuclear phagocyte system (MPS), and then eliminated by the reticuloendothelial system (RES). Therefore, they need to be modified to improve the pharmacokinetics of drugs (Lee and Thompson, 2017). In addition to being used for drug delivery, liposomes can also be used for other purposes after modification of their composition and charge (Nisini et al.,2018).

At present, the stability of liposome is an important factor affecting its wide clinical application. The oxidation and hydrolysis of phospholipids are the two main factors for the instability of liposome. In addition, the selection of sterilization methods for liposome preparations is also a challenge (Wang et al., 2023). The stability of liposomes plays a decisive role in the loading rate and release rate of encapsulated drugs in a series of processes from preparation to metabolism (Wang et al., 2023). The use of phospholipids, such as polyethylene glycol (PEG), with longer tails and lower degrees of tail unsaturation and ether linkage, can improve the stability of liposomes (Guimarães et al., 2021). For example, the use of different PEGs on the surface of liposomes can extend the half-life of blood circulation from a few minutes to several hours (Guimarães et al.,2021).

2. Solid Lipidnanoparticles（SLNs）

Solid lipid nanoparticles are solid, nanoscale colloidal nanocarriers with particle sizes mainly between 150 and 300 nm, which are usually composed of lipids in solid forms such as high purity triglycerides, complex glyceride mixtures, free fatty acids, free fatty alcohols, waxes etc. (Sheoran et al., 2022). Its size can be changed by adjusting the prescription composition and production method (German-Cortés et al., 2023). The concept of SLN has been proposed as early as the 1990s in order to combine the advantages of emulsions, solid particles and liposomes to overcome their respective limitations (Müller et al., 2002). SLNs replace the liquid lipids in the emulsion with solid lipids, which enables SLNs to remain solid at both room temperature and body temperature (Müller et al., 2002). The solid matrix in the SLN can provide greater stability by limiting the movement of drugs (Kathe et al., 2014). At the time of initial development, the appearance of SLNs was presented as tiny spherical particles capable of accommodating drugs or other molecules between fatty acid chains (Scioli Montoto et al.,2020).

Compared with other colloidal carriers, SLN has the same advantages as liposomes, such as the ability to load hydrophobic and hydrophilic drugs, biocompatibility, non-toxicity and mass production (Borges et al., 2020), but SLN has stronger stability and the ability to load hydrophobic drugs than liposomes (Satapathy et al., 2021). In addition to the

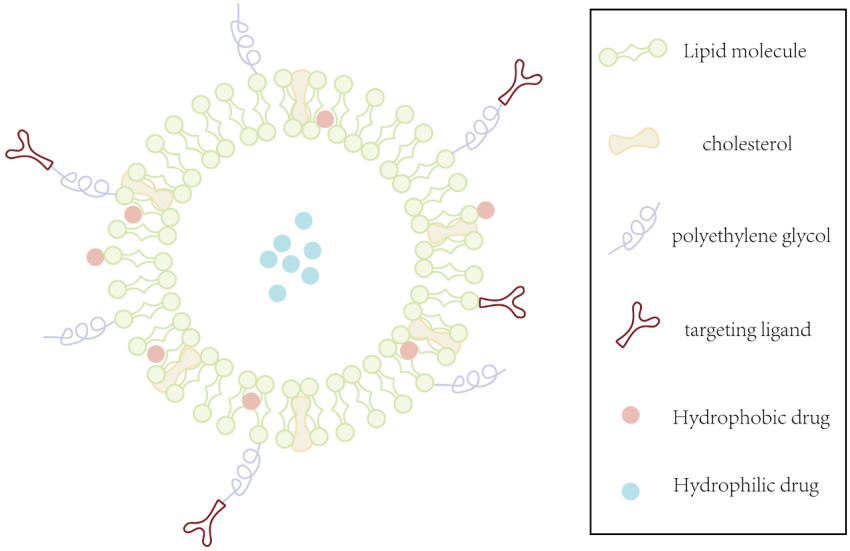

Figure 1. Structural composition of liposomes

above advantages, SLN can avoid the filtration of spleen and liver because its size cannot be taken up by reticuloendothelial system (RES), and can fix drug molecules in solid matrix to reduce the degradation and leakage of drugs; it can also improve the bioavailability of poorly soluble drugs (Mishra et al., 2018). However, due to the tight packing of the lipid matrix of SLN, the drug encapsulation space is very small, the drugloa dingisrelativelylow,andtheloadingandencapsulationofdrugsareal sovulnerabletoa variety of factors (Mishra et al., 2018).

SLNs can be classified into three types according to the composition of lipid and active ingredient, the nature and concentration of surfactant, and the type and temperature of manufacture, type I (homogeneous matrix model), type II (drug-enhanced shell model), type III (drug-enhanced core model) (Sivadasan et al., 2023). Like liposomes, SLNs can be surface-functionalized using PEGs, which can improve the targeting efficiency of drugs and genes and reduce immunogenicity (German-Cortés et al., 2023). The coating of PEG is also able to extend the time of systemic circulation of SLN (German-Cortés et al., 2023).

3. Nanostructured Lipidcarriers（NLCs）

A second generation of nanostructured lipid carriers began to emerge 10 years after the introduction of SLNs (Scioli Montoto et al., 2020). Nanostructured lipid carriers (NLC) was developed to overcome some of the shortcomings of SLN (Borges et al., 2020). Therefore, NLC has similar advantages to SLN, such as biocompatibility, biodegradability, controllability of drug release, high stability and easy preparation. NLC consists of a solid lipid matrix and a liquid lipid with a size of approximately 200-400 nm (Syed Azhar et al., 2022). NLC also includes multiple types, imperfect NLC (type 1), amorphous NLC (type 2), and polytypic NLC (types 3) (Tekade et al., 2022). The type depends on the ratio of solid to liquid lipid and determines its physicochemical properties and its interaction with drugs (Keck et al.,2021).

Compared with SLN, NLC has the advantages of higher drug loading, greater drug solubility, less water content and prevention of drug excretion during long-term storage (Ahmad et al., 2020; Chutoprapat et al., 2022). NLC has high dispersibility in water phase, so it has high encapsulation efficiency for hydrophilic drugs and lipophilic drugs; through the control of particle size, it has high permeability (Tekade et al., 2022). The matrix of NLC is not perfect, so it has higher drug loading, and the liquid lipid in the matrix has higher drug solubility (Chutoprapat et al., 2022). NLC has a strong drug immobilization capacity, and because its matrix is a solid matrix, it can prevent agglomeration between particles (Azhar et al., 2022). Drug release can be modulated by adjusting the type and amount of liquid lipid or surfactant (Chutoprapat et al., 2022). Although NLC has many advantages, its safety is still of concern, and studies have shown that NLC formulated with cationic surfactants can induce cell death and release of inflammatory mediators (Hwang et al., 2015).

Lipid-based Nanoparticles for HCC Diagnosis

As the most widely studied lipid nanosystem, liposomes can

Table 1. Comparison of solid lipid nanoparticles and nanostructured lipid carriers

	Solid lipid nanoparticles vs	Nanostructured lipid carriers	References
Ingredient	solid lipids	the combination of both solid and liquid lipids	(Akbari et al., 2022)
Types	homogenous matrix model (Type I), drug-enriched shell model (Type II), drug-enriched core model (Type III)	imperfect crystal (Type I), amorphous (Type II), multiple type (Type III)	(Borges et al., 2020; Viegas et al., 2023)
Drug loading capacity	low	high	(Duong et al., 2020)
Limitations	low loading capacity and drug expulsion during storage；irreversible conversion of low viscosity SLN dispersion to viscous gel；irritation and sensitizing potential of some surfactants.	irritation and sensitizing potential of some surfactants	(Borges et al., 2020; Jain et al., 2017; Jaiswal et al., 2016)
Common advantages	sustained drug release capabilities, biodegradation, low toxicity potential, drug protection against harsh environments, prevention of organic solvents during manufacturing		(Parvez et al., 2020; Rahman et al., 2020)
Route of administration	oral administration, parenteral administration, pulmonary administration etc.		(Madkhali, 2022)
Manufacturing methods	high pressure homogenization technique, microemulsion template, cold homogenization, solvent emulsification, solvent diffusion, reverse micelle-double emulsion, homogenization followed by ultrasonication, solvent injection, membrane contractor techniques		(Tapeinos et al., 2017; Thatipamula et al., 2011)
Size range	10-1000nm		(Mehnert and Mäder, 2001)

not only carry a variety of therapeutic drugs, but also deliver a variety of diagnostic reagents, such as ^{64}Cu and ^{14}C isotopes, quantum dots (QDs), gadolinium-based contrast agents (CA) and fluorescent probes, etc. (Silva et al., 2019). The imaging agent can be loaded at different positions and configured in various shapes depending on the type of the selected nanoscale basic lipid carrier. For example, in liposomes, the imaging agent can be loaded in an aqueous core or a bilayer lipid shell. In SLN, the imaging agent is intercalated in a solidified lipid matrix. In NLC, it is dispersed in the imperfections of a mixed matrix of oil and solid lipids. And in nanoemulsions, it is embedded in oil globules targeted to specific tumor sites (Bukhari et al.,2021).

MRI is widely used in the diagnosis of HCC. However, in most cases, MRI requires gadolinium-based contrast agents to enhance the difference between diseased tissue and normal tissue and help better visualization (Li et al., 2022). Some groups have combined CA with liposomes to prepare rare-earth-doped nanoparticles (GdREs@Lips), which can not only enhance the signal difference between diseased tissues and normal tissues, but also non-toxicity is another advantage over traditional CA (Li et al., 2022).

Ultrasound molecular imaging, as a diagnostic method at the molecular level, can detect tumor lesions earlier and diagnose diseases more accurately by using molecular probes with smaller particle size as imaging media (Montesi et al., 2019). However, the molecular probes

currently used in clinic are ultrasound microbubbles, which cannot penetrate the tumor site due to their large particle size, so it is difficult to enrich in the tumor site and make accurate diagnosis (Min et al., 2015). IRGD is a cyclic peptide of 9 amino acids, which can actively target tumor cells and promote membrane penetration. Therefore, the combination of iRGD polypeptide and liposome nanoparticles can not only actively target tumor cells, but also promote nanoparticles to penetrate tumor cell membranes and enter tumor cells (Li et al., 2021). Phase-change lipid nanoparticles coated with liquid perfluoropentane (PFP) have been shown to cause liquid-gas phase change, which can transform PFP coated lipid nanoparticles into lipid microbubbles, thus enhancing ultrasound imaging (Li et al., 2021).

Photo acoustic (PA) is an emerging non-invasive imaging method without radioactivity (Manohar and Gambhir, 2020). Indocyanine green (ICG) is a light absorber approved by the Food and Drug Administration of the United States. It can be completely metabolized in vivo and has no toxic side effects. It is an excellent choice for the development of PA probes. Therefore, there are studies on the encapsulation of ICG in liposome nanoparticles, which is conducive to PA imaging (Egloff-Juras et al., 2019).

Lipid-based Nanoparticles for the Treatment of HCC

1. Application of Liposomes

In recent years, liposomes have been widely used for drug delivery in various diseases due to their biocompatibility and biosolubility (Fan et al., 2021). When liposomes enter the circulation, they are not easily degraded, release slowly, prolong the retention time, and the anti-cancer drugs embedded in them maintain the bioavailability of drugs after reaching the site of action (Tahover et al., 2015). Especially in recent years, the size, composition and surface charge of liposomes have been gradually optimized, and the anti-tumor drugs encapsulated in liposomes have obviously avoided the uptake of RES (Klibanov et al., 1991). At the same time, liposomes are also considered to be an excellent vector for vaccines because they can enhance the production of antibodies and increase cytotoxic T lymphocytes (CTL) (Kwon et al.,2012).

Many drugs, such as Triptolide (TP), have effective anticancer effects on HCC, but they cannot play an effective role due to systemic toxicity, solubility and other problems. In order to overcome these problems, photoactivatable liposomes (LP) were prepared by combining photosensitizer Ce6 with TP (Yu et al., 2021). TP/Ce6-LP can induce cell apoptosis through calpain I/PARP signaling pathway under the condition of light. At the same time, after the liposome-encapsulated TP accumulates in the tumor site, laser irradiation makes Ce6 release ROS and further oxidize unsaturated phospholipids, destroying liposomes and releasing TP. This not only improves the therapeutic efficiency, but also reduces the toxicity of free TP (Yu et al., 2021). L-miR-375/DOX-NPs were successfully prepared by combining liposome nanoparticles with miR-375 and doxorubicin (Fan et al., 2017). This can not only inhibit the malignant phenotype of HCC cells by simultaneously exerting the anti-cancer effects of miR-375 and doxorubicin, but also partially reverse the doxorubicin resistance by down-regulating the expression of MDR1 by targeting AEG-1 (Fan et al., 2017). Glycyrrhetinic acid (GA) receptor is overexpressed in HCC, and hyaluronic acid (HA) modified nanoparticles can specifically bind to CD44 receptor overexpressed in hepatocytes. Therefore, HA and GA co-modified liposomes (CUR-APR/HA & GA-LPs) were prepared for co-delivery of curcumin and apripitant to inhibit the development of HCC (Li et al.,2022).

2. Application of Solid Lipid Nanoparticles

SLN is a physiological lipid colloid nanocarrier, which can avoid drug metabolic inactivation, protect drugs from degradation and ensure controlled drug release compared with other traditional alternative carriers (Vischio et al., 2022). It has been proved to improve the bioavailability and tissue

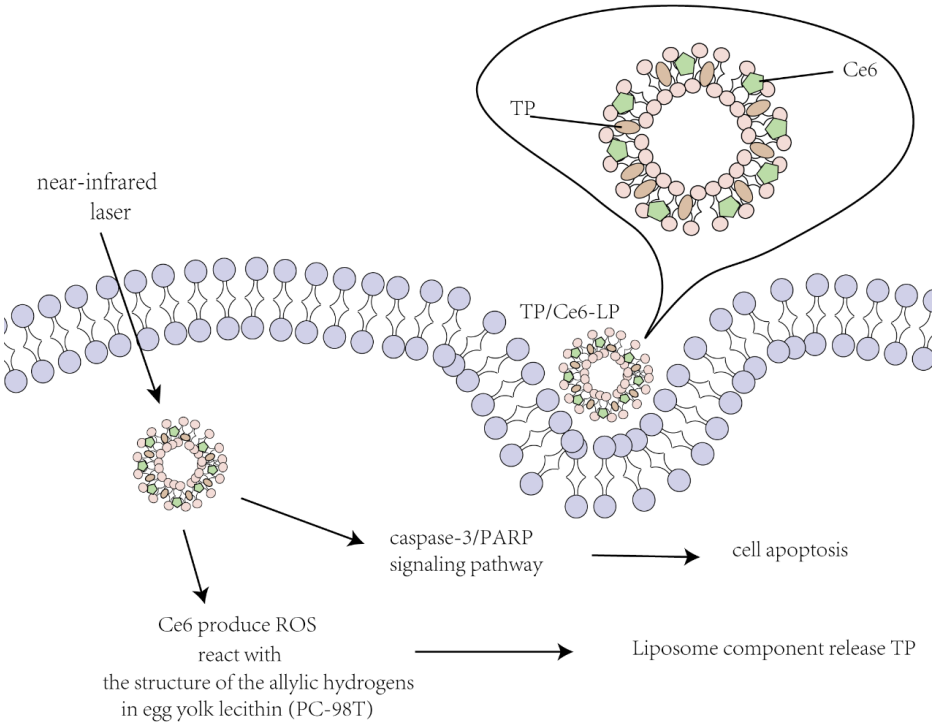

Figure 2. Mechanism of action of TP/Ce6-LP

distribution of drugs by changing the dissolution of drugs (Bayón-Cordero et al., 2019; Lazăr et al., 2019). SLN can be divided into cationic SLN and anionic SLN according to the surface charge (Le et al., 2018). Surface charge plays an important role in the stability of transport system and determines the extension of nanoparticle adsorption to the biofilm (Rasmussen et al., 2020). Cationic SLNs have greater affinity for cell membranes and serum proteins than SLNs with negatively charged surfaces (Doktorovová et al., 2014).

pH-sensitive PEGylated cationic SLN-entrapped camptothecin was prepared using stearylamine as a positive charge inducer and was found to inhibit the proliferation of human hepatocellular carcinoma (HCC36) (Chuang et al., 2017). In clinical animal experiments, it has been shown that SLN can accumulate in cancer tissues, reduce tumor volume, and rapidly distribute into tissues (Chuang et al., 2017). Rahman et al Prepared SLN composed of compritol®ATO 888 as solid lipid, tween 80 as surfactant, and soybean lecithin as cosurfactant embedded with dimethoxamine, which was detected on HepG2 cells. Configured SLNs exhibited relatively higher cytotoxicity than drug-free SLNs (Rahman et al., 2021). In animal models, drug-loaded SLN can significantly reduce the size and number of liver nodules, and enhance the activity of endogenous antioxidants to scavenge harmful free radicals (Rahman et al., 2021).

Surface modification of SLN can improve its ability to target specific receptors (Kumar et al., 2020). Solid lipid nanoparticles modified with polyethylene glycol (PEG) and loaded with organic superparamagnetic iron oxide nanoparticles (SPIONs) and sorafenib, the superparamagnetic SLN has been extensively characterized in terms of magnetic properties, colloidal stability, drug entrapment efficiency and drug loading, which can improve the safety and efficacy of sorafenib in the treatment of HCC (Vischio et al., 2022). Xu et al designed SLNs modified by GA and/or folate (FA) to encapsulate cantharidin (CTD), and confirmed that SLNs loaded with CTD could efficiently enrich drugs in tumor tissues, prevent damage to normal tissues or organs (Xu et al., 2022).

3. Application of Nanostructured Lipidcarriers

Some SLNs application limitations were observed, such as low drug loading efficiency and drug escape during storage (Salvi and Pawar, 2019). NLC was introduced in the late 1990s as a second generation SLN to overcome the problems with SLN (Naseri et al., 2015). NLC is the modification of SLN, which overcomes the unexpected kinetics of polymorphic transformation of SLN and the low drug incorporation capacity due to the crystalline structure of solid lipids (Bondì et al., 2007). Compared with liposomes, NLC has higher physical stability during storage, is easier to expand and commercialize, and is relatively cost-effective. Compared with SLN, NLC produces an imperfect crystalline lipid matrix due to the incorporation of liquid lipids, which reduces potential drug excretion during storage and increases the load of therapeutic drugs (Salvi and Pawar,2019).

10-hydroxycamptothecin (HCPT) is a drug for the treatment

of liver cancer. However, its multidrug resistance (MDR), severe systemic toxicity and renal clearance problems have seriously affected its application in chemotherapy. To solve this problem, Liu et al prepared a nanostructured lipid carrier, XG-NLC/HCPT, based on soybean oil and xyloglucan and containing galactose residue side chains (Liu et al., 2016). Studies have shown that XG-NLC/HCPT produces a more effective therapeutic effect in vivo, and shows superior cytotoxicity to HepG2 cells, and reduces systemic toxicity. XG-NLC/HCPT can reverse the drug resistance of HepG2 cells and is considered to be a promising drug delivery vehicle for liver cancer (Liu et al., 2016). Tupal et al in order to enhance the therapeutic effect of doxorubicin, α-tocotrienol was encapsulated in nanostructured lipid carriers, which were composed of pripreol®ATO5 as a solid lipid, Miglyol®812 as a liquid lipid and poloxam407 as a surfactant (Tupal et al., 2020). NLCs were successfully internalized in Huh7 cells, decreased the expression of the antiapoptotic proteins survivin and mcl-1 RNA, and enhanced the expression of Bax and Bid proapoptotic genes. Nanostructured lipid carriers loaded with α-tocotrienol significantly enhanced apoptosis of Huh7 when co-treated with DOX, with the lowest cell survival compared to free DOX and blank NLC (Tupal et al.,2020).

Bondì et al demonstrated that sorafenib loaded into NLC can greatly enhance its anti-tumor activity compared with free drug (Bondì et al., 2015). In a study by Sun et al, a nanostructured lipid carrier coated with hyaluronic acid, loaded with oleanolic acid (OA), ursolic acid (UA) and ginsenoside Rg3 (OUR) showed stronger antitumor activity than 5-fu in the treatment of liver cancer (Sun et al., 2020). Olerile et al (Olerile et al., 2017) prepared a nanostructured lipid carriers loaded with quantum dots and paclitaxel, which can monitor and track the growth of cancer cells and simultaneously inhibit liver cancer cells in a mouse tumor model ofHCC.

Summary and Outlook

HCC is a disease with high morbidity and mortality in the world, and its diagnosis and treatment have attracted much attention. The current clinical diagnosis and treatment methods have their own shortcomings. In particular, the early diagnosis of HCC plays an important role in improving the survival time and therapeutic effect of HCC. Hepatectomy, liver transplantation, transcatheter arterial chemoembolization and ablation are limited by drug resistance and recurrence rate. Nanoparticles, as a new field, provide many new ideas for the diagnosis and treatment of HCC, and can also make up for the shortcomings of some current clinical diagnosis and treatment methods.

There are various types of nanoparticles, among which lipid-based nanoparticles are one of the most widely used. Lipid-based nanoparticles have many forms, such as solid lipid nanoparticles, liposomes, nanostructured lipid carriers and so on. Lipid-based nanoparticles are composed of physiological lipids, so they have low toxicity and high safety, so they are called nano-safe carriers, which are one of the most common carriers for drug delivery systems. Lipid-based nanoparticles, from the initial liposomes to the later solid lipid nanoparticles and then to the second generation of nanostructured lipid carriers, have the advantages of low systemic toxicity and high biosolubility that inorganic nanoparticles and polymer nanoparticles cannot surpass.

Although there are many advantages, there are also some problems that cannot be ignored. Stability and safety are still important factors affecting the wide clinical application of lipid-based nanoparticles. Researchers are also trying to solve these problems, provide more effective and safer methods for the diagnosis and treatment of HCC, and strive to improve the therapeutic effect of HCC prolong the survival ofpatients.

Declarations

Ethics Approval and Consent to Participate

Not applicable.

Consent for Publication

Not applicable.

Availability of Data and Materials

Not applicable.

Competing Interests

The authors declare that they have no competing interests.

Funding

This work was supported by The National Key Research and Development Program (2022YFC2603500, 2022YFC2603505); The capital health research and development of special (2022-1-2172); Beijing Hospitals Authority Clinical medicine Development of special funding support (XMLX 202127, ZLRK202301); The Digestive Medical Coordinated Development Center of Beijing Hospitals Authority (XXZ0302); Beijing Municipal Health

Commission high-level public health technical personnel construction project, discipline leader-03-26.

Author Contributions

ML and YX contributed to study concept and design. ZZ, XL, TJ, SW, WD, WC, XB, YL, LY, YL and LZ collected and sorted out the literature. ZZ, XL and TJ drew pictures. ZZ, XL, TJ, SW and DW wrote the first draft. ML edited the English version. ML and YX approved the submitted version after modification. All authors contributed to the article and approved the submitted version. All authors read and approved the final manuscript.

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Acknowledgements

Not applicable.

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