Overcoming Biological Barriers with Stealth Liposomes: Trends in Design and Clinical Performance

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***Abstract*—Liposomes are phospholipid-based vesicular systems widely used for drug delivery due to their biocompatibility, ability to encapsulate both hydrophilic and lipophilic compounds, and flexibility in structural modification. However, conventional liposomes face major limitations, including rapid clearance by the mononuclear phagocyte system (MPS), instability in plasma, and limited circulation time. The development of polyethylene glycol (PEG)-coated "stealth liposomes" represents a major advancement in nanocarrier design, allowing prolonged blood circulation, reduced opsonization, and improved passive tumor targeting through the enhanced permeability and retention (EPR) effect. This review summarizes the fundamental concepts of liposomes, PEGylation strategies, physicochemical properties influencing stability, characterization techniques, advantages of stealth liposomes, and their cancer and non-cancer biomedical applications. Special emphasis is placed on clinical relevance, pharmacokinetic improvements, and targeted delivery approaches.**

***Index Terms*— Liposomes, PEGylation, Stealth liposomes, Drug delivery, Nanomedicine, Targeted therapy, EPR effect.**

I. INTRODUCTION

Liposomes have been extensively studied since the 1960s and continue to gain prominence as drug carriers due to their biocompatibility, low toxicity, and ability to encapsulate diverse therapeutic agents. Conventional liposomes, however, undergo rapid opsonization and clearance by the MPS, significantly limiting their therapeutic efficiency. To overcome these challenges, PEGylated or "stealth" liposomes were developed in the 1990s, providing steric stabilization and prolonged systemic circulation. Stealth liposomes have become a foundational technology in cancer chemotherapy, targeted drug delivery, and therapeutic protein transport. This review integrates fundamental knowledge with advanced research insights to provide a comprehensive overview of stealth liposomes.[1]

The inclusion of the synthetic polymer poly-ethylene glycol (PEG) in liposome composition plays a significant role in the development of stealth liposome. The PEG coating on liposome surface results in prolonged blood circulation time and it helps to reduce mononuclear phagocyte system uptake. By using stealth liposome, we can administer active molecules or drugs which have high target efficacy and activity. Stealth liposomes can be actively targeted with monoclonal antibodies or ligands by causing synthetic modification of the terminal PEG molecule.

The concept of stealth liposome i.e. coating of liposomal surface with poly-ethylene glycol was first introduced in 1990s. Poly-ethylene glycol is the widely used compound for modification of liposome for stealth effect. PEG helps to reduce their opsonization and subsequent immune cell-mediated clearance and reduce the rapid clearance of liposome by MPS. Antibodies can be attached to stealth liposome for achieving therapeutic effect and in presence of antibody stealth liposome does not affect on its ability of retain in blood circulation for prolonged tie duration. The ability of antibody which bounded to stealth liposome to recognize its target is depends on molecular weight of PEG which coated on liposome. The coating of PEG on liposomal surface proves as effective therapeutic drug delivery system which helps to improve stability of liposome in a

biological milieu which is related to inhibition of serum protein adsorption and cell adhesion.[2]

II. BASICS OF LIPOSOMES

Liposomes are spherical vesicles composed of one or more phospholipid bilayers encapsulating an aqueous core. They can be classified based on:

2.1 Lamellarity

- Uni-lamellar vesicles (ULVs)
- Oligolamellar vesicles
- Multilamellar vesicles (MLVs)

2.2 Size

- Small (<100 nm)
- Intermediate (100–250 nm)
- Large (>250 nm) [3]

2.3 Preparation Methods

- Thin-film hydration
- Reverse-phase evaporation
- Microfluidic techniques
- Vesicle extrusion

2.4 Physicochemical Properties

Key properties governing liposomal behavior include:

- Surface charge
- Bilayer fluidity
- Hydrophobicity
- Lipid packing
- Size distribution

These factors affect stability, biodistribution, and protein binding profiles. Liposomes offer the ability to encapsulate hydrophilic molecules in the core and lipophilic compounds within the bilayer, making them versatile drug carriers.[4]

III. STEALTH LIPOSOMES

Stealth liposomes incorporate polyethylene glycol (PEG) on their surface, creating a hydrated steric barrier that reduces interactions with plasma proteins and prevents rapid clearance by the MPS. Stealth liposome helps to improve stability and decreases nonspecific toxicity. But it also reduces trisection efficiency by decreasing internalization into cells and hampering endosomal release. Stealth liposome shows "enhanced permeability and retention (EPR) effect", in which they are accumulated in solid tumors by virtue of the highly permeable angiogenic blood vessels characteristic for growing tumor tissue. [5]

Stealth liposome has advantage that they improve localization of drug at site of tumor and this can be used for nucleic acid delivery as it contains coating of PEG and it protect against immune recognition and blood clearance. Stealth liposomes are hollow and spherical phospholipids and they are coated with poly ethylene glycol. This are considered as a successful drug delivery system for IV administration of anticancer drugs. The use of stealth liposome as a therapeutic drug delivery system which enclosed a drug within liposome helps to reduce plasma level of free drug and decreases the drug delivery to normal tissue which results in reduced toxicity.[6]

3.1 Role of PEGylation

PEG:

- Forms a hydrophilic cloud around liposomes
- Reduces opsonization and immune detection
- Enhances systemic circulation time
- Facilitates passive tumor targeting via the EPR effect

3.2 Mechanism of Stealth Behavior

PEGylation minimizes:

- Plasma protein adsorption
- Macrophage uptake
- Vesicle aggregation
- Premature drug leakage [7]

3.3 PEG Conformations

PEG on liposome surface forms two main structural models:

- Mushroom model (low PEG density)
- Brush model (high PEG density)

These configurations affect steric repulsion, stability, and protein interactions.

3.4 Targeted Stealth Liposomes

Ligands such as monoclonal antibodies, peptides, or folate can be attached to PEG terminals, enabling:

- Active targeting
- Enhanced intracellular uptake
- Receptor-specific delivery [8]

IV. ADVANTAGES OF STEALTH LIPOSOMES

Compared to conventional liposomes, stealth liposomes offer:

4.1 Prolonged Circulation

PEG provides steric hindrance, reducing MPS clearance and allowing circulation for hours to days.

4.2 Enhanced Tumor Targeting

Through the EPR effect, stealth liposomes preferentially accumulate in tumor tissue due to:

- Leaky vasculature
- Poor lymphatic drainage

4.3 Improved Stability

PEG inhibits vesicle aggregation and protects bilayers from enzymatic degradation.[9]

4.4 Reduced Toxicity

Encapsulation decreases free drug exposure to healthy tissues, lowering:

- Cardiotoxicity (e.g., with doxorubicin)
- Myelosuppression
- Organ-specific toxicity

4.5 Controlled and Sustained Release

PEGylation allows predictable drug release kinetics and improved therapeutic index.[10]

V. CHARACTERIZATION STUDIES

Accurate characterization ensures quality control, stability, and efficacy of stealth liposomes.

5.1 Spectroscopic Techniques

195Pt NMR

Used for cisplatin-loaded liposomes to study:

- Drug-lipid interactions
- Encapsulation efficiency
- Release behavior

31P NMR

Assesses:

- Lipid bilayer integrity
- Phase transitions
- Effect of PEG-lipids on bilayer structure [11]

5.2 Atomic Absorption Spectroscopy

Quantifies metal-based drug content (e.g., platinum in cisplatin formulations).

5.3 Photon Correlation Spectroscopy

Determines:

- Particle size
- Polydispersity index
- Zeta potential

5.4 Fractal Analysis

Evaluates morphological complexity and PEG-lipid interactions.

5.5 TLC and Lipid Degradation Studies

Used to monitor lipid hydrolysis and formation of free fatty acids.[12]

VI. BIOMEDICAL APPLICATIONS OF STEALTH LIPOSOMES

6.1 Cancer Applications

Stealth liposomes are primarily used for cancer drug delivery due to enhanced tumor accumulation and reduced toxicity.

Examples:

- Pegylated liposomal doxorubicin (PLD/DOXIL)– Reduced cardiotoxicity– Improved pharmacokinetics– Enhanced blood–brain barrier penetration
- Cisplatin-encapsulated PEG-liposomes– Higher tumor cell uptake– Lower systemic toxicity
- Paclitaxel-loaded stealth liposomes– Avoid Cremophor EL toxicity
- Vincristine + quinacrine liposomes– Efficacy in multidrug-resistant cancers [13]

6.2 Non-Cancer Applications

6.2.1 Oxygen Therapeutics

Liposome-encapsulated hemoglobin demonstrates:

- Prolonged half-life (~65 hrs)
- Reduced hemodynamic effects

6.2.2 Gene Delivery

Stealth liposomes effectively transport:

- siRNA
- Oligonucleotides
- DNA plasmids

6.2.3 Infectious Diseases

Used for diseases such as:

- Visceral leishmaniasis
- Resistant bacterial infections

6.2.4 Neurological Disorders

Chlorotoxin-modified stealth liposomes enhance delivery of:

- Levodopa to the brain (Parkinson's therapy)

6.2.5 Bone Disorders

Zoledronic acid-loaded stealth liposomes show promise in:

- Bone metastasis
- Neuropathic pain management [14]

VII. FUTURE PROSPECTS

Stealth liposomes continue to evolve through innovations such as:

- Stimuli-responsive PEGylation
- Dual-targeted PEG-liposome hybrids
- Microfluidic precision manufacturing
- Immunomodulatory PEG architectures
- CRISPR/Cas gene delivery

The challenge of accelerated blood clearance (ABC phenomenon) must still be addressed, particularly in repeated dosing scenarios.

VIII. CONCLUSION

Stealth liposomes represent a transformative platform in targeted drug delivery, offering superior stability, prolonged circulation, and reduced toxicity compared to conventional liposomes. Their ability to accumulate in tumor tissues, deliver chemotherapeutics efficiently, and provide controlled release profiles makes them highly valuable in oncology. Additionally, their applicability in gene therapy, infectious diseases, neurological disorders, and oxygen therapeutics highlights their broad potential. Continued advancements in PEGylation chemistry, ligand conjugation, and nanoengineering will further expand the clinical utility of stealth liposomes.

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X. CONFLICT OF INTEREST

The authors declare no conflict of interest.

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