# Virosomes as Novel drug delivery System: An Overview

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**ABSTRACT**

Virosomes are reconstituted viral envelopes that can fill in as vaccines and as vehicles for cell conveyance of different macromolecules. The prospect of drug delivery and targeting systems utilizing virosomes is an intriguing innovative work field. Since virosomes are biocompatible, biodegradable, non-poisonous and non-autoimmunogenuic; endeavors have been made to use them as antibodies or adjuvants and also conveyance frameworks for drugs and organic for remedial purposes. The achievement of virosomal medicate conveyance relies on upon strategy used to set up the typified bioactive materials and fuse them into the virosomes. Virosome innovation could conceivably be utilized to convey peptides, nucleic acids or, then again qualities and medications like anti-toxins, anticancer agents, and steroids.

**Keywords:** Virosomes, Drug delivery, characterization, Application.

## INTRODUCTION

The Novel invention of therapeutics against neurodegenerative or cancer disorders involve delivery systems to facilitate target drugs toward particular host tissues and cell types by controlled release and receptor-mediated uptake. Virosomal technology present a novel difficult delivery system toward convene these confront. Toward enhance the effectiveness of gene delivery through the beginning of molecules directly into cells. Virosomes has been developed in combining various agents like antigen, drug and DNA among fusion genic viral cover proteins.[1]

The Enhancement of delivery competence in vivo is a major objective in the field of virosomes research. In spite of improvements in non-viral and viral vector systems, major hurdle in the delivery of drugs and other macromolecules into the preferred cell category is crossing the permeability barrier imposed by the plasma membrane followed by the controlled release inside the cytoplasm. Virosomes are regenerate empty influenza virus envelopes. Wherever infectious neuclocapside be replaced by compound of choice. Virosomes are not replicate but they are pure fusion activity vesicles thus deliver the incorporated compound such as drug, antigen, genes inside the target cell. Virosomes protect pharmaceutically active substances from proteolytic degradation and low pH within endosomes, allowing their contents to remain intact when they reach the cytoplasm.[5] The virosome carrier system in excess of newly drug delivery vehicles like proteo liposomal and liposomal carrier systems.

## VIROSOME STRUCTURE

Virosomes are consist of spherical or unilamellar phospholipid bilayer vesicle having mean diameter in the range of 120-180 nm. Influenza virus is most commonly used for virosome production and genetic material of the source virus. Virosomes are not capable to replicate other than pure fusion active vesicles are presents.

### Structure of Virosome

The Virosome mainly constituents of Immune stimulating Regenerate Influenza Virosomes (IRIVs) consist of naturally occurring phosphatidyl choline (PC) and phospholipids (PL). PC forms around 70% of the virosomal structure. The left behind 30% of membrane components has the envelope phospholipids originating from the influenza virus to facilitate provide haemagglutinin (HA) and neuraminidase (NA) glycoproteins. Virosomes can be optimized used for maximal inclusion of the drug or for the greatest physiological effect by modifying the content or else type of membrane lipids used. It is even possible to generate carriers for antisense-oligonucleotides or other some genetic molecules depending on whether positively or negatively loaded phospholipids are included into the

membrane. Various ligands like peptides, cytokines, and monoclonal antibodies (MAbs) can be incorporated into the virosome. It also displayed on the virosomal surface. Tumor-specific monoclonal antibody fragments (Fab) might be linked to virosomes to direct the carrier to selected tumor cells.

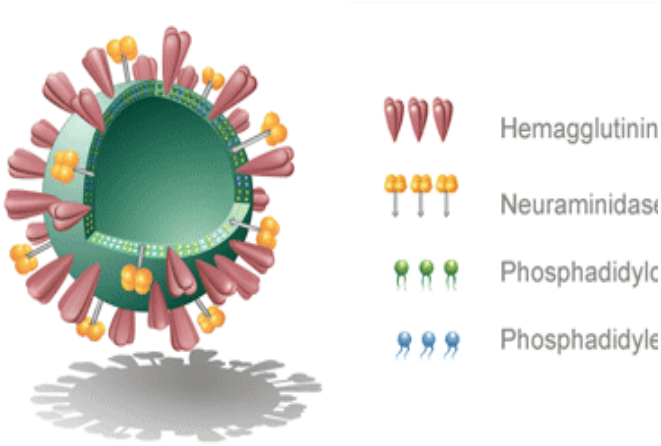


Figure no.1: Structure of Virosome

## PREPARATION OF VIROSOME

**Selection of virus**

Virosome are reconstituted viral envelop which have consequent from different virus. Influenza virus envelope is most often used to produce virosome but virosome can also be made from Sendai virus, sindbis, Epstein-burr virus, friend murine leukemia virus, herpes simplex virus.[2]

**Selection of antigen**

Antigen is preferred as per our necessities. Antigens which are used like a bacterium parasite, carcinogenic cell, or whole cell is used as antigens. Cell components include RNA, DNA, or plasmid could also be used as antigen.[3]

**Reconstituted of virosome**

Virosome solubilized with detergent such as (octaglucoside, nonidert p-40). Due to solubilization with detergent genetic material and internal viral protein will sediment afterward detergent is removed by different method like hydrophobic resins and dialysis from supernatant. By uses ultracentrifugation development viral matrix protein and nuclei capsid is removed. Antigen which has coupled to lipid anchor is mixed with surfactant or polymer solution. This solution is process among virosome mover antigen bound virosome is achieved.[4]

**Advantages of virosomal drug delivery [6]**

- Enable medication conveyance into the cytoplasm of target cell
- Virosomes are biodegradable.
- Protects drugs against corruption.
- Biocompatible, and non-lethal
- No ailment transmission hazard
- No autoimmunogenity
- Broadly material with terrifically imperative medications (anticancer drugs, proteins, peptides, nucleic acids, anti-infection agents, fungicides)
- Promotes combination movement in the endolysosomal pathway
- Target-specific delivery of antigens and amplification of the immune response.
- Extended uptake, distribution and elimination of drug in body.
- Up scaling according to standard procedure.
- Virosome allow patient specific modular vaccine regimen.
- Can be regulated by injection or nasally.
- Release of active substance in cytosol of selected cell. The antigen is partially protected from extra cellular degradation and the resulting depot effect greatly facilitates immune potentiation.
- Target-specificdeliveryofantigensandamplificationof theimmuneresponse.
- Virosome allow tolerant specific modular vaccine regimen.

## CHARACTERIZATION OF VIROSOMES

**Protein detection**

Virosome preparation could be usually consequence in the comparatively consistent protein to lipid ratios. For the confirm the presence of HA protein in the virosomes the agent Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) are used.[7]

**Structure and size**

For the Negative stain electron microscopy should be normally be used to determine the ultra structure and size of virosomes. The Prepared staining

solutions could be rather be of neutral pH for avoid acid induced conformation alteration of HA.[8]

**Fusion activity**

Regularly virosomes show pH dependent film combination movement like local flu infection. Virosomal combination with organic or counterfeit target films can be surveyed in vitro with an excimer measure utilizing pyrene-named lipids, where the decline of surface thickness of the pyrene-phosphatidyl choline-name on combination with an unlabeled layer compares to a decrease of excimer fluorescence. Combination action additionally can be in a roundabout way checked by deciding hemolytic action, which relates nearly to combination action and displays pH reliance indistinguishable with that of combination.

## EVALUATION OF VIROSOMES [9]

A) Surface morphology and vesicle shape: - Transmission electron microscopy, Freeze break electron microscopy.

B) Size dispersion and vesicle size:- Dynamic light scrambling, Transmission electron microscopy, Zeta sizer, Photon connection spectroscopy, Laser light diffusing, Gel saturation and gel avoidance.

C) Surface charge: - Free stream electrophoresis.

D) Surface pH and electrical surface potential: - Zeta potential estimations and pH touchy tests.

E) Lamellarity: - Small edge x-beam dissipating, Freeze break electron microscopy, 13p-NMR.

F) Phase conduct: Freeze crack electron microscopy, Differential checking colorimetry.

G) Percent of free medication: Mini section centrifugation, Gel avoidance and Ion trade chromatography, Protamine accumulation, Radiolabelling.

H) Drug discharge: Diffusion cell/dialysis.

I) Pyrogenicity: Rabbit fever reaction test or Limulus ambeocyte lysate (LAL) test.

J) Animal poisonous quality: observing survival rates, histology and pathology.

K) Chemical examination of surface: Static auxiliary particle mass spectrometry.

## APPLICATIONS OF VIROSOME TECHNOLOGY

Viruses are committing intracellular parasites as they are fundamentally subordinate upon particular host cells for their survival. This guideline leads to the improvement of a medication conveyance framework that copies the viral example of cell disease. Virosomes are made out of a phospholipid bilayer with the viral surface glycoproteins distending from the surface of these vesicles. The synthesis of the vesicular layer empowers the virosomes to be biocompatible and biodegradable. They are productively consumed and circulated to the objective site without being modified by the physiological procedures of the body. Additionally, the plan and structure of virosomes is to such an extent that medication particles of various nature can be consolidated in them. The lipid bilayer can effortlessly incorporate the hydrophobic medications in it. Hydrophilic medications, on the other hand, turn into a piece of the focal lacunae [10-15].Virosomes can be coupled to an immune response to guarantee the focused on conveyance of a helpful operator to upgrade the tissue specificity. These antibodies tie to the particular receptors of cells supporting the conveyance of medication atoms to these objectives. This property can, particularly, be used for conveying the medication particles with limit security profiles. Disease chemotherapeutic operators, for example, can be conveyed particularly to the tumors by marking the virosomes with antibodies [16-17]. Virosomes have appeared to viably transport macromolecules including drugs, nucleic acids and proteins to different cell sorts including hepatocytes, erythrocytes, safe cells and glioma cells.Various virosome based items have been affirmed by the United States Food and Drug Administration (FDA) for human utilize .The surface glycoproteins of Influenza infection, hepatitis infections and vesicular stomatitis infection have been effectively consolidated in various antibody and medication conveyance system [18-22]. Virosomes containing malignancy chemotherapeutic specialists, antimalarial, antibacterial and antifungal operators have appeared effective discharge profiles in vitro and in vivo. In view of the same standard, bacterial apparitions have been created. These vesicles contain the external shell or the encompass protein of different gram negative microscopic organisms [23-26]. These bacterial apparitions emulate a comparative design as is seen in the event of a characteristic disease. The virosome based sedate conveyance is, in any case, quick, sheltered and viable rather than other related system.

**VIROSOMES AS IMMUNOPOTENTIATING AGENTS**

Virosomes are the agents that can serve the function of delivering antigens and drugs to specific cell types. The chief property exploited through the virosome design is the interaction amongst the antigenic proteins of the virus with the cellular receptors [38]. Moreover, the identification, uptake and representation of the antigen incorporated in the virosome by the relevant antigen presenting cells helps in stimulating the immune system. As a result efficient regulatory and effector immune responses are generated. They initiate both cell mediated and humor alarms of immune system. Additionally, virosomes induce both cytotoxic and helper T-cell responses [27-29]. Virosomes cannot only serve as a means to transfer the immunogen to the body but can also act as adjuvants for directing the immune response to the particular antigen. They, being of particulate nature, can easily attract the dendritic cells and other antigen presenting cells for attaining immunological benefits. The composition of the virosome ensures that the antigen, whether intercalated into the lipid bilayer, conjugated to the surface proteins or present in the central cavity, is delivered continuously in a sustained manner to the immune system [30-31]. This delay in the release of the antigen can act as a tool for focusing the immune response to the particular antigen in order to gain a depot-like effect. Furthermore, the combined delivery of the antigen and the adjuvant can help in the attainment of an exaggerated immune protection against various diseases. Recent studies on murine models have exhibited up to four-fold improved humoral response in case of virosome based product in comparison to that observed on the delivery of nascent antigen [32-33].

**VIROSOMES AS AGENTS OF TARGETED DRUG DELIVERY**

one of the imperative essentials of a medication conveyance framework is to transport a remedial specialist successfully to the objective site in an auspicious way. Keeping in mind the end goal to help the focused on sedate conveyance, drugs should be either changed or bundled in such a way, to the point that restoratively successful amounts of medication atoms achieve the site of activity. Change might include the modification of physical as well as synthetic parameters of the medication bringing about the generation of new synthetic elements, blending with other substance constituents to alter their in vivo discharge profiles or, on the other hand change of physical structures of the medication particles. Virosomes can bundle medications of an assortment of nature in themselves [33-34]. They can fill in as fantastic intends to convey hydrophilic and hydrophobic medication atoms to a particular kind of tissue. The water-cherishing or hydrophilic medications are epitomized in the focal compartment amid the virosome generation prepare. The lipophilic drugs, then again, can't be epitomized in this way and are, along these lines, installed in the lipid bilayer. The moderate breaking down what's more, disintegration of the virosomes inside the cell can fill in as a method of conveying these medication particles to the expected site of activity. The exemplification of different types of hereditary material in the virosome, to be utilized for prophylactic or helpful purposes, has been accomplished in various examinations. The lipid bilayer of the virosome makes a difference in the assurance of these remedial operators from different nucleic corrosive corrupting proteins including DNAases and RNAases. The viral glycoproteins subsequent to perceiving the particular cell sorts help in the combination of the films. The hereditary material once conveyed can, at that point, be used by the cell hardware for the creation of the encoded qualities [35-36].

**VIROSOME-CELL INTERACTION**

The main preferred standpoint of the virosome innovation is their ability to mimic an in vivo contamination express that can be useful in drawing in the resistant players and the arrangement of macromolecules to the separate site of activity. Virosomes perceive and tie to the same receptors that are used if there should be an occurrence of a characteristic viral disease. Sialic corrosive receptors, for example, are used by the flu virosomes. After the cell receptor acknowledgment by the infection, combination of viral and endosomal layer is watched. If there should arise an occurrence of flu virosomes, for instance, the hemagglutinin (HA) viral protein uses its dipartite get together for a similar reason [37-38]. Furthermore, the neuraminidase (NA) is likewise incorporated into the virosome get together

as it can upgrade the immunogenicity and focusing of the virosome to a specific tissue. The virosome-receptor connection has been explored for the treatment of various sicknesses including parasitic illnesses, viral maladies, neurological clutters and numerous other metabolic issue [39]. In every one of the cases, the principle point is the arrangement of a nano-sized protein, nucleic corrosive or a medication particle to the expected site of activity. Peptides and proteins have been effectively conjugated with the virosome-surface glycoproteins. Immunizations have been produced against the Respiratory Syncytial Virus (RSV) utilizing the flu virosomes by intertwining the monitored proteins of the Hepatitis C infection surface proteins with the virosomal proteins positive enlistment of cytotoxic White blood cell insusceptible reaction. Correspondingly, epitopic locales of B-cell have been conceived utilizing flu virosomes particularly against intestinal sickness [40]. Moreover, numerous pathogens have been focused on utilizing the virosome framework. This system, along these lines, helps in maintaining a strategic distance from rehashed and different dosing for vaccination purposes.

## PHARMACOKINETICS OF VIROSOMES

Pharmacokinetics data can be utilized to translate the distinctions in the pharmacological impact of liposomal-entangled medication and free medication, consequently can be abused for measurements planning. Pharmacokinetics manages time course of retention, dissemination and debasement of the virosomal transporters in vivo. The pharmacokinetics of virosomes requires the information of conceivable available locales after intravenous organization as this is the most acknowledged course for different virosomal details misused for clinical therapeutics with the exception of topical plans. Virosomes modifies both the tissue dissemination and the rate of leeway of a medication as they are influenced by the pharmacokinetics parameters. Under ideal conditions the medication has been conveyed inside the virosomal fluid stage amid flow and it spills at adequate rate to wind up noticeably bioavailable on landing in tissue or other particular destinations. Bioavailability in the event of virosomal transporters can be characterized as the measure of free medication that can get away from the bounds of the bearer and in this manner end up plainly accessible for redistribution to neighboring tissue [41].

## MECHANISM OF ACTION OF VIROSOMES

Virosomes act both as a medication conveyance transporter and furthermore as an adjuvant with different capacities amid the enlistment of a safe reaction to body's. The transporter work contains the beneficial outcomes of implanting the antigen into a higher structure, the virosome molecule. The adjuvant capacity identifies with immune stimulating properties of the virosomes and their segments on the resistant framework. Above all, virosomes prevail with regards to invigorating particular invulnerability without causing nonspecific aggravation [42].

### CARRIER FUNCTION RESPONSE

The mix of the antigen into the higher structure of the virosome molecule balances out the antigen to jam the local status of B cell epitopes and shields the antigen from debasement. The antigen shown at first glance virosomals which mirrors the first pathogen or target cell and along these lines supports the era of antibodies important for security. The introduction of the antigen as a tedious surface structure upgrades its acknowledgment by immunizer creating B-cells. The size and surface structure of the virosome particles make them an alluring focus for take-up and handling by safe cells and which is a vital stride in the start of an invulnerable reaction [43].

### VIROSOMES ADJUVANT FUNCTION

The adjuvant capacity of virosomes depends on the nearness of flu determined envelope proteins, specifically the overwhelming haemagglutinin (HA). Prior antibodies against flu tie to virosomes and label them productively for fast take-up and preparing by antigen displaying cells (APC). The common capacity of antibodies is to tie infection and piece disease. Previous flu particular partner T cells are enacted by those antigen exhibiting cells (APC) showing the prepared sections of the flu proteins. Flu proteins enacted aide cells quickly multiply and discharge cytokines to help and improve the enlistment of impact or safe cells, e.g. immune response creating cells [44].

## DIFFERENCE BETWEEN VIROSOMES AND LIPOSOMES

Viral envelope glycoproteins with receptor-official and layer combination properties that empower the phone liposomes have been viewed as promising vehicles for focusing on and conveyance of organically dynamic atoms to living cells both in vitro and in vivo. Liposomes can possibly combine with cells and by and large neglect to give obvious conveyance of epitomized atoms to the cell cytoplasm. Virosomes contains practical conveyance of epitomized particle [45].

## VIROSOME UPTAKE BY CELLS

**a. Attachment**

This includes official of the virosomes by means of HA to the cell receptors that are a film glycoprotein or glycolipid with terminal sialic corrosive. If there should be an occurrence of particular virosomes, Fab sections are coupled by a cross-linker with a spacer arm to the virosomal surface. Particular virosomes will also perceive antigenic structures on the focusing on cell surface and bringing about a connection to target cells by two distinctive restricting components. Along these lines, particular virosomes apply selectivity for extraordinary cell sorts.

**b. Penetration**

After **Penetration** entry of virosomes happens by receptor-intervened endocytosis. The virosomes are caught in endosomes, acidic combination of the virosomal film with endosomal layer. The combination is interceded by the viral spike glycoprotein hemagglutinin (HA). The layer combination response in the endosome librates the virosomes from its lipid envelope and gives access to the embodied medications to the cytosol.

**c. Functions by the Carrier**

The reconciliation of the antigen into the higher structures of the virosomes molecule balance out the antigens, safeguards the local status of B cell epitopes and shields the antigens from debasement. In addition, the introduction of the antigen as a monotonous surface structure improves its acknowledgment by counter acting agent creating B cells.

**d. Memory Support**

The nearness of flu inferred hemagglutinin (HA) incites a memory reaction as a greater part of individuals have a level of common, prior insusceptibility against flu. This contains both humeral and cell invulnerability: previous flu particular antibodies tag virosomes proficiently for fast take-up and handling by antigen introducing cells (APC). Memory T partner cells rapidly multiply besides discharge cytokines to help and upgrades Target-specific delivery of antigens and amplification of the immune response.

Table no.1: Current Study of formulation and recovery

| Molecules | Development or Research Indications | Ref. |
|---|---|---|
| Mumps F/HN DNA plasmid | Research | [46] |
| PTH-r P DNA plasmid | Research | [47] |
| CEA and CD40L DNA plasmid | Research | [48] |
| Antisense L-myc OPT | Research | [49] |
| F RSV protein | Research | [50] |
| Fab' Her-2 Neu and doxorubicin | Research | [51] |
| mAb anti epithelial glycoprotein 2 | Research | [52] |
| *Plasmodium falciparum* SPf66 peptide | Research | [53] |
| *Plasmodium falciparum* AMA-1 peptide | Phase I clinical trial (PEVION) | [54] |
| HCV peptides | Research | [55] |
| Amyloid Ab peptides | In development (PEVION) | [56] |

Table no.2: Virosome Based Products Approved and under review by regulatory authorities.

| Therapeutic/Prophylactic Purpose | References |
|---|---|
| Influenza virus vaccine | [57] |
| Hepatitis A virus vaccine | [58] |
| Hepatitis B virus vaccine | [59] |
| Hepatitis C virus vaccine | [60] |
| Antifungal agents | [61] |
| Cancer chemotherapy | [62] |
| Antiparasitic agents | [63] |

## CONCLUSION

Virosomes can be used to deliver an antigen in the host body through altered routes like intranasal, intradermal, and intramuscular, depending on the aim of immunization deprived of side effects. Virosomes could also be abused as bearers for targeted drugs and for immunomodulating molecules particularly in cancer treatment. An attractive feature of virosomes is the possibility to target them to choose cells utilizing Fab pieces of monoclonal antibodies particular for the binding. Influenza virosomes can be an abundant device for delivering antigens and atoms of various nature, for example, proteins, peptides, plasmids, oligonucleotides and even medications, to cells and because of their flexibility and it is conceivable to abuse them in various logical divisions.

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