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Review Article

Chitosan Non-Particulate Vaccine Delivery System: A Review

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ABSTRACT

Several preclinical and clinical trials have used chitosan, a material produced from shellfish. Results show it's well-tolerated, stimulates the immune system, and has positive outcomes across various infections. Notably, chitosan has been found effective as an adjuvant for nasal vaccinations, particularly in clinical trials for a norovirus vaccine. When combined with monophosphoryl lipid, chitosan (ChiSys) elicited strong immune responses and provided protective immunity against norovirus challenges. This article summarizes the significant evidence supporting the development of chitosan-adjuvanted vaccines, including key unpublished data. Chitosan is a naturally occurring polysaccharide that has gained significant attention in the pharmaceutical industry. Its unique properties, such as being non-toxic, biodegradable, and sticky, make it an excellent candidate for delivering vaccines and medications. Chitosan also has antiviral properties, making it a promising component for vaccines. Additionally, its chemical structure can be modified to create new versions with improved properties, enhancing its effectiveness as a vaccine adjuvant. Overall, chitosan's versatility and benefits make it a valuable tool in the development of vaccines and medications. Chitosan, a natural and versatile material, has been widely researched for delivering medications and vaccines. Its ability to enhance the immune response makes it a valuable component in vaccine development. This review focuses on using chitosan as a vaccine adjuvant, exploring its inherent ability to stimulate the immune system. We'll also discuss various forms of chitosan-based delivery systems, such as; Thermosensitive hydrogels, Microneedles, Conjugates. Additionally, we'll examine the benefits of using chitosan as a coating material for vaccine carriers.

INTRODUCTION

Vaccines can be grouped into three generations:

1. **Whole/Inactivated Vaccines*:** These vaccines use the entire disease-causing pathogen, which is inactivated to prevent infection. They trigger a strong immune response but can have safety concerns.
2. **Acellular/Subunit Vaccines*:** These vaccines use only specific parts of the pathogen, such as proteins or sugars. They are

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safer and more reliable than whole-cell vaccines.

3. **Nucleic Acid-Based Vaccines*:** These vaccines use genetic material (DNA or mRNA) to instruct cells to produce specific proteins, triggering an immune response.

Each generation has its advantages and limitations, but they all aim to protect against diseases. The rise of new viruses and microbes poses a significant threat to human and animal health, resulting in substantial financial losses. Vaccines are the most effective way to combat these pathogens. Vaccines work by boosting the body's natural defenses and controlling the type and strength of the immune response. They typically consist of two main components: antigens and adjuvants. Adjuvants enhance the immune response and protect the antigens. Common adjuvants include mineral oils, bacterial components, aluminum salts, and plant extracts. However, some adjuvants have limitations. For example, mineral oil and aluminum salts are less effective against certain types of immunity and may only stimulate a specific immune response. Other adjuvants, such as those derived from bacterial components, nucleic acids, and oligopeptides, may break down quickly in the body, reducing vaccine effectiveness. As a result, researchers are exploring alternative adjuvants, particularly those derived from plants and animals, which may offer improved safety and effectiveness [1].

What is Chitosan

Chitosan, a natural polymer derived from chitin, has gained significant attention in biomedical research due to its unique properties and potential applications. Chitosan is found in the exoskeletons of crustaceans and insects and is known for its:

- Biocompatibility
- Biodegradability
- Low toxicity
- Antimicrobial properties

These characteristics make chitosan an attractive material for various biomedical applications, including:

- Drug delivery systems
- Tissue engineering

Chitosan's positive charge allows it to interact with mucosal surfaces, making it suitable for mucoadhesive drug delivery systems. However, its poor mechanical properties and low water solubility require modifications to make it more suitable for biomedical applications. Chemical modifications can improve chitosan's properties without altering its fundamental structure. These modifications enable efficient drug delivery and protect the active agent from degradation. Chitosan's biocompatibility, stability, and non-toxicity make it suitable for use with various organs, tissues, and cells. Chitosan molecules can be tailored for specific applications through chemical or enzymatic modifications, making them physiologically and physically active [2].

Chitosan's Structure:

Chitosan is a type of polysaccharide with a linear structure and semi-crystalline properties. It's composed of two main building blocks:

1. N-acetyl D-glucosamine (a modified glucose molecule)
2. D-glucosamine (a glucose molecule with an amino group)

These units are linked together in a specific pattern, forming the backbone of the chitosan molecule.

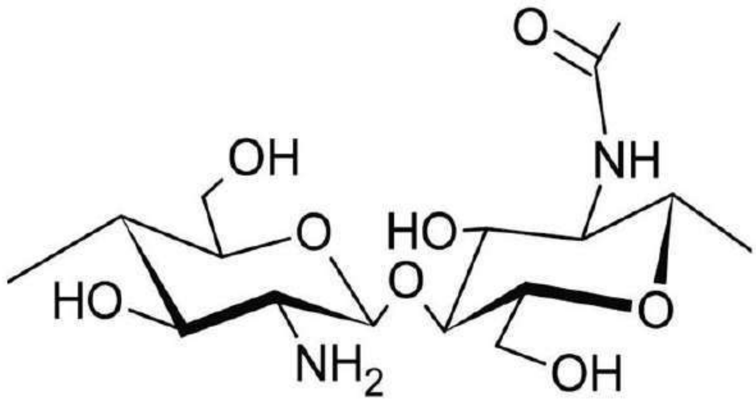

Figure 1: Chemical Structure of Chitosan

Chitosan's Molecular Weight:

To determine the molecular weight of chitosan, researchers use various techniques, including:

1. Gel-permeation chromatography (GPC)
2. Light scattering
3. Viscometry (measuring the viscosity of a solution)

The Mark-Houwink equation is used to calculate the molecular weight:

H = KM^α
Where:
- η is the inherent viscosity
- K and α are constants
- M is the molecular weight

For chitosan, the constants K and α have been determined in a specific solution, and the equation becomes:

[n] = 1.81 × 10^−3 M^0.93

This equation helps researchers calculate the molecular weight of chitosan based on its viscosity.

Chitosan: Chemical and Physical Properties:

Chitosan is a type of polysaccharide, a long chain of sugar molecules, derived from the shells of crustaceans or the cell walls of fungi. Its properties and uses depend on:

-Molecular weight
- Degree of de-acetylation (charge density)
- Distribution of acetyl groups

Natural chitosan salts have limited solubility in water, especially at neutral or high pH levels, which can be a problem for delivering vaccine antigens. To overcome this, scientists modify chitosan's molecular structure to improve its solubility. In solution, chitosan's amino groups become positively charged, making it a cationic polysaccharide. This property gives chitosan its mucoadhesive qualities, which are essential for nasal drug and vaccine delivery applications [3,4].

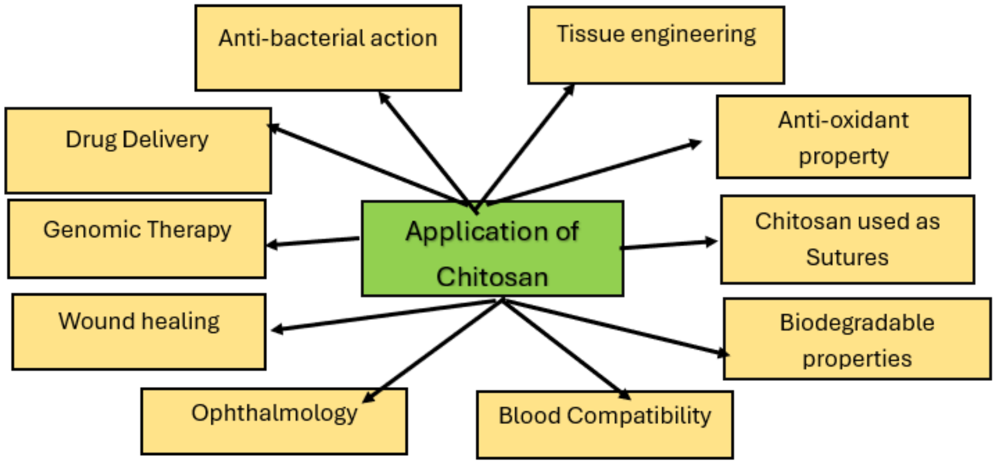

Figure 2: Applications of Chitosan

Chitosan Safety:

Advantages & Disadvantages

Chitosan is generally considered a safe, non-toxic, and non-allergenic material, making it suitable for medical and pharmaceutical applications. However, its safety can vary depending on its molecular weight, degree of de-acetylation, and chemical modification. Numerous studies have tested the safety of chitosan salts, derived from shellfish, in various animal species and through different routes of administration. The results support its safe use. Chitosan's stability in the body depends on its molecular weight and degree of de-acetylation. However, it is generally accepted that chitosan is not broken down in the human intestine and acts as a dietary fiber, excreted through feces. The oral toxicity of chitosan is low, with an LD50 of 16 g/kg in mice, similar to that of sugar. For pharmaceutical and medical applications, highly purified, GMP-grade chitosan is required. Researchers have found no allergic responses in individuals with shellfish allergies when challenged with shellfish-derived glucosamine. Chitosan dressings have also been used safely, without any allergic reactions or adverse events, even in people with shellfish allergies. Overall, chitosan is considered a safe and non-allergenic material for medical and pharmaceutical application [5].

Table 1: Advantages & Disadvantages

Advantages	Disadvantages
• High density of amino groups as active sites • Highly suitable for sustainable catalysis • Easy accessibility of active sites • Facile functionalization of amino groups to anchor wide ranges of metal/metal complexes • Commercially available, low cost, environmentally benign • Widely used in pharmaceutical applications	• Absence of size selectivity due to the lack of pores • Poor thermal/chemical stability • Cascade catalysis are limited • Precise control metal NPs size is difficult • High possibility of metal leaching • Reactions at higher temperatures are difficult

Mode of Action of Chitosan

Chitosan's Immune Response, Chitosan has been shown to trigger both cellular and humoral immune responses when administered through various routes, including injection, mucosal, and skin applications. Studies have demonstrated that chitosan activates key immune cells, such as:

- Dendritic cells (DCs)

- Macrophages
- Lymphocytes

Additionally, research has found that chitosan enhances the activity of natural killer (NK) cells, which play a crucial role in fighting infections and cancer. Interestingly, low molecular weight chitosan (LMWC) was found to be more effective in stimulating NK cell activity and cytotoxicity against cancer cells compared to high molecular weight chitosan (HMWC). These findings suggest that chitosan has potent immunomodulatory effects, making it a promising component for vaccine development and immunotherapy." "Chitosan's Immune Effects Studies have investigated how chitosan affects the immune system, including:

- How it's taken up and distributed in the body
- The type of immune cells it recruits
- The production of cytokines (signaling molecules)
- The activation of T cells

In rats, oral administration of chitosan enhanced the natural immune response at the mucosal level, promoting a Th2/Th3-biased microenvironment. In humans, chitosan selectively enhanced the induction of Th2 cells after intranasal immunization with a diphtheria toxin. Other studies have explored the effects of chitosan oligosaccharides (COS) on dendritic cells and T cells. The results suggest that COS can:

- Activate dendritic cells
- Promote the proliferation of T cells
- Depend on the molecular size of COS for its biological activity

The combination of chitosan with other adjuvants has also been investigated to enhance the immunogenicity and protective efficacy of vaccines. For example, combining chitosan with a mucosal adjuvant (LTK63) and a conjugate vaccine (CRM-MenC) induced high bactericidal antibody titers in mice. The use of chitosan-based particle systems for vaccine delivery will be reviewed in the sections that follow, with a brief overview of each mode of administration (mucosal, cutaneous, and parenteral).

Mucosal Delivery:

Mucosal immunization focuses on vaccines that protect against infections at entry points like the mouth, nose, and lungs. This approach is crucial since most pathogens enter the body through mucosal surfaces. The mucosal Immune system is a network that protects the body from harm. It connects organized lymphoid tissues (like lymph nodes) with diffuse mucosal tissues (like the lining of the gut). Through innate and adaptive immunity, the mucosal immune system maintains balance and protects the body from infections. Mucosal immune responses start when specialized cells (M cells) in the mucosa take in antigens. These antigens are then processed by immune cells (APCs) and presented to T cells. Activated B cells produce IgA antibodies, which travel to mucosal sites to fight infections. Recent research shows that Toll-like receptors (TLRs) play a key role in recognizing pathogens and activating immune responses. TLRs are pattern recognition receptors that detect specific microbial components. With its ease and acceptability, the oral route is considered the most suitable for mucosal immunization.

A. Oral Delivery:

Oral Vaccination using Chitosan/DNA Nanoparticles Researchers explored the use of chitosan/DNA nanoparticles to create an oral vaccine against Der p1, a major allergen responsible for triggering dust mite allergies [6].

The study found that the nanoparticles:

– Stimulated a strong Th1 immune response against Der p
– Induced specific antibodies against Der p1
– Were more effective than traditional injection-based immunization

Chitosan for Sublingual Immunotherapy (SLIT), Chitosan has also been investigated for its potential in SLIT, a non-invasive treatment for respiratory allergies.

Two types of chitosan microparticles were tested for their ability to:

- Enhance allergen uptake by immune cells
- Promote tolerance to allergens when administered sublingually to mice with asthma.

The study aimed to develop a more effective and convenient treatment for respiratory allergies."

B. Nasal Delivery:

Chitosan Enhances Nasal Immune Response Numerous studies have shown that chitosan and its derivatives boost the immune response when administered through nasal immunization.

The immune response is influenced by:

- Particle size
- Degree of deacetylation (DD)
- Molecular weight of the chitosan used

Researchers created nanoparticle systems using differently charged chitosan derivatives:

- TMC (positively charged)
- MCC (negatively charged)

These nanoparticles were used to deliver tetanus toxoid (TT) through nasal immunization in mice. The results showed enhanced immune responses, demonstrating the potential of chitosan derivatives for mucosal immunization.

C. Pulmonary Delivery:

Improving immunity against airborne viruses using pulmonary immunization is a growing area. The study used an HLA-A2 transgenic mouse model to test the effectiveness of a DNA vaccine containing HLA-A*0201-restricted T-cell epitopes of Mycobacterium tuberculosis and formulated in chitosan nanoparticles. The chitosan/DNA formulation stimulated DC maturation, whereas the chitosan solution alone did not. This suggests that the DNA released by the particles can stimulate DCs. The toxicity and kinetics of nanoparticles (350 nm) made from hydrophobically modified glycol chitosan (HGC) were assessed following intratracheal instillation into mice.

- **Cutaneous Delivery**

The application of vaccine antigens and adjuvants to the skin, also known as transcutaneous, has been shown to be a good target organ for generating both cellular and humoral immune responses in several animal species and clinical trials in humans. The skin contains a high density of DCs in the dermis and Langerhans cells (LCs), which are mostly found in the epidermis.

- **Parenteral Delivery**

Non-invasive immunization is a high goal for public health authorities because existing immunization techniques are dangerous, especially in impoverished countries where non-sterile syringes are frequently reused. However, parental delivery is still the most effective option. Studies are being conducted employing chitosan for parenteral administration. C57BL/6 mice were immunized subcutaneously with Ag85B-MPT64-Mtb8.4 (AMM), a fusion protein from three Mycobacterium tuberculosis genes, loaded into chitosan microspheres (6 mm), IFA, or PBS [7].

Nanotechnology:

Nanotechnology in Medicine:

Nanomedicine involves using tiny systems, measuring 1-100 nanometers, to diagnose, treat, or prevent diseases. These systems combine at least two components, including an active ingredient. Nanoparticles, such as particles, liposomes, and dendrimers, play a crucial role in delivering drugs safely and efficiently. They provide various benefits over bigger particles, including;

- Improved drug encapsulation
- Better pharmacokinetics
- Enhanced bioavailability
- Increased therapeutic efficacy

Nanotechnology has revolutionized the field of drug delivery. Chitosan Nanoparticles for Drug Delivery Chitosan is an ideal material for creating nanoparticles and microparticles for controlled drug release. Chitosan nanoparticles offer several benefits:

- Stability
- Low toxicity
- Simple and mild preparation methods
- Versatile routes of administration

These nanoparticles can control the release of active agents and avoid the use of hazardous organic solvents. Chitosan's properties make it an excellent drug delivery carrier:

- Soluble in aqueous acidic solutions
- Can be cross-linked with other molecules
- Cationic nature allows for ionic cross-linking with anions.

Preparation of Chitosan Nanoparticles:

1) Ionotropic Gelatin method
2) Microemulsion method
3) Polyelectrolyte complex (PEC) method
4) Emulsification-cross linking method
5) Complex coacervation method
6) Solvent evaporation method
7) Coprecipitation method [8].

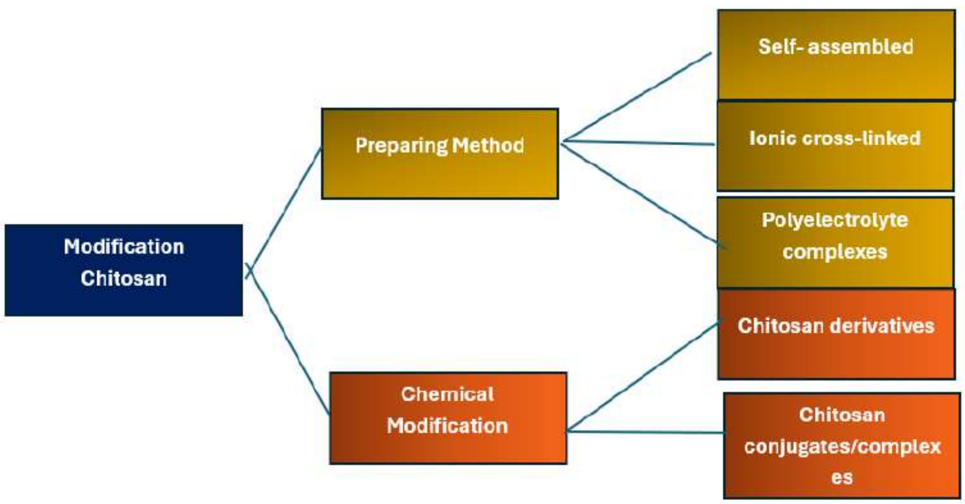

Figure 3: Preparation and Modification of Chitosan Nanoparticles

Application of Chitosan nanoparticles:

1. Chitosan Nanoparticles in Medicine and Pharmaceutics

Chitosan nanoparticles (NPs) have been widely researched for their potential in medicine and pharmaceutics. The benefits of using chitosan NPs include:

- Biocompatibility
- Ability to encapsulate and modify drugs and active ingredients
- Protecting drugs from enzymatic breakdown
- Reduced damage to non-targeted tissues or cells.

These features make chitosan NPs valuable for

- Drug delivery
- Cancer treatment
- Biological imaging and diagnosis

Furthermore, chitosan nanoparticles have been proven to

- Biodegrade slowly, allowing for controlled and continuous drug release
- Maintain stability due to their positive surface charge

These properties enable chitosan NPs to effectively carry and deliver substances within the human body.

2. Agriculture:

Chitosan Nanoparticles in Agriculture Research on using chitosan nanoparticles (NPs) in agriculture has grown rapidly, driven by the need for sustainable and eco-friendly farming practices.

Chitosan NPs are used as:

- Nanocarriers to stabilize active ingredients
- Pesticide and fertilizer release systems are controlled.

These benefits enable:

- Lower doses of agrochemicals to be used
- Fewer treatments to be applied

As a result, the risk of environmental contamination and harm to non-target organisms is reduced. Chitosan NPs offer a promising solution for sustainable agriculture, minimizing the impact on the environment while maintaining crop health and productivity.

3. Water waste management:

The need for a cost-effective, sustainable, and efficient alternative to activated carbon has led researchers to explore bio-based options. Chitosan nanoparticles (NPs) are promising candidates due to their:

- Functional amino and hydroxyl groups, which enable them to remove various pollutants, including:
- Heavy metals
- Pesticides
- Dyes

Chitosan NPs have several advantages:

- Higher surface area, allowing for greater pollutant absorption
- Potential for higher capacity than conventional micro-sized sorbents

In wastewater treatment, ultrafine nanoparticles (smaller than 100 nm) are preferred due to their:

-Increased surface area, enabling better pollutant removal

Chitosan NPs offer a sustainable and effective solution for waste water treatment, addressing the need for innovative and eco-friendly technologies.

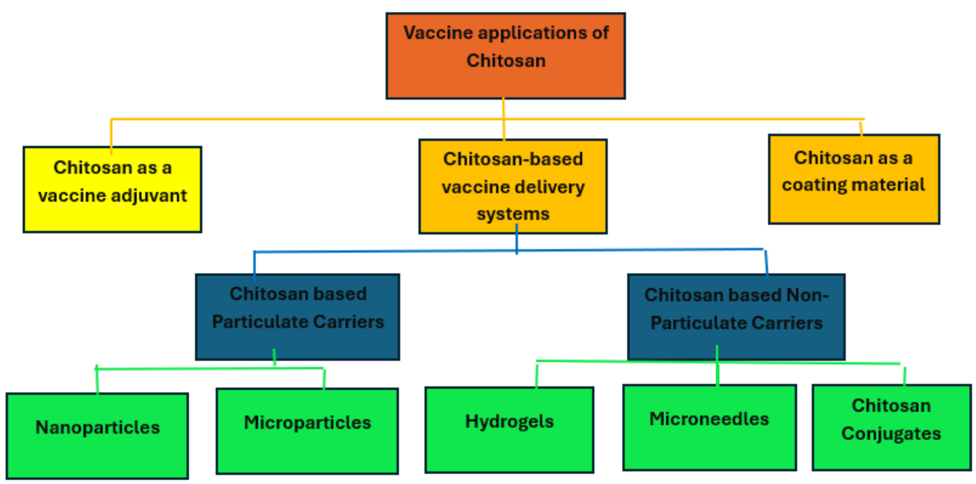

Figure 4: Vaccine Application of Chitosan & It's Derivative in Different Forms

Antioxidant Activity of Chitosan

Antioxidants are gaining attention due to their potential to prevent or treat various diseases, including;

- Neurodegenerative diseases (Alzheimer's, Parkinson's, Huntington's)
- Cancer
- Diabetes complications

Chitosan, a natural polymer, has antioxidant properties due to its:

- Amino group
- Hydroxyl groups

These groups can react with free radicals, neutralizing their harmful effects. Some chitosan derivatives, such as:

- Chitosan sulfates
- N-2 carboxyethyl chitosan Have shown improved antioxidant activity.

Chemical modifications, like attaching gallic acid or phenolic compounds, can also enhance the antioxidant properties of chitosan.

Researchers use various methods to measure antioxidant activity, including:

- DPPH assay
- ABTS assay
- FRAP assay
- ORAC assay

These tests evaluate the ability of antioxidants to neutralize free radicals and protect against oxidative stress.

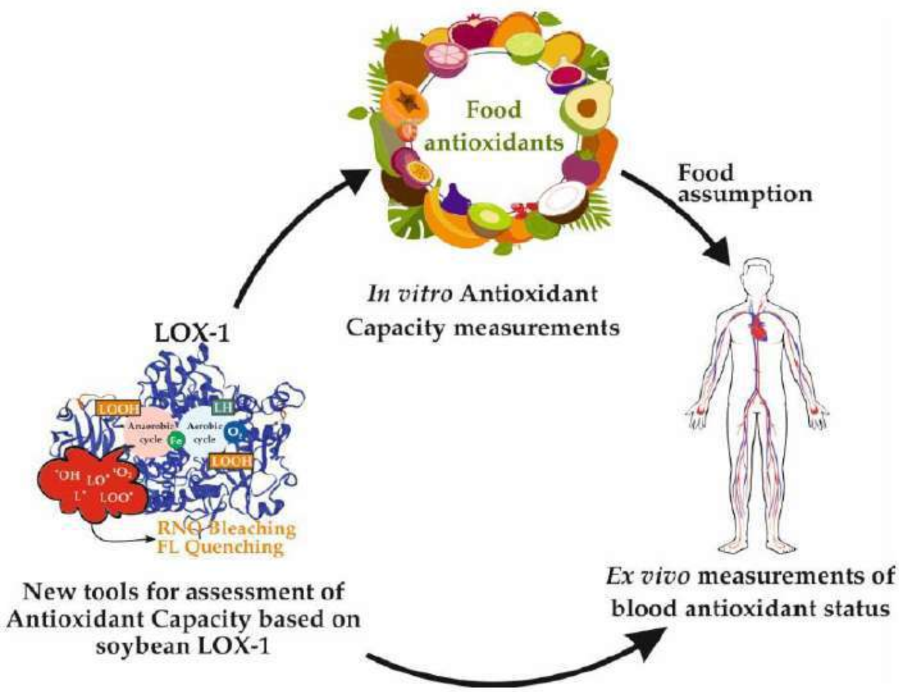

Figure 5: Assessment of Antioxidant Capacity & Putative Health

Antiviral properties:

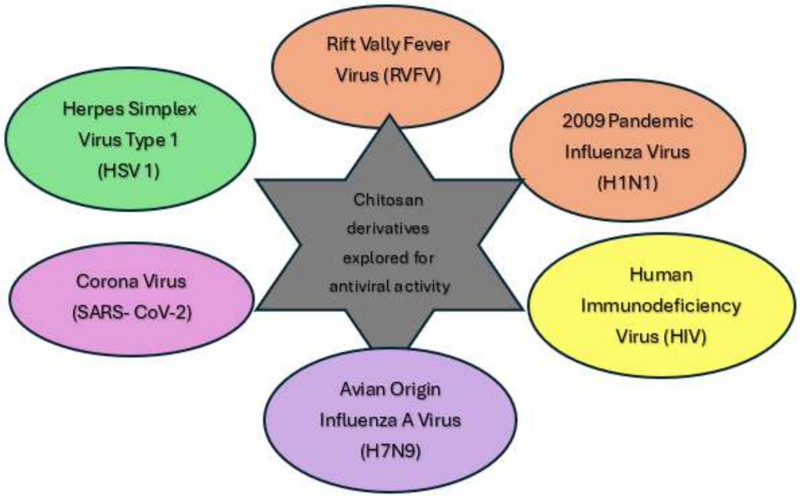

Figure 6: Chitosan Derivative Exploring Myriad Antiviral Properties

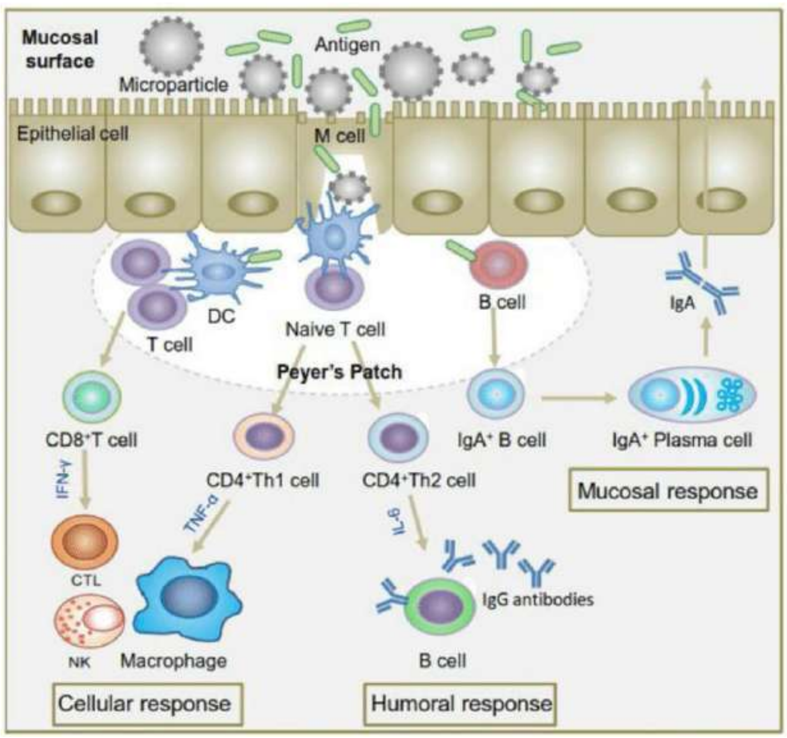

Figure 7: Various Immune Response Induced by Particulate Vaccine

Chitosan in Cancer Treatment:

Chitosan nanoparticles have shown promise in cancer treatment due to their:

- High cytotoxicity against cancer cells and tumors
- Low systemic toxicity in laboratory and some animal studies

These nanoparticles are being explored for:

- Cancer diagnosis
- Targeted drug delivery
- Reduced side effects

Chitosan nanoparticles have:

- Positive surface charge
- Mucoadhesive properties, allowing them to stick to mucous membranes and release drugs gradually

These properties make them suitable for:

- Cancer therapy

- Gene delivery
- Buccal, pulmonary, mucosal, ocular, nasal, and salivary drug delivery
- Vaccination delivery, for example, researchers have used chitosan nanoparticles to deliver the anticancer drug doxorubicin, reducing its toxicity and improving its effectiveness.

Other studies have shown that:

- Chitosan oligosaccharides conjugated with doxorubicin can suppress tumor growth more effectively
- Chitosan nanoparticles can enhance the anticancer activity of curcumin, a natural compound with therapeutic potential

Overall, chitosan nanoparticles hold promise for improving cancer treatment outcomes and reducing side effects [9-12].

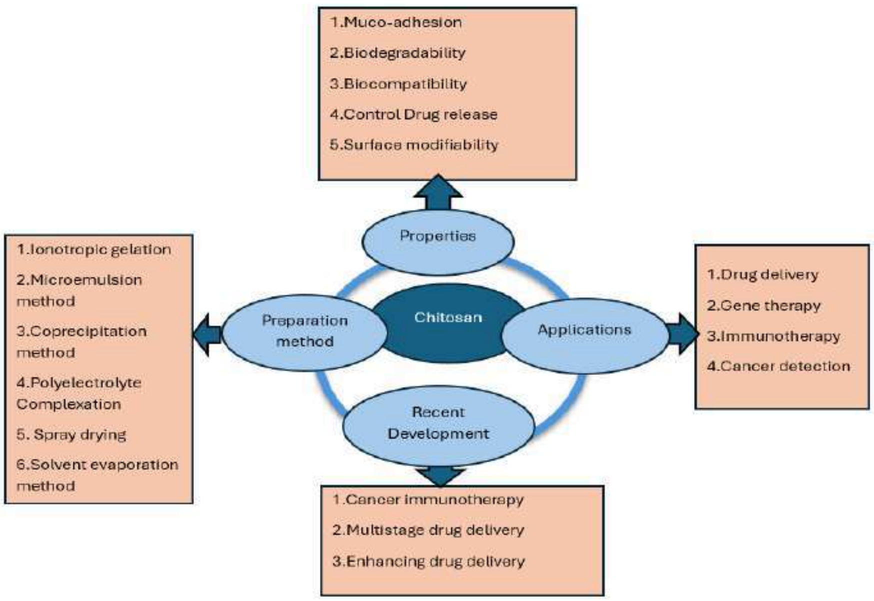

Figure 8: Multifaceted Chitosan Drug Delivery

CONCLUSION:

The present review focuses on the recent development of chitosan-based nanocomposites for biomedical applications. Chitosan, a polysaccharide biopolymer, has a distinct chemical structure as a linear polycation with high charge density, reactive hydroxyl and amino groups, and significant hydrogen bonding. Unlike synthetic polymers, chitosan has exceptional physicochemical and biological properties such as low toxicity, biodegradability, biocompatibility, low cost, haemostatic, mucoadhesive, and so on, making it an appropriate biomaterial for a wide range of biomedical applications including tissue regeneration, wound dressing, drug delivery, and more. Recent research indicates that chitosan has considerable potential in a variety of biomedical applications. Chitosan's molecular weight and deacetylation level influence its varied qualities. Furthermore, chitosan can be easily changed through a variety of reactions, resulting in the production of novel derivatives. Chitosan nanoparticles are best suited for controlled drug delivery, mucosal drug delivery, and improving the stability of medicines, genes, or proteins when synthesized as chitosan nanocarriers. They are also a better option for tissue engineering applications. This research studied and assessed different chitosan changes that can increase vaccine transport to APCs, potentially boosting immune responses. Chitosan's stability, membrane permeability, muco-adhesiveness, and controlled release behaviour have been improved as a result of functional changes, suggesting its promise as a vaccine carrier system.

REFERENCES

1. Rasim Masimov & Ellen k. Wasan, Chitosan Non-Particulate vaccine delivery system of pharmacy & nutrition, university of Saskatchewan Saskatoon, SK, Canada, journal of Pharmacy & pharmaceutical sciences 24 July 2024
2. Girish Kumar, Tarun Virmani, Shashi Kiran Misra, Ashwani Sharma, Kamla Pathak exploration of Chitosan and it's modified derivative as vaccine adjuvant: A review carbohydrates polymer technologies & applications 8 (2024)100537,www.sciencedirect.com/journal/
3. Harsh Yadav, Rishabha Malviya, Niranjan Kaushik, Chitosan in Biomedicine: A comprehensive review of recent developments carbohydrates polymer technologies and application 8 (2024) 100551, www.sciencedirect.com/journal/
4. Alan Smith, Michael Perelman & Michael Hinchliffe Chitosan A promising safe & immune enhancing adjuvant for intranasal vaccines Human vaccine & Immunotherapeutics 10:3,797-807; March 2014, lands bioscience.
5. https://images app.goo.gl/MsgdTDlxMjpqwunt8
6. Sevda Senel chitosan-based particulate systems for non-invasive vaccine Delivery department of pharmaceutical technology, faculty of pharmacy, Hacettepe University, 06100: https://www.research gate.net/publication/278177579.
7. Kalpana Nagpal, Shailendra Kumar Singh, and Dina Nath Mishra, Chitosan Nanoparticles: A promising system in novel drug delivery,nov.2010, chem.Pharm.Bull58(11)1423-1430(2010)
8. Yedi Herdiana, Nasrul Wathoni, Shaharum Shamsuddin, Muchtaridi Muchtaridi Drug release study of the Chitosan - based nanoparticles Heliyon 8(2022)8674.www.cell.com/Heliyon.
9. Murat Yanat, Karin Schroen, preparation method & application of Chitosan nanoparticles within outlook towards enforcement of biodegradable packaging reactive & functional polymers 161(2021)104849, www.elsevier.com/locate/reast.
10. Inmaculada Aranaz, Andres R. Alcantara, Maria Conception Civera, Conception Arias, Begona Elorza, Angles Ceras Caballero, & Niuris Acosta.
11. Qingye Meng, Ying Sun, Hailin Cong, Hao Hu, Fu-Jian xu- An overview of Chitosan & it's application in infectious disease, Drug Delivery & Translational research (2021)11:1340-1351.
12. Anil Kumar Grewal, Raj Kumar Salar, Chitosan nanoparticle delivery system: An effective approach to enhancing efficacy & safety of anti-cancer drugs.

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