ISSN : 2350-0743

International Journal of Recent Advances in Multidisciplinary Research

Vol. 11, Issue 04, pp.9714-9724, April, 2024

REVIEW ARTICLE

LOCAL DRUG DELIVERY USING CHITOSAN MICROSPHERES – A REVIEW OF LITERATURE

***Aastha Moza, Titus Thomas and Srinivasa Prasad, T.**

Department and Institution: OMFS Department, MeenakshiAmmal Dental College, Alapakkam main road, Maduravoyal, Chennai, Tamil Nadu, 600095, India

ARTICLE INFO

Article History

Received 08th January, 2024
Received in revised form
20th February, 2024
Accepted 27th March, 2024
Published online 30th April, 2024

Keywords:

Chitosan, electrolyte, chitosan nanoparticle, drug delivery, cancer therapy, emulsion, gelation, growth factor, nano particle.

****Corresponding author: Aastha Moza,***

ABSTRACT

Background: Drug discovery is the process by which therapeutic agents are identified and developed while drug delivery refers to the processes and technologies employed to provide vehicles through which therapeutic agents are administered to patients ensuring safety, efficacy, and quality. Synthetic materials were favoured against natural materials due to challenges of standardization and extensive characterization required for natural materials. ***Objectives:*** Purpose of the study is to review the application of chitosan as a nano particle and its significance as a local drug delivery system. ***Methodology:*** The search study for the review paper is based on current literature having 15 articles which has been beneficial in title and research work done on local drug delivery. ***Results:*** This is a detailed literature review so the results will be in terms of the work done as discussed in review of literature. ***Conclusion:*** This paper reviews the structural characteristics and various synthesis methods used to produce chitosan nanoparticles and nanostructures, such as ionic gelation, micro emulsion, polyelectrolyte complexing, emulsification solvent diffusion, and the reverse micellar method. Various characterization techniques and analyses are also discussed.

Citation: Aastha Moza, Titus Thomas and Srinivasa Prasad, T. 2024. *"Local drug delivery using chitosan microspheres – A Review of Literature", International Journal of Recent Advances in Multidisciplinary Research,* 11, (03), xxxx-xxxx

INTRODUCTION

Drug discovery is the process by which therapeutic agents are identified and developed while drug delivery refers to the processes and technologies employed to provide vehicles through which therapeutic agents are administered to patients ensuring safety, efficacy, and quality. Synthetic materials were favoured against natural materials due to challenges of standardization and extensive characterization required for natural materials. General problem of drug targeting consists of a basic pitfall, certifying that the most effective interaction of drugs with target cells, including their proper binding on cell membranes and intracellular transport; effectively deliver drugs towards certain target cells, avoiding unfavourable drug distribution makes it a challenging arena. Therefore to overcome these aspects of drug delivery, need for drug targeting to site of action, drug device-cell interaction, time-based drug release and bioavailability came into existence (1). Drug delivery refers to the methods, formulations, technologies, and processes involved in transporting a pharmaceutical substance in the body to achieve the desired therapeutic effect. It encompasses the approaches of administering medicinal compounds in humans and animals to attain therapeutic effectiveness. Recent developments in drug delivery systems are primarily been focused on smart targeteddelivery, focusing on drug administration at the appropriate time, dosage, and location with maximum safety and efficacy.

Enhancement of drugs using polysaccharides, natural polymers derived from sugar units bonded by glycosidic linkages in a linear or branched pattern, are gaining attractions as drug carriers, building blocks for drug delivery, bioactive materials, and excipients. Polysaccharides are found abundantly in the human body, animals, plants, microorganisms, and marine organisms such as algae. The polysaccharides are devitalized to produce semisynthetic polysaccharides such as carboxymethylcellulose, starch acetate and chitosan sulphate. Targeted drug delivery system differ from conventional or traditional drug delivery in that they acquire site-specific release of drugs from a dosage form, while the former depends on drug absorption through biological membranes (3). Ideally, a drug-targeting complex is expected to be non-toxic, non-immunogenic, biochemically inert, biodegradable, biocompatible and physio-chemically stable in vivo and in vitro. This system brings an opportunity for controlled release of drugs, allowing sufficient time for drugs to act with enhancedtherapeutic action and respond to specific stimuli, such as pH, light (2). Nanomedical approaches to drug delivery concentrate on the development of nanostructure devices like the microcapsules or nanospheres to improve the bioavailability of the drug and target it to the specific site of interest (5). A pharmaceutical drug carrier, chitosan is currently used with several drug delivery systems based on routes of administration. Oral route is the most popular and the most practical way to administer a therapeutic agent, particularly from the point of view of the patient. Oral chitosan microsphere preparations have been prepared

extensively using a suspension or emulsion cross-linking procedure. Chitosan is well tolerated by living tissues and is biodegradable and non-toxic recently envisaged for the controlled release of drugs administered parenterally in the form of cross-linked microspheres. Parented administration of drug products offers a number of advantages over the oral route. Microspheres are well known to be useful as drug delivery systems due to their possible localization to the target site (4). Drug release is characterized by an initialburst effect. Since chitosan combines unique physicochemical characteristics, in vivo biodegradability, biocompatibility, and antimicrobial action, it has been widely investigated over the last few decades for potential applications as a drug carrier for many possible routes of administration. Biodegradable microspheres synthesized to avoid their degradation and denaturation by pepsin and hydrochloric acid in the stomach, vaccine antigens have been incorporated into antigen delivery systems including gelatine capsules soluble in alkaline pH, liposomes and microspheres. Context of drug carrier resorption and wound healing, chitooligomers and monomers, generated bylysozyme, N-acetylglucosamines and human chitinase, activate macrophages and stimulate fibroblasts, respectively (6).

General review of Oral local Drug delivery

Knowledge of the permeability features of oral mucosa is crucial in selecting the most appropriate formulation so that a drug is absorbed and it reaches the deeper layers of the oral epithelium, according to local variations in mucosal thickness, epithelial keratinization, and lipid composition. These are collectively known as barrier region features. It is generally accepted that the connective tissue of oral mucosa is not an effective barrier to the penetration of substances. One of these is localized at the basal complex, and the second is in the intercellular spaces of the superficial epithelial layers as shown in Figure 1. Permeability measurements suggest that different substances may permeate oral epithelium at different rates, depending on the chemical nature of the molecule and the histologic features of the tissue being traversed. There are considerable regional differences in the permeability pattern of the oral mucosa. In general, the permeability of oral mucosa decreases gradually from the sublingual through to the buccal and palatal mucosa. The rank order is related to the relative thickness and degree of keratinization between these regions: the sublingual mucosa is relatively thin and nonkeratinized, the buccal mucosa is thicker and nonkeratinized, and the palatal mucosa is intermediate in thickness and it is keratinized. A single drug can permeate through oral mucosa, using both routes simultaneously, but the route offering the least resistance to penetration is usually preferred, depending on the physicochemical properties of the drugs. Mucosa of the oral cavity comprises of keratinized (gingiva and palate) and nonkeratinized epithelial tissues (sublingual and buccal). Buccal absorption bypasses, hepatic first pass metabolism. Buccal tissue is readily accessible and localization of a dosage form with a defined surface area over extended periods maximizes absorption, in addition to a higher degree of control and reproducibility relative to other mucosal delivery routes.

The epithelium of the oral mucosa is approximately 40 to 50 cell layers thick. In particular, the presence of saliva in association with swallowing, chewing, and phonation acts to wash away most of thedrug from the site of application, resulting in a short retention time of the dosage forms and, consequently, low therapeutic efficacy (7). Overcoming the various pitfalls can be achieved by:

1. Rapid drug loss from the site of absorption by salivary scavenging and mechanical stress
2. Non-uniform distribution of drugs in saliva on release from delivery systems
3. Poor patient compliance because of an unpleasant taste and sensation in the mouth
4. Relative permeability of the oral mucosa and potential barrier region to drug absorption

Two main pathways seem to be implicated in passive diffusion across membranous tissues, however, the intracellular (or transcellular) pathway, and the intercellular (or paracellular) pathway. Drug delivery via the oral mucosa can be subdivided into 2 different approaches: drug delivery via keratinized mucosa, and drug delivery via nonkeratinized mucosa as shown in Figure 2. Transcellular pathway encompasses the transport of molecules through apical and basolateral membranes. The drug transfer here is through the cell itself. The mechanism of action is through transcytosis and active carrier mediated transportation. Presence of a concentration gradient between the regions facilitates the transport pathway. Paracellular pathway occurs through the transport across the epithelium through the intercellular voids.

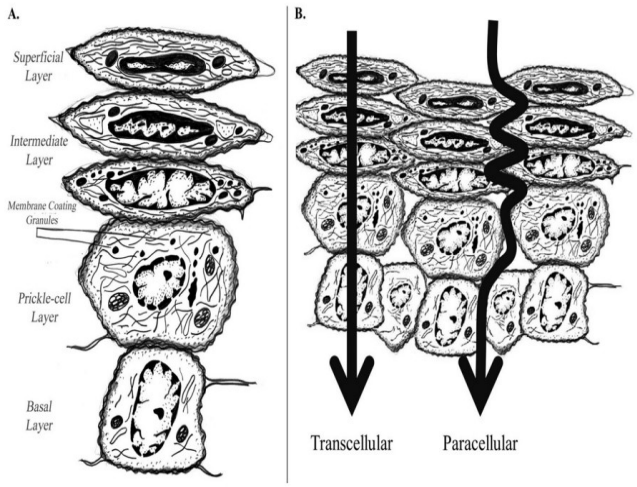

Figure 1. Ultrastructural oral buccal epithelium & drug transport (7)

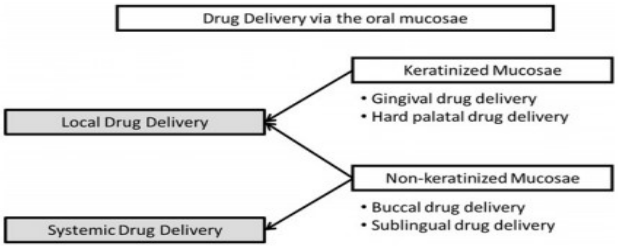

Figure 2. Schematic representation of possible approaches (7)

Therefore, it lacks the need for energy requirement. In the availability, of drug absorption through gastrointestinal tract it is pivotal. It facilitates movement through the lipid membrane.Diffusion via either of the pathways can be summarised with the similarity of the transport to be through capillaries of the body. Currently, many biologically active drugs for the treatment of oral conditions have been administered systemically with possible serious side effects that limit their use only tosevere andrefractory cases. The development of effective systems for local drug delivery offers a more targeted therapeutic option, thereby reducing the required drug doses and the risk of systemic side effects.

Relationship of drug delivery and drug targeting

Drug delivery (DD) refers to the methods, formulations, technologies, and processes involved in transporting a pharmaceutical substance in the body to achieve the desired therapeutic effect. It encompasses the approaches of administeringmedicinal compounds in humans and animals to attain therapeutic effectiveness. Recent developments in drug delivery systems (DDSs) are primarily been focused on smart DD, which focuses on drug administration at the appropriate time, dosage, and location with maximum safety and efficacy.

There are five generations of DDSs, as shown in Figure 3 and targeted delivery belongs to the fourth generation. TDDSs are where a drug is delivered to a specific location, rather than the whole body or organ, and combine diverse fields of science, such as polymer science, pharmacology, bioconjugate chemistry, and molecular biology. TDD is aimed at managing and controlling the pharmacokinetics, pharmacodynamics, aspecific toxicity, immunogenicity, and biorecognition of therapeutic agents (7). The end goal is improving treatment effectiveness while reducing side effects. TDDSs differ from conventional or traditional DDSs in that they acquire site-specific release of drugs from a dosage form, while the former depends on drug absorption through biological membranes.

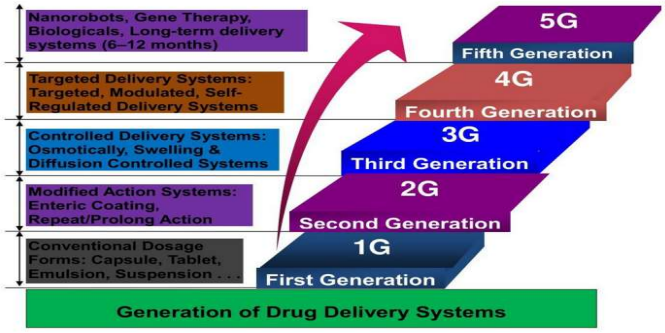

Figure 3. Generation of drug delivery (2)

Figure 4. Targeted drug delivery (2)

Basic aim of targeted drug delivery scheme is to counteract the disadvantages in the administration of a drug by conventional drug delivery. Orally administered drug once absorbed into the body undergoes systemic circulation followed by distribution and absorption. This causes a higher concentration of drug to be administered to ensure therapeutic concentration is delivered to the site of action, ensuring a lower dose is used, nonspecific tissues or organs aren't exposed to the drug and the drug bears minimal side effects on the body.

Main strategies of drug targeting include

1. Direct administration to target site
2. Passive targeting through leaky vasculature
3. Stimuli responsiveness
4. Vehicles with high affinity for target site

Nano drug delivery at a nanoscale proffered solutions to challenges that a conventional drug delivery cannot tackle. Challenges include poor bioavailability, poor distribution, intracellular penetration and controlled release. This Nanoscale drug administration is best achieved by polysaccharides as they bear innate biological advantages, cell recognition and interactions, resemblance to extracellular matrix and hemocompatibility and enhanced permeability. Concept of the Magic Bullet- Paul Ehrlich envisioned the concept of selectively targeting a pathogen without harming the host organism using "magic bullets." According to Ehrlich, drugs should go straight to their anticipated targets in the body and only interact with the target molecule. The basic principle behind drug targeting as shown in Figure 4 is delivering a high concentration of drug to the targeted site. Ideally, a drug-targeting complex is expected to be atoxic, nonimmunogenic, biochemically inert, biodegradable, biocompatible. Considerable parameters include drug concentration, particulate location and distribution, molecular weight, physicochemical properties, enzymes, electric fields, physiological environment, concentration of polymers.

PCFTDDS are the backbone of PDDS. They have found extensive applications in drug delivery because they offer unique properties that have not been attained by any other material. Polymers play an important role in advanced DDSs, as they can be used to assist delivery and as excipients, they allow controlled and targeted drug release. Micro and nanospheres fabricated from abiodegradable polymer enable controlled drug release at desired sites. Polymeric nanocarriers show promising pharmacokinetics at both the whole body and cellular levels. Generally, polymer-based drug nanocarriers can significantly increase the solubility of hydrophobic drugs, reduce their cytotoxicity toward normal tissue, prolong the circulation time of drugs in blood (2). During the 20th century, Paul Ehrlich introduced the idea of drug targeting. In the 1960s, Peter Paul synthesized the first NPs for drug targeting (8). In 1963, the use of magnetic nanocarriers was introduced. Meyers *et al* used an externally applied magnet to compile small iron particles to be injected into the leg veins of dogs.

These microspheres were enclosed with Adriamycin as an anticancer drug. Nanocarriers are defined as small entities with size 500 nm. Various nanocarriers such as polymers, micelles, liposomes, dendrimers, gold, carbon nanotubes, silicon, and iron oxide are used. They have been developed and employed as carriersfor drugs / vehicles for the controlled release of drugs, especially for anticancer medicines. NPs are known as prospective and profitable drug carriers over conventional drugs for cancer therapy due to their promising characteristics such as the ability to be functionalized with drugs, increased therapeutic efficacy, enhanced drug stability, and capability to entrap lipophilic, hydrophilic, lipophobic and hydrophobic drugs (9).

Drug carriers, sometimes called drug vectors, are the most important entity required for successful transport of the loaded drug to the intended target. They transport, retain, and deliver the drug within or at the location of the target. They are capable of performing such specific functions by slight virtue of structural modification. There are different types of drug carriers, such as colloidal, polymers, monoclonal Abs, NPs, and cell. Colloidal DDSs are nano scaled targeting vesicles of particulate or vesicular dosage form. They include liposomes, nanospheres, multiple emulsions, and ceramics. These type of drug vectors sequester, transport, and retain the active drug, while they elute or deliver it within or in the vicinity of the target, with the ability to modify the distribution profile. Controlled-release formulations guarantee sustained release of a drug in a modulated or contained pattern, thereby facilitating the availability of the drug over a prolonged period to provide therapeutic action. Controlled release of therapeutic agents is desired to prevent dose dumping, reduced side effects, provide constant and/or continuous delivery over a prolonged period, reduce frequency of dosing, and subsequently promote patient adherence. Various conventional and unconventional drug delivery systems are formulated to ensure that the minimum therapeutic concentration is available in circulation and possible, target sites in order to elicit therapeutic responses. When a drug is administered orally for instance, it is absorbed, goes into the systemic circulation, and isdistributed throughout the body. Systemic distribution of a drug implies that the drug is exposed to the cells, tissues, or organs that have no need of it. Consequently, a higher dose of the drug is administered in order to ensure that therapeutic concentration is delivered to the target site actually requiring the drug (1).

Drug targeting facilitates the delivery of drug and its therapeutic concentration to the site of action. Such localization ensures that a lower dose is used so that other tissues or organs are not exposed to the drug and consequently there are minimal side effects. Strategies to drug targeting include direct administration to the target site, passive targeting through leaky vasculature, stimuli responsiveness, and use of vehicles with high affinity for the target site. Drug targeting can be facilitated by pH, temperature, and magnetic responsiveness. Owing to their biogenicity and cell recognition, polysaccharides are favourable components of drug targeting delivery systems. A targeted drug delivery system comprises the drug, the drug carrier, and the targeting moiety (1). Active targeting entails the binding of a ligand such as peptides, and vitamins to the surface of nanocarriers. The function of these ligands is to attach an individual receptor on the surface of the cell, via RME. This RME mechanism consists of three steps. First, the ligand attaches to a suitable receptor on the cell, subsequently resulting in the formation of endosomes. Endosomes are compartments of a plasma membrane, which include the receptor and ligand complex. Finally, theendosomes transfer to the desired site, and the drug is released under the influence of pH difference or enzymes (10).

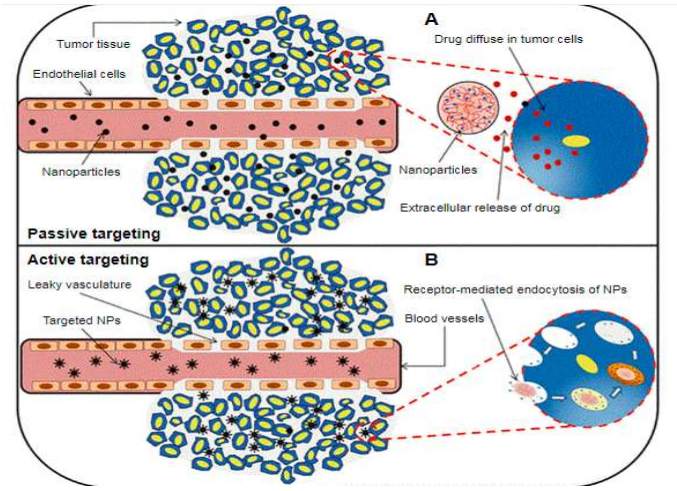

Figure 5. Targeted drug delivery (10)

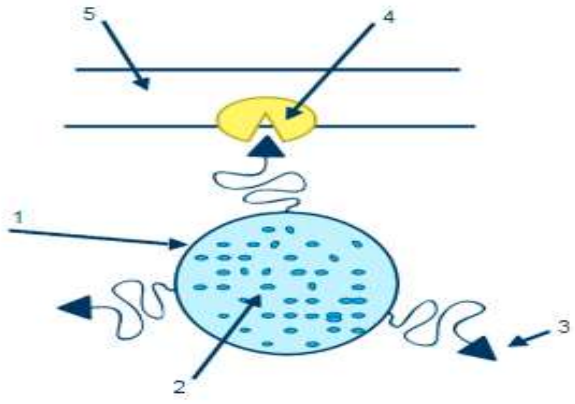

Figure 6. Ligand mediated target delivery (10)

Passive targeting depends mainly on the leaky tumour vessels, while active targeting depends on the presence of ligands on the nanocarrier surface. In Figure 5, (A) drug targeting can be described as passive when nanoparticles diffused via the leaky tumour vessels. while in active targeting (B), drug delivery took place when the ligands of the nanocarriers were attached to the receptor on the tumour cells.In Figure 6, a carrier(1) loaded with the drug (2) is treated with a ligand (3) capable of recognizing the binding positions (4) on the surface of the cell (5) (10). Administering these drugs in a drug delivery system may induce either therapeutic / toxic effects. Furthermore, some drugs do not remain in the targeted sites, long enough to cause the desired therapeutic action.

MATERIALS AND METHODS

History of Chitosan: The first explicit description of chitin was written in 1811 by Professor Braconnot (13). In 1823, Odier found a material with the same general properties as fungine in the cuticle of beetles and designated this material “chitin” after the Greek word “chiton” meaning “coat of mail”. Rouget boiled chitin in potassiumhydroxide and found that it became soluble in organic acids leading to the discovery of chitosan in 1859. Chitin is a cheap and economically wise option used as a biopolymer for its multitude applications. Although chitin is a natural organic polymer, its rate of biodegradation is quite slow in these shell wastes, thereby yielding huge quantities of processing discards. Crab and shrimp shell wastes are the primary source of biomass for the industrial production of chitin and chitosan. Polymers are semi-synthetically derived amino polysaccharides which can be degraded, making it environmentally friendly.

Chitosan: Unique Polysaccharide Structure: Chitin is one of the most abundant organic materials, second only to cellulose in the amount produced annually by biosynthesis. It occurs in animals, particularly in crustacea, molluscs and insects where it is an important constituent of the exoskeleton, and in certain fungi where it is the principal fibrillar polymer in the cell wall.

Chitosan – A Natural Biomaterial: Biomaterial is ‘‘any substance or combination of substances, other than drugs, synthetic or natural in origin, which can be used for any period of time, which augments or replaces partially or totally any tissue, organ or function of the body, in order to maintain or improve the quality of life of the individual’’ as given by the American National Institute of Health (11). Among these polymers, metals, ceramics and composites are most commonly employed. Chitin is a semi-crystalline homopolymer of β-(1→4)-linked N-acetyl-D-glucosamine as shown in Figure 7. The shell of crustaceans consists of 30-40% protein, 30-50% calcium carbonate and calcium phosphate and 20-30% chitin. In chitin, the polysaccharide framework is reinforced and modified bya protein matrix, obtained upto 14-27% and 13-15% of the dry weight of shrimp and crab processing wastes respectively.

Structure of chitosan: Although chitin distributes widely in nature but fungi, Algae, Protozoa, arthropod shells (exoskeletons) are the most easily accessible sources of chitin. Chitin exists in tightly bound complexes with other substances in the cuticles of crabs and shrimps, and some portions of polypeptides are suggested to be linked covalently to a small number of the C-2 amino groups as shown in Figure 8. Chitin exists in tightly bound complexes with other substances in the cuticles of crabs and shrimps, and some portions of polypeptides are suggested to be linked covalently to a small number of the C-2 amino groups. Furthermore, the molecular packing of b-chitin is less tight than that of ordinary a-chitin, and b-chitin can be isolated under similar, but milder conditions. When chitin is treated withconcentrated aqueous sodium hydroxide and mixed with crushed ice, an alkali chitin solution form is the result. In the alkaline homogeneous solution, N -deacetylation proceeds smoothly, and a product with about 50% deacetylation is soluble even in neutral water (15). Chitosan, a partially deacetylated product of chitin, is a copolymer consisting of β-(1→4)-2- acetamido-D-glucose and β-(1→4)-2-amino-D-glucose units, with the latter usually exceeding 80% (16). Biopolymer is a glycosaminoglycan and is composed of two common sugars, glucosamine and N-acetylglucosamine, the proportion of which depends on the alkaline treatment. Chitosan has one primary amine and two free hydroxyl groups for each monomer with a unit formula of C6H11O4N. Being structurally analogous to cellulose, chitosan shows a similar linking in which the hydroxyl at carbon-2 has been replaced by amino groups. Exhibiting

polymorphism, there are 3 forms of crystal types namely α, β and γ known to occur in nature, which differ in the packing and polarities of adjacent chains in successive sheets. Chitin is soluble in very few solvents whereas most of the aqueous acids dissolve chitosan. The multitude of cationic sites formed due to protonation of amino groups by acids along the chitosan chain increases its solubility by increasing both the polarity and the degree of electrostatic repulsion.Crystal structures of two polymorphs of chitosan, tendon (hydrated) and annealed (anhydrous) polymorphs as shown in Figure 9, have beenreported by Clarkand Smith in 1936, later called tendon chitosan prepared from a crab tendon chitin by similar deacetylation reported by Clark and Smith.

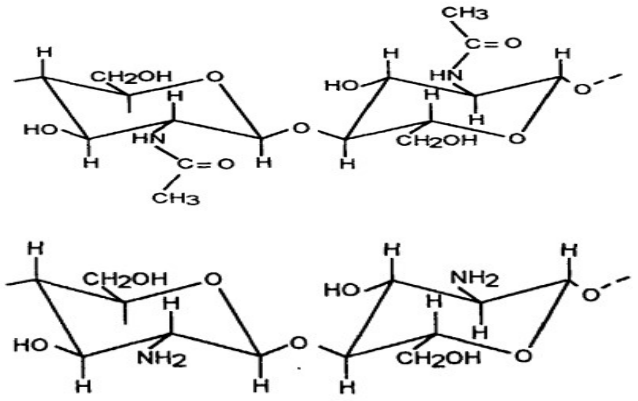

Figure 7. Structure of chitin and chitosan (4)

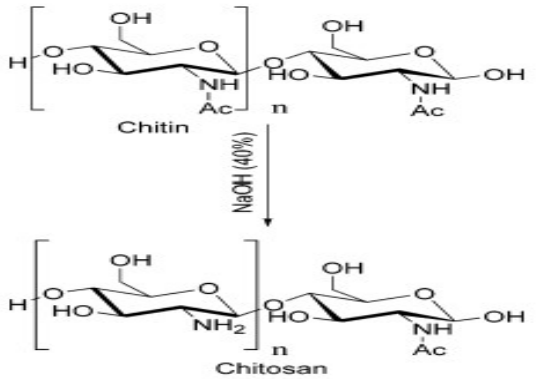

Figure 8. Chitin and its derivatives (14)

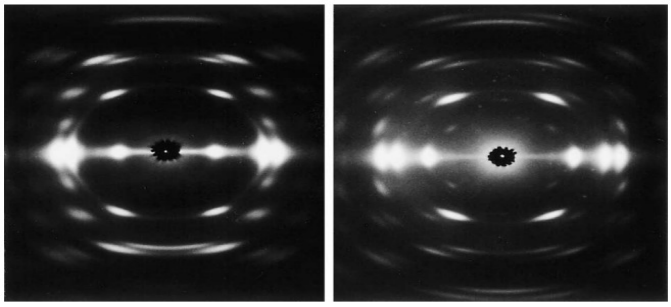

Figure 9. Fibre patterns of tendon and annealed chitosan (17)

In both crystals, chitosan molecule takes up similar conformation (Type I form) to each other, an extended two-fold helix stabilized by intramolecular hydrogen bond, which is also similar to the conformation of chitin or cellulose. Three chitosan conformations other than Type I form as shown in Figure 10, have been found in the crystals of chitosan-acid salts. Type I salts are mostly anhydrous, and in these crystals the backbone chitosan chains retain the extended two-fold helix of the unreacted chitosan molecule. Also conformational change ofchitosan molecule takes place by salt formations of Types II and III (17).

Properties of Chitosan: Chitosan is the only alkaline polysaccharide in nature, whereas the others like cellulose, dextran, pectin are either neutral or acidic. It is nontoxic, odourless, biocompatible and biodegradable. Chitosan is hydrolyzed in vivo by lysozyme to oligomers that activate macrophages to produce N-acetyl-D-glucosamidase, which catalyzes production of NAG, D-glucosamine, and substituted glucosamines from oligomers. Chitosan is characterized by the number of sugar units per polymer molecule (n), which defines its MW. Theaverage MW of chitosan may range from 50 to 2000 kDa. Deacetylation process of chitosan brings about a change in MW, which is inversely proportional to membrane crystallinity. The acetyl content in chitosan is measured by DDA. DDA of chitosan has been shown to correlate with its solubility in acidic solution and the crystallinity of its membrane. It is known that the charge density along the chain increases with an increase in the DDA, and that chain flexibility of chitosan molecules can be manipulated by changing the DDA. Commercially available chitosan has degree of deacetylation ranging from 40 to 98 %. With an increase in deacetylation, the chitosan chain becomes more flexible, thus tending to form a random coil with more intramolecular hydrogen bonds within the chain. Chitosan chains are less entangled and more ellipsoid in shape, and their mechanical properties generally weaker than those of less deacetylated microspheres. In contrast, the less deacetylated chitosan chain was more extended and had stronger intermolecular interactions, which made the chains more entangled (18).Viscosity of chitosan solution increases with concentration and degree of deacetylation and decreases with temperature.

Chitosan is pseudoplastic material, since viscosity decreases with increasing rate of shear. Solubility depends on the pKa and strength of the acidic solvent.At pH value below 4, the amino groups of chitosan are protonated which leads to electrostatic repulsion between charged groups and polymer swelling. At pH 5.2, an unstable structure is generated which upon neutralization with an excess of NaOH, the ionic strength of the solution increases and therefore the size of the aggregates decreases due to compaction of the macromolecular coils.The free amino groups form intermolecular hydrogen bonds with the oxygen of the adjacent chains. At pH above 6.5 (pKa of the amino group), the size of the aggregates increases and phase separation occurs. The polymer coagulates and becomes an amorphous solid. Amino groups make chitosan a cationic polyelectrolyte (pKa ≈ 6.5), one of the few found in nature.

Chitosan is protonated upon dissolution in aqueous acidic medium at pH < 6.5, but when dissolved possesses high positive charge on –NH3 + groups and the resultant soluble polysaccharide is positively charged. Chitosan aggregates with polyanionic compounds and chelates heavy metal ions. Both the solubility in acidic solution and aggregation with polyanions impart chitosan with excellent gel-forming properties (20).

Chitosan Microsphere preparation method (Figure 11)

Emulsion cross linking: This method as shown in Figure 12utilizes the reactive functional amine group of chitosan to cross-link with aldehyde groups of the cross-linking agent. In this method, w/o emulsion is prepared by emulsifying the aqueous solution of chitosan in the oil phase. Aqueous droplets are stabilized using a suitable surfactant. The stable emulsion, thus formed, is cross-linked by using an appropriate cross-linking agent such asglutaraldehyde to harden the droplets. Microspheres are filtered and washed repeatedly with n-hexane followed by alcohol and then dried. This method allows us to control the particle size of the microspheres. The size of final product depends upon the extent of cross-linking agent used while hardening aswell as the speed of stirring during the formation of emulsion.The degree of stirring determines the size of dispersed droplets. By varying any one or both of these parameters, the size of droplets can be changed to obtain the chitosan microspheres.

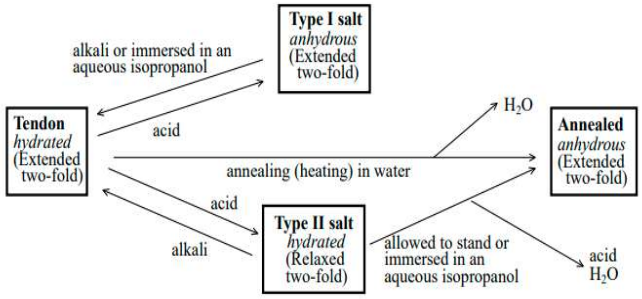

Figure 10. Crystalline Transformation of Chitosan

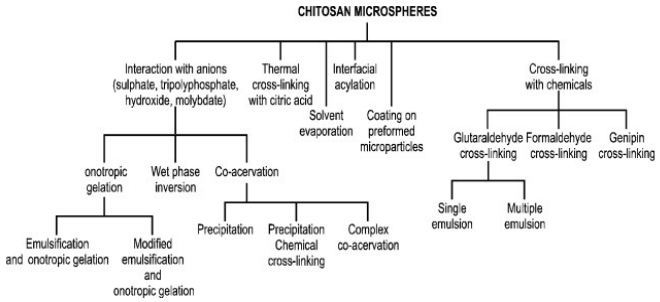

Figure 11. Methods for preparation of chitosan microsphere (21)

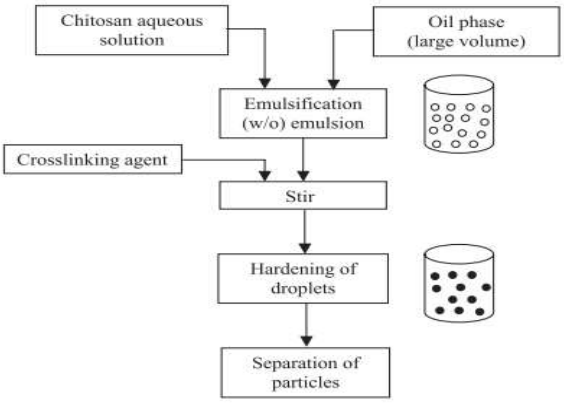

Figure 12. Preparation of chitosan particulate systems by emulsion cross-linking method (22)

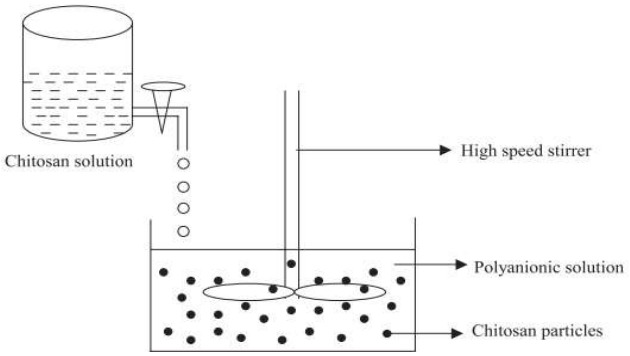

Figure 13. Preparation of chitosan particulate systems by ionic gelation method (22)

Ionotropic gelation method: TPP is a polyanion, which can interact with the cationic chitosan by electrostatic forces as shown in Figure 13. This forms the primary basis of the method of preparation. In the ionic gelation method, chitosan is dissolved in aqueous acidic solution to obtain the cation of chitosan. Complexation between oppositely charged species forms the spherical precipitate however limiting their usage in drug delivery. A modified ionotropic gelation method is reported which makes use of a high voltage electrostatic field to prepare protein-loaded chitosan microspheres using BSA as a model protein. Microspheres prepared by this technique exhibited good sphericity and dispersity (5).

Coacervation / Precipitation method: It utilizes the physicochemical property of chitosan. It is insoluble in alkaline pH medium, but precipitates/coacervates when it comes in contact with alkaline solution as shown in Figure 14. Particles are produced by blowing chitosan solution into an alkali solution like sodium hydroxide, NaOH-methanol or ethane diamine using a compressed air nozzle to form coacervate droplets.

Emulsion-droplet coalescence method: The novel emulsion-droplet coalescence method was developed by Tokumitsu. It utilizes the principles of both emulsion cross-linking and precipitation as shown in Figure 15. However, in this method, instead of cross-linking the stable droplets, precipitation is induced by allowing coalescence of chitosan droplets with NaOH droplets.

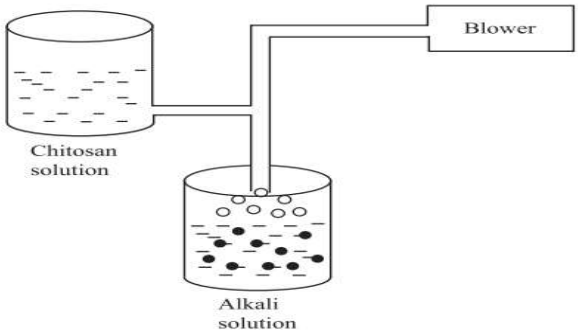

Figure 14. Preparation of chitosan particulate systems by coacervation/precipitation method (22)

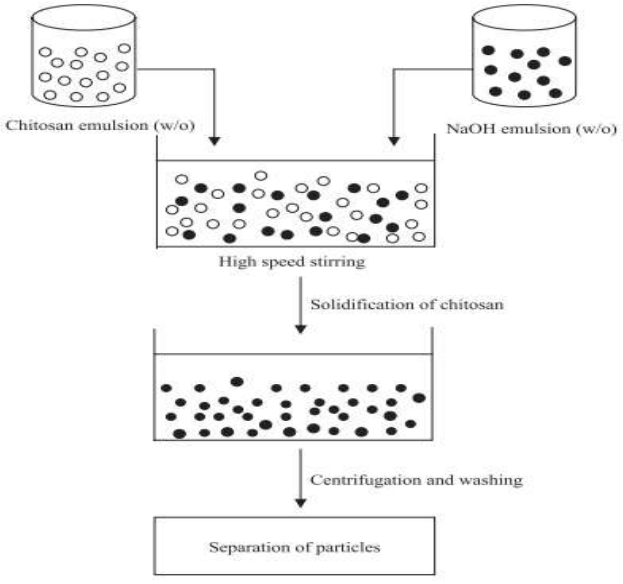

Figure 15. Preparation of chitosan particulate systems by emulsion-droplet coalescence method (22)

Spray drying technique: In this method, chitosan is first dissolved in aqueous acetic acid solution, drug is then dissolved or dispersed in the solution and then, a suitable cross-linking agent is added as shown in Figures 16 and 17. This solution or dispersion is then atomized in a stream of hot air. Atomization leads to the formation of small droplets, from which solvent evaporates instantaneously leading to the formation of free-flowing particles (5).

Reverse micellar method: In this method, the surfactant is dissolved in an organic solvent to prepare reverse micelles as shown in Figure

18. Reverse micelles are thermodynamically stable liquid mixtures of water, oil and surfactant. To this, aqueous solutions of chitosan and drug are added with constant vertexing to avoid any turbidity (5).

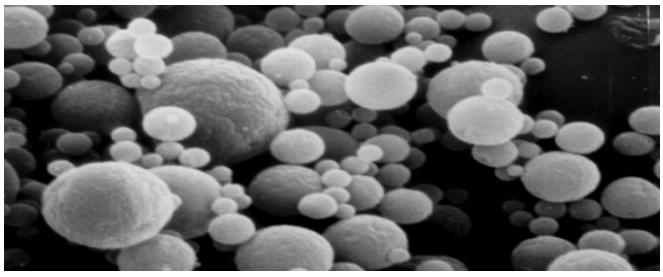

Figure 16. Spray drying producing chitosan microsphere (22)

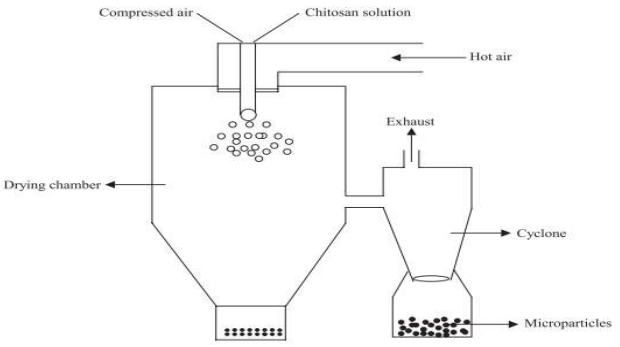

Figure 17. Preparation of chitosan particulate systems by spray drying method (22)

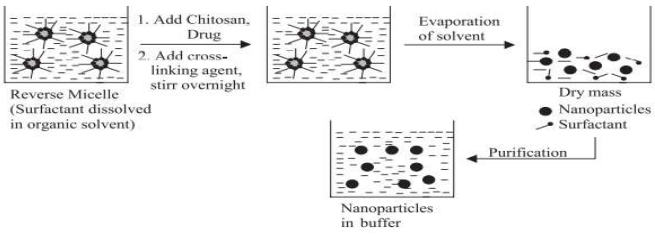

Figure 18. Preparation of chitosan particulate systems by reverse micellar method (22)

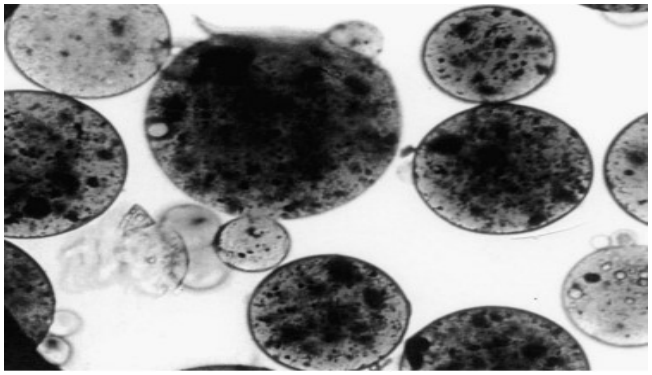

Figure 19. Image representative of optical chitosan microsphere

Characterization of Microspheres: Microspheres are defined as monolithic spheres distributed throughout the matrix either as a molecular dispersion of particles. Their diameter ranges from 1 micron meter to 1000-micron meter. The use of microsphere-based therapy allows drug release curetted based on formulation of various drug–polymer combinations. The total dose of medication and the kinetics of release are the variables, which can be manipulated to achieve the desired result using innovative microencapsulation technologies, and by varying the copolymer ratio, molecular weight of the polymer (21).

Microsphere based systems may increase the life span of active constituents and control the release of bioactive agents. Microspheres have large surface to volume ratios thereby used for controlled release of insoluble drugs. Microspheres can broadly be classified into the following categories:

1. Bio-adhesive microspheres are based on the sticking property as a drug delivery device, exhibiting intimate contact time and therapeutic application at site of application.
2. Magnetic microspheres localize a magnetically targeted drug. Magnetic carriers are used as the support material and they can be easily separated from the reaction medium and stabilized in a fluidized bed reactor by applying a magnetic field. Magnetic carriers can be manufactured using inorganic materials or polymers (23).

Magnetic chitosan microspheres used in targeted drug delivery are expected to be retained at the target site capillaries under the influence of an external magnetic field according to the successful attempt as per the study conducted by Gallo and Hassan, 1988 (21). Morphological and magnetic properties of the microspheres werecharacterized by different techniques ranging from SEM, optical microscopy as shown in Figure 19 to magnetometry. The results demonstrated that the stirring rate of the suspension medium and the Fe3O4 /chitosan ratio are the most effective parameters for the size/size distribution while the chitosan MW has no significant effect on these properties for the given MW range (i.e. 150 to 650 kDa) as shown in Figures 20 and 21. The best magnetic quality of the magnetic chitosan microspheres is around 9.1 emu/g microsphere at 10 kG magnetic field intensity (23). Suspension crosslinking is used for production of chitosan microsphere using glutaraldehyde as a cross linking agent. Magnetic chitosan microspheres were evaluated using parameters like size and distribution and stirring rate.

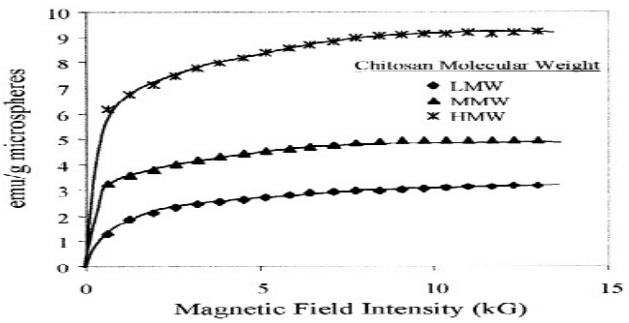

Figure 20. Effects of chitosan molecular weight on the magnetic properties (23)

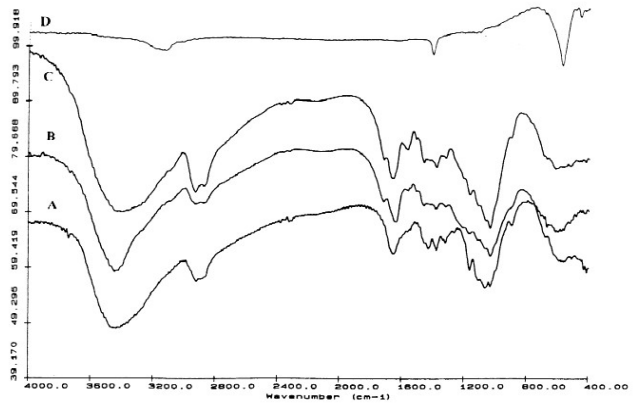

Figure 21. Spectra of (A) chitosan polymer, (B) magnetic and (C) non-magnetic chitosan microspheres, and (D) Fe O particles (23)

RESULTS

This is a detailed literature review so the results will be in terms of the work done as discussed below in review of literature.

Table 1. Biodegradability of chitosan and its derivatives (26)

Sample	Ds	Solubility in h2o	Biodegradation,a%
Chitosan	0.00	No	1.6
30	0.18	YES	67.3
30	0.27	YES	62.5
30	0.46	YES	51.3
31	0.44	YES	8.6
32	0.26	YES	5.0
33	0.29	NOb	24.8
34	0.38	YES	33.6
35	0.24	NO	27.8
36	0.49	NO	7.7
		a Time-21 days	bWater- Insoluble after lyophilization

REVIEW OF LITERATURE

Muzzarelli r *et al* (1987) worked on the biological activity of chitosan as an ultrastructure. He explained the injury of a tissue can be repaired which is possible by the interaction of cellular and extracellular matrix. Chitosan is a polysaccharide structure that exerts morphological functioning similar to extracellular glycoprotein, the inductive capacity of chitosan on connective tissue rebuilding is considered of prime significance. Suranjana Roy *et al* in 1992 investigated the novel procedure of controlled release preparation of microspheres. In vitro determination of the release kinetics of one water soluble and one water insoluble polymer i.e., ethyl cellulose and methylcellulose were studied. A double emulsification technique was used to prepare the microsphere. After dissolution, size of channels has increased considerably, while size of microsphere appears intact (31). Emir bakidenkbas *et al* (2002) studied the magnetic property of chitosan and method of preparation. Manufacturing of chitosan spheres can be done using inorganic materials which provides good mechanical resistance, resistance of microbial attack which lacksenough functional polymers for binding.Overcoming this pitfall, polymers can be used which provide variety of surface functional groups which can be tailored to specific applications. Micrographs were studied to deduce the morphology of fe3o4 and magnetic chitosan microspheres determining their spherical shape rather than a smooth surface. Varying size ranges of the magnetic microspheres were obtained and parameters like stirring rate, molecular weight, fe3o4 and chitosan ratio were determined using a vibrating sample magnetometer. Sinha v r *et al* (2004) studied Chitosan microsphere as a potential carrier for drug delivery. The study proves interaction of chitosan with low molecular counterions such as polyphosphates, sulphates and crosslinking with glutaraldehyde. the size of the chitosan microsphere can be changed according to the site for oral, nasal and parenteral delivery. Loading of the drug is done by using the swelling properties of the microspheres to the drug solution. The release of drug from chitosan microsphere is multifactorial ranging from properties like molecular weight, concentration of chitosan, crosslinking ability. Therapeutic benefits such as anticancer, anti-inflammatory, antibiotics are integrated into chitosan microspheres to achieve a controlled drug release.

Agnihotri s *et al* (2004) studied on advances on chitosan based on its nanoparticle structure. Methods of preparation range from spray drying, emulsion crosslinking and precipitation. Trimethyl chitosan chloride, a quaternized chitosan derivative proved to increase theefficiency i.e enhance the permeation of hydrophilic macromolecular drug across mucosal epithelia via tight junctions.

As per the study conducted by Berthold *et al* in 1996, incorporation of the drug onto previously formed chitosan microspheres was performed, prednisolone sodium phosphate was adsorbed to previously manufactured chitosan microspheres. The drug adsorption was found to be dependent upon the initial drug concentration. A higher initial concentration led to a higher loading efficiency. It was also observed that lipophilic steroid was adsorbed in lower amounts as compared to their hydrophilic derivatives. Since the encapsulation of isoniazid tends to be limited by its hydrophilic characteristics, therefore, the drug was adsorbed onto pre-formed chitosan alginate microspheres prepared by complex coacervation as per Lucinda-Silva and Evangelista, 2003 (21). Jalehvarshosaz *et al* (2007) studied the numerous applications in field of waste water treatment, agriculture, fabric and textiles, based on the physiochemical and biological properties in addition to its biocompatibility, biodegradability properties. The author talks about the nasal delivery of drug obtained via microspheres composed of sodium alginate, chitosan hydrochloride or both using a spray drying method, based primarily on the penetration enhancing properties of chitosan via the opening of tight junctions. Magnetic carrier technology for carrier delivery, marks a possibility for drug targeting. Chitosan microspheres of 100–250-micron meter have been prepared by suspension crosslinking method. Conclusively it has been used widely for its mucoadhesive property, and to maintain antibacterial activity of the drug. Hejazi and amiji in 2002, used two different methods for drug loading. In the first method, tetracycline was mixed with chitosan solution before the simultaneous crosslinking and precipitation. In the second method, the drug was incubated with pre-formed microspheres for 48 hours. When the drug was added to the polymer solution before crosslinking and precipitation, only 8% was optimally incorporated. On the other hand, when the drug was incubated with the pre-formed microspheres, a maximum of 69% could be loaded. This signifies that the drug can be adsorbed on to the chitosan microspheres to a greater extent using the latter method (21).

Prabhakaran m *et al* (2008) studied the chitosan particle as a colloidal structure and analysed the evidence of its efficacy in improving the transport of associated molecule through the mucosa and epithelia. The study concluded its importance in drug delivery and gene therapy. Rebecca R. Klaussner *et al* in 2008 provided evidence on the corelation of chitosan's rheological property and its ability to Electro spin. TFA is most often used as the solvent to successfully electro spin chitosan. The attempt was made to electro spin pure chitosan (molecular weight 148000 g/mol) with a DD of 75-85% in a concentrated acetic acid solvent system. Additionally, chitosan-PEO blends were electro spun into defect free nanofiberswith diameters in the range of 62 to 129 nm. Time studies were also carried out to determine the effect on the $\eta 0$, and the electro spinnability of chitosan blended solutions. Electrospinning chitosan with high degrees of deacetylation (DD) is especially challenging because it is a cationic polyelectrolyte, resulting in a much higher solution viscosity than a neutral polymer of similar chain length. As the charge on the chain increases, the conformation of the chain in solution expands and the viscosity increases substantially. Therefore, the degree of deacetylation is an extremely important parameter to consider when attempting to electro spin chitosan. Multiple attempts proved Chitosan-PEO blends can be electro spun to create a mesh constructed of defect-free, nanofibers with diameters ranging from 62 to 129 nm, which would be suitable for use in biomedical applications. Of the samples studied, chitosan with the lowest molecular weight (148000 g/mol) was most suitable to electro spin. Increasing the total polymer (chitosan + PEO) concentration in solution reduces the number of beads, while increasing the chitosan concentration decreases fibre diameter. For the chitosan-PEO blends, reducing the acetic acid concentration from 45% to about 30% reduced the number of bead defects in the electro spun fibres, possibly by altering the conformations of the polymers and the conductivity of the solutions. Over time, the chitosan-PEO blended solutions phase separate and are unable to be electro spun. As a result, blended solutions should be electro spun within 24 hours of initially being blended to

minimizecomplications. The addition of NaCl stabilizes the blended solutions and increased the time the blended solutions could be stored before electrospinning (19).

Wang *et al* (2011) worked on the preparation of Chitosan – ibuprofen sustained release microsphere. The microspheres were prepared using crosslinking and samples were characterized by Ems (electron microscope) and UV spectrophotometer. Tarunkumarvarun *et al* (2017) talk about the methods of chitin and chitosan extraction using commercially and antimicrobial properties of chitosan oligomers. It involves process of degradation of shrimp shell waste into powder followed by demineralization, deproteination and deacetylation to chitosan. The study exhibited antimicrobial action of chitooligomers against many gut pathogens and thereby enhance the gut health. Thus, chitooligomers as feed additive may replace antibiotics in the animal feed which in turn helps in dealing with problems of antibiotic residue in the animal products. Ava M. Vargason *et al* (2021) put forth in their paper the classes of therapeutic and delivery paradigms, the three core paradigms of drug delivery that span all classes of therapeutics: drug modifications, microenvironment modifications and drug delivery system. Drug delivery has evolved along generations of therapeutics ranging from small molecules to proteins and peptides, to nucleic acids and, most recently, to live-cell therapies. Sathyanmyana M. Upadrashta *et al* (1992) evaluated chitosan as a binder for chlorpheniramine maleate tablets in comparison with other cellulose binders. The effects of binder concentration on the mechanical properties of granules and tablets as well as on disintegration time and dissolution profiles were studied. Results showed that granules prepared with methylcellulose had lowest percentage of fines and friability. Chitosan tablets showed best dissolution profiles. The rank order correlation for binder efficiency was: hydroxypropyl methylcellulose > chitosan > methylcellulose > sodium carboxymethylcellulose. Kumaresh S. Soppimath *et al* (2000) studies the contributions in the field of biodegradable polymeric nanoparticles used as drug delivery systems. Preparation of the nanoparticle is done by either polymerization of the monomers or dispersion of preformed polymers. The various processes of drug modification and opsonization responsible for coating of nanoparticles and their passage through penetration barriers was dealt with(24).

Vladimir E. Tikhonov et al (2005) evaluated chitosan as a starting material, a low molecular weight (LMW) chitosan with a high solubility and low viscosity in water at physiologically acceptable pH values. LMW chitosan and its DDC-derivatives showed high activity against bacteria, yeast and fungus. Suppressed fungal colony growth and inhibited fungal spore germination at 0.01% (w/v) concentration was observed (25). Jeong *et al* in 2005 evaluated the Chitosan structural formula to be a linear amino polysaccharide randomly distributed (1–4) linked D-glucosamine, N-acetyl-D-glucosamine units, found in the exoskeleton of lobster, crabs, and shrimp (28). Lehr *et al* in 1992 and his colleagues studied the in vitro mucoadhesive nature and solubility of pH dependant polymer chitosan and its amino group formed by electrostatic interaction between a polyion and counterions which become uneven in gastric fluid. This issue was resolved by irretrievable chemical cross-linking by cross-linker GA used at different concentrations to modulate the release of drugs from NPs (29). Tae Hwan Park *et al* studied Artecoll, a second-generation PMMA based filler, which is widely used in the permanent correction of prominent nasolabial folds, marionette wrinkles, and deep glabellar wrinkles using a carrier as microsphere filler. Most significant complication following PMMA microsphere implantation is granuloma formation. Great research using similar microsphere fillers is being done for aesthetic plastic surgery (30).

DISCUSSION

Drug delivery systems using chitosan as a carrier NP are in enormous usage, since they are able to load a wide range of natural and chemical agents including proteins, oligosaccharide, and anticancer drugs. Various routes of administration occur ranging from nasal, intravenous, oral and ocular. NPs can potentially target specific tissues, hence cell specific targeting of NPs seems to be a promising way to increase local drug concentration, prevent non-specific interactions and decrease the toxicity and side effects of systemic administration. The concentration of the medicinal formula should lie between minimal toxic concentrations and the minimal effective concentration. Moreover, drug carriers enhance the pharmacokinetic effect, protect the medicinal agent from degradation via enzymes, and carry lipophilic and hydrophilic drugs to meet the intended usage of the system. Targeted drug delivery is an approach to deliver the therapeutic agents to an intended organ or tissue to increase the efficacy and reduce toxicity. Two essential requirements must be fulfilled to have a successful drug delivery system. First, the system must have a minimal loss of activity and dose in the blood circulation system. Second, the therapeutic formula should act only on the desired tissues without harming other healthy cells. Drug delivery systems based on diffusion mechanisms are driven and controlled via a concentration gradient. These strategies of targeting NP's, not only cause the lower dosage of required drugs, but also lead them directly to the receptors. ElhamRostami *et al* 2020 presented an extensive view of chitosan NPs, discussing their types such as antibody, magnetic and pH sensitive conjugated chitosan. MNPs have been used for medical purpose during the last two decades (32). The structure of MNPs used for drug delivery systems is mainly referred to its magnetic core and surface coating. The surface coatings minimize the aggregation of MNPs under physiological conditions major application being cancer therapy and have been shown to increase the efficacy of chemo- and/ or radio-therapy in clinical trials (10). Chitosan a biodegradable natural polymer in form of a microsphere offers enormous pharmaceutical applications due to its biocompatibility, high charge density, non-toxicity and mucoadhesion. It not only improves the dissolution of poorly soluble drugs but also exerts a significant effect on fat metabolism in the body. Various techniques used for preparing chitosan microspheres and evaluation of these microspheres, have also been reviewed. In this review we highlight the biocompatibility, biodegradability, its wound healing and its hemoadsorbant properties. In 2004, M. N. V. Ravi Kumar *et al* studied the biodegradability which was enhanced by chemical modification compared with original chitosan. As shown in the table 1, various modified chitosan forms were included in the study and compared to the original chitosan.

Varied forms ranging from fluorinated, trimethylated to sugar modified chitosan were compared. Excellent biodegradability was shown in 30 with various DS, although it gradually decreased with increasing DS. Derivatives 33, 34, and 35 modified with PEG, quaternary ammonium, and amido groups also showed good biodegradability. Moderate biodegradation was shown by 31 and 36 bearing hydroxyethyl and nitrile groups. These results suggest that biodegradation was associated with the chemical structure of chitosan derivatives (26). Sinha VR *et al* in 2004, worked on the factors that affect the entrapment efficiency and release kinetics of drugs from chitosanmicrospheres. Commercially, chitosan is available in the form of dry flakes, solution and fine powder. It has an average molecular weight ranging between 3800 and 2,000,000 and is from 66 to 95% deacetylated. Chitosan microspheres are used to provide controlled release of many drugs and to improve the bioavailability of degradable substances such as protein or enhance the uptake of hydrophilic substances across the epithelial layers.

Mizuno et al. studied the stability of bFGF incorporated into a chitosan film as a delivery vehicle for providing sustained release of bFGF. The therapeutic effect of this system on wound healing in genetically diabetic mice was determined as an animal model for treating clinically impaired wound healing. Growth factor was incorporated into chitosan films before drying by mixing bFGF solution with the hydroxypropyl-chitosan solution. Chitosan film or bFGF-chitosan film was applied to full-thickness wounds created on the backs of diabetic mice. Results showed that the wounds were

smaller in day 20 in the bFGF-chitosan group than in chitosan alone group. Proliferation of fibroblasts and an increase in the number of capillaries were observed in both groups, but granulation tissue was more abundant in the bFGF-chitosan group (27). Growth factors instrumentally affect the inflammatory, proliferation and migratory phases of wound healing. A variety of growth factors have been reported which participate in the process of wound healing including: EGF,PDGF, FGF, TGF-β1, IGF-1, human growth hormone and granulocyte–macrophage colony-stimulating factor. Studies have shown that polyethylene oxide containing chitosan microspheres were used as potential blood compatible hemoadsorbants(21). Hemcon dental dressing seems to reduce postoperative side effects and obtain rapid soft tissue healing. Their results showed better and early wound healing and reduced incidence of post extraction complications because of its biocompatibility, isolating and anti-inflammatory ability and supporting the formation of blood clot in the tooth socket. Akshat Gupta *et al* in 2008 studied chitosan's ability to promote wound healing and induce bone formation. Chitosan is effective in promoting wound healing and early osteogenesis in erupted tooth socket after extraction. Chitosan's properties of binding, with red blood cells allows it to rapidly clot blood, thereby marking its haemostatic actions.

Chitosan has pronounced antimicrobial effects due to destabilization of the outer membrane of gram-negative bacteria and permeabilization of the microbial plasma membrane. In addition, chitosan modulates the functions of inflammatory cells and subsequently promotes granulation and organization. Chitosan and its derivatives accelerate wound healing by enhancing the functions of inflammatory cells such as PMNleukocytes, macrophages, fibroblasts or osteoblasts. It has also been reported that chitosan could increase the tensile strengthof wounds. Noel *et al* evaluated chitosan film as a potential localized drug delivery carrier, which does not require later removal owing to the biodegradability of chitosan. Currently, chitosan derived haemostatic agents have been used for post-extraction bleeding control in patients. It has been reported that chitosan increased its haemostatic capability. The anticoagulant activity of chitosan seems to be positive charge dependent due to amino groups on chitosan which actually interact with negatively charged membranes of red blood cells. Molecular weight of chitosan may also affect the binding or clumping of red blood cells. During a study conducted by Yang et al, when blood sample was mixed with chitosan solution, the erythrocytes were distorted after aggregation. Degree of deacetylation of chitosan may also affect wound's haemostasis (4). Chitin and chitosan and their derivatives have analgesic effect on inflammatory pain but in the later years, others have studied that analgesic effect by chitosan is due to intra-peritoneal administration of acetic acid. Moreover, it was proposed that chitosan showed a greater analgesic ability than chitin due to its polycationic nature. The difference in analgesic activity of chitosan involves the absorption of proton ions by chitosan which are released in the inflammatory area. The free primary −NH2 groups on chitosan protonatein the presence of acidic conditions which reduce the pH of effected area and cause the effective analgesic influence (4).

Drug delivery systems using chitosan NPs are in enormous usage, since they are able to load a wide range of natural and chemical agents including proteins, oligosaccharide, and anticancer drugs. In additions, chitosan NPs can be applied via various routes of administration as nasal, intravenous, oral and ocular. NPs can potentially target specific tissues, hence cell specific targeting of NPs seems to be a promising way to increase local drug concentration, prevent non-specific interactions and decrease the toxicity and side effects of systemic administration. The concentration of the medicinal formula should lie between minimal toxic concentrations and the minimal effective concentration. Moreover, drug carriers enhance the pharmacokinetic effect, protect the medicinal agent from degradation via enzymes, and carry lipophilic and hydrophilic drugs to meet the intended usage of the system. Targeted drug delivery is an approach to deliver the therapeutic agents to an intended organ or tissue to increase the efficacy and reduce toxicity. Two essential requirements must be fulfilled to have a successful drug delivery system. First, the system must have a minimal loss of activity and dose in the blood circulation system. Second, the therapeutic formula should act only on the desired tissues without harming other healthy cells. Drug delivery systems based on diffusion mechanisms are driven and controlled via a concentration gradient. Targeted delivery using carriers at the site of action can be achieved by modifying NPs using peptides, antibodies or small molecules. These strategies of targeting NP's not only cause the lower dosage of required drugs, butalso lead them to the receptors. ElhamRostami *et al* 2020 presented an extensive view of chitosan NPs, discussing their types such as antibody, magnetic and pH sensitive conjugated chitosan. MNPs have been used for medical purpose during the last two decades (32). The structure of MNPs used for drug delivery systems is mainly referred to its magnetic core and surface coating. The surface coatings minimize the aggregation of MNPs under physiological conditions major application being cancer therapy and have been shown to increase the efficacy of chemo- and/ or radio-therapy in clinical trials (10).

CONCLUSION

Novel drug delivery is comprised of three major components: a drug, a targeting moiety, and a carrier system. A therapeutic agent is either encapsulated via passive absorption or chemical conjugation into the carrier as the drug carrier system is significantly produced, its effects on the pharmacokinetics and pharmacodynamics of the drug, so it must be selected with great care. Specific drug delivery functionalities can be therefore achieved using polysaccharides that have shown stimuli responsiveness as native and/or derivatized. As the entrapment efficiency of drugs in the chitosan microspheres is directly dependent upon the chitosan concentration, multifunctionality can be conferred on them, enhancing their ability to respond to stimuli, diagnose, image, and treat with controlled release of therapeutic agents over time as single devices (1). Chitosan being a versatile polymer offers the targeted drug delivery by improving the dissolution rate of poorly soluble drugs and thus can be exploited for bioavailability enhancement of such drugs. Reacting chitosan with controlled amounts of multivalent anion results in crosslinking between chitosan molecules and forms the basis of most of the preparation techniques. (21). Apart from crosslinking, chitosan microspheres have also been prepared by a number of other processes, coacervation, multiple emulsion method, solvent evaporation, etc as mentioned in ourreview. The particle size of chitosan microspheres can be modified approximately for application through the oral, nasal and parenteral route of delivery.

Conclusively since chitosan combines unique physicochemical characteristics, in vivo biodegradability, biocompatibility, and antimicrobial action, it has been widely investigated over the last few years for potential applications as a drug carrier for many possible routes of administration. Nasal drug delivery represents an interesting alternative to the parenteral route for administration of drugs that show poor oral bioavailability, such as peptides and proteins. Chitosan is able to swell and form a gel-like layer in an aqueous environment which is favourable for interpenetration of polymer and glycoprotein chains of the mucus. Parenteral, nasal and transdermal route of delivery surpasses first pass metabolism however the oral route as explained in this review remains the primary mode of administration due to the gel-forming property of the polysaccharide at low pH, along with its antacid and antiulcer properties along with it ease of administration.

Source(s) of support/ Funding statement: NA

Conflicting Interest (If present, give more details): No conflict of interest.

REFERENCES

Agnihotri S.A., Mallikarjuna N.N., Aminabhavi T.M. November 2004. Recent advances onchitosan-based micro- and nanoparticles in drug delivery. Journal of Controlled Release.100(1):5–28.

Albinali K.E., Zagho M.M., Deng Y., Elzatahry A.A. March 2019. A perspective on magnetic core–shell carriers for responsive and targeted drug delivery systems. *International Journal of Nanomedicine.* 14:1707–23.

Barakat N.S. October 2009. Magnetically modulated nanosystems: a unique drug-delivery platform. *International Journal of Nanomedicine.* 1;4(7):799–812.

Bergmann C.P., Stumpf A. 2013. Dental ceramics, topics in mining, metallurgy and materials engineering.Springer, Berlin.

Braconnot H. 1881. British nutrition foundation journal.79:265–304.

Chen Y.l.2008. Preparation and characterization of water-soluble chitosan gel for skin hydration. University Sains Malaysia Thesis. 1 - 48.

Dai T., Tanaka M., Huang Y.Y., Hamblin M.R. July 2011. Chitosan preparations for wounds and burns: antimicrobial and wound-healing effects. *Expert Review of Anti Infective Therapy.* 9(7):857–879.

Denkbaş E.B., Kiliçay E., Birlikseven C., Öztürk E. February 2002. Magnetic chitosan microspheres: preparation and characterization. Reactive and Functional Polymers. 50(3):225–232.

Felt O., Buri P., Gurny R. January 1988. Chitosan: A Unique Polysaccharide for Drug Delivery. Drug Development and Industrial Pharmacy. 24(11):979–993.

Habibi Y., Lucia L.A. 2012. Polysaccharide Building Blocks: A Sustainable Approach to the Development of Renewable Biomaterials. John Wiley & Sons.pp430.

He P., Davis S.S., Illum L. May 1998. In vitro evaluation of the mucoadhesive properties of chitosan microspheres.International Journal of Pharmacy. 166(1):75–88.

Klossner R.R., Queen H.A., Coughlin A.J., Krause W.E. October 2008. Correlation of Chitosan's Rheological Properties and its ability to electrospin. Biomacromolecules.9(10):2947–2953.

Kruk M.G., Veit K., Huebner F. 2015. History and Possible Uses of Nanomedicine Based on Nanoparticles and Nanotechnological Progress. *Journal of Nanomedicine Nanotechnology*. [cited 2021 Nov 7];06(06).

Kumar M.R., Muzzarelli R. A., Muzzarelli C., Sashiwa H., Domb A.J. December 2004. Chitosan chemistry and pharmaceutical perspectives. Chemical Reviews. 104(12):6017–6084.

Kurita K. June 2006. Chitin and Chitosan: Functional Biopolymers from Marine Crustaceans. *Marine Biotechnology journal.* 8(3):203–26.

Mahmoudi K., Bouras A., Bozec D., Ivkov R., Hadjipanayis C. 2018. Magnetic hyperthermia therapy for the treatment of glioblastoma: a review of the therapy's history, efficacy and application in humans. International Journal of Hyperthermia. 34(8):1316–28.

Mitra A., Dey B. 2011. Chitosan Microspheres in Novel Drug Delivery Systems. *Indian Journal of Pharmaceutical Sciences.* 73(4):355–366.

MuzzarelliA. February 1997. Human enzymatic activities related to the therapeutic administration of chitin derivatives: Cellular and Molecular Life Sciences. 53(2):131–40.

Ngwuluka NC. 2018 Responsive polysaccharides and polysaccharides-based nanoparticles for drug delivery.Stimuli Responsive Polymeric Nanocarriers for Drug Delivery Applications. *Woodhead Publishing;* [cited 2021 Nov 12], 1: 531–554.

Ogawa K., Yui T, Okuyama K. April 2004. Three D structures of chitosan. *International Journal of Biological Macromolecules.* 34(1):1–8.

Paderni C., Compilato D., Giannola L.I., Campisi G. September 2012. Oral local drug delivery and new perspectives in oral drug formulation.Oral Surgery Oral Medicine Oral Pathology Oral Radiology Journal. 114(3):25–34.

Park T.H., Seo S.W., Kim J.K., Chang C.H. April 2012. Clinical Experience with Polymethylmethacrylate Microsphere Filler Complications. *Aesthetic Plastic Surgery.* 36(2):421–426.

Rabea E.I., Badawy M.T., Stevens C.V.., Smagghe G, Steurbaut W. November 2003 Chitosan as Antimicrobial Agent: Applications and Mode of Action. Biomacromolecules. 4(6):1457–65.

Roy S., Pal M., Gupta BK. September 1992. Indomethacin-Loaded Microspheres: Design and Preparation by a Multiple-Emulsification Technique and Their in Vitro Evaluation. PharmaceuticalResearch.9(9):1132–6.

Sang Y. H., Eun Lee J., Chung H., Chan Kwon I., Young Jeong S. March 2005. Self assembled nanoparticles containing hydrophobically modified Glycolchitosan for gene delivery. *Journal of Controlled Release.* 103(1):235–43.

Sinha V.R., Singla A.K., Wadhawan S., Kaushik R., Kumria R., Bansal K, et al. 2004. Chitosan microspheres as a potential carrier for drugs. *International Journal of Pharmacy*. 274(1–2):1–33.

Siri JGS., Fernando C. N., Silva ND. July 2017.Review: Chitosan Nanoparticles for Effective and Safe Drug Delivery:Potential Big Deal in Intellectual Property Business. Journal of Science and Technological ResearchSharda University.7(1):1–9.

Soppimath K.S., Aminabhavi T.M., Kulkarni A.R., Rudzinski W. E. January 2001. Biodegradable polymeric nanoparticles as drug delivery devices. *Journal of Controlled Release*. 70(1–2):1–20.

Tewabe A., Abate A., Tamrie M., Seyfu A., AbdelaSirajE.July 2021, Targeted Drug Delivery — From Magic Bullet to Nanomedicine: Principles, Challenges, and Future Perspectives. *Journal of Multidisciplinary Healthcare,* 14:1711–1724.

Tikhonov V.E., Stepnova E.A., Babak V.G., Yamskov I.A., Palma-Guerrero J., Jansson H.B. et al. 2006. Bactericidal and antifungal activities of a low molecular weight chitosan and its N-/2 (3)-(dodec-2-enyl) succinoyl/-derivatives.Carbohydrate Polymers.64(1):66–72.

Varun T.K., Senani S., Jayapal N., Chikkerur J., Roy S., Tekulapally V.B, et al. February 2017. Extraction of chitosan and itsoligomers from shrimp shell waste, their characterization and antimicrobial effect. Veternary World. 10(2):170–175.

Yoon H.G., Kim H.Y., Lim Y..H, Kim H.K., Shin D.H., Hong B.S., et al. September 2000. ThermostableChitosanase from Bacillus sp. Strain CK4: Cloning and Expression of the Gene and Characterization of the Enzyme. Applied and Environmental Microbiology.66(9):3727–3734.

****