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A REVIEW ON: CHITOSAN MICROSPHERE FOR COLONIC DRUG DELIVERY: INFLAMMATORY BOWEL DISEASE

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ABSTRACT

The objective of following review is to elaborate chitosan microsphere, chitosan is natural polymer used as potential carrier for many drugs and also for many drug delivery such as topical, parenteral, buccal, targeted drug delivery. The colonic drug delivery is highly desirable for local treatment of a variety of bowel diseases such as ulcerative colitis, Crohn's disease, ameabiosis, colonic cancer, and systemic delivery of many drugs which degrades in stomach and small intestine, such as proteins and peptides. Colon is the distal organ of the GIT, hence difficulty arises in colonic drug delivery. However there are many techniques for colon specific drug delivery namely prodrugs, pH dependent, time dependent, microbially triggered system, pressure controlled, osmotic controlled, multiparticulates system such as microparticle, nanoparticle, pellets etc. Microparticles is preferred over unit dosage form as the system enables the drug to reach colon quickly & retained in it for long period of time. Using different techniques of microparticles targeted drug delivery achieved. Due to this dose of drug and side effects minimizes.

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INTRODUCTION

Colon targeted drug delivery systems, have been the focus point of formulation because the colon is considered as a suitable site for delivery both conventional & labile molecules & it is also a site for some specific diseases such as, ulcerative colitis, Crohn's disease, bowel cancer, some infections & constipation[1]. Various approaches have been used for oral delivery of drug to the colon which include time dependent delivery, pH dependent system, biodegradable polymers. Colonic delivery is a good candidature for delivery of proteins peptides and vaccines where the enzymatic degradation and the hydrolysis of proteins can be minimized and increases the systemic bioavailability. Drug delivery to the colon is beneficial not only for the drug delivery of proteins & peptide drugs (degraded by digestive enzymes of stomach & small intestine) but also for the delivery of low molecular weight compounds used to treat diseases associated with the colon or large intestine. Targeted drug delivery to the colon, by means of combination of one or more controlled release mechanisms, hardly release drug in upper part of GIT but release in colon following oral route. Specifically delivering drug to the colon, a lot of benefits would be acquired in terms of improving safety & reducing toxicity when treating local & systemic chronic diseases.

Appropriate local targeting is the main challenge in the treatment of ulcerative colitis. For improved localization, a well designed drug delivery system is beneficial to enhance therapeutic efficacy. The drugs commonly used are anti-inflammatory agents, 5-aminosalicylates and corticosteroids for the treatment of moderate and severe IBD and immunosuppressive agents for the treatment of the disease in severe stages. A major challenge in the therapy of IBD is the prevention and reduction of drug-related side effects. As described above there are many approaches used in CTDDS (Colon targeted drug delivery system). Among all, microparticulate system is one of the best approaches for controlled drug delivery in specific site of inflammation. Microparticles are small free flowing particles consisting of natural or synthetic polymers having particle diameter ranging from 1 to 1000 µm.

Pharmaceutical invention and research are increasingly focusing on delivery systems which enhance desirable therapeutic objectives while minimizing side effects. The last two decades there has been a remarkable improvement in the field of novel drug delivery systems. Carrier technology offers an intelligent approach for drug delivery by coupling the drug to a carrier particle such as microspheres, nanoparticles, liposomes, etc which modulates the release and absorption characteristics of the drug.

Factors affecting colon targeted drug delivery:

Generally includes physiological factor & pharmaceutical factor.

Physiological factor:

- Gastric emptying.
- pH of colon.
- Colonic microflora.

Pharmaceutical factor:

a) Drug carrier
- Partition coefficient
- Chemical nature
- Stability

b) Polymers used in colon targeting

Inflammatory bowel diseases (IBD):

Inflammatory bowel diseases are the idiopathic chronic multifactorial inflammatory diseases of gastrointestinal tract, which mainly include ulcerative colitis and Crohn's disease. Small intestine and large intestine or colon are the main regions involved in inflammatory bowel diseases, which are marked by the chronic inflammation in specific mucosal or transmural locations. At present, the etiology of disease is not fully understood but it has been hypothesized that various factors such as genetic, gut environmental, psychosomatic, autoimmune, epidemiological are responsible for the development of ulcerative colitis. Gut environmental factors include immune/epithelial interactions, bacterial infections, and epithelial barrier functions. Epidemiological studies include dietary habits, smoking habits, intake of drugs, hormonal status, variations due to different climates, and changes due to social circumstances[5].

Signs & Symptoms:

The chief initial symptoms of onset of ulcerative colitis consists of severe abdominal pain, tenesmus, anorexia, bloody or mucous diarrhea in contrast to Crohn's disease, which does not show any bloody diarrhea. The severe symptoms include weight loss, tachycardia, anemia, rectal bleeding, and bowel distension.[5]

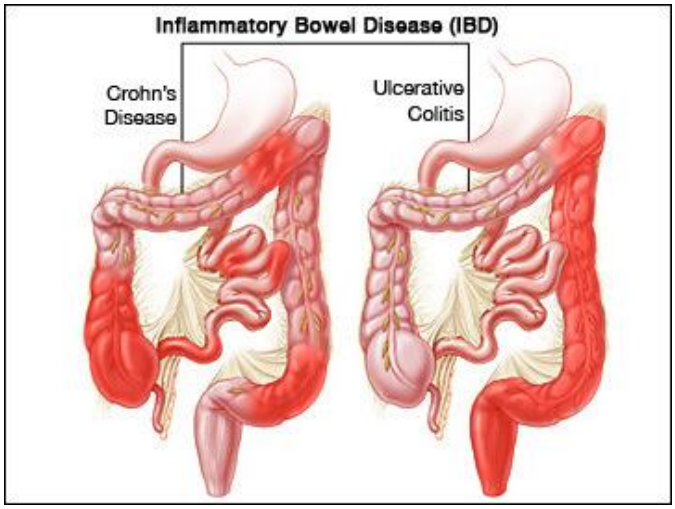


Fig: Shows difference between Crohn's disease & ulcerative colitis.

Table No. 1 Comparison between Ulcerative colitis & Crohn's disease.

Features	Ulcerative colitis	Crohn's disease
Site of infection	Extending from rectum to entire colon	Any part of GIT (mainly ileum is involved)
Pathology & complication	Mucosal inflammation, if get severe can cause colon cancer. Fistulas, abscesses and strictures absent	Transmural inflammation, non caseating granulomas, fistulas, abscesses, perianal involvement and strictures are common.
Distribution	Continuous inflammation	Discontinuous
Symptoms	Diarrhea, wt. loss, malnutrition and other Extra intestinal manifestations, smoking improves condition	Diarrhea, abdominal pain, wt. loss, malnutrition, growth failures in kids, smoking make it worse
Drugs	Prednisolone, Budesonide, Metronidazole, Sulfasalazine.	Azathioprine, Osalazine, Budesonide.

Chitosan

Chitosan is a cationic unbranched polysaccharide consisting of copolymers of d-glucosamine and N-acetyl-d-glucosamine units linked by β-(1–4)-glycosidic linkages. It is obtained by deacetylation of chitin generally extracted from crustacean shells [1].

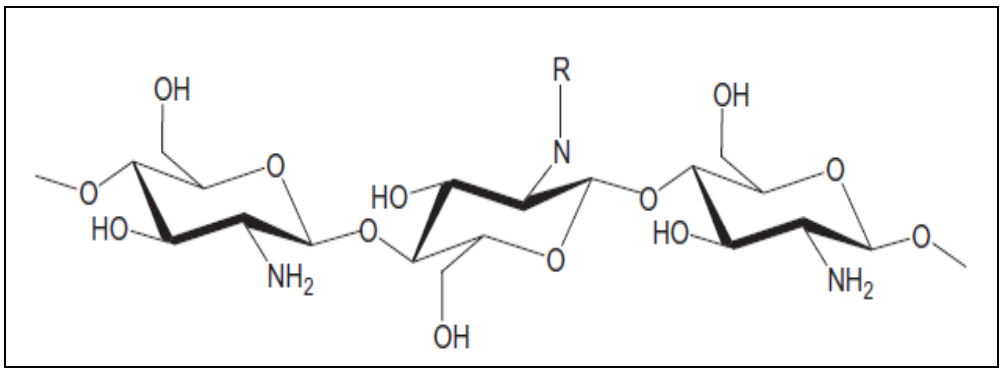


Fig No.2 Chemical Structure of chitosan R=H or COCH3.

Chitosan (CS), is the second most abundant polysaccharide next to cellulose. Both are made by linear β-(1,4)-linked monosaccharide. However, an important difference to cellulose is that CS composed of 2-amino-2-deoxy-β-D-glucan combined with glycosidic linkages[1]. The primary amine groups render special properties that makes CS very useful in pharmaceutical application. However applications of chitin are limited compared to CS because chitin is structurally similar to cellulose, but chemically inert. Acetamide group of chitin can be converted into amino group to give CS, which is carried out by treating chitin with concentrated alkali solution. The chitosan molecule is a copolymer of N-acetyl-D-glucosamine and D-glucosamine and it differs in the degree of N-acetylation (40-98%) and molecular weight (50-2000 kDa). Several studies demonstrated that the way to prepare chitosan from chitin is determinant to define its characteristics such as molecular weight, deacetylation degree and crystallinity. The deacetylation degree is referred in literature as one of the most important characteristics involved in the chitosan characterization. The molecular weight is determinant to define the viscosity of chitosan. These two characteristics have a significant role in the biological chitosan properties. The chitosan biodegradability is important for example for the controlled release of active substances. Some authors verify that decreasing the deacetylation degree the biodegradability increases. However, a uniform distribution of the acetylated groups contributes to decreasing the biodegradability. Another interesting property is the biocompatibility, but this property depends on the characteristics of the sample (natural source, method of preparation, molecular weight, and deacetylation degree). Chitosan and its composite polymeric hydrogels are being formulated as controlled-release drug carriers and make an interesting alternative for pH dependent release in gastrointestinal tract (GIT) [3,2].

Chitosan based delivery system have been widely studied for colonic drug targeting since this system can protect therapeutic agents from the hostile condition of upper GIT & release the entrapped agents specifically at the colon through degradation of glycosidic linkages of chitosan by colonic microflora. Chitosan has been widely studied as an excipient for drug delivery because of its following advantages. A chitosan dispersed system was newly developed for colon-specific drug delivery which was composed of drug reservoir and the outer drug release-regulating layer dispersing chitosan powder in hydrophobic polymer. It was observed that the thickness of the outer layer controls the drug release rate. Since the dispersed chitosan dissolves easily under acidic conditions, an additional outer enteric coating was also provided to prevent the release of drug from chitosan dispersed system in the stomach[2].

Advantages:

1) It is natural in origin.
2) Nontoxic substance.
3) Biocompatible polymer.
4) Inexpensive.

Disadvantages:

It is insoluble at physiological pH hence restricted its application in drug delivery.

Microsphere:

Microspheres is the system in which the drug substance is either homogenously dissolved or dispersed in a polymeric matrix. It constitute an important part of this particulate drug delivery system by virtue of their small size and efficient carrier characteristics. However, the success of this novel drug delivery system is limited due to their short residence time at the site of absorption. It would therefore be advantageous to have means for providing an intimate contact of the drug delivery system with absorbing gastric mucosal membranes. Microspheres are characteristically free powders consisting of proteins or synthetic polymers that are biodegradable in nature and ideally having a particle size less than 200µm[6].

Different forms chitosan microsphere can be used for colon targeted drug delivery system. These are given as follows.

Modified chitosan

Although chitin and chitosan are attractive biomacromolecules, they are water-insoluble materials. Chitosan is soluble only in acidic solutions because of its rigid crystalline structure and deacetylation, which limit its application to bioactive agents such as drug carriers. However it is possible to modify the chitosan structure to form water soluble chitosan which is easily soluble in neutral aqueous solutions. This advantage is useful for drug carriers and for food industrial applications. In order to develop water soluble chitosan, many attempts have been made to modify the molecular structure of chitosan, and thereby improve or control its properties, with carboxymethyl, dihydroxyethyl, sulfuryl, or phosphoryl groups. The solubility can be improved by chemical modifications involving the introduction of hydrophilic functional groups. Many chitosan derivatives can be produced, such as carboxymethyl chitosan, N-sulfofuryl chitosan, 5-methyl pyrrolidinone chitosan, dicarboxymethyl chitosan, and quaternized chitosan. For example, carboxylation consists in the incorporation reaction of one or more carboxyl group into the chitosan molecule. The localization of these new groups in the molecule of chitosan can change with the type and conditions of the reaction involved and the type of reagents used.

Nacher. A et al developed N-succinyl chitosan microparticles loaded with 5-amino salicylic acid for colon specific drug delivery. NS chitosan were prepared by introducing succinic group into chitosan N-terminal of the glucosamine group. The microparticles were prepared by spray drying technique. It was found that highest swelling ration and drug release at pH 7.4 where microparticles were able to deliver more than 90% 0f 5-aminosalicylic acid during 24hrs experiment[10].

Crosslinked chitosan:

One important use of chitosan consists in its application in controlled release systems, usually as nanocapsules or microcapsules, hydrogels, nanofibres, films and tablets. Chitosan has one important advantage over other encapsulating agents, which is the possibility to establish covalent or ionic bonds with the crosslinking agents, building a sort of network, where the active substance stays retained. Consequently, these chemical bonds carry advantages in terms of controlled release. Depending on the nature of the crosslinker, the main interactions forming the network are covalent or ionic bonds. Covalent crosslinking leads to the formation of hydrogels or microparticles with a permanent network structure, because irreversible chemical bonds are formed. This type of linkage allows absorption of water and/or bioactive compounds without dissolution and allows the drug release by diffusion. The addition of a second polymer as encapsulating agent makes possible the pH controlled drug delivery. Crosslinkers agents used to establish covalent bonds are molecules with at least two reactive functional groups that allow the formation of bridges between polymeric chains. The most common crosslinkers used with chitosan are dialdehydes, such as glyoxal and in particular glutaraldehyde. However, the main disadvantage of this kind of crosslinking agents, dialdehydes, is that they are generally considered toxic for food application purposes. For example, glutaraldehyde is known to be neurotoxic and glyoxal is known to be mutagenic. Ionically crosslinked microparticles or hydrogels are generally considered as biocompatible and well tolerated. Their network is non-permanent and forms reversible links. Ionically crosslinked chitosan hydrogels or microparticle. Example of ionic crosslinker is sodium tripolyphosphate, calcium phosphate[7].

As the crosslinking density increases, water content, swelling capacity and the mesh size of the network decrease. Indeed, increasing the amount of crosslinker decreases the ability of chitosan to form hydrogen bonds with water molecules. Moreover, the higher the crosslinking density, the lower the swelling ability of chitosan hydrogels due to the slower relaxation time of the polymeric chains, which results in a decreased drug-release rate[8]. Concern over disintegration of chitosan microsparticles has led to their by crosslinking to make a rigid polymer for use as a core material in controlled drug release.

Mucoadhesive microsphere

The combination of the mucoadhesion concept with CTDDS would avail a more efficient colonic targeting for treatment of IBD. However, the microparticulates system is limited due to their short residence time at the site of absorption. This can be improved by coupling bioadhesion characteristics to microparticulates & developing mucoadhesive microparticles. Compared to the stomach and small intestine, the colon is deemed a more suitable site for mucoadhesion due to its thicker mucus layer and lower disruptive colonic motility. Thiolated polymers such as thiolated chitosan, have been developed as a mucoadhesive polymers and shown excellent mucoadhesive properties by forming disulphide bonds with cysteine-rich domains of mucus glycoproteins. Chitosan has been shown to possess mucoadhesive properties due to molecular attractive forces formed by electrostatic interaction between positively charged chitosan and negatively charged mucosal surfaces. These properties may be attributed to[11]:

a) Strong hydrogen bonding groups like –OH, –COOH
b) Strong charge
c) High molecular weight
d) Sufficient chain flexibility.
e) Surface energy properties favoring spreading into mucus

Kurniawan et al formulated thiolated chitosan using cysteine as thiomer. In which chitosan dissolved in 1% acetic acid and also added cysteine. To this solution 1-ethyl-3-(3-dimethylamino propyl) carbodiimide (EDAC) added. The mixture was incubated for 3hrs with continuous stirring. Then solution is lyophilized gives thiolated chitosan[16].

Haogang Duan et al prepared mucoadhesive composite microparticles based on N-acetyl cysteine chitosan & sodium alginate for dual drug delivery of 5-aminosalicylic acid & curcumin for inflamed colon. They found that more mucoadhesion occurred as compared to plain chitosan[11].

Chitosan & alginate microparticles:

Chitosan and alginate have been widely studied as drug delivery platforms targeted to the colon because the drug release from these systems is triggered by the enzymatic activity of the colonic resident micro-biota. Additionally, chitosan shows high mucoadhesive properties that have been used to prolong chitosan residence in the gastrointestinal tract. Size & charge are major determinant of the particles ability to passively target the inflamed intestinal mucosa and reach maximum retention times in the tissue. These microspheres prevent or reduce the release rate of small molecules in conditions simulating the gastric environment and they control the release in conditions simulating the intestinal environment, thus permitting higher drug absorption and better therapeutic effect.

Wang et al formed chitosan/alginate icariin microsphere & described that, Icariin with the poor solubility and low bioavailability limited the treatment of many diseases in clinic. In this study, the protective mechanism of chitosan-alginate microspheres loaded with icariin were investigated with trinitrobenzene sulfonic acid (TNBS)/ethanol induced colonic mucosal injury in rats [15].

Microsphere formed by different techniques.

1) Ionotropic gelation
2) Emulsion crosslinking
3) Spray drying
4) Emulsion solvent evaporation
   a) Single emulsion
   b) Double emulsion
5) Interfacial polymerization
6) Solvent Evaporation

Ionotropic gelation:

It involves simply the interaction of an ionic polymer with oppositely charge ion to initiate cross linking. The chitosan solution in acetic acid was extruded dropwise through a needle into different concentrations of aqueous solutions of magnetically stirred tripolyphosphate or some other anion. The beads were removed from the counter ion solution by filtration, washed with distilled water and dried. Unlike simple monomeric ions, the interaction of polyanion with cations (or polyanion with polycation) cannot be completely explained. Microparticles can be formed by two ways External ionotropic gelation & internal gelation. Externally crosslinked micopellets were also capable of greater drug encapsulation efficiency & slower drug release. Vajpayee et al developed curcumin microsphere using natural polymers i.e. guar gum, xanthan gum, sodium alginate by ionotropic gelation method for treating colonic cancer. Also coated microsphere using Eudragit s100 polymer which retard drug release in stomach & small intestine[12].

Dario Leonardi et al formed albendazole microparticles by ionic interaction method in which they sprayed drug & polymer solution over ionic solution by keeping temperature constant while continuous stirring was maintained to complete ionic interaction[14].

Emulsion crosslinking:

In this process chitosan solution (in acetic acid) is added to liquid paraffin containing a surfactant resulting in formation of w/o emulsion. A crosslinking agent of varying amount is added depending upon the crosslinking density require. The microspheres formed are filtered, washed with suitable solvents and dried.

Wang. J et al developed microencapsules of norfloxacine in chitosan and chitosan oligosaccharide by dissolving norfloxacine in chitosan and chitosan oligosaccharide in acetic acid solution following dispersing this solution into liquid paraffin containing span 80 under mechanical stirring[15].

Emulsion solvent evaporation:

Single emulsion technique:

In this method, drug is dissolved in aqueous medium, which acts as dispersed phase, is dropped into oil phase, which acts as continuous phase. A single emulsion will form, which is further stabilized by addition of either crosslinker or by heating. The most commonly used chemical crosslinkers are formaldehyde, glutaraldehyde, epichlorohydrin, etc.

Garud et al prepared microsphere of mesalamine by using emulsion crosslinking method for the treatment of ulcerative colitis[13].

Double emulsion technique:

In this technique, two emulsions are formed, one is primary and other is secondary. For the preparation of primary emulsion, protein aqueous solution is dispersed in lipophilic continuous phase for the encapsulation of protein contained in dispersed aqueous phase. This primary phase is then homogenized and dropped into aqueous solution of poly vinyl alcohol, which acts as secondary phase. Addition of primary phase to secondary leads to the formation of double emulsion.

Spray drying:

This technique is used for the preparation of polymeric blended microspheres having the size range from 1 to 100 µm. In this method, polymer is dissolved in volatile organic solvent like DCM or acetone. Then core material is dispersed in this polymeric solution. This dispersed phase is atomized in a spray chamber under hot air, which leads to the formation of fine mist from which solvent evaporates instantaneously. The polydispersity of microparticles obtained from conventional spray drying is due to the broad size range of droplets and random droplet trajectories generated by conventional atomisers. The process is flexible, offering substantial variation in microencapsulation matrix, is adaptable to commonly used processing equipment and produces particles of good quality.

Shendge et al formulated budesonide microsphere using spray drying technique & cross linked chitosan polymer with glutaraldehyde. Chemical modification of chitosan was carried out by using glutaraldehyde which causes crosslinking between chitosan molecules rendering them water insoluble. Drug, polymer and crosslinking agent were spray dried to form microparticles[17].

Interfacial polymerization:

In this method two reactive monomers, one is dissolved while other is dispersed separately in two immiscible liquid. A brisk reaction occurs at the interface between two solutions, which create a thin layer interfacial film.

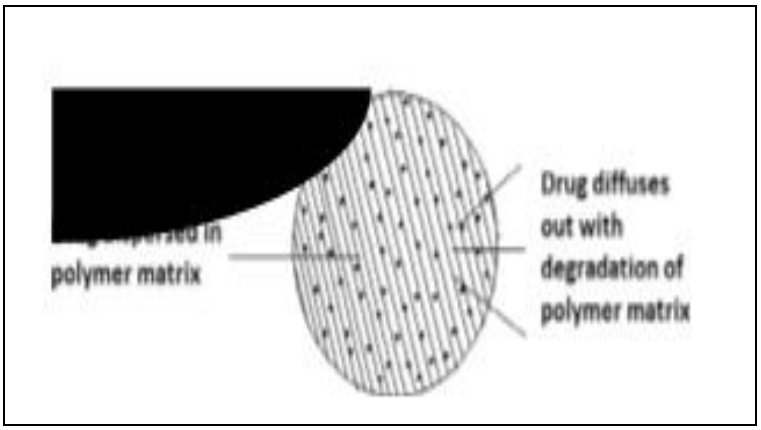
Table No: 2 Comparison of different techniques of microparticles:

Sr.No.	Name of Technique	Advantages	Disadvantages	Application
1.	Spray drying	Rapid process Completely evaporation of solvent, formation of porous microparticle,	Costly process, high temperature is required, less yield obtained.	Budesonide microparticles by Shendge et al [17]
2.	Ionotropic gelation	Simple & rapid process More entrapment efficiency.	Mechanical strength of formed particles is less.	Albendazole microparticles developed by Dario Leonardi et al [14].
3.	Interfacial polymerization	Fast rapid & efficient.	Due to large w/o interface, enzymes or proteins get inactivated. It's hard to control the polymerization reaction.	Polyaniline nanoparticles by interfacial polymerization. (B. Han et al) [20]
4.	Emulsion crosslinking	Simple method, convenient	Washing is required for removing excess crosslinker.	Microencapsulation of norfloxacin by Wang .J et al [15]
5.	Single emulsion	Reproducible process, less time requires, less operation skill requires.	Poor encapsulation efficiency.	This method is used to prepare microspheres containing protein and peptide drugs. 5-Flourouracil microsphere developed by Murthy. R et al [18]
6.	Double emulsion	Controlled drug release Used for hydrophilic drugs proteins, peptides, vaccines	Stability problem, Coalescence	Poly(lactide-co-glycolide) microparticles containing serum albumin was prepared by w/o/w emulsion technique. (C. Yan et al) [21]

Mechanism of drug release:

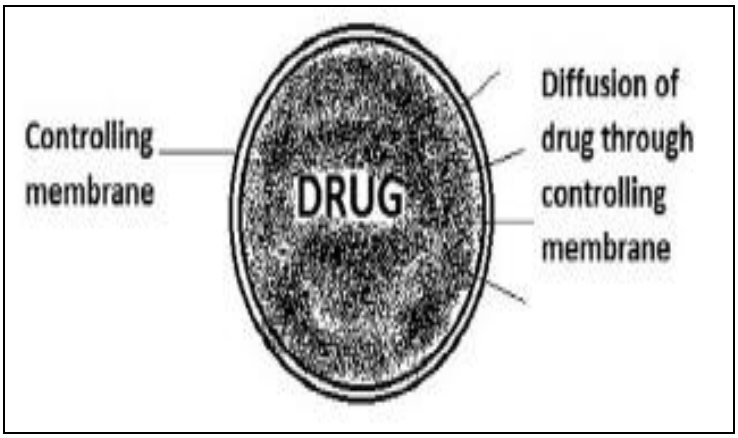
Diffusion controlled reservoir system:

In this system, a controlling membrane is present around drug, through which drug starts diffusing out. Here, release rate is not affected by matrix degradation. After complete diffusion.[3]



Diffusion controlled monolithic system:

In this system, drug is dispersed in polymer matrix. With degradation of polymer matrix, drug starts diffusing out. Here, release rate is highly affected by matrix degradation. Fig. 3 shows diffusion of drug through polymer matrix.[3]



Degradation controlled monolithic system:

In this system, drug is uniformly dispersed in polymer matrix and rate of diffusion depends upon degradation of matrix. Generally, rate of diffusion is slow as compared with degradation of matrix[3]. The formula for calculating release of sphere is governed by following equation:

$$Mt/M\infty = 1-[(1-t/t\infty)]^3$$

Where Mt is amount of drug released at time t;
M1 is amount at time t1 for total erosion

CONCLUSION

Chitosan play important role in colon specific drug delivery. As it is biodegradable polymer which is degraded by colonic microflora. In case of inflammatory bowel diseases (IBD) crosslinked, modified & mucoadhesive microparticles of chitosan produces local effects & helps in curing colonic diseases. Colon is the most suitable region of GIT for systemic as well as local drug delivery. Those drugs which are degraded in stomach and small intestine can be given by this type of drug delivery. Hence widely used for treatment of many diseases. Also the residence time of colon is more which is also useful in controlled release delivery, hence long lasting effect produces. Among different techniques of colon targeted drug delivery, microparticles system is very simple and convenient.

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