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Ionotropic Gelation: A Promising Cross Linking Technique for Hydrogels.

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ABSTRACT

Microencapsulation is by far the most commonly exploited technique for the generation of multiparticulate drug delivery system. But serious drawbacks of this technique like non-uniform coating, non-reproducible release kinetics and more importantly, the use of more or less harsh conditions in the formulation process limits the encapsulation of the many substances such as proteins, enzymes and live cells etc. Besides this, the regulatory authorities, such as U.S.FDA are restricting to greater degrees the amount of additional components allowed such as organic solvents. The solution of this problem is presented by a technique, which involved neither the use of harsh chemicals nor elevated temperature and based on principle of ionotropic gelation (polyelectrolyte complexation). This review emphasizes the importance of ionotropic gelation and its cross linking potential to formulate hydrogel beads using different natural polymers.

INTRODUCTION

There are a variety of disease states (Cancer, AIDS, Cardiac diseases) where systemic drug administration has poor efficacy due to low solubility, random body distribution and toxicity problems. Therefore, drug entrapped in polymeric beads offer novel drug delivery approach for an intended application, such as targeted locations and long-term delivery times (days, weeks, months, and even years). Multiparticulate drug delivery systems have various well known advantages over single unit dosage forms. The potential use of microspheres in pharmaceutical industry has been considered since the 1960s for the following applications ^[1].

- Taste and odor masking
- Conversion of oils and other liquids to solids for the ease of handling
- Protection of drug against the environment (moisture, light heat)
- Improvement of flow properties of powders
- Separation of incompatible materials
- Production of sustained release, controlled release and targeted medications.
- Reduced dose dumping potential compared to large implantable devices.

One of the most highly exploited techniques to formulate Multiparticulate drug delivery systems is Microencapsulation. Different microencapsulation techniques available have been extensively reviewed as follows ^[2].

- Solvent evaporation
- Phase separation Coacervation
- Solvent extraction
- Spray Drying
- Spray Coating

Although all methods offer many significant advantages, it is only at the sake of some drawbacks. Some of the important drawbacks of these techniques include non-uniform coating, Non-reproducible release kinetics and more importantly, the use of more or less harsh conditions in the formulation process which limits the many substances such as proteins, enzymes and live cells etc. as core materials for encapsulation. Besides this, the regulatory authorities, such as U. S. FDA are restricting to greater degrees the amount of additional components allowed such as organic solvents. The solution of this problem is presented by a technique, which involved neither the use of harsh chemicals nor elevated temperature. The technique proposed is based on principle of ionotropic gelation (polyelectrolyte complexation). This technique involves interaction of a cation (or an anion) with an ionic polymer to generate a highly cross linked structure. The ability of such highly cross linked structure to sustain the drug release is exploited in this method. Moreover, the simple and mild Condition under which it is achieved is also an advantage [3].

Ionotropic Gelation (Polyelectrolyte Complexation)

Ionotropic gelation involves simply the interaction of an ionic polymer with oppositely charge ion to initiate cross linking. Unlike simple monomeric ions, the interaction of polyanion with cations (or polyanion with polycation) cannot be completely explained by the electro-neutrality principle. The three dimensional structure and presence of other groups influence the ability of cations (or anions) to conjugate with anionic (or cationic) functionalities and some kind of selectivity is found. (Fig 1)

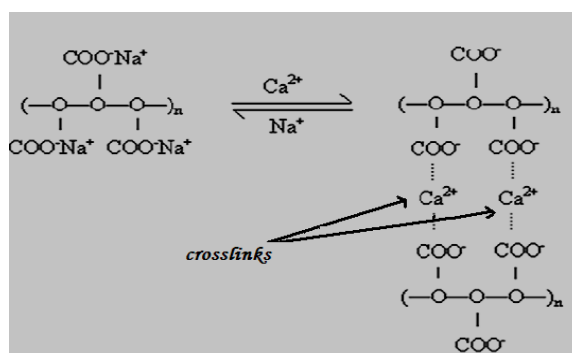


Figure 1: Electrostatic interaction between -COO groups of alginate and cross linker Ca++ ions.

Methods of ionotropic gelation

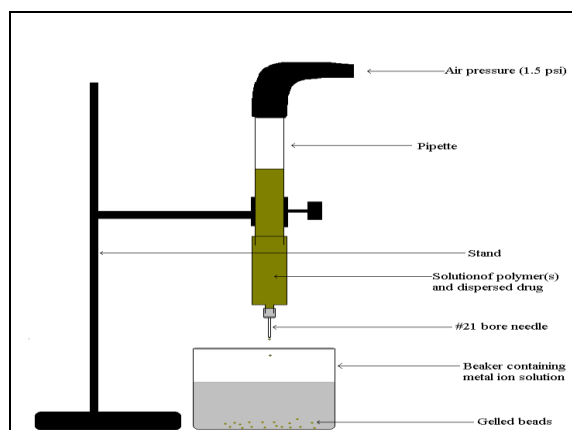


Figure 2 External ionotropic Gelation

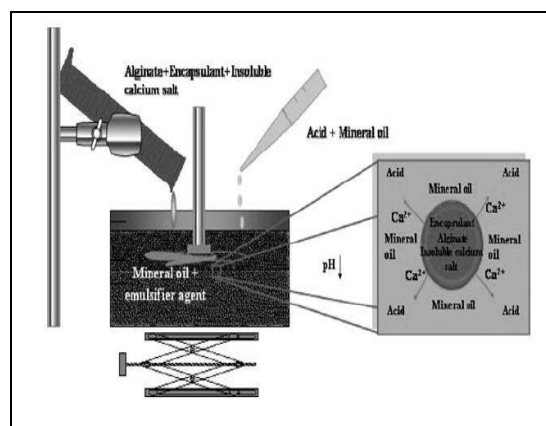


Figure 3: Internal Gelation/Emulsification

There are two methods by which hydrogel beads can be generated using ionotropic gelation technique. These methods differ from each other in the source of the cross linking ion. In one of the methods, the cross linker ion is positioned externally shown in Fig 2. [4] whereas in the other method, the cross linker ion is incorporated within the polymer solution in inactive form shown in Fig 3. [5] External cross- linking produced thinner films with smoother surface, greater matrix strength, stiffness and permeability than internally cross-linked films. Externally cross-linked micropellets were also capable of greater drug encapsulation efficiency and slower drug release rate. There are variety of natural and synthetic polymeric systems that have been investigated for the controlled release of drug. Hydrophilic polyionic carbohydrates such as alginate and chitosan have been paid much attention in recent years [6,7]. Since the preparation of beads by these materials involves the use of aqueous solvents, environmental problems associated with organic solvents would be minimized. A variety of natural polymers and their

derivatised products have been successfully employed in hydrogel system for various pharmaceutical applications. In this review the potential of sodium alginate and chitosan to form the highly cross linked structure and its pharmaceutical applications is discussed. As compared to other natural polymers, sodium alginate and chitosan shows no variations in viscosity and hence produces more uniform gel structure which forms stronger cross linked structure and more loading of entrapped material.

Sodium Alginate

Sodium alginate is sodium salt of alginic acid, a naturally occurring polysaccharide obtained from marine brown algae. Alginic acid is a linear copolymer composed of D-mannuronic acid (M) and L-guluronic acid (G) shown in Fig 4,5 [8]. Alginates are linear unbranched copolymers of -d-mannuronic acid (M) and -l-guluronic acid (G) units. The M and G monomers are 1→4 linked by glycosidic bonds, forming homopolymeric M- or G-blocks and heteropolymeric MG blocks. In the presence of polyvalent cations such as Ca²⁺ or Al³⁺, cross-linking occurs to form gels. The cations act as bridges between the anionic polymer chains, constituting junction zones, forming a hydrogel network. Ca²⁺, a commonly used cross-linker, preferentially interacts with G blocks due to structurally favorable chelation sites formed by the corrugated chains [9].

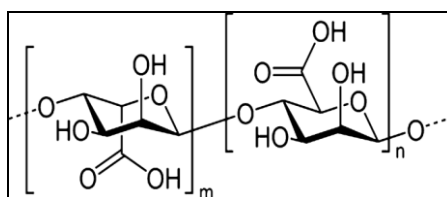


Figure 4: Structure of alginic acid

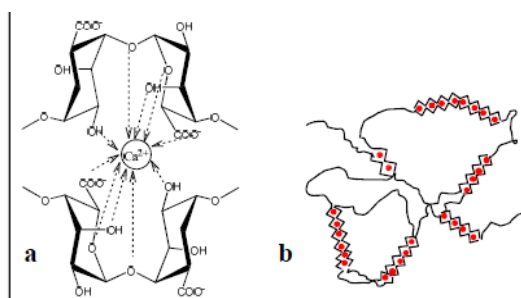


Figure 5: a) Cross linked structure of sodium alginate with Calcium ions (circle)
5-b) "Egg box" model where GG blocks represented by zig-zag portions and MM and MG blocks represented by smooth parts of the polymer chains

Many researchers reported calcium cross linked alginate beads for different applications like controlled release, mucoadhesive action. As sodium alginate is anionic in nature, it can be cross linked with different cations like calcium, aluminum, barium but degree of cross linking varies according to valency and ionic radius of cross linking ion. Gel network structure of alginate with different cations is shown in Fig 6. Alginate is commonly employed as cross-linked matrices or coats in drug delivery systems [10,11,12,13]. It is also extensively used to encapsulate living cells, such as yeasts for fermentation and pancreatic islets for clinical applications [14,15]. Sodium alginate is a biopolymer that is widely used as an encapsulation matrix due to its ability to form hydrogels upon cross-linking. Its ability to gel under mild conditions makes alginate the polymer-of-choice in food, pharmaceutical and biotechnological applications. Its unique property of forming water insoluble calcium alginate gel through ionotropic gelation with calcium ions is a simple, mild and eco-friendly condition has made possible to encapsulate macromolecular bio-active agents like cell, enzyme, protein and vaccine.

Chitosan

Chitosan is a hydrophilic cationic polyelectrolyte obtained by alkaline N-deacetylation of chitin shown in Fig 7. Chitin is the most abundant natural polymer next to cellulose and is obtained from crab and shrimp shells. Chitin and CS represent long-chain polymers having molecular mass up to several million Daltons. Chitosan is relatively reactive and can be produced in various forms such as powder, paste, film, fiber, etc. The primary amine groups render special properties that make CS very useful in pharmaceutical applications. Compared to many other natural polymers, chitosan has a positive charge and is mucoadhesive. Chitosan, being a cationic polysaccharide in neutral or basic pH conditions, contains free amino groups and hence, is insoluble in water. In acidic pH, amino groups can undergo protonation thus, making it soluble in water.

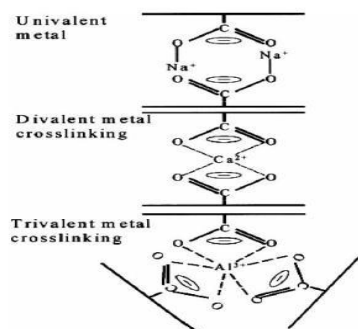


Figure 6: Alginate gel network formation by multivalent ions

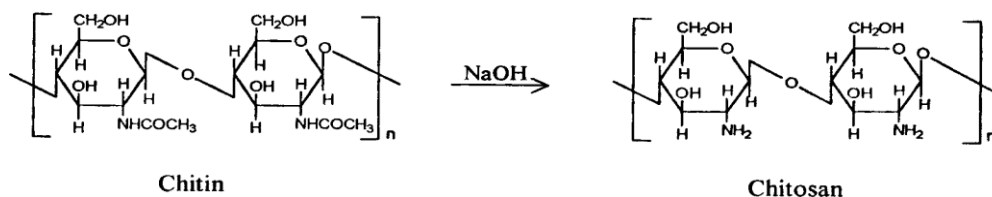


Figure 7: Conversion of chitin to chitosan.

Table 1: List of various natural polymers employed in Cross linked Hydrogel System.

Polymer	Properties	Applications
Sodium alginate	Natural Biodegradable Biocompatible Hydrophilic	Effective carrier for GIT sensitive enzymes, proteins. Entrapment of water soluble drugs.
Chitosan	Polyaminosaccharide obtained by Deacetylation of chitin. Bioadhesive & Antimicrobial properties.[33] Anionic polymer	Interpenetrating network (IPN) Hydrogel system for sustained release.[25,26,27] Improved Enzyme activity and enzyme Loading. Treatment of oral mucositis[28]
Pectin	Long chains of α -1,4-linked galacturonic acid units with varying degrees of esterification with methyl groups. The degree of esterification (DE) affects solubility and gelation properties; pectins with DE above 50% are labeled high-methoxyl and below 50% DE low-methoxyl pectins polysaccharide	Floating pulsatile drug delivery for Diclofenac sodium. Improved entrapment of Metronidazole in core & surface cross linked beads [29,30]
Konjac	Neutral Polysaccharide with biocompatible biodegradable	Synergistic effect with xanthan for controlled drug delivery. Drug delivery, Gel filler materials for prosthetic implants, Cellular therapy. [31,32]

Chitosan is biocompatible with living tissues since it does not cause allergic reactions and rejection. It breaks down slowly to harmless products (amino sugars), which are completely absorbed by the human body. It possesses antimicrobial property and absorbs toxic metals like mercury, cadmium, lead, etc. In addition, it has good adhesion, coagulation ability, and immunostimulating activity. The use of complexation between oppositely charged macromolecules to prepare CS microspheres has attracted much attention because the process is very simple and mild [16,17]. In addition, reversible physical cross-linking by electrostatic interaction, instead of chemical cross-linking, has been applied to avoid the possible toxicity of reagents and other undesirable effects. Tripolyphosphate (TPP) is a polyanion, which can interact with the cationic CS by electrostatic forces [18].

Pharmaceutical applications of sodium alginate and chitosan beads

Combination of alginate- Konjac Glucomannan-Chitosan

Zhimin He and et al studied alginate (ALG)-konjac glucomannan (KGM) - chitosan (CHI) beads as a controlled release matrix. In this Bovine serum albumin and insulin were used as model proteins for in vitro assessments. The use of KGM system for BSA encapsulation appears to increase the protein loading within the matrix compared to the classical system of protein. Entrapment by calcium ALG gelation, and the highest loading efficiency was obtained from ALG-KGM-CHI system. It is also observed that the leaking of protein from ALG beads was more than ALG-KGM beads, an ion cross linked ALG delivery system when exposed to low pH can undergo a reduction in ALG molecular weight, which results in faster degrading and release of a molecule. KGM was not hydrolyzed in acid solution and it stabilizes the gel [19].

Effect of internal and external gelation on alginate and its use as a coat and drug delivery system

Comparable extent of cross-linkages was produced by the two gelation methods as indicated by comparable CaCl_2 contents between externally and internally cross-linked films but the distributions of cross-linkages were different. External cross-linking produced thinner films with smoother surface, greater matrix strength, stiffness and permeability than internally cross-linked films. Externally cross-linked micropellets were also capable of greater drug encapsulation efficiency and slower drug release rate. The differences in the properties observed were due to the different gelation mechanisms involved and the physical form of the matrix produced. External gelation is the preferred method in producing cross-linked alginate for coating and encapsulation purposes [20].

Controlled release of diclofenac sodium from sodium alginate beads

Tejraj M. Aminabhavi et al prepared sodium alginate beads by precipitation of Na-Alg in alcohol followed by crosslinking with glutaraldehyde in acidic medium. Preparation of the beads was optimized by considering the percentage entrapment efficiency, swelling capacity of beads in water and their release data. The beads produced at higher temperatures and longer times of exposure to the crosslinking agent have shown the lower entrapment efficiency, but extended release of DS from the beads. Diclofenac sodium, being a salt of an acid, is insoluble in acidic media and hence, an increase in the percentage entrapment efficiency was observed with an increase in % HCl content in methanol. The percentage entrapment efficiency decreased with an increase in the time of exposure to the crosslinking agent may be due to the increased release of the drug from the matrix at longer time of exposure [21].

Water-based ionotropic external gelation technique for Furosemide loaded alginate microspheres

M. K. Das prepared furosemide loaded alginate microspheres by external gelation technique. Previously reported, the furosemide microspheres prepared by emulsion-solvent evaporation method utilize a larger volume of organic solvents, which are costly and hazardous because of the possibility of explosion, toxicity and air pollution. The pulsatile release pattern was observed from all the formulations investigated. The alginate microspheres swelled and eventually disintegrated in phosphate buffer of pH 7.4 [22].

Effective oral drug delivery system for Bovine serum albumin

Sevgi Takka et al prepared the Bovine serum albumin loaded beads by ionotropic gelation of alginate with calcium chloride and chitosan. Oral administration of peptide and protein drugs requires their protection from degradation in the gastric environment and the improvement of their absorption in the intestinal tract. Alginate and chitosan are natural polymers that they are biocompatible, biodegradable, and produce no systemic toxicity on administration. The optimum condition for preparation alginate-chitosan beads was alginate concentration of 3% and chitosan concentration of 0.25% at pH 5. The resulting bead formulation had a loading efficacy of 98.5% [23].

Retention of lipase activity using chitosan hydrogel beads

Low enzyme activity and low enzyme loading have been reported by several researchers and attempts are being made to optimize enzyme loading and minimize the loss of enzyme in the reaction medium, while still maintaining or improving the enzyme activity. On the basis of the activity of an amount of free enzyme equivalent to the actual lipase entrapped, about 50% retention of activity was seen with lipase entrapped in these chitosan beads. Lipase-loaded chitosan hydrogel bead also offers advantages with respect to re-use and stability of the immobilized enzyme [24].

CONCLUSION

Biopolymers based hydrogel is an effective drug delivery system with various pharmaceutical applications like site specific drug delivery, protection of enzymes, proteins and peptide from gastric environment, controlled drug delivery system, immunogenicity and bioadhesive properties etc. Various cross linking methods are available for the development of natural or synthetic polymer based hydrogel beads or microcapsules but ionotropic gelation (polyelectrolyte complexation) is a simple, mild and cost effective technique. Since the preparation of beads by sodium alginate, Konjac and chitosan involves the use of aqueous solvents, environmental problems associated with organic solvents would be minimized.

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