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THERMOREVERSIBLE MUCOADHESIVE INSITU GEL: A REVIEW

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ABSTRACT

The main aim of pharmacotherapeutic is the attainment of therapeutic drug concentration at the intended site of action for sufficient period of time to elicit the pharmacological action. A major problem faced by conventional formulation in various routes of administration viz. nasal, ocular, vaginal, rectal, etc is drainage of instilled formulation, poor patient compliance as in the case of vaginal and rectal delivery system. Also, blurred vision and irritation to mucosa is observed in the case of ocular and nasal delivery respectively. The drainage of the formulation results in inaccurate dose delivery thus decreasing the bioavailability of the drug which demands an increase in the frequency of the administration. To overcome these limitations, there is need of novel delivery system such as thermoreversible mucoadhesive *insitu* gel which provides an ease of administration and improved patient compliance as these smart delivery system is free flowing liquid at ambient temperature and gels at physiological temperature which is higher than the LCST (low critical solution temperature) of thermoreversible polymer. Further this novel formulation can be made more beneficial by inclusion of mucoadhesive polymer which facilitates the adhesion of the formulation and permeation of the drug through the mucous membrane, thereby increasing the residence time and hence the bioavailability of the drug. The *insitu* gel forming polymeric formulation offers sustained as well as controlled drug delivery which is the prerequisite of the modern pharmaceutical design. The article presents the detailed review of thermoreversible as well as mucoadhesive polymers, mechanism of gelation and mucoadhesion and the factors affecting the same, evaluation methods of thermoreversible mucoadhesive *insitu* gel and finally various thermoreversible mucoadhesive *insitu* gel delivery systems (injectable, intramuscular, nasal, ocular, vaginal, rectal, intraoral, periodontal).

KEY WORDS: *Insitu* gel, Thermoreversible polymers, Mucoadhesive polymers, Low critical solution temperature, Transition temperature, Poloxamer.

INTRODUCTION

Over the past 30 years, greater attention has been focused on development of controlled and sustained drug delivery systems. Amongst the extensive research has been carried in designing of polymeric drug delivery systems, the development of *in situ* gel systems has received considerable attention [1]. Smart polymeric systems represent promising means of delivering the drugs; these polymers undergo sol-gel transition, once administered, i.e. they are in solution phase before administration, but gels under physiological condition [1, 2]. These systems are injectable fluids that can be introduce into the body in a minimal invasive manner prior to solidifying or gelling with in the desired tissue, organ or body cavity. There are various physical and chemical stimuli leading to in situ gel formation viz. temperature, pH, electric field, magnetic field and light [3, 4]. Stimuli responsive polymer mimics biological system in a crude way where an external stimulus (pH and temperature) result in a change in the properties of the formulation. These changes may be in the conformation, solubility, alteration of hydrophilic/hydrophobic balance or release of a drug molecule. This also includes a combination of several responses at the same time [2].

In the past few years, increasing number of *in situ* gel forming systems have been investigated and many patents for their use in various biomedical applications including drug delivery have been reported. This novel drug delivery system promotes an ease and convenience of administration, deliverance of accurate dose as well as

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prolongs the residence time of the drug in contact with mucosa thereby increasing the bioavailability of the drug [1, 2].

Among *in situ*-forming systems, temperature-induced phase transitions from free flowing liquids at ambient temperature, to gels at body temperature, have gained considerable attention today in drug delivery research [1, 5] as shown in figure 1.

The use of biomaterial whose transitions from sol-gel is triggered by increase in temperature is an attractive way to approach in-situ formation. The ideal critical temperature range for such system is ambient and physiologic temperature, such that clinical manipulation is facilitated and no external source of heat other than that of body is required to trigger the gelation. A useful system should be tailorable to account for small differences in local temperature, such as might be encountered in appendages at the surface of skin or in the oral cavity [1].

In the early 1980s, the concept of mucosal adhesives, or mucoadhesives, was introduced into the controlled drug delivery area. Mucoadhesives are synthetic or natural polymers that interact with the mucus layer covering the mucosal epithelial surface and main molecules constituting a major part of mucus, thereby increasing the dosage form contact time and residence time with the mucous membranes resulting in increase in the bioavailability of the drug delivery. The concept of mucoadhesives has alarmed many investigators to the possibility that these polymers can be used to overcome physiological barriers in long-term drug delivery [6].

The combined effect of mucoadhesion and thermoreversible property will enhance prolongation of residence time of dosage form and the accuracy of the dose in body cavity respectively, leading to increased bioavailability of the drug.

ADVANTAGES OF THERMOREVERSIBLE MUCOADHESIVE INSITU GEL [7]

1. Ease of administration.
2. Reduction of taste impact (in nasal delivery)
3. Reduction of irritation (in nasal delivery)
4. Improved patient compliance.
5. Accuracy of dosing as leakage of drug is prevented.
6. Prolonged residence time.
7. Improved bioavailability.
8. Target delivery to mucosa for better absorption.
9. Sustained and controlled drug delivery.

THERMOREVERSIBLE POLYMER

Thermoreversible polymers are a novel state of matter having both solid and liquid like properties. Thermosetting systems are in the sol form when initially constituted, but upon heating, they set into their final shape. This polymer can be delivered as a fluid and solidifies within the body's microenvironment where the temperature is higher than the sol-gel transition temperature. This sol-gel transition is known as curing. But if this cured polymer is heated further, it may lead to degradation of the polymer. Curing mainly involves the formation of covalent cross links between polymer chains to form a macromolecular network as shown in figure 2. Such a system has both fluidity and elasticity [2].

MECHANISM OF GELATION

Thermoreversible polymers undergo sol to gel above its LCST. The lower critical solution temperature (LCST) or lower consolute temperature is the critical temperature below which the components of a mixture are miscible for all compositions. The word *lower* indicates that the LCST is a lower bound to a temperature interval of partial miscibility, or miscibility for certain compositions only. Temperature-induced aqueous gelation mechanism and gel structure has been elucidated using probe techniques, light scattering, rheometry, small angle neutron scattering, nuclear magnetic resonance measurements [5].

Different thermoreversible polymers exhibit the thermo-responsive gelation by various mechanisms. The following classification of thermoreversible gels, although not profuse, can be given on the basis of their crosslink formation, with an example for each of them [5]:

a) Crystallization: Poly (vinyl chloride).
b) Liquid-liquid phase separation in solution: Atactic polystyrene: When a solution of high molecular weight polystyrene in carbon disulfide (CS_2) is quenched to low temperatures, gelation will occur, due to liquid-liquid phase separation.
c) Liquid-liquid phase separation in bulk: Styrene–isoprene–styrene triblock copolymers.
d) Complex formation: Isotactic and syndiotactic poly (methyl methacrylate).
e) Helix formation: Gelatin.
f) Hydrogen bonding: Poly(vinyl alcohol) + Congo Red.
g) Chemical equilibrium: Poly (vinyl alcohol) + borate, partly hydrolysed poly (acrylamide) + chromium.
h) Interaction of side-chains in side-chain liquid crystalline polymers: polyacrylates with discotiside-chains.

Copolymer

The copolymer is in unimer form at low temperature or concentration. Block copolymers containing one block with a LCST at a temperature range where the other block is soluble, self assemble in response to temperature increase. Morphology of the self assembled structure depends on copolymer architecture and molecular Weight (MW) [5].

Micelle formation occurs as a result of PPG dehydration and hydrophobically driven self-assembly temperature (at a critical micellar temperature (CMT) and at concentrations above the critical micellar concentration (CMC) [2, 5]. At high enough concentrations, the high density of micelles leads to locking in crystalline structures

of hard spheres, a process which is usually, but not accurately, referred to as 'gelation' as shown in figure 3 [5].

A recently reported, alternative approach was based on interpenetrating networks of poly (N-isopropylacrylamide) (PNIPAM) and poly(acrylic acid) (PAAc), formulated in nanoparticles. The collapse of PNIPAM above its LCST triggered the bonding of the nanoparticles into a network while the repulsion between the charged PAAc chains prevented agglomeration [5].

THEORY

A number of polymers exhibit abrupt changes in their physical properties like solubility and viscosity with increases in temperature; the resulting sol-gel transition occurring at the lower critical solution temperature (LCST) is characterized by minimal heat production and absence of byproducts. Let us consider the free energy of association (G) between the polymer chains [5]:

$$G = H - T.S$$

where, H is the enthalpy term, S the entropy term and T temperature.

Increase over a critical temperature results in a larger value of T S than the positive enthalpy term (H), and thus a negative G favoring polymer association: chain-chain interactions (hydrophobic effects, hydrogen bonding) dominate over chain-water hydrogen bonding [5].

Several theories related to formation of thermoreversible gel have been suggested. Individual polymer followed a different theory of gel formation [2].

1. **Cubic crystalline phase** :

It was demonstrated by Wanka et al [2].

2. **Micellar aggregation :**

Jeong et al. demonstrated the gel formation through this mechanism. There was an abrupt change in micellar diameter and aggregation number when the temperature was above 30°C, and the second virial coefficient decreased sharply at this temperature, indicating a sudden increase in polymer- polymer attraction [2].

3. **Micelle-micelle cluster formation :**

Kwon et al. established that through hydrophobic interaction between core phases was responsible for the gel formation due to temperature change. E.g. As gelation of methylcellulose and hydroxypropyl methylcellulose is caused by the hydrophobic interaction between molecules containing methoxy substitution. The molecules of these polymers are hydrated at low temperature and there is little polymer-polymer interaction other than simple entanglement. As the temperature is raised, polymers lose their water of hydration and become some gel like structure with low viscosity. Eventually, when sufficient dehydration of the polymer occurs, polymer-polymer association takes place, and the system approaches an infinite network structure [2].

We can say the phase transition in term of the biologically relevant intermolecular forces depends on several different interactions as (I) Van-der Walls interaction (II) Hydrophobic interaction (III) Hydrogen bonding with change in ion interaction and (IV) Attractive ionic interaction [2].

INFLUENCE OF THIRD COMPONENT ON THE TRANSITION TEMPERATURE

Since the thermo-responsive behaviour depends on the solvent interaction with the polymer and the hydrophilic/hydrophobic balance within the polymer molecules, it is not surprising that additives to polymer/solvent system can influence the position of the volume phase transition. Three interesting "additives" are salts, surfactants and a co-solvent. All additives can alter the solvent quality and therefore can alter the polymer– solvent (+ additive) interactions. Therefore, the transition temperature can be shifted to a large extent or it can even disappear. Other aggregation forms such as micellisation can also occur (in contrast to a coil-to globule transition) [5].

e.g. PNIPAM and PVCa differ in their response to addition of a surfactant. Where PNIPAM shows a monotonous increase in the hydrodynamic radius upon addition of an ionic surfactant like sodium dodecyl sulfate (SDS), where as that of PVCa initially decreases. In both the cases, the transition temperature is proportionate to surfactant concentration until it levels out at a certain surfactant concentration [5].

CLASSIFICATION OF THERMOREVERSIBLE POLYMER

SYNTHETIC POLYMERS

Cellulose

Methylcellulose

Methylcellulose (MC) is water-soluble polymer which is known to undergo thermoreversible gelation in aqueous solution upon heating [5].

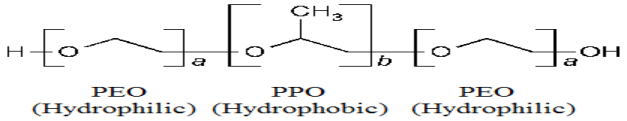
Mechanism of gelation of methyl cellulose

At low temperature, water molecules are presumed to form "cagelike" structures to surround the hydrophobic methoxyl groups, causing the MC to become water-soluble. Upon heating, these structures distort and break to expose the hydrophobic regions, inducing the formation of aggregates. Thus, the gelation is a manifestation of the hydrophobic effect and co-solutes that are readily soluble in water [5].

Hydroxypropyl methylcellulose

Transition temperature can be lowered by reducing the hydroxy propyl molar substitution [5].

Poloxamer (Pluronic)



Poloxamers or pluronic (marketed by BASF Corporation) are the series of commercially available difunctional triblock copolymers of non-ionic nature. They are regarded as PEO-PPO-PEO copolymers. Chemically they are Oxirane, methyl-, polymer with oxirane or α-Hydro-ω- hydroxypoly(oxyethylene)a poly(oxypropylene)b poly(oxyethylene)a block copolymer [1, 8]. It is biocompatible and has been approved by FDA for use in human body [5, 9].

Mechanism of gelation of poloxamer

Due to the PEO/PPO ratio of 2:1, when the poloxamer is placed into cold water, at low concentration; hydration layer surrounds the poloxamer molecule as hydroxyl groups of the copolymer forms hydrogen bond which surrounds the hydrophobic portions are separated due to hydrogen bonding. They undergo change in solubility with change in environment temperature. With increasing temperature, desolvation of the hydrophilic chains occurs as the result of breakage of hydrogen bonds. This results into hydrophobic interactions amongst the polyoxypropylene domains and gel gets formed. The molecular weight and percentage of hydrophobic portion are determinant factors for gelling behaviour. The gel formation occurs only when concentration is above critical micellar concentration [1, 5].

Reverse thermal gelation is the unique property of pluronic copolymers which makes it useful in the various drug delivery systems such as oral, ocular, nasal, topical, dental, and other biomedical fields. Poloxamer 407 forms transparent gel without syneresis, and also undergoes quick dissolution [5].

Because the length of the polymer blocks can be customized, the pluronic triblock copolymers are available in various grades differing in molecular weight and physical form [2, 5].

Coding of poloxamer

For the *Pluronic tradename*, it starts with a letter to define its physical form at room temperature (L = liquid, P = paste, F = flake (solid)) [2, 5] followed by two digits :

first digit x 300 = approximate molecular weight of the hydrophobe (POP)

last digit x 10 = % polyoxyethylene content

e.g. L61 =Pluronic with a polyoxypropylene molecular mass of 1,800 g/mol and a 10% polyoxyethylene content. In the example given, poloxamer 181 (P181) = Pluronic L 61 [5].

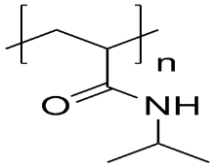
For the *poloxamer genericname,* these copolymers are commonly named with the letter "P" (for poloxamer) followed by three digits [5]:

- first two digits x 100 = approximate molecular mass of the hydrophobe (POP)
- last digit x 10 = % polyoxyethylene content

e.g., P407 = Poloxamer with a polyoxypropylene molecular mass of 4,000 g/mol and a 70% polyoxyethylene content [5].

Acryl amides

Poly-(N-isopropylacrylamide) PNIPAM



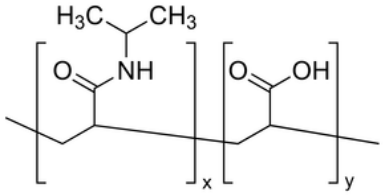
Poly-(N-isopropylacrylamide) , a typical thermo-sensitive polymer, has been widely studied, chiefly because of its phase transition at 32-33°C [9]. The LCST of PNIPAM can be changed by copolymerization with a second monomer which shifts upon shifting the hydrophilic/hydrophobic balance [5]. Hydrophobic co monomers (isopropyl) increase the LCST, whereas hydrophilic co-monomers (amide) have the opposite effect [9].

Mechanism of gelation of PNIPAM

Wang et al. have investigated the coil-to-globule transition of PNIPAM in water. They found a hysteresis between the radius of gyration during the heating and the cooling curve. Two intermediate states were observed, which gives in total four different, thermodynamically stable states *viz.* coil, crumpled coil, molten globule, and globule [5].

The toxicity of PNIPAAm in the body is unknown and the lack of compatibility with cells and blood, thus its application in drug delivery systems may be extremely restricted [10].

Poly (N-isopropylacrylamide-co-acrylic acid) PNIPAM-co-AA



Copolymerization of acrylic acid (AA) prevents syneresis of PNIPAM [5].

Poly(acrylic acid-co-acrylamide)

An interpenetrating network of poly (acrylic acid) and polyacrylamide is one of the few examples of a system with UCST behavior within the biomedical setting. The transition temperature is at 25 °C. The UCST behavior is caused by the cooperative effects coming from the hydrogen bonding between AAc and AAm units [5].

Various other acryl amides are Poly(N-isopropylacrylamide)/poly (ethylene oxide) ,Poly-(R)-N-(1-hydroxybutan-2-yl)acrylamide, Poly N-isopropylacrylamide -polylactide copolymers, Poly (N-isopropyl acrylamide-co-N-hydroxymethyl acrylamide), Poly(ethoxypropyl acrylamide), Poly (N-isopropylacrylamide-co-methyl

methacrylate-comathacrylic acid), Poly (N-isopropylacrylamide-co-hydroxyethylmethacrylate) [2,8, 9].

Poly(methyl vinyl ether) PMVE

Poly (methyl vinyl ether) has a transition temperature exactly at 37°C, which makes it very interesting for biomedical application. The polymer exhibits a typical type-III demixing behavior, which is in contrast to the thermal behavior of PNIPAM [5].

Poly(N-vinyl caprolactam) PNVCL

It also possesses very interesting properties for medical and biotechnological applications, e.g. biocompatibility, high absorption ability and a transition temperature within the settings of these applications (32-34°C) [5, 9].

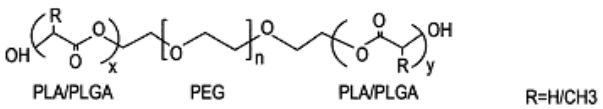
Poly(N-ethyl oxazoline) PEtOx

Poly(N-ethyl oxazoline)s have a transition temperature around 62°C, which is too high for any drug delivery application. However, recently a double thermo-responsive system by graft polymerization of EtOx onto a modified PNIPAM backbone has been prepared. Currently, these systems are explored for their potential in drug delivery, because they tend to aggregate micelles above the LCST. Unfortunately the poly (oxazoline) chemistry has the disadvantage that it is not very tolerant against unprotected functionalities [5].

Elastin-like oligo and polypeptides (ELP)

Polypeptides can also show LCST behavior, when hydrophilic and hydrophobic residues are balanced well. A polymer made out of the pentapeptide GVGVP as repeating unit exhibits a volume phase transition at 30°C, which is the hydrophobic folding and assembling transition [5, 10]. Below the phase transition, water molecules are structured around the polymer molecule; the attractive forces weaken upon heating and they finally go into the bulk phase. Above their phase transition temperature, there is the stabilization of secondary supra-molecular structure, i.e. a twisted filament structure of β-spirals, which have type II β-turns. The phase transition of these protein- based polymers can be described in terms of an increase in order [5]. Conjugation of ELP and Doxorubicin had demonstrated enhanced delivery of drug due to combined action of EPR (passive targeting) and hyperthermia (active targeting) [10].

Poly (ethylene oxide)/poly (D,L-lactic acid-co-glycolic acid)



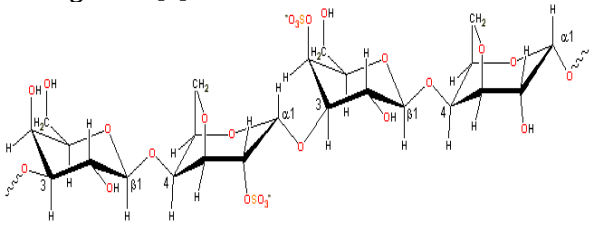
Due to its LCST in the range of 30-35°C, biodegradability properties and maintenance of gel integrity for > 1 month in rats, it is ideal for an injectable sustained release insitu drug delivery system [9].

Various other thermoreversible polymers are Poly (ε-caprolactone)-poly(ethylene glycol)-poly(ε-caprolactone), Poly (methyl 2-acetamidoacrylate-co-methyl acrylate), Poly(organophosphazene) hydrogels, Poly(organophosphazenes) with lactic acid ester and methoxyethoxyethoxy side groups, Poly(*N*-acryloyl pyrrolidine), Polyvinyl acetone [8].

NATURAL POLYMERS

Gelatin [2]

Carrageenan [2]



Xyloglucan

Xyloglucan is a polysaccharide derived from tamarind seeds and is composed of a (1-4)-β-D-glucan backbone chain, which has (1-6)-α-D xylose branches that are partially substituted by (1-2)-β-D-galactoxylose. When xyloglucan is partially degraded by β- galactosidase, the resultant product exhibits thermally reversible gelation by the lateral stacking of the rod like chains. The sol-gel transition temperature varies with the degree of galactose elimination. It forms thermally reversible gels on warming to body temperature. Its potential application in oral delivery exploits the proposed slow gelation time (several minutes) that would allow in-situ gelation in the stomach following the oral administration of chilled xyloglucan solution. Xyloglucan gels have potentially been used for oral, intraperitoneal, ocular and rectal drug delivery [1].

Xanthum gum

Xanthan gum is a high molecular weight extra cellular polysaccharide produced by the fermentation of the gram-negative bacterium *Xanthomonas campestris*. The primary structure of this naturally produced cellulose derivative contains a cellulosic backbone (β- D-glucose residues) and a trisaccharide side chain of β-D-mannose-β-D-glucuronicacid-α-D-mannose attached with alternate glucose residues of the main chain. The anionic character of this polymer is due to the presence of both glucuronicacid and pyruvic acid groups in the side chain [1]. It shows thermoreversible gelation with locust bean gum at appropriate gum concentration. [11].

Chitosan

Chitosan is a biodegradable, thermosensitive, polycationic polymer obtained by alkaline deacetylation of

chitin, a natural component of shrimp and crab shell. Chitosan is a biocompatible pH dependent cationic polymer, which remains dissolved in aqueous solutions up to a pH of 6. Neutralization of chitosan aqueous solution to a pH exceeding 6.2 leads to the formation of a hydrated gel like precipitate. The pH gelling cationic polysaccharides solution are transformed into thermally sensitive pH dpendent gel forming aqueous solutions, without any hemical modification or cross inking by addition of olyol salts bearing a single anionic head such as lycerol, sorbitol, fructose or glucose phosphate salts to chitosan aqueous solution [1, 5]**.**

Gellangum

Gellan gum (commercially available as Gelrite TM or Kelcogel TM) is an anionic deacetylated exocellular polysaccharide secreted by Pseudomonas elodea with a tetrasaccharide repeating unit of one α-L-rhamnose, one β-D-glucuronic acid and two β-D-glucose residues. It has the tendency of gelation which is temperature dependent or cations induced. This gelation involves the formation of double helical junction zones followed by aggregation of the double helical segments to form a three-dimensional network [1].

A list of developed materials exhibiting thermally-driven phase transitions is given in Table 1 [5].

CLASSIFICATION OF TEMPERATURE SENSITIVE HYDROGEL BASED ON ITS ENGINEERING

NEGATIVE THERMOSENSITIVE

Negative temperature-sensitive hydrogels have a lower critical solution temperature (LCST), such hydrogel *contract upon heating* above the LCST. Polymers with low critical temperature (LCST) transition between ambient and physiologic temperature is used for this purpose. e.g. poly(N-isopropylacrylamide) (PNIPAAm). It is a water soluble polymer at its low LCST, but hydrophobic above LCST, which result on precipitation of PNIPAAm from the solution at the LCST [1].

POSITIVE THERMOSENSITIVE

A positive temperature sensitive hydrogel has an upper critical solution temperature (UCST), such hydrogel *contracts upon cooling* below the UCST. e.g. poly(acrylic acid) (PAA), polyacrylamide (PAAm)/poly(acrylamide-co-butyl methacrylate) have positive temperature dependence of swelling [1].

THERMALLY REVERSIBLE GELS

Polymer solution is a free flowing liquid at ambient temperature and gels at body temperature. E.g. Pluronics **[**1**].**

THERMALLY IRREVERSIBLE GELS

Cappello et al. developed novel "protein polymers" ProLastins, which undergo an irreversible sol gel transition. When injected as a solution into the body, the material forms a firm, stable gel within minutes. It remains at the site of injection providing absorption times from less than one week to many months. Such a system would be easy to administer into desired body cavity [1].

FACTORS AFFECTING THERMOREVERSIBLE GEL FORMATION

It is very necessary to understand the following factors before making a thermoreversible formulation to avoid the failure of the formulation [2].

PHYSIOLOGICAL CONDITION

1. Membrane transport
2. pH of tissue fluid
3. Mucocilliary clearance (as in case of nasal administrations).

PHYSICOCHEMICAL PROPERTIES OF POLYMERS

1. Concentration of thermoreversible polymer
2. Molecular weight
3. Transition temperature
4. Hydration value
5. Polymer morphology
6. Crystal state and polymorphism of polymers
7. Phase separation behavior of polymers.

FORMULATION FACTORS

1. Clarity
2. pH
3. Gelation temperature
4. Viscosity
5. Osmolarity
6. Spreadability.

MUCOADHESIVE POLYMERS

Various mucoadhesive polymers can be classified on the basis of their source as shown in table 2.

CLASSIFICATION OF MUCOADHESIVE POLYMERS ON THE BASIS OF THEIR ADHESION TO MUCIN-EPITHELIAL SURFACE [6]

1. Polymers that become sticky when placed in water and owe their mucoadhesion to **stickiness.**

2. Polymers that adhere through nonspecific, noncovalent interactions which are primarily **electrostatic** in nature (although hydrogen and hydrophobic bonding may be significant).

3. Polymers that **bind to specific receptor site** on tile self surface.

CHARACTERISTICS OF AN IDEAL MUCOADHESIVE POLYMER [6**]**

1. It should be nontoxic, nonirritant and nonabsorable from the gastrointestinal tract.
2. It should preferably form a strong noncovalent bond with the mucin-epithelial cell surfaces.
3. It should adhere quickly to most tissue and should possess some site-specificity.
4. It should allow incorporation of the drug, and should offer no hindrance to its release.
5. The polymer must not decompose on storage or during the shelf life of the dosage form.
6. The cost of polymer should not be so high that the prepared dosage form remains competitive.

Molecular characteristics **[**6**]**

1. Strong hydrogen bonding groups (-OH, -COOH).
2. Strong anionic charges; although an anionic nature is preferable for a good mucoadhesive, a range of nonionic molecules (e.g., cellulose derivatives) and some cationic (e.g., Chitosan) can be successfully used.
3. Sufficient flexibility to penetrate the mucus network or tissue crevices.
4. Surface tension characteristics suitable for wetting mucus/mucosal tissue surface.
5. High molecular weight (up to 100,000) [2, 6].

The relative mucoadhesive force of some potential bio (muco) adhesive pharmaceutical polymers is tabulated in table 3 [12].

MECHANISM OF MUCOADHESION

There are two steps which are responsible for the mucoadhesion as shown in figure 4 [2].

Contact stage: An intimate contact (wetting) occurs between the polymer and mucus membrane. It has been demonstrated that the intimacy of contact between the polymer and mucus membrane is improved when the surface of latter is rough. The surface roughness is defined by the aspect ratio (d/h) of the maximum depth (d) to the maximum width (h). The polymers with the aspect ratio less then 1/20 exhibit poor mucoadhesion [2].

Consolidation stage: Various physicochemical interactions occur to consolidate and strengthen the adhesive joint, leading to prolonged adhesion, which improves the mucoadhesive strength [2].

THEORY OF MUCOADHESION

1. Wetting Theory of Mucoadhesion

The wetting theory is perhaps the oldest established theory of adhesion. It is best applied to liquid or low-viscosity bioadhesives. It explains adhesion as an embedding process, whereby adhesive agents penetrate into surface irregularities of the substrate and ultimately harden, producing many adhesive anchors. Free movement of the adhesive on the surface of the substrate means that it must overcome any surface tension effects present at the interface. The wetting theory calculates the contact angle and the thermodynamic work of adhesion [13]:

a. The adhesive work done is related to the surface tension of both the adhesive and the substrate, as given by Dupre's equation:

$$\omega_A = \gamma_b + \gamma_t - \gamma_{bt} \quad (1)$$

where, ω_A is the specific thermodynamic work of adhesion and γ_b , γ_t, and γ_{bt} represent, respectively, the surface tensions of the bioadhesive polymer, the substrate, and the interfacial tension between both phases.

b. Contact angle is inversely proportional to the affinity of the liquid mucoadhesive to the mucous membrane [6]. Figure 5 shows a drop of liquid bioadhesive spreading over a soft-tissue surface:

Horizontal resolution of the forces as shown in figure 6 gives the Young equation 2 [13]:

$$\gamma_{ta} = \gamma_{bt} + \gamma_{ba} \cos\theta \quad (2)$$

where θ is the angle of contact, γbt is the surface tension between the tissue and polymer, γba is the surface tension between polymer and air, and γta is the surface tension between tissue and air.

If $\gamma_{ta} \geq \gamma_{bt} + \gamma_{ba}$, then $\theta = 0$, thus there will be wetting.

If a bioadhesive material is to successfully adhere to a biological surface, it must first dispel barrier substances and then spontaneously spread across the underlying substrate, either tissue or mucus. The spreading coefficient, S_b, can be defined as shown in Equation 3 which states that bioadhesion is successful if *Sb* is positive:

$$S_b = \gamma_{ta} - \gamma_{bt} - \gamma_{ba} > 0 \quad (3)$$

2. Electrostatic theory

Transfer of electrons occurs across the mucoahesive polymer and mucous membrane which results in the formation of the electrical double layer at the interface and a series of attractive forces responsible for mucoahesion [13].

3. Adsorption Theory

The mucoadhesive polymer adheres to the mucin epithelial surface because of one or more secondary surface forces (van der Waal's forces, hydrogen bonding, and hydrophobic bonding) acting between the chemical structures at the two surfaces. When polar molecules or groups are present, they reorientate at the interface. Chemisorption can occur when adhesion is particularly strong [2, 13].

4. Diffusion theory

The concentration gradient will drive the polymeric chains of the bioadhesive to interpenetrate into glycoprotein mucin chains and reach a sufficient depth within the opposite matrix to allow formation of a semipermanent bond as shown in figure 7 [13]**.**

The process can be visualized from the point of initial contact. Depth of diffusion is dependent on the diffusion coefficient of both phases. Reinhart and Peppas reported that the diffusion coefficient depends on

the molecular weight of the polymer strand and that it decreases with increasing cross-linking density [13].

The mean diffusional depth of the bioadhesive polymer segments, *S* , may be represented by Equation 4:

$$S = \sqrt{2tD} \quad (4)$$

Where *D* is the diffusion coefficient and *t* is the contact time. The exact depth needed for good bioadhesive bonds is unclear, but is estimated to be in the range of 0.2-0.5 ˝m.

Duchene used Equation 4 to give Equation 5, which can be used to determine the time, *t*, to bioadhesion of a particular polymer:

$$t = l^2 / D \quad (5)$$

in which *l* represents the interpenetrating depth and *Db* the diffusion coefficient of a bioadhesive through the substrate.

5. Fracture Theory

This theory describes the force required for the separation of two surfaces after adhesion [2, 13] as shown in figure 8. The maximum tensile strength {Sm} produced during detachment can be determined by dividing the maximum force of detachment {Fm} by total surface area involved in adhesion interaction [14]:

$$Sm = Fm/Ao \quad (6)$$

The fracture strength is equivalent to the adhesive strength as per equation 7. This theory is useful for the study of bioadhesion by tensile apparatus [13].

$$\sigma = \sqrt{(E \times \epsilon / L)} \quad (7)$$

where σ is the fracture strength, ϵ fracture energy, *E* young modulus of elasticity, and *L* the critical crack length.

6. Mechanical Theory

This theory assumes that adhesion arises from an interlocking of an irregular adhesive on a rough surface. Surface roughness provides a large surface area as well as large contacts between interfaces [2].

FACTORS AFFECTING MUCOADHESION

1. POLYMER-RELATED

a. Hydrophilicity

Bioadhesive polymers possess numerous hydrophilic functional groups, such as hydroxyl and carboxyl. These groups allow hydrogen bonding with the substrate, swelling in aqueous media, thereby allowing maximal exposure of potential anchor sites. In addition, swollen polymers have the maximum distance between their chains leading to increased chain flexibility and efficient penetration of the substrate [13].

b. Molecular weight

The interpenetration of polymer molecules is favored by low-molecular-weight polymers, whereas entanglements are favored at higher molecular weights**.** The optimum molecular weight for the maximum mucoadhesion depends on the type of polymer. Bioadhesive forces increases with increase in the molecular weight of the polymer ≥ 100,000. Beyond this level, there is no further gain. For example, polyethylene glycol (PEG), having 20,000 molecular weight has little adhesive character, whereas PEG with 200,000 molecular weight has enhanced, and a PEG with 400,000 has superior adhesive properties [13, 15].

c. Cross linking and swelling

Cross-link density is inversely proportional to the degree of swelling. The lower is the cross-link density, the higher the flexibility and hydration rate; the larger the surface area of the polymer, the better the mucoadhesion. To achieve a high degree of swelling, a lightly cross-linked polymer is favored [13]. However, if too much moisture is present and the degree of swelling is too great, a slippy mucilage results and this can be easily removed from the substrate. Maximum bioadhesion in vitro occurs with optimum water content during the dynamic process of bioadhesion [13, 15]. The mucoadhesion of cross-linked polymers can be enhanced by the inclusion in the formulation of adhesion promoters, such as free polymer chains and polymers grafted onto the preformed network [13].

d. Spatial conformation

Despite a high molecular weight of 19,500,000 for dextrans, they have adhesive strength similar to that of polyethylene glycol (PEG), with a molecular weight of 200,000. The helical conformation of dextran may shield many adhesively active groups, primarily responsible for adhesion, unlike PEG polymers, which have a linear conformation [13].

e. Concentration of active polymer

The effect of polymer concentration is dependable on the physical state (solid / liquid) of the bioadhesive drug delivery systems. In Solid BDDS, as the polymer concentration is increased, bioadhesive strength also increases; while an optimum concentration is required for best bioadhesion in liquid BDDS. It was shown by Duchkne that, for solid dosage forms such as tablets, the higher the polymer concentration, the stronger the mucoadhesion. In highly concentrated liquid BDDS, beyond the optimum concentration, the coiled molecules become solvent-poor and the chains available for interpenetration are not numerous; thereby the adhesive strength drops significantly [13].

f. Drug Excipient Concentration

Blanco Fuente studied the effect of propranolol hydrochloride to Carbopol® (a lightly cross-linked poly (acrylic acid) polymer) hydrogels adhesion. Author demonstrated increased adhesion when water was limited in the system due to an increase in the elasticity, caused by the complex formation between drug and the polymer. While in the presence of large volume of water, the complex precipitated out, leading to a slight decrease in the adhesive character. Increasing toluidine blue O (TBO) concentration in mucoadhesive patches based on Gantrez® (poly (methylvinylether/maleic acid)) significantly increased mucoadhesion to porcine cheek tissue. This was attributed to increased internal cohesion within the patches due to

electrostatic interactions between the cationic drug and anionic copolymer [13].

g. Flexibility of Polymer Chains

It is critical for interpenetration and entanglement. Mobility of individual polymer chains decrease as water-soluble polymers become crosslinked and thus the valuable length of the chain that can penetrate into the mucus layer decreases, which reduces mucoadhesive strength [15].

2. ENVIRONMENT RELATED

a. Applied Strength

The adhesive strength increases with the applied strength or with the density of its application up to an optimum. The depth of interpenetration is affected by the pressure initially applied to the mucoadhesive tissue contact site. If high pressure is applied for a satisfactory longer period of time, polymers become mucoadhesive even though they do not have attractive interaction with mucin [13, 15].

b. pH at Polymer Substrate Interface

The pH at the bioadhesive to substrate interface can influence the adhesion of bioadhesives possessing ionizable groups [13]. Mucus will have a different charge density on the basis of pH due to the difference in dissociation of functional groups on the carbohydrate moiety and the amino acids of the polypeptide backbone [15]. Many bioadhesives used in drug delivery are polyanions possessing carboxylic acid functionalities. If the local pH is above the p*Ka* of the polymer, it will be largely ionized and vice versa. The approximate p*Ka* for the poly (acrylic acid) family of polymers is between 4 and 5. The maximum adhesive strength of these polymers is observed around pH 4-5 and decreases gradually above a pH of 6. A systematic investigation of the mechanisms of mucoadhesion clearly showed that the protonated carboxyl groups, rather than the ionized carboxyl groups, react with mucin molecules, presumably by the simultaneous formation of numerous hydrogen bonds [13].

c. Initial Contact Time

Contact time between the mucoadhesive and mucus layer determines the extent of swelling and interpenetration of the mucoadhesive polymer chains. More mucoadhesive strength increases as the initial contact time increases [13, 15].

3. PHYSIOLOGICAL VARIABLES

a. Mucin Turnover

The natural mucin turnover from the mucus layer is important for at least two reasons.

- Due to mucin turnover, mucoadhesives are detached from the surface; no matter how high the mucoadhesive strength is, thereby decreasing the residence time of the mucoadhesive on the mucus layer [2, 15]. The turnover rate differs in the presence of mucoadhesive, blood and disease state [13, 15].
- Substantial amount of soluble mucin molecules results from mucin turnover. Before these mucin molecules have a chance to interact with mucus layer, they first interact with mucoadhesives [15].

b. Disease States

Physicochemical properties of mucus are known to change during diseased states, such as common cold, gastric ulcers, ulcerative colitis, cystic fibrosis, bacterial and fungal infections of the female reproductive tract and inflammatory conditions of the eye. Under these conditions, the exact structural changes taking place in mucus are not clearly understood. The mucoadhesive property must be evaluated when the mucoadhesives are used in the diseased state [2, 15].

Thus a good understanding of influence of all these factors can serve to minimize the problem during the designing and development of new delivery systems.

FORMULATION OF THEMOREVERSIBLE MUCOADHESIVE IN-SITU GEL

Thermoreversible mucoadhesive *insitu* gel is prepared by cold method [16-19].

Incorporation of thermoreversible polymer

Preliminarily concentration of thermoreversible polymer is screened at various concentration (% w/v) for determination of lowest possible concentration that gives thermorevesible gelling property below physiological temperature. Thermoreversible poylmer is added to solution containing drug, and is left at 4°C in refrigerator until a clear solution is obtained [16-18]. Active substances that are insoluble in water are dissolved prior to addition in Ethanol, Isopropyl alcohol or Propylene glycol at 5˚C to form a homogeneous mass [19].

Incorporation of mucoadhesive polymer: Mucoadhesive polymer is screened at various concentrations in thermoreversible polymer solution under gentle stirring for formulation of optimum thermosensitive and mucoadhesive gel. Mucoahesive polymer is slowly added to the above solution with continuous agitation, and is stored in refrigerator until clear solution is obtained [16-17].

EVALUATION OF THEMOREVERSIBLE MUCOADHESIVE IN-SITU GEL

1. **pH of Formulation**

1ml quantity of each formulation was transferred to the 10ml volumetric flask and diluted by using distilled water to make 10ml. pH of resulting solution was determined by using pH meter [20].

2. Viscosity and rheology

This is an important parameter for the *in situ* gels, to be evaluated. The viscosity and rheological properties of the polymeric formulations, either in solution or in gel made with artificial tissue fluid (depending upon the route of administrations) instead of 5% mannitol, were determined with Brookfield rheometer or some other type of

viscometers such as Ostwald's viscometer. The viscosity of these formulations should be such that no difficulties are envisaged during their administration by the patient, especially during parenteral and ocular administration [1].

3. Measurement of gelation temperature

a. Visual inspection : 10 ml volume of solution is transferred to 20 ml transparent vial containing a magnetic stirrer bar. The vial is heated at an increasing rate of 1°C/min with constant stirring at 100 rpm. The temperature at which rotation of bar stopped is taken as the gelation temperature [16, 17].

b. Rheological method

A rheological study is performed with a thermostatically controlled Brookfield Programmable Rheometer fitted with CP-52 spindle. The cone/plate geometry is used. The shear stress is controlled to maintain a shear rate of 10/sec shear rate for precise determination of the gelling temperature. The temperature is increased in steps of 1°C/min, from 20-40°C to locate the solution/gel transition point. The gelling temperature is determined graphically as the inflection point on the curve of the apparent viscosity (mPas) as a function of the temperature (°C) [16].

4. Gel-Strength

This parameter can be evaluated using a rheometer as shown in figure 9 [2, 20]. Depending on the mechanism of the gelling of gelling agent used, a specified amount of gel is prepared in a beaker, from the sol form. The probe is slowly pushed through the gel by placing the weights on the probe, resulting in rising of gel in a beaker at certain rate. The changes in the load on the probe can be measured as a function of depth of immersion of the probe below the gel surface [1]**.**

5. Evaluation of the mucoadhesive strength

The mucoadhesive potential of each formulation is determined by measuring the force required to detach the formulation from mucosal tissue by using a modified chemical balance as shown in figure 10 [2, 17, 20].

A section of mucosa is instantly fixed with mucosal side out onto each glass vial using a rubber band. The vials with mucosa are stored at 37°C for 5 mins. Then next vial with a section of mucosa is connected to the balance in inverted position while first vial is placed on a height adjustable pan. Fixed amount of sample of each formulation is placed onto the mucosa of first vial. Then the height of second vial is adjusted so that mucosal surfaces of both vials come in intimate contact. Then weight is kept rising in the pan until vials get detached. The bioadhesive force is determined from the minimal weights that detached the tissues from the surface of each formulation using following equation [2, 17].

Detachment stress (dyne/cm^2) = m × g /A,

where, m = Weight required for detachment of two vials (gms)

g = Acceleration due to gravity [980cm/s^2]

A = Area of tissue exposed

6. Effect of initial contact time on mucoadhesive strength

Formulation is allowed to be in contact with mucosa for varying contact times, and the bioadhesive force is determined as discussed above. Contact time that resulted in maximum bioadhesive strength is selected as optimum contact time required for adequate adhesion [16].

7. Spreadibility

As evident from the theory of mucoadhesion, a mucoadhesive formulation that is having high spreadability and high surface tension will adhere strongly to the mucus membrane. The spreadability in terms of flow ability of various mucoadhesive thermoreversible gels is determined using a rectangular, hollow, glass chamber (10cm×6cm×4cm) with inlet and outlet of hot water. The mucosa from serosal side is pasted on chamber. Hot water is circulated for acquiring physiological temperature. One drop of formulation is placed on mucosa at an angle of 120° and the distance traveled by drop before it gets converted into gel is determined [17].

8. Texture analysis

The firmness, consistency and cohesiveness of formulation are assessed using texture analyzer which mainly indicates the syringe ability of sol so the formulation can be easily administered in-vivo [1].

9. Fourier transform infra-red spectroscopy

During gelation process, the nature of interacting forces can be evaluated using this technique by employing potassium bromide pellet method [1].

10. Thermogravimetric analysis

It can be conducted for *in situ* forming polymeric systems to quantitate the percentage of water in hydrogel [1].

11. Differential scanning calorimetry

It is used to observe if there are any changes in thermograms as compared with the pure ingredients used thus indicating the drug- excipient interactions [1].

12. In-vitro drug release studies

a. For the *in situ* gel formulations to be administered by oral, ocular, nasal or rectal routes

The drug release studies are carried out by using the plastic dialysis cell. The cell is made up of two half cells, donor compartment and a receptor compartment. Both half cells are separated with the help of cellulose membrane. The sol form of the formulation is placed in the donor compartment. The assembled cell is then shaken horizontally in an incubator. The total volume of the receptor solution can be removed at intervals and replaced

with the fresh media. This receptor solution is analyzed for the drug release using analytical technique [1].

b. For injectable *insitu* gels

The formulation is placed into vials containing receptor media and placed on a shaker water bath at required temperature and oscillations rate. Samples are withdrawn periodically and analyzed [1].

13. Histopathological studies

Two mucosa tissue pieces (3 cm^2) are mounted on in vitro diffusion cells. One mucosa is used as control (0.6 mL water) and the other is processed with 0.6 mL of optimized organogel (conditions similar to in vitro diffusion). The mucosa tissues are fixed in 10% neutral carbonate formalin (24 hours), and the vertical sections are dehydrated using graded solutions of ethanol. The subdivided tissues are stained with hematoxylin and eosin. The sections under microscope are photographed at original magnification ×100. No change on the ultrastructure of mucosa morphology and the epithelial cells on microscopic observations indicate that the organogel has no significant effect on the structure of the mucosa [1].

APPLICATIONS OF THEMOREVERSIBLE MUCOADHESIVE IN-SITU GEL

Thermoreversible polymers with LCST at biologically relevant temperature are having wide biomedical applications. Drug loading to these polymeric delivery systems is readily achieved by simple mixing; however, an inconvenience is the rapid dissolution and release, limiting the use for delivery periods of maximum a few days. This can be modified by the incorporation of a sustain release polymer to increase the duration of release and action [5].

Thermoreversible drug delivery is considerable in the treatment of cancer, pain, transdermal delivery of insulin, gene therapy, tissue engineering and peptide delivery. Transparency of some polymers like Poloxamers makes them ideal for ophthalmic applications. Some of the applications of thermoreversible drug delivery system are discussed below [5].

INJECTABLE DRUG DELIVERY SYSTEMS

Pluronic F127 (PF-127) shows thermoreversible property, which is of the utmost interest in optimizing drug formulation and delivery. However, its hitherto unresolved drawback of a low phase transition temperature [T (tr)] has limited its application in injectable drug delivery systems. This can be overcome by synthesizing a new type of PF-127 copolymers with higher T (tr) using a simple oxidative method. Ma y et al studied the drug-releasing feature of oxidized PF-127 and oxidized PF-127-containing silver nanoparticles (SNPs), carrying arsenic trioxide (ATO), in a subcutaneous model of rats. Injectable hydrogels prepared with oxidized PF-127 were less viscous and easier to inject, at the same concentration, than their precursor. Addition of SNPs further elevated T (tr), resulting in even lower viscosity of the injectable hydrogel prepared from SNP-containing oxidized PF-127. The oxidized PF-127 copolymers did not differ significantly in ATO-releasing ability, compared with parental PF-127, but the addition of SNPs altered the ATO-releasing feature of oxidized PF-127 to some extent. The results presented herein warrant further investigation of the modified PF-127 copolymers to deliver ATO or other drugs in the form of injectable hydrogels [21].

Presently, the treatment of arterial aneurysms has been limited to invasive technique i.e. either surgical clipping or endovascular coiling (for brain aneurysms) and replacement of the weakened section of the vessel by a bypass graft in (aortic and peripheral aneurysms). Novel non invasive technique had been investigated for the treatment of aneurysms by using thermoreversible gelation polymer (TGP) as an embolic material. The right common iliac artery of rats was surgically ligated and an experimental aneurysm was created by applying exogenous elastase. Seven days later, aneurysms were harvested and used. It was concluded that TGP mixed with both dermal fibroblasts and bFGF induced the most advanced thrombus organization in the experimental aneurysms followed by TGP mixed only with dermal fibroblasts. TGP may be useful as a delivery device to deploy dermal fibroblasts and cytokines into experimental aneurysms in rats [22].

INTRAMUSCULAR DEPOT

In addition to systemic targeted administration, localized drug delivery may be accomplished by introducing a drug depot directly at the target site. Implantation of preformed hydrogels has drawbacks viz. their size, a source of potential risks and patient discomfort. To overcome this limitation, design focus is being placed on injectable materials with the ability to form in a mild manner 3-dimensional elastic matrices under physiological conditions such as physiological temperature [5].

OCULAR DELIVERY

For *in situ* gels based ocular delivery, natural polymers such as gellan gum, alginic acid and xyloglucan are most commonly used polymers [1]. The major problem in the drug delivery to the ocular region is the drug loss due to the lacrimal drainage. Additionally, the dilution of the drug released in the lacrimal secretions creates a subtherpeutic concentration which causes failure of therapy. This may be rectified by the use of thermo-responsive gelation systems with additional mucoadhesive property, which enhances the residence time of a drug in the ophthalmic mucosa as shown in table 4. The *in situ* gel formation sometimes may create a blurred vision, but this can be corrected by the use of transparent polymeric gel like pluronic [5].

INTRAORAL DELIVERY

Thermoreversible mucoadhesive *insitu* gel finds its application in the local delivery of mouth, as shown in table 5, where it prolongs the residence time of the drug and

hence provides effective pharmacological action; which is the major drawback of the conventional formulation.

NASAL DELIVERY

Due to easier instillation of a liquid to nasal cavity than that of a solid or powdered medication, thermoreversible sol-gel systems find a good place in targeting the drug to nasal epithelium as well as brain (table 6) via nasal drug delivery. The nasal thermoresponsive solutions, due to their adjusted isotonicity and particle size, cause less irritation to nasal mucosa [5].

VAGINAL AND RECTAL DRUG DELIVERY

One of the major drawbacks of vaginal and rectal suppositories and pessaries is the leakage from the site of action as they melt in the cavities. Also these routes have poor patient compliance. Thermoreversible mucoadhesive insitu gel can be an alternative means of administering the drug with the help of a syringe device. The residence time is increased by the reduction in the leakage. Thus it enhances the bioavailability and effectiveness of the drug. Drugs such as an antifungal, an antiseptic, an antibiotic, a contraceptive or a combination of two or more of these can be delivered through vaginal and rectal formulations in the form of a gel [5]. Various delivery systems are tabulated in table 7.

PERIODONTAL GELS

Temperature-sensitive *in situ* gel containing 0.1% w/v Chlorhexidine hydrochloride was formulated using thermoreversible polymer Poloxamer 407 and Carbopol 934P as mucoadhesive polymer. The formulation showed sustained drug release for a period of 6 h, which satisfied to treat periodontal disease [33].

Bansal et al developed satranidazole thermo reversible mucoadhesive gel for the treatment of periodontitis using thermoreversible polymer sodium carboxymethylcellulose (SCMC) and mucoadhesive polymer carbopol 934P. Gel was evaluated for its clinical effectiveness along with marketed metronidazole gel. At the end of the study (42 days of clinical studies), both formulations were found to significantly reduce the probing depth, plaque index, gingival index, calculus criteria, and bleeding index. However, the developed gel was more effective in reducing the above parameters than marketed metronidazole gel. This study confirmed the acceptability and effectiveness of satranidazole gel for treatment of periodontitis [34].

COMMERCIAL FORMULATIONS OF THERMO REVERSIBLE IN-SITU GEL

Timoptic-XE

It is a timolol maleate ophthalmic gel formulation of Merck and Co. Inc., supplied as a sterile, isotonic, buffered, aqueous gel forming solution of timolol maleate. This formulation is available in two dosage strengths 0.25% and 0.5% in market. The pH of the solution is approximately 7.0, and the osmolarity is 260-330 mOsm. Each ml of Timoptic-XE 0.25% contains 2.5 mg of timolol (3.4 mg of timolol maleate). Inactive ingredients include gellan gum, tromethamine, mannitol, and water for injection and the preservative used is benzododecinium bromide 0.012%. Timoptic- XE, when applied topically on the eye, reduces the elevated, as well as normal intraocular pressure, whether or not accompanied by glaucoma [1].

Regel:depot-technology

Regel is one of the Macromed's proprietary drug delivery system and based on triblock copolymer, composed of poly (lactide-co-glycolide)-poly (ethylene glycol)-poly(lactide-co-glycolide). It is a family of thermally reversible gelling polymers developed for parenteral delivery that offers a range of gelation temperature, degradation rates and release characteristics as a function of molecular weight, degree of hydrophobicity and polymer concentration. Following injection, the physical properties of polymer undergo a reversible phase change resulting in formation of a water insoluble, biodegradable gel depot [1].

- **Oncogel** is a frozen formulation of paclitaxel in Regel. It is a free flowing liquid below room temperature which upon injection forms a gel in-situ in response to body temperature [1].
- **hGHD-1** is a novel injectable depot formulation of human growth hormone (hGH) utilizing Regel drug delivery system for treatment of patients with hGH deficiency [1].
- **Cytoryn** is a novel, peritumoral, injectable depot formulation of interleukin-2 (IL-2) for cancer immunotherapy using Regel drug delivery system. It is a free flowing liquid below room temperature that instantly forms a gel depot upon injection from which the drug is released in a controlled manner. Cytoryn enhances the immunological response by safely delivering four times the maximum tolerated dose allowed by conventional IL-2 therapy. Cytoryn also activates the systemic antitumor immunity. Regel system stabilizes and releases IL-2 in its bioactive form. The release of drugs is controlled by the rate of diffusion from and degradation of the depot [1].

Table 1. Phase transition of various thermoreversible polymers [5].

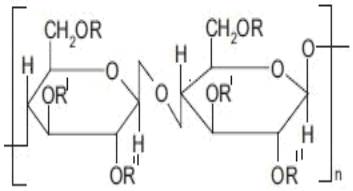
	Thermoreversible polymers	Concentration (%)	Phase transition range (°C)
PNIPAM	PNIPAM	3-5	~32
	PNIPAM-co-AA	>3	30-40
	Nanoparticles of PNIPAM	>2.5	32-35

Poloxamer	Poloxamer 407	20-30	~25
	Poloxamer-co-PAAc	0.5-5	~25
	Oligo (Poloxamers)	20-30	20-30
Cellulose	Methylcellulose	1-5	25-50
	Hydroxylpropyl Methylcellulose	1-5	75-90
	Ethyl hydroxyethylcellulose with surfactants	1-5	30-40
PLGA-PEG	PEG-PLGA-PEG	15-30	~30
	PEG-g-PLGA	15- 3	~30
Miscellaneous	Chitosan	2%	~40
	Peptide	2%	25-65
	Poly(organophosphazenes)	5-10%	~37

Table 2. Mucoadhesive polymers [2, 6].

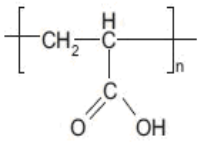
(I) SYNTHETIC POLYMERS

Cellulose derivatives

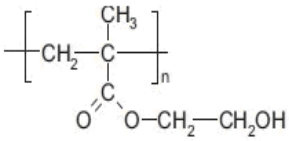


	R	R″	R′
Methylcellulose	H	H	CH_3
Ethylcellulose	H	H	CH_2CH_3
Hydroxy-ethylcellulose	H	H	CH_2CH_2OH
Hydroxyl propyl cellulose	H	H	$CH_2CH(OH)CH_3$
Hydroxy propyl methylcellulose (Hypromellose)	H	CH_3	$CH_2CH(OH)CH_3$
Sodium carboxy methylcellulose	H	H	CH_2COONa

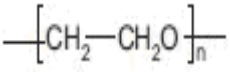
(b) Poly (acrylic acid) polymers (carbomers, polycarbophil, carbopol)



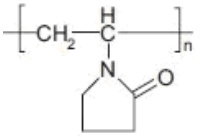
(c) Poly hydroxyethyl methylacrylate



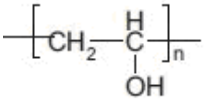
(d) Poly ethylene oxide (PEO) or Poly ethylene glycol (PEG)



(e) Poly vinyl pyrrolidone (PVP



(f) Poly vinyl alcohol (PVA)



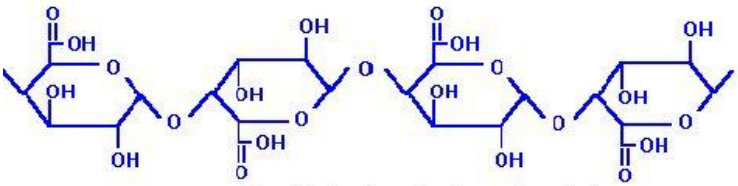
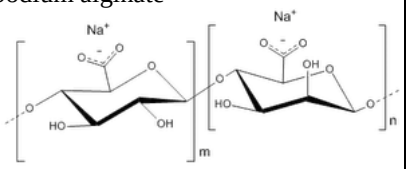
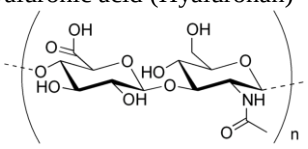
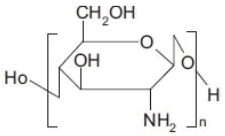
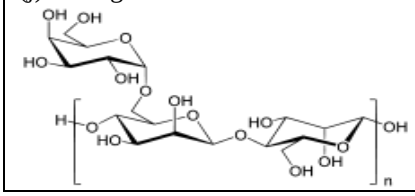
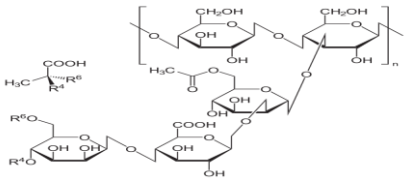
<table>
<tr><td colspan="3">(II) NATURAL POLYMERS</td></tr>
<tr><td>(a) Tragacanth
(c) Soluble starch
(d) Lectin
(e) Karaya gum
(f) Gelatin</td><td colspan="2">(b) Pectin
 
Pectin (polygalacturonic acid)</td></tr>
<tr><td>) Sodium alginate
  </td><td>(h)Hyaluronic acid (Hyaluronan)
  </td><td>(i) Chitosan
  </td></tr>
<tr><td>(j) Guar gum
  </td><td colspan="2">(k) Xanthan gum
  </td></tr>
</table>

Table 3. Mucoadhesive force of various mucoadhesive polymer.

Mucoadhesive Polymer	Relative Mucoadhesive Force	Qualitative Bioadhesion Property
Carboxy methyl cellulose	193	Excellent
Carbopol	185	Excellent
Polycarbophil	-	Excellent
Tragacanth	154	Excellent
Sodium alginate	126	Excellent
HPMC	125	Excellent
Gelatin	116	Fair
Pectin	100	Poor
Acacia	98	Poor
Povidone	98	Poor

Table 4. Ocular drug delivery systems.

Drug	Thermoreversible polymer	Mucoadhesive polymer	Conclusion
Fluconazole	Pluronic 407	HPMC K4M, HEC, PVP K30	The order of drug permeation through the membrane was HEC > PVP K30 > HPMC K4M [23].
Moxifloxacin hydrochloride	Poloxamer 407 and 188	0.5% and 1% HPMC K4M	Sustained release of drug (98%) over 12 hr [24].

Table 5. Intraoral drug delivery systems.

Drug	Thermoreversible polymer	Mucoadhesive polymer	Conclusion
Mebeverine hydrochloride	Polaxamer 407	2% of HPMC K100M	Prolong residence time [25].
Naproxen	Pluronic F127 (PF127)	Carbopol 934P	Improve oral residence time and absorption of naproxen [26].

Table 6. Nasal drug delivery systems.

Drug	Thermoreversible polymer	Mucoadhesive polymer	Conclusion
Sumatriptan	Pluronic F127	Carbopol 934P	Increase in residence time and better patient compliance [16].
Amitryptylline HCl	Pluronic PF127	HPMCK4M	1. Sustained delivery 2.Increased bioavailability as compared to oral route (40%) [27].
Venlafaxine HCl	Lutrol F127 (18%)	0.3 % PVP K30	Drug was more effective in comparision to oral administration of equivalent dose [28].
Nanoparticles loaded with levodopa	Pluronic PF127	chitosan	Improved brain uptake of levodopa alone, thus avoiding the use of carbidopa unlike conventional dosage form [29].
Raloxifene HCl in thermoreversible mucoahesive microemulsion insitu gel (tmmig)	14 % Pluronic F127	0.3 % carbopol 934P	1.Controlled delivery. 2.Increase in bioavailability as compared to oral route (B.A. 2%) [30].

Table 7. Vaginal and rectal drug delivery systems

Drug	Thermoreversible polymer	Mucoadhesive polymer	Conclusion
Clotrimazole as inclusion complex with β-cyclodextrin 1:1 ratio	Pluronic F127 (20%)	0.2 % HPMC	Continous and prolong release of drug, better therapeutic efficacy and patient compliance in the treatment of vaginitis [31].
Mebeverine hydrochloride	poloxamer 407 (23%) + poloxamer 188 (7 %)	1.5% MC	1. Increase in bioavailability. 2. Restrict its absorption to only the lower rectum [32].

Fig 1. Transition of thermoreversible insitu gel from sol – gel.

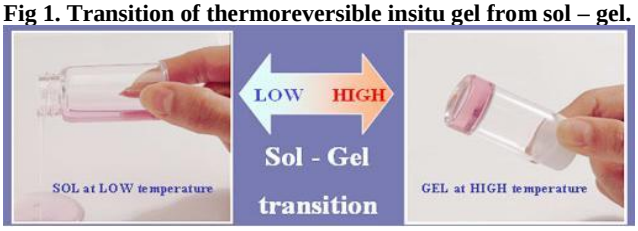


Fig 2. Curing of thermoreversible polymer

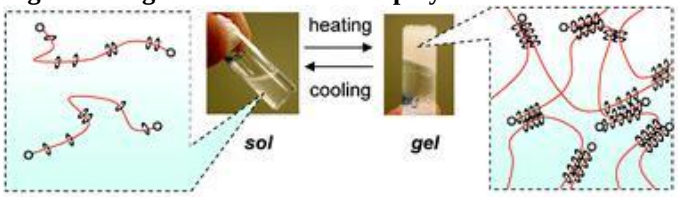


Fig 3. Poloxamer structure (A) and self-assembly (B).

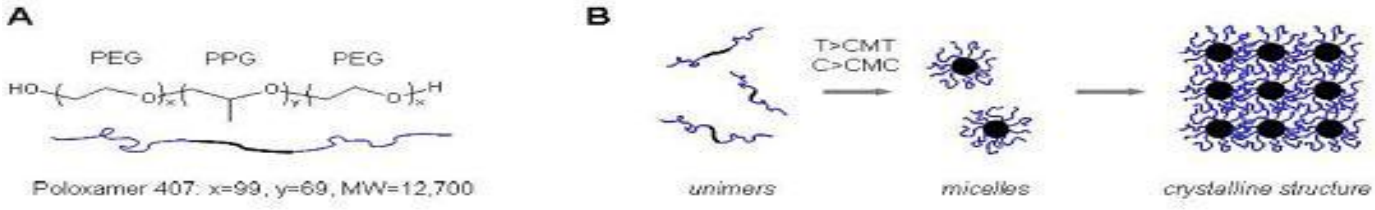


Fig. 4. Two steps of mucoadhesion

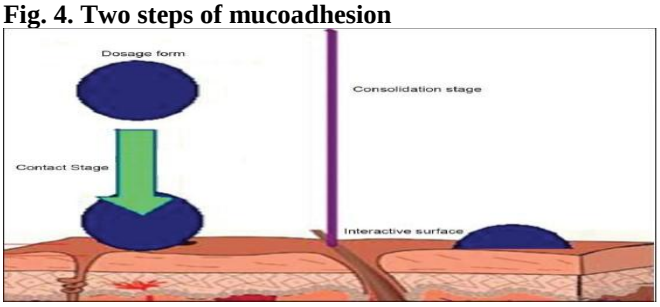


Fig 5. A liquid bioadhesive spreading over a tissue surface.

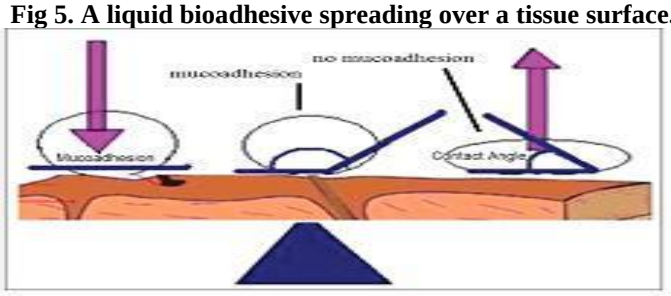


Fig 6. Forces affecting the contact angle.

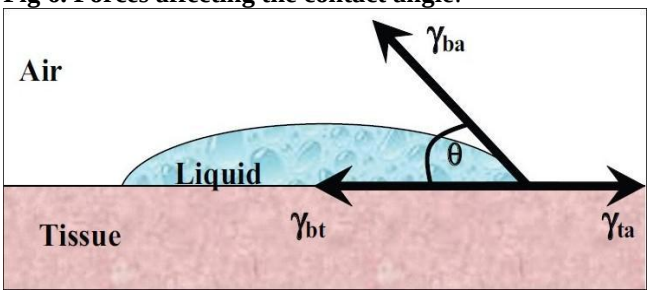


Fig. 7. Schematic representation of the diffusion theory of bioadhesion.

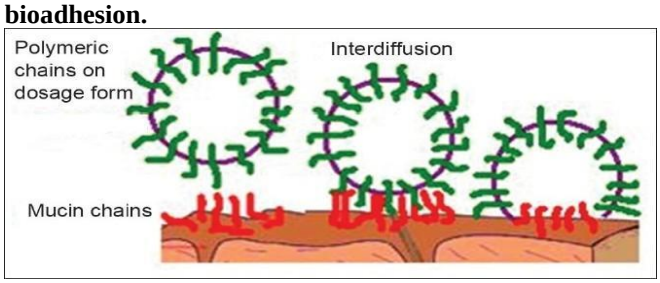
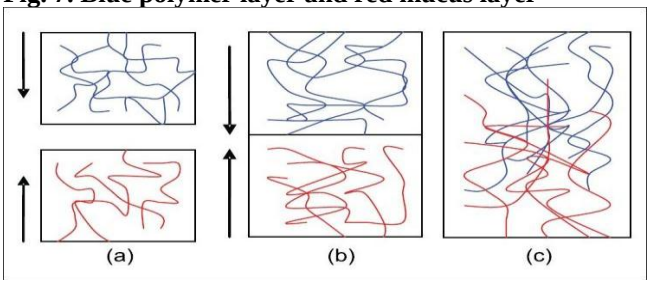


Fig. 7. Blue polymer layer and red mucus layer



Blue polymer layer and red mucus layer (a) before contact; (b) upon contact; (c) the interface becomes diffuse after contact for a period of time.

Fig 8. Mucoadhesion fracture theory.

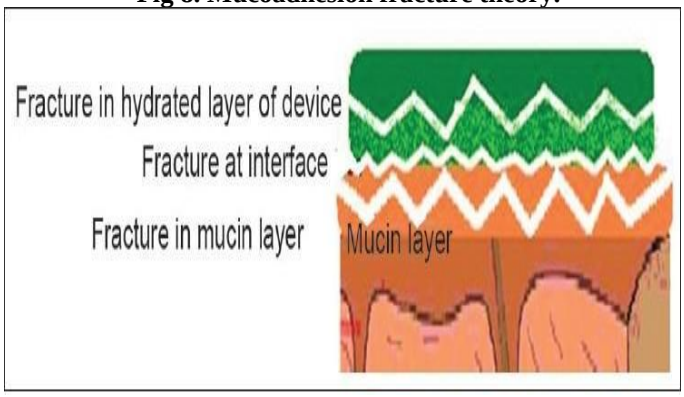


Fig 9. Rheometer

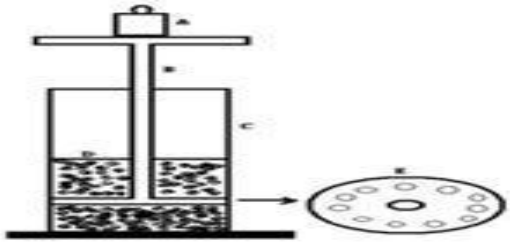
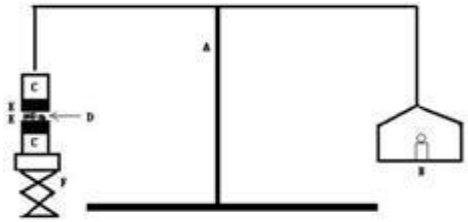


Fig 10. Modified chemical balance for measuring the mucoadhesive force.



CONCLUSION

The primary requirement of a successful drug administration focuses on increasing patient compliance which the thermo responsive *in situ* gelling systems offer. Development of polymeric *in situ* gels for delivery of various drugs provides a number of advantages over conventional dosage forms. Instead of acting passively as pure drug carriers, they interact and respond to the environmental setting. Sustained and prolonged release of the drug, good stability and biocompatibility characteristics makes the *in situ* gel dosage forms more acceptable and excellent drug delivery systems. Due to their adjustable target gelation temperature, thermoreversible drug delivery system can be used for many routes of administration. While due to compatibility with the wide range of controlled release polymers and other pharmaceutical excipients, the drug release rate can be adjusted very easily. Thus, the stimuli sensitive, thermoreversible mucoahesive drug delivery system become a reliable, patient compliant method with increased residence time and bioavailability of many drugs due to mucoadhesive property.

FUTURE PROSPECTS

Various biodegradable polymers are used for formulation of *in situ* gels, but there are fabrication problems, difficult processability, use of organic solvents for their preparation (especially for synthetic polymer based systems), burst effect and irreproducible drug release kinetics. Natural polymers satisfy the characteristics of an ideal polymer but batch to batch reproducibility is difficult therefore synthetic polymers are used. The work may be done for the minimization of these limitations. Properties of both the types of polymers may be changed by physical and chemical methods for the improvement in the performance of the polymers. The numbers of thermoreversible polymers are very low and the performances of existed classes are not satisfactory.

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