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Polymer Based Microgels/Nanogels: Development and Application In Drug Delivery

Ganga Srinivas[1]*, Pooja Ramnathkar[1]

1.Department of Pharmaceutics, Vivekanand Education Society's College of Pharmacy, Chembur, Mumbai – 400078

ABSTRACT

Microgels/nanogels are crosslinked polymeric particles, which can be considered as hydrogels if they are composed of water soluble/swellable polymer chains. These polymeric particles possess high water content, biocompatibility, and desirable mechanical properties. Offers unique advantages for polymer-based drug delivery systems (DDS) such as a tunable size from nanometers to micrometers, a large surface area for multivalent bioconjugation, and an interior network for the incorporation of biomolecules. This review article describes the recent developments of microgel/nanogel particles as drug delivery carriers for biological and biomedical applications and also various synthetic strategies for the preparation of microgels/nanogels.

Keywords: Microgels, nanogels, polymers based drug delivery systems.

*Corresponding Author Email: gangach@rediffmail.com

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INTRODUCTION

Microgels are defined as cross-linked polymeric particles which are confined to smaller dimensions (100nm-1µm). Nanogels are defined as submicron sized microgels. These polymeric particles are also termed as hydrogels, if they are composed of water soluble or water swellable polymeric chains[1]. Microgel/nanogels possess tunable chemical and three-dimensional (3D) physical structure, good mechanical properties, high water content and biocompatibility[1]. Thus the design and preparation of these polymeric particles have attracted a wide interest in various fields such as biomedical engineering, pharmaceutical application, biomaterial science, bio nanotechnology etc.

Types of Microgels/nanogels

Based on origin of polymer used for the preparation of microgels/nanogels

i. Synthetic microgels/nanogels: Generally prepared by heterogeneous polymerization of hydrocarbon based hydrophilic or water soluble monomers in presence of multi-functional crosslinkers[2].

   Examples of monomers, N-isopropylacrylamide, 2-hydroxyethyl methacrylate, 2-hydroxypropyl methacrylate, poly acrylamide etc.

ii. Biopolymer based microgels/nanogels: Possess high content of functional group such as hydroxyl, amino, carboxylic acid group. These functional group are utilized in crosslinking for bio conjugation with cell targeting agents[2].

   Examples of biopolymers, chitosan, hyaluronan, dextran, cellulose, alginate etc.

Based on stimuli response from external environment

These can be further classified into two main categories such as physical stimuli e.g. change in temperature, application of light, electric or magnetic field and chemical stimuli which includes change in pH, ionic strength or presence of chemical or biological compounds[3]. These external stimulus bring about changes in physico-chemical parameters of the polymer inducing alteration of the hydrophilicity of polymer functional group.

i. Temperature sensitive microgels/nanogels: Generally poly(N-isopropylacrylamide) is used as network forming polymer. This polymer exhibits a lower critical solution temperature (LCST i.e the critical temperature below which the components of a mixture are miscible for all compositions[4]). Thus the mechanism underlies an endothermic, entropically driven phase transition from a random coil of the polymer

to a collapsed globule at temperature above 32^0C[3].

ii. pH and ionic strength sensitive microgel/nanogels: These are prepared by crosslinking of weak polyelectrolytes which undergoes a pH-dependent phase transition. The mechanism involves swelling/deswelling of materials as response to change in pH of the environment due to protonation/deprotonation of the weakly acid and weakly basic groups along the chain of network forming polymers[3].

iii. Light sensitive microgels/nanogels: In this approach, several chromophores change their polarity upon irradiation leading to phase transition. Co-polymers of acrylamide and poly(n-isopropyl acrylamide) can be used as photo-sensitive polymers[3].

iv. Microgels/nanogels based on cleavable crosslinking points: The concept is based on either complete or partial cleavage of crosslinking point resulting in increase swelling or total dissolution of the polymeric particles. Different triggers that have been applied to induce crosslinkers degradation are hydrolytically degradable cleavable crosslinker, enzymatically cleavable cross-linkers, photo degradable cross-linkers[3].

Properties of microgels/nanogels

i. Microgels/nanogels have high water content, adjustable chemical and physical properties[1,2]

ii. Due to their small size have large surface area for multivalent bio conjugation[1, 2]

iii. Have an interior network for incorporation of bio molecules[1,2]

iv. Physical entrapment of bioactive molecules such as proteins, drugs and DNA as well as their *in vitro* release behaviour, have been widely investigated as drug delivery carriers for biomedical application[1, 2]

Important parameters based on properties of microgels/nanogels

Several parameters are to be considered to design and develop effective microgel/nanogel based drug delivery for *in vivo* application

i. Stability for prolonged circulation in the blood stream: This parameter is important because the instability of microgels/nanogels may result in premature release of therapeutics causing adverse side effects[1]

ii. Novel functionality on the surface of polymeric particle for bio conjugation with specific ligands that recognizes receptors on diseased cells: For e.g. the effective ligand for cancer cells which are bio conjugated with polymeric particle's surface include folic acid derivatives, peptides, proteins etc which helps in active targeting of cancer cells[1]

iii. Control over the dimension below 200nm in diameter: This facilitate cellular uptake of nanogels through a receptor-mediated endocytosis cross cell membranes as well as reduce their uptake by mononuclear phagocyte system (MPS), consequently increasing their blood circulation time[1]

iv. Biodegradability of microgels/nanogels: Biodegradation should not only control the release of drug but also enable to remove empty device after release of drug. Thus development of cross-linkers functionalized with bio degradable linkages such as peptides, anhydrides, oligo esters, disulfidesetc have been done where these linkages were degraded to water soluble polymers in external environments[1]

Preparation of microgels/nanogels

Several methods have been developed for preparation of microgels/nanogels. These methods are classified as follows,

1. Photolithographic and micro molding methods
    i. Photolithographic technique
    ii. Micro molding technique
2. Microfluidic preparation
3. Fabrication of biopolymers
    i. W/O heterogeneous emulsion method. This method is further classified as inverse (mini) emulsion method, reverse micellar method and membrane emulsification method.
    ii. Aqueous homogenous gelation method
    iii. Spray drying method
    iv. Chemical crosslinking of dextran which involves Michael addition reaction and free radical polymerization.
    v. Heterogeneous free radical polymerization which includes methods such as dispersion polymerization, precipitation polymerization, inverse (mini) emulsion polymerization and inverse micro emulsion polymerization.

Of the above described methods for preparation of microgels/nanogels, in this review we will be describing the most widely used methods for the preparation of microgels and nanogels.

Photolithographic technique

This technique involves a top-down process called ‘Particle Replication In Non wetting Templates (PRINT) Figure1, which fabricate submicron sized microgels with control over shape, particle size and composition. Replica of photocurable perfluropolyther (PFPE) was used as

moulding material[1,5]. This offers advantage of elimination of the formation of residual interconnecting film between moulded objects which allows for the preparation of isolated objects. In this approach, the master templates of silicone were made using electron beam lithography[1].

Using PRINT process, mono dispersed microgels of poly(ethylene glycol diacrylate), triacrylate resins and polypyrrole were prepared in various shapes such as bar, conical, trapezoidal and arrow.

Figure1 illustration of PRINT process[1]

Microfluidic preparation

This approach have been recently developed for the preparation of monodispersed micron sized microgel. The microfluidic device is fabricated using elastomeric materials such as poly(dimethyl siloxane) or polyurethane elastomer as building blocks. The device consist of inlets for monomers and continuous liquid and microchannels with tapered junction where two immiscible liquids merge. The mechanism involves emulsification of monomers breaking up liquid threads to droplets and then *in situ* crosslinking of the resulting droplets by photopolymerization or polycondensation[1]. The shape and morphology of the polymeric particles is governed by variation of flow rates of liquid and reaction time.

Several approaches for the microfluidic preparation of micron sized polymeric particle is based on gelation methods in microchannel such as chemical geltion, physical gelation by temperature change, reversible shear thinning and ionic crosslinkers, and coalescence induced gelation[1,6] In this review article physical gelation by ionic crosslinking will be discussed.

Physical gelation with ionic crosslinking involves two approaches Figure 2, internal gelation where droplet consisting of a gelling polymer and a crosslinking agent precursor which is in inactive form. The liquid phase containing crosslinking activator diffuses into droplet and react with crosslinking agent precursor leading to gelation[1, 6]. Second approach involves external gelation where droplets are emulsified in continuous phase that contains crosslinking agent. Diffusion of crossliknking agent to emulsified droplets lead to gelation. Thus the crosslinking

agent should be soluble in aqueous droplet. E.g. microfluidic preparation of alginate Micro gels crosslinked with calcium (Ca^{2+}) ions[1, 6]

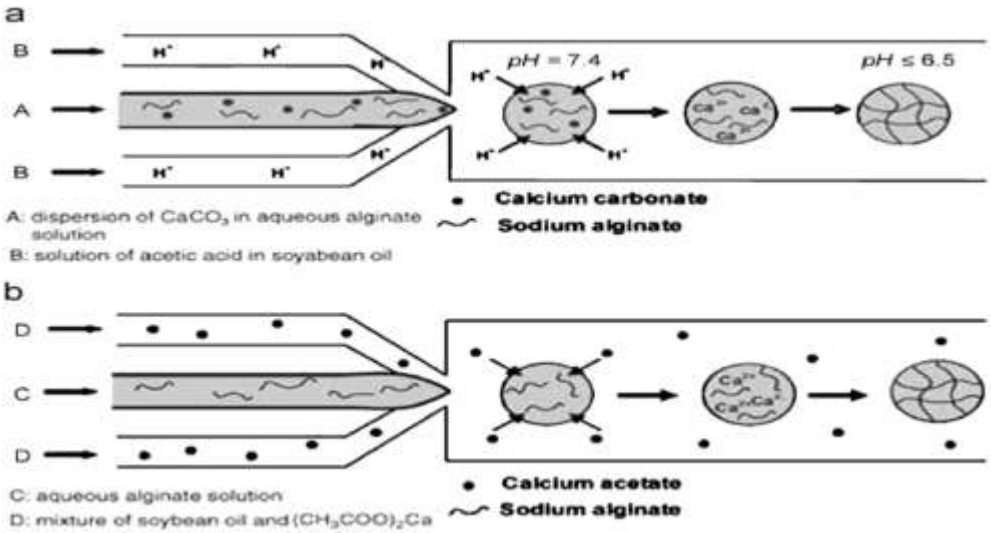

Figure 2 Illustration of microfluidic preparation of alginate microgels using internal gelation (a) and external gelation (b), where $CaCO_3$ is used as crosslinking agent precursor and acetic acid as crosslinking agent activator[6].

Fabrication of biopolymer

The naturally occurring biopolymers are non-toxic and offer high water solubility, biocompatibility and biodegradability. Thus many methods have been developed for the preparation of microgels/nanogels of these biopolymers.

i) *W/O heterogeneous emulsification method.* This approach involves generally two steps; step 1. Emulsification of aqueous droplets of biopolymers in continuous oil phase with an aid of oil soluble surfactants, step 2. Crosslinking of biopolymers with water soluble crosslinkers. The approaches include inverse (mini) emulsion, reverse micelle method and membrane emulsification. Of the methods described we will be discussing inverse (mini) emulsion method[1].

In inverse (mini) emulsion method a w/o emulsion is formed, where aqueous biopolymer droplets are dispersed in a continuous oil phase using a homogenizer or a high-speed mechanical stirrer[1,2]. Generally mineral oil and hexane are used as continuous phase, whereas span 80 and aerosol OT (sodiumbis (2-ethylhexyl) sulfosuccinate) are used as oil-soluble surfactants. Various drugs are physically incorporated into aqueous droplet and the resulting droplets are then crosslinked with crosslinking agents[1]. These crosslinked particles are prepared as dispersion and purified by centrifugation, washing with organic solvents, lyophilization or

precipitation. E.g. preparation of gelatin based Microgels crosslinked with glutardialdehyde[2] Figure 3.

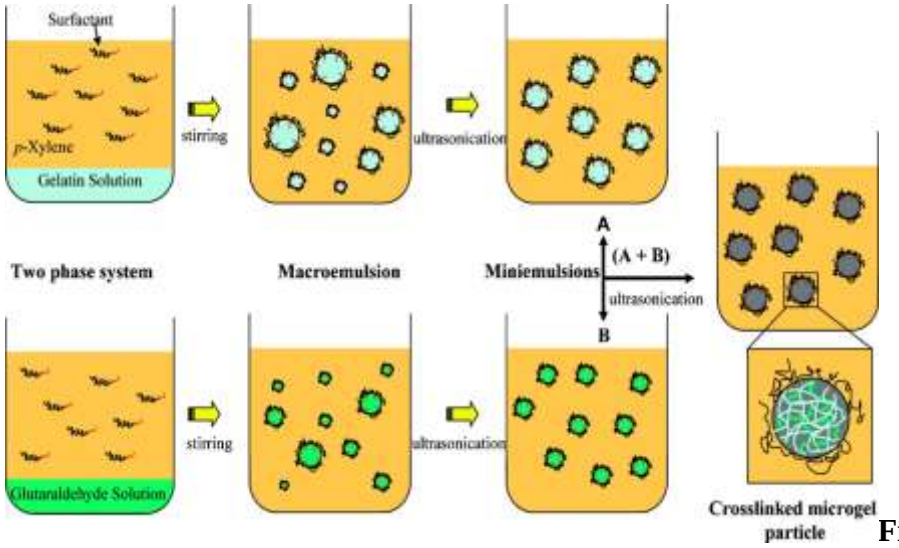

Figure 3

Illustration of synthesis of gelatine-based microgelscrosslinked with glutardialdehyde[2].

ii) *Aqueous homogenous gelation method*. The method involves two approaches covalent chemical crosslinking utilises carbodiimide couple reaction of biopolymer with water soluble crosslinker, and reversible physical crosslinking involves ionic gelation based on electrostatic interaction of cationic biopolymer such as chitosan with polyanion in water[1]. This method is advantageous as it prevents possible toxicity of crosslinker agents and reagents that are involved in chemical crosslinking approach.

iii) *Spray drying method*. This method is widely used in pharmaceutical sciences. The method involves use of a spray dryer which consists of atomizer and drying chamber. Solution and suspension of drugs, polymers are atomized to fine droplets through the nozzle of the atomizer[1,2]. A stream of hot air in drying chamber induces quick evaporation of solvents from droplets resulting in formation of microgels/nanogels. The obtained particles are dried in vacuum chamber and collected. Nozzle size, spray flow rate, atomization speed and extent of crosslinking determine the size of the particle. This method has been developed to prepare microgles of chitosan and hylurauon polymers as biodegradable drug delivery carriers[1].

iv) *Chemical crosslinking of dextran*. Biodegradable dextran based micogels/nanpgels are prepared using various method based on chemical crosslinking such as

carbodiimide coupling, Michael addition reaction which involves modification of hydroxyl group of dextran with thiol-acrylate networks and free radical polymerization which involves modification of hydroxyl group of dextran with different methacrylate precursors e.g. glycidyl methacrylate, 2-hydroxyethyl methacrylate[1].

v) *Heterogeneous free radical polymerization.* This method involves reaction of hydrophilic or water-soluble monomers in presence of either difunctional or multifunctional crosslinkers. The various approaches include dispersion polymerization, inverse (mini) emulsion polymerization and inverse micro emulsion polymerization utilizing an uncontrolled free radical polymerization approach[1]

Of the above mentioned methods we will be discussing inverse microemulsion polymerization. In this approach, aqueous droplet of biopolymer are stably dispersed in a continuous organic medium with the aid of large amount of oil soluble surfactants. Polymerization occurs in aqueous droplet, producing stable hydrophilic and water-soluble colloidal nanoparticles. Well defined nanoparticles of poly(N-isopropylacrylamide), polyacrylamide and poly(dimethylacrylamide) as well as magnetic nanoparticles containing iron oxide are prepared using this method[1].

vi) *Heterogeneous controlled/living radical polymerization(CRP).* This is a versatile method for preparation of polymeric particles with controlled molecular weight, narrow molecular weight distribution, well defined structure and useful end functionalities. This method is widely used to prepare polymer-protein/peptide bioconjugates. Various methods for CRP have been developed, includes atom transfer radical polymerization (ATRP), stable free radical polymerization (SFRP), and reversible additional fragmentation chain transfer polymerization (RAFT). However only few successful CRP preparations have been reported. A ATRP in inverse mini emulsion for synthesis of crosslinked nanogels of well-controlled water soluble polymers in presence of a disulfide-functionalized dimethacrylate was reported.

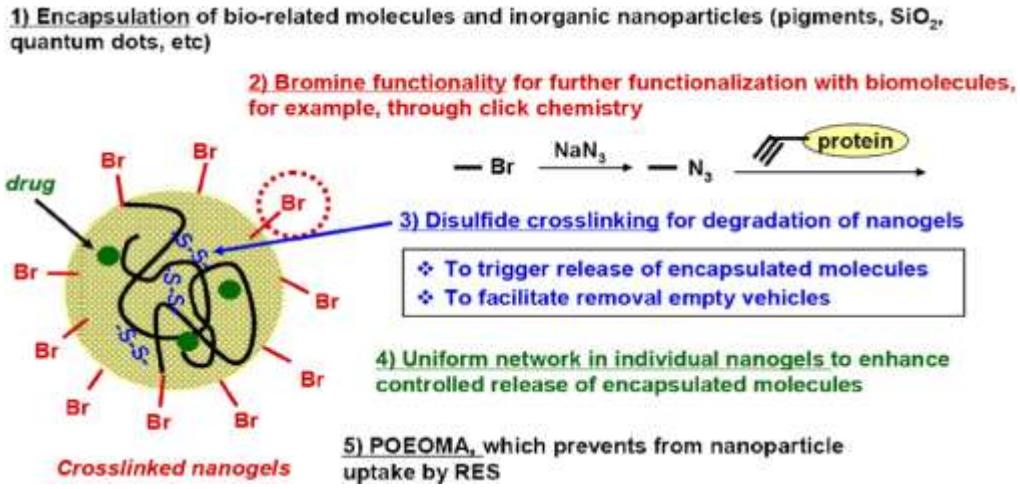

Figure 4 Illustration of unique features of stable biodegradable nanogels of well-controlled water-soluble polymers in presence of a disulfide functionalized dimethacrylate for effective targeted drug delivery[1].

This approach utilized various features[1]as illustrated in Figure 4 such as 1. The surface of particle has a high degree of halide functionality for functionalization with biomolecules. 2. Incorporation of a high loading level of drugs such as anticancer drugs. 3. Degradable disulfide linkage which triggers release of encapsulated molecules and removal of empty device. 4. The degraded individual polymeric network in individual particles. This improves control over the release of encapsulated agents. 5. The coating of polyethylene glycol prevents nanoparticle uptake by reticuloendothelial system (RES) thus enhancing blood circulation time of the nanoparticles. The stability of nanoparticle prepared by ATRP is superior to those prepared by convectional free radical polymerization.

A well defined functionalized nanogel prepared by this method provides great potential as drug delivery carriers to target-specific cells for biomedical application.

Pharmaceutical consideration

i. *Drug loading*: The incorporation of active compounds into microgels/nanogels matrices can by two methods[3]

- Post-formation loading, functional compounds are incorporated in preformed gel networks either by soaking the gel networks in concentrated solution of respective substance or by adsorption[3]
- *In situ* loading, incorporation of substances is achieved during the network formation[3]

ii. *Drug release:* the release of incorporated substances from the gel network of microgels/nanogels is governed by passive diffusion and the rate-determining step of the release is categorized in three different mechanism[3]

1. Diffusion-controlled: in this approach two concepts are distinguished based on internal structure of the gel network. Frist approach, the pore size of gel network is larger than the hydrodynamic diameter of the incorporated compounds thus the diffusion of the substance is governed by porosity and tortuosity of the gel than the internal network structure. Second, the porous gel with pore sizes comparable to the dimensions of the incorporated substances than diffusion is governed by the polymeric chains in the crosslinked network, which provides stearic hindrance to the embedded compounds[3].
2. Swelling-controlled: the mechanism is based on swelling of gel as the rate limiting step for release i.e the diffusion of incorporated compounds is faster than the swelling of the gel matrix[3].
3. Chemically-controlled: the release mechanism is based on chemical reaction in the gel matrix. The chemical reaction includes the cleavage of network forming polymer chains resulting in surface or bulk erosion of the microgel/nanogel particle or the cleavage of the pendant chains between the polymeric network and the compound to be delivered[3].

Characterization

1. Particle size and particle morphology. Morphology can be characterized using a laser scanning confocal microscopy (LSCM). LCMS equipped with a digital camera allows real-time observation of hydrogels[7,19].
2. Water uptake. Thermal gravimetric analyzer (TGA) is used to determine the amount of aqueous solution taken up by the hydrogel particles. The weight of ten dried hydrogels were determined and then soaked in distilled water for 3hrs to allow swelling of the particles. The water uptake is calculated from the difference in weight of fully hydrated hydrogel (Wh) particles and dried particle (Wd)[19],

$$\text{Water uptake} = (Wh - Wd/Wd) \times 100$$

3. Drug loading efficiency: The amount of model drug loaded in hydrogel particle or loading efficiency is determined using the equation[19];

$$\text{Loading efficiency} = \frac{\text{Amount of drug loaded in the particle}}{\text{Dried weight of the hydrogel particles}}$$

4. Hydrophobicity: hydrophobicity governs transport, distribution, metabolism of molecules. Protein folding etc. thus is determination is done using two-phase partition chromatography, adsorption hydrophobic dyes, adsorption of radiolabelled probes[20].
5. Protein adsorption: two dimensional polyacrylamide gel electrophoresis (2D-PAGE) can be used to determine the microgels/nanogels plasma protein interaction,quantitively[7]

Advantages of microgels/nanogels

i. Offers biocompatibility, controllable biodegradability, absorbability and low toxicity of the degradation end products[1, 2]
ii. Sustained release potentials and ease of administration[1,2]
iii. Stimulus response to effect of temperature and pH[1,2,3]
iv. Due to their small size they can pass through the sinusoidal spaces in the bone marrow and spleen as compared to other systems like microspheres and liposomes. Circulation time in the blood can be increased[1,7]
v. Due to their larger surface area, they have higher loading capacity[1,7]
vi. Nanogels can act as controlled release system depending on their polymeric compositions[1,2,9]
vii. Targeting moieties can be attached to enhance their specificity[1,2,9]
viii. These are effective in site-specific and targeted drug delivery[1,2]

Limitations

i. High surface area that may lead to high aggregation in biological system[1,7]
ii. Can be quickly scavenged by RES system of body resulting in low biological half-life[1,7,9]
iii. High immunogenicity or foreignness[1,9]
iv. Possibility of remaining empty device after the drug has been released[1]
v. High specialized techniques required to control the dimension of particles in range of micro meter to nanometer[1,7]

Application of microgels/nanogels as in drug delivery

i. Microgels/nanogels can act as a carrier system for drug delivery. For e.g. multi-responsive nanogels containing motifs of ortho ester, oligo(ethylene glycol) and disulphide linkage act as carriers of hydrophobic ant-cancer drugr such as doxorubicin[10]
ii. The incorporation of magnetic particles in nanogels/microgels and subsequent placement of external field around the desired site of action can improve the efficacy of the drug regimen by magnetic targeting[1]

iii. Microgels can also be used as a carrier for delivery of drug into deep lungs[11]

iv. Microgels can be used for delivery bio macromolecular such as proteins, peptides[12]

v. Polymeric nanogels are used as vaccine drug delivery. Polymeric nanogels protects vaccines antigens from degradation in-vivo, and surface conjugates with antibodies or specific ligands, could increase active targeting specificity[13]

vi. Microgels prepared by the emulsion co-polymerization of bifunctional and monofuctional monomers had good shelf stability and dispersed inorganic pigment showed excellent film appearance[17]

vii. Novel core-shell poly(acrylamide) magnetic nanogels suggested promising potential applications in targeted radiopharmaceutical carriers for cancer therapy, and in biological and medical studies[14]

viii. Self-rupturing microcapsules which consist of a biodegradable dextran-based microgel surrounded by a polyelectrolyte membrane showed pulsed drug delivery of the drug[16]

ix. Encapsulation of cells in microgels has been demonstrated as an effective approach for cell immobilization, cell transplantation, artificial cell, cell therapy, bioassay and diabetes treatment[21]

x. Microgels has also been used for tissue engineering[21]

xi. Hybrid micro-/nanogels for optical sensing and intracellular imaging[18]

CONCLUSION

Recent development of microgel/nanogel particle as drug delivery carriers for biomedical applications were reviewed in this report. Major synthetic strategies for the preparation of microgels/nanogels were described. However from the report we can conclude that the design and development of effective micro gel-based DDS for in-vivo applications require a high degree of control over properties. These include excellent stability for prolonged circulations in the blood stream; novel functionality for further bioconjugation; dimension less than 200 nm in diameter and biodegradability for facile removal of the empty device and sustained release of drugs. Thus the future goal of research on this area is to improve design of microgels/nanogels with specific targeting residue to enable highly selective uptake into particular cells, especially targeting to cancer cells. Also there is a need to elucidate the specific interactions between biomolecules and cellular integral receptors. In this way, careful control over stability, size, biodegradability and functionality for bioconjugation will guide the development of the next

generation of microgels/nanogels.

REFERENCES

1. Jung Kwon Oh *et al.* The development of microgels/nanogelsfor drug delivery applications. Progress in polymer science 2008; 33:
2. Jung Kwon Oh *etal.* Biopolymer-based microgels/nanogels for drug delivery applications. Progress in polymer science 2009; 34:1261-1282.
3. Daniel Klinger, Katharina Landfester. Stimuli-responsive microgels for the loading and release of functional compounds: Fundamental concepts and application. Polymer 2012; 53:5215-5225.
4. M. Constantin, M. Cristea*et al*. Lower critical solution temperature versus volume phase transition temperature in thermo responsivedrug delivery system. EXPRESS Polymer Letters 2011; Vol.5:10:839-848.
5. Byron D. Gates. Nanofabrication with molds and stamps. Review feature 2005:44-49.
6. Rong-Cong Luo, Chia-Hung Chen. Structured Microgels through Microfluidic assembly and their biomedical applications. Review article 2012;1-23.
7. N.K.Jain. An introduction to novel drug delivery systems, 1st ed, Vallabh Prakashan; 2010:145-153.
8. Reuben T. Chacko*et al.* Polymer nanogels: A versatile nanoscopic drug delivery platform. Advanced drug delivery reviews 2012;64:836-851.
9. Serguel V Vinogradov. Nanogels in the race for drug delivery. Nanomedicine 2010;5(2): 165-168.
10. Zeng-Ying Qiao*et al.* Multi-responsive nanogels containing motifs of ortho ester, oligo(ethylene glycol) and disulfide linkage as carriers of hydrophobic anti-cancer drugs. Journal of controlled release 2011;152; issue 1: 57-66.
11. Prinda Wanakula*et al.* Nano-inside-micro: Disease-responsive microgels with encapsulated nanoparticles for intracellular drug delivery to the deep lung. Journal of controlled release 2012;162; issue 2:429-437.
12. Helena Bysell *et al*. Microgels and microcapsules in peptide and protein drug delivery. Advance drug delivery review 2011;63(13):1172-1185.
13. Silvia A Ferreira *et al.* Polymeric nanogels as vaccine delivery systems. Nanomedicine: Nanotechnology, Biology and Medicine (2012).
14. Hanwen Sun *et al.* Novel core-shell magnetic nanogels synthesized in an emulsion-free

aqueous system under UV irradiation for targeted radiopharmaceutical application. Journal of Magnetism and Magnetic material 2005;294(3): 273-280.

15. Jhon Erik Wong *et al*. Layer-by-layer assemble on stimuli-responsive microgels. Current opinion in colloid & interface science 2008;13(6):403-412.
16. B.G De Geest*et al.* Layer-by-layer coating of degradable microgels for pulsed drug delivery. Journal of controlled release 2006;116(2):159-169.
17. Keizou Ishii. Synthesis of microgels and their application to coatings. Colloids and Surface A; Physicochemical and Engineering Aspects 1999 ;153; issues1-3:591-595.
18. Weitai Wu and Shuiqin Zhou.Hybrid micro-/nanogels for optical sensing and intracellular imaging. Review Article 2010.
19. Tatiya Trongsatikul, Bridgette M. Budhlall. Microgels or microcapsules. Role of morphology on the release kinetics of thermo responsive PNIP Am-co-PEGM hydrogels. Polymer chemistry 2012.
20. Pietro Cozzini and Francesca Spyrakis. Hydrophobicity in drug design. Medicinal chemistry 2006:1-3.
21. Rong-Cong Luo, Chia-Hung Chen. Structured Microgels through Micro fluidic assembly and their biomedical applications. Review article 2012:1-23.