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Perspective

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Recent Advances in Graft Copolymerization and Applications of Chitosan: A Review

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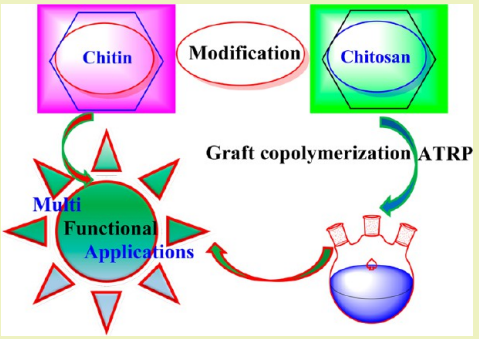
ABSTRACT: Chitosan is among one of the most important and most studied natural polymers. The cationic nature of chitosan makes it a polymer of high importance from environmental and biomedical point of views among the other natural polysaccharides. However, it also suffers from a few disadvantages and requires further development to achieve the targeted results and desired range of efficiency. To overcome some of the disadvantages of the pristine chitosan, it is most imperative to functionalize it with suitable functional groups. Therefore, it is highly desired to understand the chemistry of the reactions used to alter the surface characteristics of chitosan. Among various techniques presently being used to tailor the surface characteristics of chitosan, graft copolymerization is of the utmost importance. The aim of the present perspective is to describe the recent advances in the graft copolymerization of chitosan with particular emphasis on atom transfer radical polymerization (ATRP). This perspective describes the synthesis, characterization, and multifunctional applications of different types of chitosan-based copolymers.



KEYWORDS: *Natural polymers, Chitosan, Grafting, ATRP, Multifunctional Applications*

INTRODUCTION

Polymer-based materials, ranging from synthetic to renewable polymers, have attracted intense interest during the last few decades in industries related to energy storage in batteries, super capacitors, structural composites, and biomedical[1−5] to name a few. However, compared to their synthetic counterparts, renewable polymer-based materials offer a number of advantages especially regarding their environmental friendliness.[6−9] Much research work is being carried out on the usage of renewable resource-based materials as one of the components in the resulting final product.[10,11] Renewable resource-based materials are also potentially user friendly, and these materials have been proven to be a virtuous component in a number applications.[12−15] Among various renewable polymers, chitosan (CS) is one of the most commercially important biocompatible polymers from an environmental or biomedical point of view.[16−18] It is a copolymer of N-acetyl-D-glucosamine and D-glucosamine.[19,20] It is procured from chitin (a nitrogenous polysaccharide), which is the main building component of crustacean shells by the hydration of chitin.[21,22] The hydration is carried out in basic solutions at temperatures ranging from 80−140 °C for a certain period of time, with the maximum time being up to 10 h.[22] A huge amount of chitin is produced every year as waste from the food industry. Chitosan is a unique cationic polysaccharide, which is distinguished among other polysaccharides in the sense that all other polysaccharides lack a cationic nature.[23] It is mainly obtained at the synthetic scale by deacetylation of chitin, and the process of deacetylation is carried out to different degrees depending upon the targeted applications. These different deacetylation degrees lead to chitosan's numerous degrees of deacetylation (DD). The physiological properties of chitosan, especially solubility, are determined by its molecular weight and degree of deacetylation. The degree of deacetylation is most accurately determined by nuclear magnetic resonance (NMR) as well as infrared (IR) spectroscopy and has been comprehensively reviewed.[24,25] Different researches have reported that the degree of solubility of chitosan is higher for higher degrees of deacetylation (DD) and lower molecular weight.[26] Chitosan has been found to be readily soluble in acidic solutions and insoluble in water. Water-soluble chitosan has been reported to be prepared using various deacetylation processes, and it has been found to be susceptible to lipases, lysozymes, and other enzymes.[22]

On the other hand, physicochemical, biological, and structural characteristics of chitosan are determined by the different functional groups present on its backbone.[26] The prime functional groups present in the chitosan polymer are hydroxyl, amino, and acetamido groups.[27] It has been reported that the amino groups of chitosan have a pK_a value of 6.5, and this value makes it a pH responsive polymer. It is protonated at pH values below physiological pH. These structural functional groups makes chitosan a unique material with inherent properties such as biocompatibility, biodegradability, and nontoxicity. Because of the biodegradability of chitosan in the human body, it is recommended for possible use in clinical development.[28]

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Different biological properties of chitosan have been found to be closely related to its solubility.[19]

Compared to the other natural polysaccharide polymers, chitosan also suffers from few disadvantages, one being its low water solubility at physiological pH. The transfection efficiency of native chitosan is also relatively low, and it lacks some functionalities that are highly desired for some applications. Thus, a number of chemical modification techniques have been used to overcome these drawbacks of chitosan. Among the various techniques, graft copolymerization is most frequently used and has been reviewed. However, limited information is available in the reviews on the atom transfer radical polymerization (ATRP) of chitosan. Therefore, in this perspective, the different studies on the usage of ATRP to incorporate the desired functionalities in chitosan are included. The role of different reaction parameters has been discussed in detail. Finally, the applications of these copolymers have also been discussed. Different physical and chemical properties as well as the applications of chitosan-based materials have been recently reviewed.[29,30] In this perspective, we will give a brief overview of these properties but will mainly focus on the atom radical polymerization (ATRP) of chitosan.

STRUCTURE AND CHEMISTRY OF CHITOSAN

Chitosan is one of the most versatile renewable polymers that has been employed frequently in a number of forms such as particles, films, gels, membranes, or scaffolds for different targeted applications.[30,20−26] Table 1 summarizes the inherent properties of chitosan.[31] These applications range from the biomedical field, including drug delivery, tissue engineering, etc., to the industrial field, including adsorption of metal ions and dye removal to name a few.[32−36] Table 2 summarizes dye removal using different derivatives of chitosan.[37] It is a highly basic polysaccharide as opposed to the other polymers such as agar, pectin, dextrin, agarose, carragenas, and cellulose. As compared to the parental material chitin from which it is derived, chitosan has been found to be more suitable because of its better solubility in water and organic solvent.[29] The chemical structure of chitosan (Figure 1a) shows that it is made up of linear β-(1 → 4) glycosidic linkages.[30] These linkages in chitosan are similar in structure to cellulose as demonstrated in Figure 1b.[29] The first and foremost step in the extraction of chitosan from chitin is the removal of acetate moiety from chitin.[38] This process is accomplished using hydration that involves amide hydrolysis carried out under alkaline conditions using concentrated sodium hydroxide (NaOH) or through enzymatic hydrolysis in the presence of chitin deacetylase.[29] Compared to other polysaccharide-based polymers, the presence of primary amino groups ($-NH_2$) makes chitosan a potential candidate for pharmaceutical applications. One of the most significant features for the use of chitosan in biomedical applications is its biodegradability.[23] Different aspects of biodegradability have been recently reviewed.[30] Figure 1c shows the biodegradation of chitosan, which is so far not well understood.

Different physical and chemical properties of chitosan have been reported to be dependent upon the molecular weight and degree of deacetylation (DD).[27] Some of the special properties of chitosan include solubility in various media, polyelectrolyte behavior, viscosity, ability to form films, mucoadhesivity, polyoxysalt formation, metal chelations, and optical and structural characteristics.[20,29] Owing to the presence of different functional groups as demonstrated in Figure 1a and b, chitosan has been found to exhibit excellent chemical as well as biological properties.[29] The presence of active functional primary amino groups in chitosan provides a specific platform for side group attachment under mild reaction conditions depending upon the targeted applications. The presence of these side groups on a chitosan backbone has been found to provide flexibility to the given material with specific functionality.[32] These groups are also responsible for changing the biological and physical properties. The chelating properties of chitosan have also lead to its widespread use in water purification and toxic ion removal. Different chemical modification techniques have been used to change the surface characteristics of chitosan. Physical and chemical modifications have been frequently used for this purpose.[29] Among the chemical modification techniques used, the most important are acylation, hydroxylation, nitration, alkylation, sulphonation, phosphorylation, xanthation, and graft copolymerization. Figure 2a and b summarizes these reactions for different applications.[29,30] Among these techniques, graft copolymerization has been most widely used for incorporating the desired functionalities.[30] Different types of reaction initiator systems, such as a redox initiator, ceric ion, fentons reagent, γ-irradiation, etc., have been used, and all have proven to be beneficial for carrying out successful polymerization. Graft copolymerization of chitosan has been mostly performed using the chemical initiator or γ radiation.[29,39,40] Different types of graft copolymerization reactions used to alter the properties of chitosan have been comprehensively reviewed,[40,41] so we will be focusing in the following section on the atom transfer radical polymerization (ATRP) of chitosan.

Table 1. Intrinsic Properties of Chitosan[a]

Physical and chemical properties	linear aminopolysaccharide with high nitrogen content
	rigid D-glucosamine structure; high crystallinity; hydrophilicity
	weak base; deprotonated amino group acts a powerful nucleophile (pK_a 6.3)
	capacity to form hydrogen bonds intermolecularly; high viscosity
	insoluble in water and organic solvents; soluble in dilute aqueous acidic solutions
	numerous reactive groups for chemical activation and cross-linking
	forms salts with organic and inorganic acids
	chelating and complexing properties
	ionic conductivity
polyelectrolytes (at acidic pH)	cationic biopolymer with high charge density (one positive charge per glucosamine residue)
	flocculating agent; interacts with negatively charged molecules
	entrapment and adsorption properties; filtration and separation
	film-forming ability; adhesivity
	materials for isolation of biomolecules
biological properties	biocompatibility
	nontoxic
	biodegradable
	adsorbable
	bioactivity
	antimicrobial activity (fungi, bacteria, viruses)
	antiacid, antiulcer, and antitumoral properties
	blood anticoagulants
	hypolipidemic activity
	bioadhesivity

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Table 2. Adsorption Capacities of Chitin and Chitosan Derivatives for Various Metal Ions and Radionuclides Removal from Water[a]

sample no.	adsorbent	adsorbate	adsorption capacity
1	chitin	Cd(II)	14.0 mg/g
2	chitosan (CS)	Cd(II)	5.93 mg/g
3	chitosan	Hg(II)	815.0 mg/g
4	chitosan	Cu(II)	222.0 mg/g
5	chitosan	Ni(II)	164.0 mg/g
6	chitosan	Zn(II)	75.0 mg/g
7	chitosan flakes	Cu^{2+}	1.8−2.2 mmol/g
8	chitosan	Cu, Zn, As, and Cr	137, 108, 58, and 124 mg/g
9	aminated chitosan beads	Hg	2.26 mmol/g
10	porous magnetic CS beads	Cd(II)	188−518 mg/g
11	chitosan coated on perlite	Cu(II)	196.07 mg/g
12	chitosan coated on perlite	Ni(II)	114.94 mg/g
13	chitosan obtained from silkworm chrysalides (ChSC)	Pb(II)	72.0 mg/g
14	chitosan obtained from silkworm chrysalides (ChSC)	Cu(II)	87.0 mg/g
15	carboxyl-grafted chitosan	Cu(II)	318.0 mg/g
16	amido-grafted chitosan	Cr(VI)	935.0 mg/g
17	chitosan coated onto polyvinyl chloride (PVC) beads	Ni(II)	120.5 mg/g
18	chitosan coated onto polyvinyl chloride (PVC) beads	Cu(II)	87.9 mg/g
19	chitosan	Al(III)	45.45 mg/g
20	chitosan	VO_4^{3-}	∼400−450 mg/g
21	glutaraldehyde cross-linked chitosan beads	VO_4^{3-}	402.5 mg/g
22	glutaraldehyde cross-linked chitosan beads	MoO_4^{-}	763.0 mg/g
23	molybdate-impregnated chitosan gel beads	As(V)	∼160.0 mg/g
24	chitosan beads	As(III)	1.83 mg/g
25	chitosan beads	As(V)	1.94 mg/g
26	chitosan flakes	As(V)	1331−14,160 μg/g
27	chitosan flakes	Cr(VI)	22.09−102.0 mg/g
28	noncross-linked chitosan	Cr(VI)	78.0 mg/g
29	cross-linked chitosan	Cr(VI)	50.0 mg/g
30	chitosan coated onto nonporous ceramic alumina	Cr(VI)	154.0 mg/g
31	quaternary ammonium salt of chitosan (QCS)	Cr(VI)	30.2−68.3 mg/g
32	cross-linked chitosan	Cr(III)	6.0 mg/g
33	cross-linked chitosan	Cr(VI)	215.0 mg/g
34	aminated chitosan	Ni(II)	30.2 mg/g
35	aminated chitosan	Cr(VI)	28.7 mg/g
36	chitosan particles	VO_4^{3-}	90 mg/g
37	chitosan	Au(III)	30.95 mg/g
38	N-carboxymethyl chitosan (NCMC)	Au(III)	33.90 mg/g
39	chitosan	Ag	42.0 mg/g
40	chitosan benzoyl thiourea derivative	Co(II)	29.47 mg/g
41	chitosan benzoyl thiourea derivative	Eu(III)	34.54 mg/g
42	cross-linked chitosan	U(VI)	72.6 mg/g

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ATOM TRANSFER RADICAL POLYMERIZATION OF CHITOSAN: CHEMISTRY AND APPLICATIONS

Among various graft copolymerization techniques used to change the surface characteristics of polymers and other materials for different applications, the technique of atom transfer radical polymerization is rapidly emerging as the first preference due to the enormous advantages it offers over other techniques.[40,41] One of the biggest disadvantages of the conventional radical polymerization technique is the lack of control over the polymer structures as it could not form the well-defined polymers that are highly desired for sophisticated applications.[40−43] Indeed, the synthesis of different types of materials with suitable well-defined architectures, functionalities, and compositions is highly desired for practical applications in different fields.[44,40,41] The advantages of ATRP over the conventional graft copolymerization technique include compatibility with both aqueous and organic media and a high tolerance toward a wide range of functional groups.[44] In ATRP, the control over radical polymerization during synthesis of desired polymer architectures is based on two prime principles. As per the first principle, initiation of the reaction should be fast enough so as to afford a persistent concentration of growing polymer chains. While as per the second principle, most of the dormant polymer chains should retain the ability to grow as a result of the formation of the established dynamic equilibrium between the growing radicals and the dormant one.[44] Figure 3 shows the mechanism of ATRP along with some examples.[45] The mechanism of radical polymerization involves the generation of a radical ($R^{\bullet}$) through the homolytic cleavage of an alkyl−halide bond (R-X) using a transition metal complex (Mt^n).[45,46] As a result of this reaction, the transition metal complex reaches a higher oxidation state Mt^{n+1}. Subsequently, $R^{\bullet}$ may propagate with suitable monomers (mostly vinyl monomers) or become a dormant species reversibly by M^{tn+1}.[46] The role of the transition metal catalyst in the polymerization reaction is to establish a reversible equilibrium between the dormant species and growing radicals.[45] It has been reported that by controlling the concentration of the propagating radicals the termination reaction can be easily suppressed to produce the desired well-defined polymer chains with narrow molecular weight distribution.[44,45] The ATRP reactions do not require rigorous experimental conditions to accomplish the process of polymerization. These reactions can also be performed with a number of functional monomers with the exception of acidic monomers.[45] The last few years have seen a tremendous increase in the usage of ATRP for different applications, and this is expected to continue in the coming years.[44,45]

The presence of suitable functional groups on chitosan, such as $-NH_2$ and $-OH$ groups, makes it susceptible to be polymerized using ATRP, as these groups provide the basis for interaction with other reaction components. Tahlawy and Hudson have reported their study on the synthesis and characterization of chitosan graft poly(methoxy polyethylene glycol methacrylate) by using atom transfer radical polymerization.[47] In this work, chitosan was modified to serve as a co-initiator to prepare synthetic polymers with well-defined chitosan functionality at specific places within each macromolecule. In this work to carry out the polymerization reaction, a chitosan macroinitiator was prepared by reacting 2-bromo-isobutyryl bromide with chitosan in the presence of pyridine as a base, after the chitosan amino group had been protected as the imine. Heterogeneous ATRP reactions were carried out in water. The effects of different

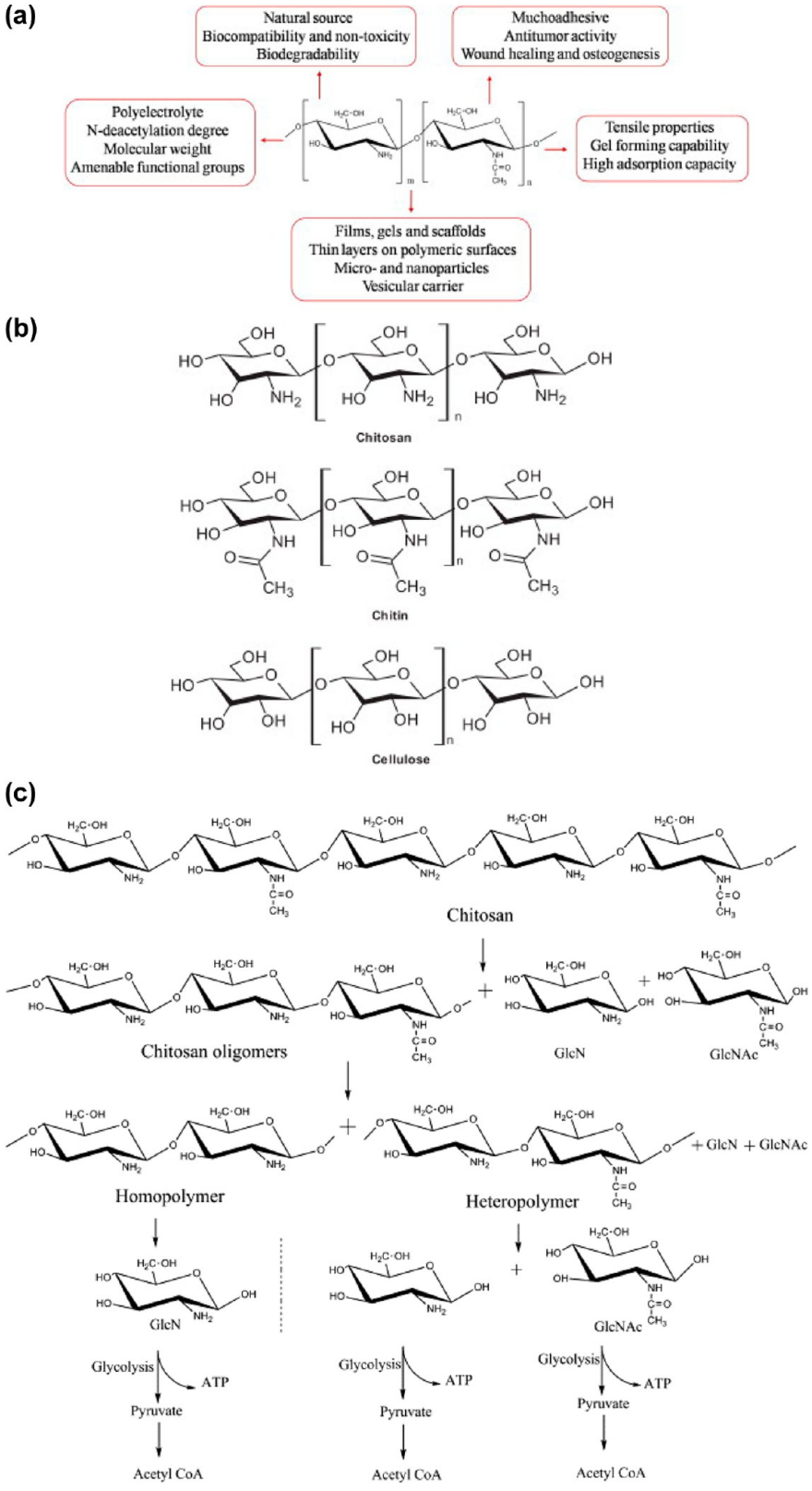

Figure 1. (a) Chemical structure of chitosan. Reprinted with permission.[30] Copyright 2014 Elsevier. (b) Comparative structure of chitosan, chitin, and cellulose. Reprinted with permission.[29] Copyright 2013 Elsevier. (c) Chitosan biodegradation. Reprinted with permission.[30] Copyright 2014 Elsevier.

reaction parameters that affect the rate of polymerization were studied in detail that also include the study of the concentration of chitosan macroinitiator and Cu (I) Br/bipyridine complex. It was revealed from the detailed experimental study that chitosan macroinitiators successfully polymerize the methoxy-poly-(ethylene glycol) methacrylate (MeO (PEG350) MA) monomer via atom transfer radical polymerization. The kinetic study of the polymerization reaction was also carried out, and it was found

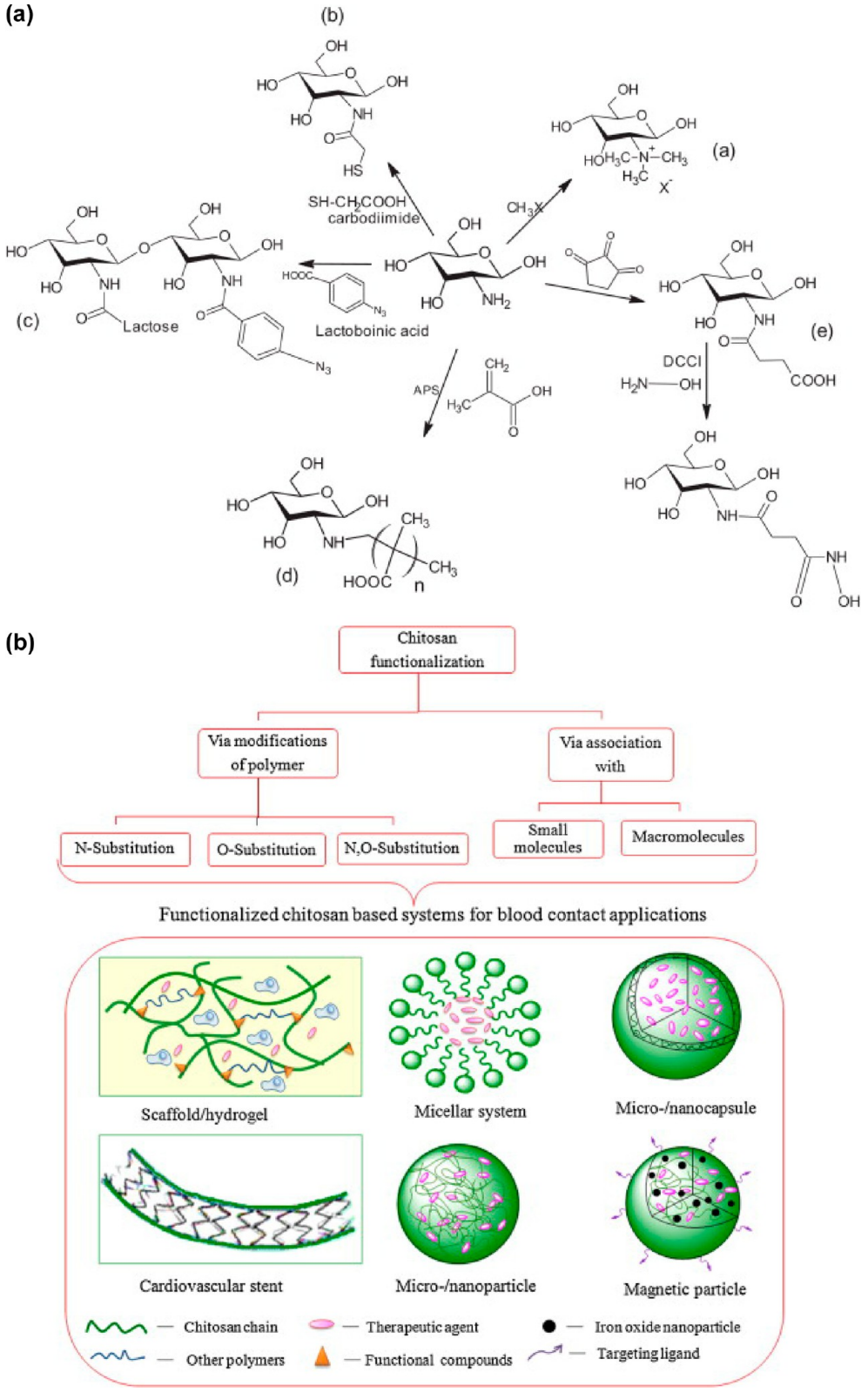

Figure 2. (a) Chemical modification of chitosan for different applications: (a) methylation, (b) thiolation, (c) azylation, (d) copolymerization, and (e) N-succinylation. Reprinted with permission.[29] Copyright 2013 Elsevier. (b) Strategies used to improve the blood compatibility of chitosan. Reprinted with permission.[30] Copyright 2014 Elsevier.

that the mechanism followed a first-order polymerization reaction.[47]

Surface-initiated atom transfer radical polymerization of chitosan was carried out for the selective adsorption of mercury ions.[48] The removal of mercury ions was accomplished by grafting chitosan beads with polyacrylamide to prepare chitosan-*g*-polyacrylamide via surface-initiated atom transfer radical polymerization. Prior to the polymerization reaction of

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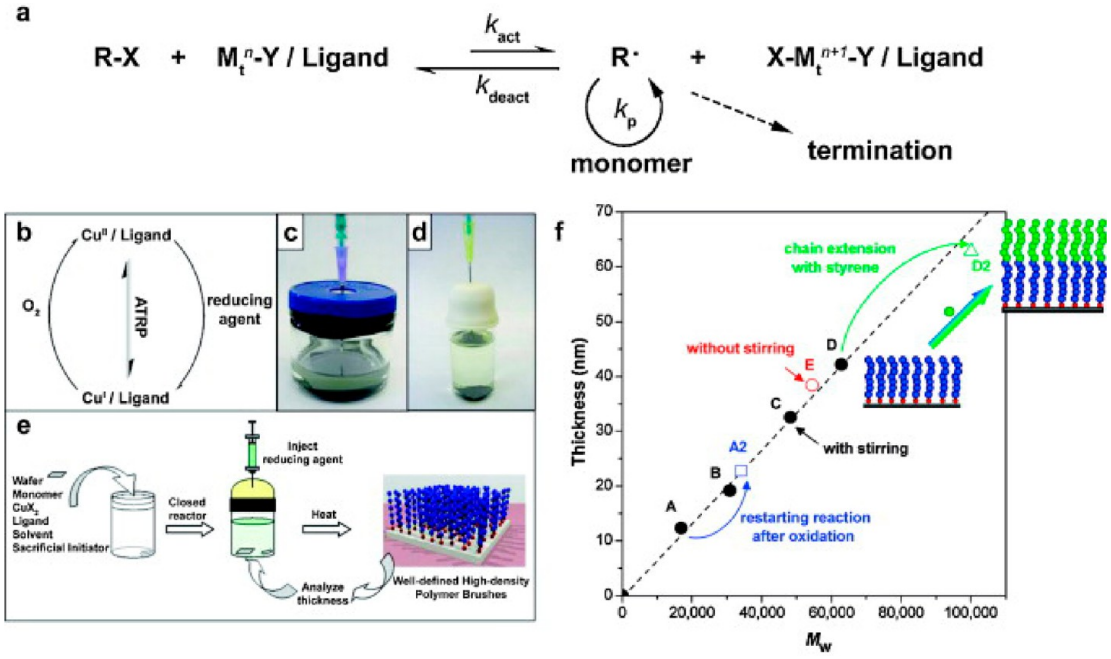

Figure 3. (a) Mechanism of transition metal-catalyzed ATRP. (b) Proposed mechanism of ARGET ATRP in the presence of limited amounts of air. Typical setup for grafting polymer chains from silicon wafers using ARGET ATRP with limited air (c) in a large jar and (d) in a sample vial. (e) Illustration of procedures for surface-initiated ARGET ATRP with limited air. (f) Relationship between the grafted poly(butyl acrylate) (PBA) brush thickness measured in air by ellipsometry and M_n of free PBA polymers. In this example, it was demonstrated that polymers could grow with or without stirring. It was also shown that the polymerization could be stopped and restarted with continued growth of PBA chains or extension of the chains by polystyrene. Reprinted with permission.[45] Copyright 2012 Elsevier.

acrylamide from the surfaces of chitosan beads, 2-bromoisobutyryl bromide was reacted to the surfaces as the polymerization initiator. It was observed that the chains of polyacrylamide on the chitosan-*g*-polyacrylamide beads grew longer with the increase in reaction time. Subsequently, the synthesized chitosan-*g*-polyacrylamide beads were subjected to the adsorption isotherm study using mercury and lead in a single component system in which the concentrations varied from 10 to 200 mg/L at pH 4. The effect of pH was studied in the range of 3−6 with a fixed initial mercury and lead concentration of 20 mg/L. It was observed from the study that compared to the pristine chitosan beads the chitosan-*g*-polyacrylamide beads demonstrated greater adsorption capacities.[48] The graft copolymers also showed faster adsorption kinetics for mercury ions compared to the parental chitosan. In comparison to the adsorption capacity of 181.8 mg/g for the chitosan beads, the chitosan-*g*-polyacrylamide beads exhibited a maximum adsorption capacity of up to 322.6 mg/g at pH 4 with initial mercury concentrations of 10−200 mg/L. Furthermore, the adsorption equilibrium time was also found to be less than 60 min in comparison to the time of more than 15 h for the chitosan beads. The selectivity of chitosan-*g*-polyacrylamide to adsorb mercury was also compared with lead, and it was found that the chitosan-*g*-polyacrylamide beads had much better selectivity in the adsorption of mercury ions in comparison to lead ions at pH < 6. On the other hand, the pristine chitosan beads did not show any clear selectivity for either of the two metal (mercury/lead) species. On the basis of the reaction mechanism, the enhanced mercury adsorption was attributed to the efficient number of amide groups grafted onto the surfaces of the beads. The higher selectivity of chitosan-*g*-polyacrylamide beads was attributed to the ability of mercury ions to form covalent bonds with the amide. A desorption study of the metal ions was also carried out, and it was found that the adsorbed mercury ions on the chitosan-*g*-polyacrylamide beads can be desorbed in a per chloric acid solution suggesting that the regenerated beads can be reused.[48] Surface-initiated atom transfer radical polymerization of chitosan particles was also carried out using styrene as the reaction monomer.[49] In this reaction, polystyrene-grafted chitosan particles (PS-*g*-chitosan) were synthesized using the macroinitiators, which are bromoacetyl chitosans having surface amino and hydroxyl groups. The reaction was carried out in a toluene system using the catalysts of 1,10-phenanthroline and Cu (I) Br. The effect of reaction time on the polymerization reaction was studied in detail, and it was found that conversion of monomer (C %) and the percentage of grafting (PG %) increases significantly with increasing polymerization time. The conversion of the monomer (C %) and the percentage of grafting (PG %) were found to reach the values of 5.31% and 19.12% after a polymerizing time of 5 h, respectively. The synthesized polystyrene-grafted chitosan particles were thoroughly characterized using elemental analysis (EA), Fourier transform infrared (FTIR), differential scanning calorimetry (DSC), and X-ray diffraction (XRD). All of these characterization techniques confirmed the characteristics of a controlled/"living" polymerization reaction. It was concluded that the all the used polystyrene was successfully bonded to the chitosan backbone with no formation of a homopolymer.[49]

Synthesis of Chitosan Nanospheres. Chitosan nanospheres were also functionalized using a surface-initiated activator generated by electron transfer (AGET) ATRP mediated by an iron catalyst in the presence of limited amounts of air.[50] In the polymerization reaction, $FeCl_3 \cdot 6H_2O$ was used as the catalyst, PPh_3 as the ligand, and ascorbic acid as the reducing agent. In this study, two types of polymers, namely poly(methyl methacrylate) (PMMA) and amphiphilic block copolymer poly(methyl methacrylate)-*b*-poly(poly(ethylene glycol) methyl

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ether methacrylate) (PMMA-*b*-P (PEGMA)), were grafted onto chitosan nanospheres. The polymerization reaction resulted in the synthesis of well-defined individual nanospheres that were composed of a chitosan core and a densely grafted outer PMMA or PMMA-*b*-P (PEGMA) layer. Figure 4a and b shows the scheme for the synthesis and photographs of the different reaction products, respectively, as a result of the ATRP synthesis. The kinetic study carried out showed that a linear kinetic plot for the homopolymer is obtained with a linear increase in molecular weight (M_n) with conversion. The linear plot of the grafted percentage for chitosan nanospheres versus time was also observed. The newly synthesized grafted materials were thoroughly characterized using different techniques, and the chemical composition of the nanospheres surfaces at different surface modification stages was authenticated using FTIR spectra and X-ray photoelectron spectroscopy (XPS). Figure 4c and d shows the transmission electron microscopy (TEM) images and XPS spectra of the different nanospheres confirming their successful synthesis, respectively. It was demonstrated that the well-defined PMMA or amphiphilic PMMA-*b*-P (PEGMA) brushes were covalently tethered on the chitosan nanospheres surfaces.[50]

Chitosan-*graft*-poly(oligoethylene glycol methacrylate) (OEGMA) copolymers were prepared by ATRP using two different approaches of "grafting from" and "grafting to" to study the effect of these two routes.[51] Figure 5a and b shows the different routes used to synthesize the graft polymers. In this work, poly(oligoethylene glycol methacrylate) (poly(OEGMA)) was used because of its advantages such as being soluble and biologically inert, which may lead to graft copolymers potentially suitable for biomaterials. In both reactions, two initiators were used, and the initiator containing the two methyl groups was found to perform better than the initiator with the one methyl group as the latter one was less stable. It was found to decompose more readily under the reaction conditions used. In the "grafting from" synthesis route, chitosan was reacted with a succinimide-containing initiator to form a macroinitiator and polymerized with OEGMA in the presence of the macroinitiator in an acidic aqueous solution. In the "grafting to" synthesis method, poly(OEGMA) with a succinimide end group was also synthesized and successively attached to chitosan to form a copolymer. The "grafted to" comb polymers were found to display a significant change in physical appearance even with a low amount of grafted polymer incorporation in the copolymer. The comparative study of the two synthesis routes demonstrated that "grafting from" copolymers could not be completely purified and were found to contain unbound oligomers even after extensive washing. On the other hand, the "grafting to" synthetic route lead to the polymers of higher purity and was more defined.[51]

A comb-like pH-sensitive graft copolymer, poly[(2-dimethylamino)ethyl methacrylate]-*graft*-chitosan, was successfully prepared using atom transfer radical polymerization (ATRP) and active ester conjugation methods.[52] The synthesis reaction was carried out using mild reaction conditions in homogeneous solutions. The graft ratio was found to reach a value of 58%, which is comparable to the traditional free radical-polymerized chitosan graft copolymers. Different characterization techniques such as laser light scattering, surface tensiometry, potentiometric titration, and transmission electron microscopy were used to study the pH-responsive association behavior of the graft copolymer in aqueous solution. The effect of pH on the association behavior was studied in detail. It was

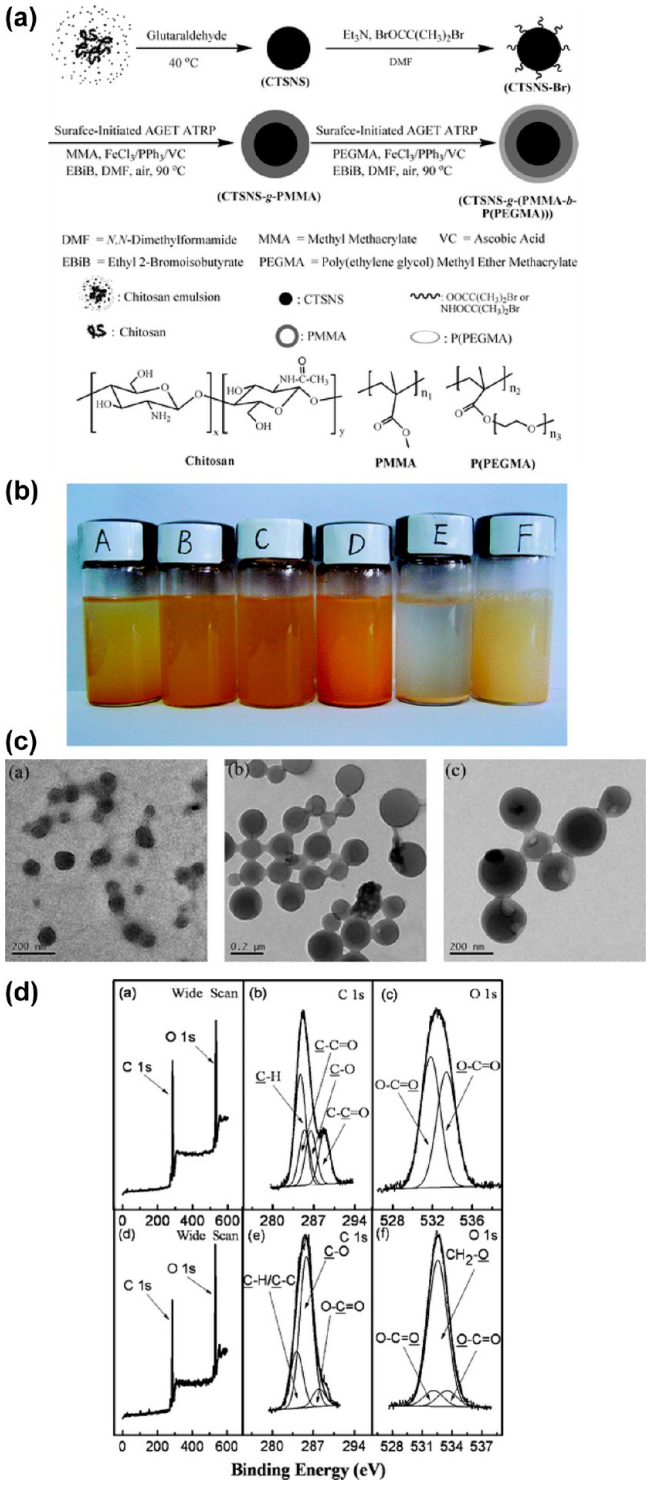

Figure 4. (a) Schematic illustration of surface modification of chitosan nanospheres (CTSNSs) by surface-initiated AGET ATRP technique mediated by an iron catalyst. (b) Photos of (A) CTSNS-Br, (B) CTSNS-*g*-PMMA, and (C) CTSNS-*g*-(PMMA-*b*-P (PEGMA)) in THF; photos of (D) CTSNS-Br, (E) CTSNS-*g*-PMMA, and (F) CTSNS-*g*-(PMMA-*b*-P (PEGMA)) in deionized water. Samples: CTSNS-*g*-PMMA obtained by surface-initiated AGET ATRP of MMA for 5 h using CTSNS-Br as the surface active initiator and CTSNS-*g*-(PMMA-*b*-P(PEGMA)) obtained by surface-initiated AGET ATRP of PEGMA for 15 h using CTSNS-*g*-PMMA as the macroinitiator. (c) TEM images of (a) CTSNS, (b) CTSNS-*g*-PMMA from 5 h of surface-initiated AGET ATRP of MMA using CTSNS-Br as the surface active initiator, and (c) CTSNS-*g*-(PMMA-*b*-P(PEGMA)) from

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Figure 4. continued

15 h of surface-initiated AGET ATRP of PEGMA using CTSNS-*g*-PMMA as the macroinitiator. (d) XPS (a) wide scan, (b) C 1s, and (c) O 1s core-level spectra of PMMA-grafted chitosan nanospheres (CTSNS-*g*-PMMA) from 5 h of surface-initiated AGET ATRP of MMA using CTSNS-Br as the surface active initiator; XPS (d) wide SCAN, (e) C 1s, and (f) O 1s core-level spectra of the amphiphilic diblock copolymer-grafted chitosan nanospheres (CTSNS-*g*-(PMMA-*b*-P(PEGMA))) from 15 h of surface-initiated AGET ATRP of PEGMA using CTSNS-*g*-PMMA as the macroinitiator. Reprinted with permission.[50] Copyright 2009, American Chemical Society.

observed that the behavior changes with changes in pH. At pH ∼ 4, the copolymer was found to exhibit the Gaussian chains behavior, while at pH 5−6, the core−shell structured micelles were obtained via self-assembly. At pH ∼ 7, the core−shell structure was found to collapse into double-layered hard spheres. At pH ∼ 8, the micelles were found to be aggregated and precipitated. This behavior was attributed to the difference in proton dissociation between the amine groups of chitosan and DMAEMA segments. Kinetic studies were used to study the control behavior of the reaction, and it was observed that the polymerization of DMAEMA initiated by NBPS proceeds in a well-controlled manner. Thus, the successful utilization of ATRP was found to endow a narrow molecular weight distribution to the PDMAEMA side chains of the copolymers. Different reaction products were characterized in detail by FTIR, ^{1}H NMR, and gel permeation chromatography (GPC). These phenomenon were further reconfirmed by the potentiometric titrations and zeta potential tests.[52]

Comb-like, dually stimuli-responsive, graft terpolymers based upon chitosan derivatives were synthesized using atom transfer radical polymerization (ATRP) and click chemistry.[53] Chitosan (CS)-grafted terpolymers were obtained with both poly[(2-dimethylamino) ethyl methacrylate] and poly(*N*-isopropylacrylamide), and the product was designated as CS (-*g*-PDMAEMA)-*g*-PNIPAM. Figure 6a depicts the synthesis route for the preparation of the CS (-*g*-PDMAEMA)-*g*-PNIPAM graft

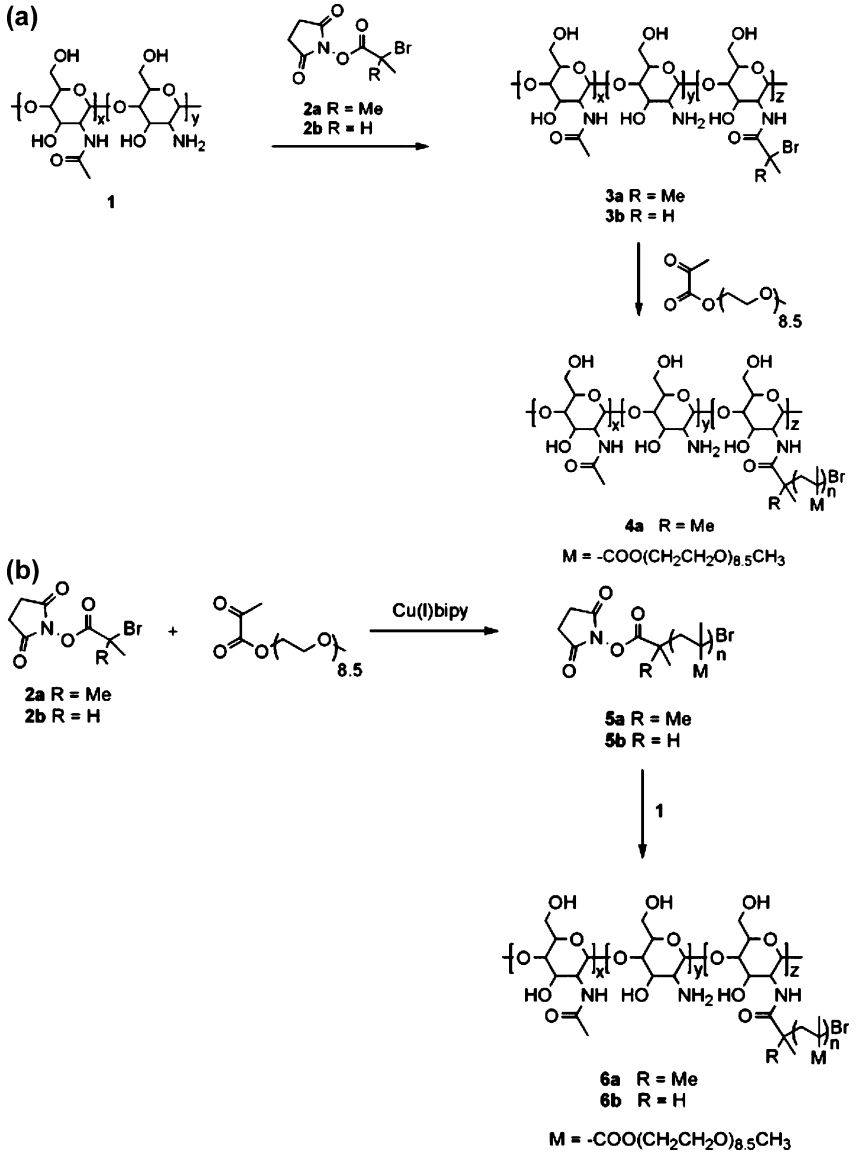

Figure 5. (a) "Grafting from" synthetic route used to prepare chitosan-*graft*-poly(OEGMA) copolymers. (b) "Grafting to" synthetic route used to prepare chitosan-*graft*-poly(OEGMA) 6 from chitosan 1 and poly(OEGMA) 5. Reprinted with permission.[51] Copyright 2009 Elsevier.

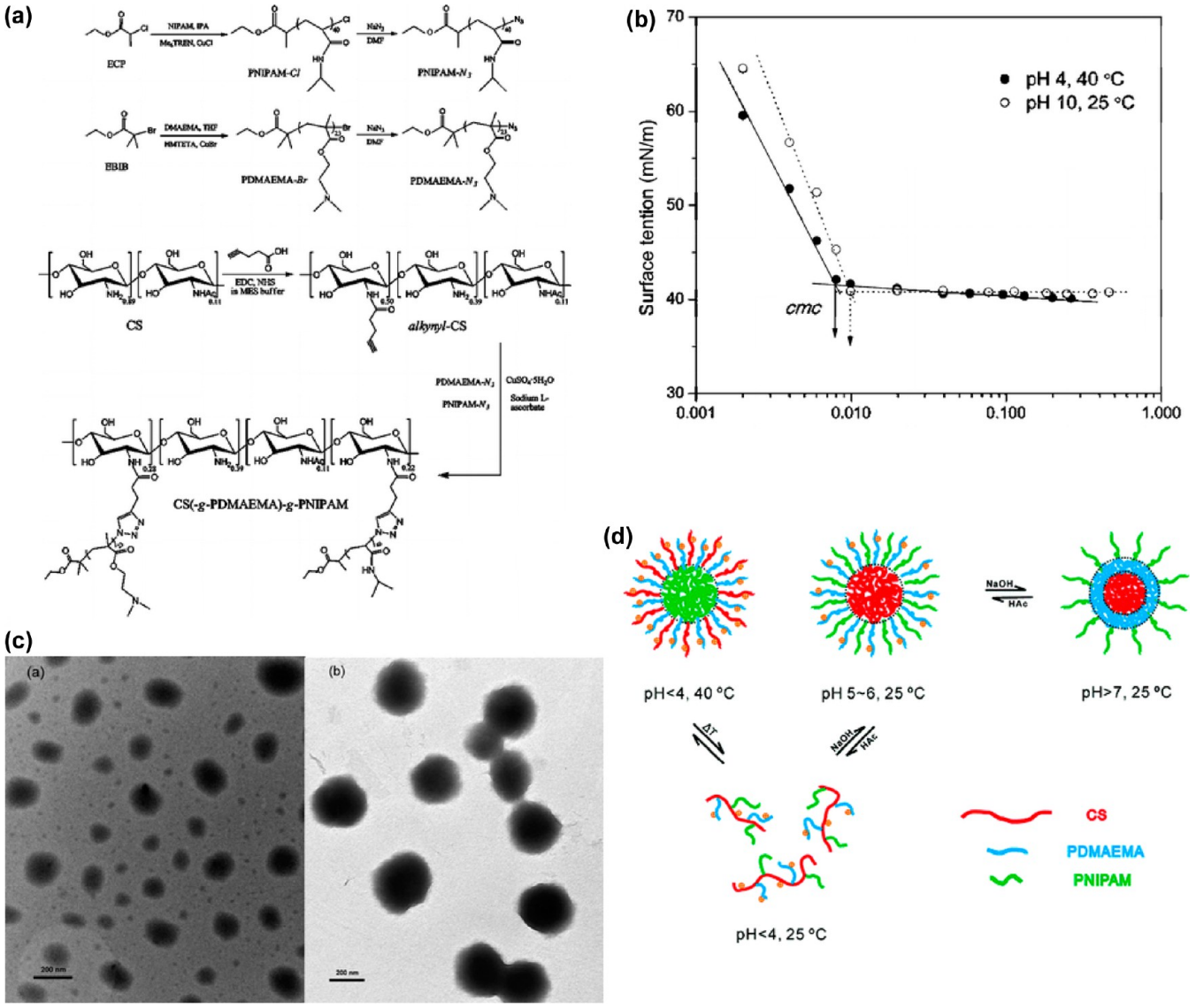

Figure 6. (a) Synthetic route for preparation of CS (-*g*-PDMAEMA)-*g*-PNIPAM graft terpolymer. (b) Surface tension versus concentration of CS (-*g*-PDMAEMA)-*g*-PNIPAM under two representative conditions: (a) pH 4 and 40 °C and (b) pH 10 and 25 °C. (c) TEM micrographs for self-assembled micelles obtained from 0.05 wt % CS (-*g*-PDMAEMA)-*g*-PNIPAM terpolymer solutions at (a) pH 4 and 40 °C and (b) pH 10 and 25 °C. (d) Schematic description for the magic association behavior of the dual hydrophilic CS (-*g*-PDMAEMA)-*g*-PNIPAM terpolymer. Reprinted with permission.[53] Copyright 2010, American Chemical Society.

terpolymer. The synthesis reaction was accomplished by first synthesizing PDMAEMA and PNIPAM via ATRP and subsequently substituting the halide end groups with azido groups. Chitosan was then converted into alkynyl-CS via amidation, and PDMAEMA-N3 and PNIPAM-N3 side chains were consequently grafted onto the chitosan backbone via click reaction. This lead to the formation of the well-defined graft terpolymer. The click reaction was found to take place in the mild and homogeneous solution and proved to be an efficient coupling method to get grafted chitosan with a higher degree. A number of characterization techniques, namely, surface tensiometry, proton nuclear magnetic resonance (^{1}H NMR), zeta potential, laser light scattering (LLS), and transmission electron microscopy (TEM), were then used to study the thermo- and pH-responsive micellization behavior of the synthesized graft terpolymers in aqueous solutions. Figure 6a and b shows the surface tensiometry and TEM results of the CS (-*g*-PDMAEMA)-*g*-PNIPAM terpolymer. It was observed that in an acidic environment (pH < 4) at elevated temperature (>38 °C), the core−shell structured micelles with PNIAPM as a core and CS/PDMAEMA as a shell were formed. However, in alkaline solutions (pH > 7) at room temperature, the unimers were turned into the micelles with CS/PDMAEMA cores. It was confirmed from the experimental study that the utilization of ATRP facilitated the modulation of the side chains flexibly as well as guaranteed their narrow molecular weight distribution contributing to a low polydispersity of the terpolymer. On the basis of the experimental results, the authors claimed that the study was the first example of a nonlinear dual hydrophilic CS copolymer that showed the magic micellization behavior. Figure 6d shows the magical association behavior of the dual hydrophilic CS (-*g*-PDMAEMA)-*g*-PNIPAM terpolymer.[53] The same research group has also reported their study on the synthesis of chitosan-*graft*-poly(*N*-isopropylacrylamide) (CS-*g*-PNIPAM) using atom transfer radical polymerization (ATRP) and click chemistry.[54] The synthesized products were characterized using

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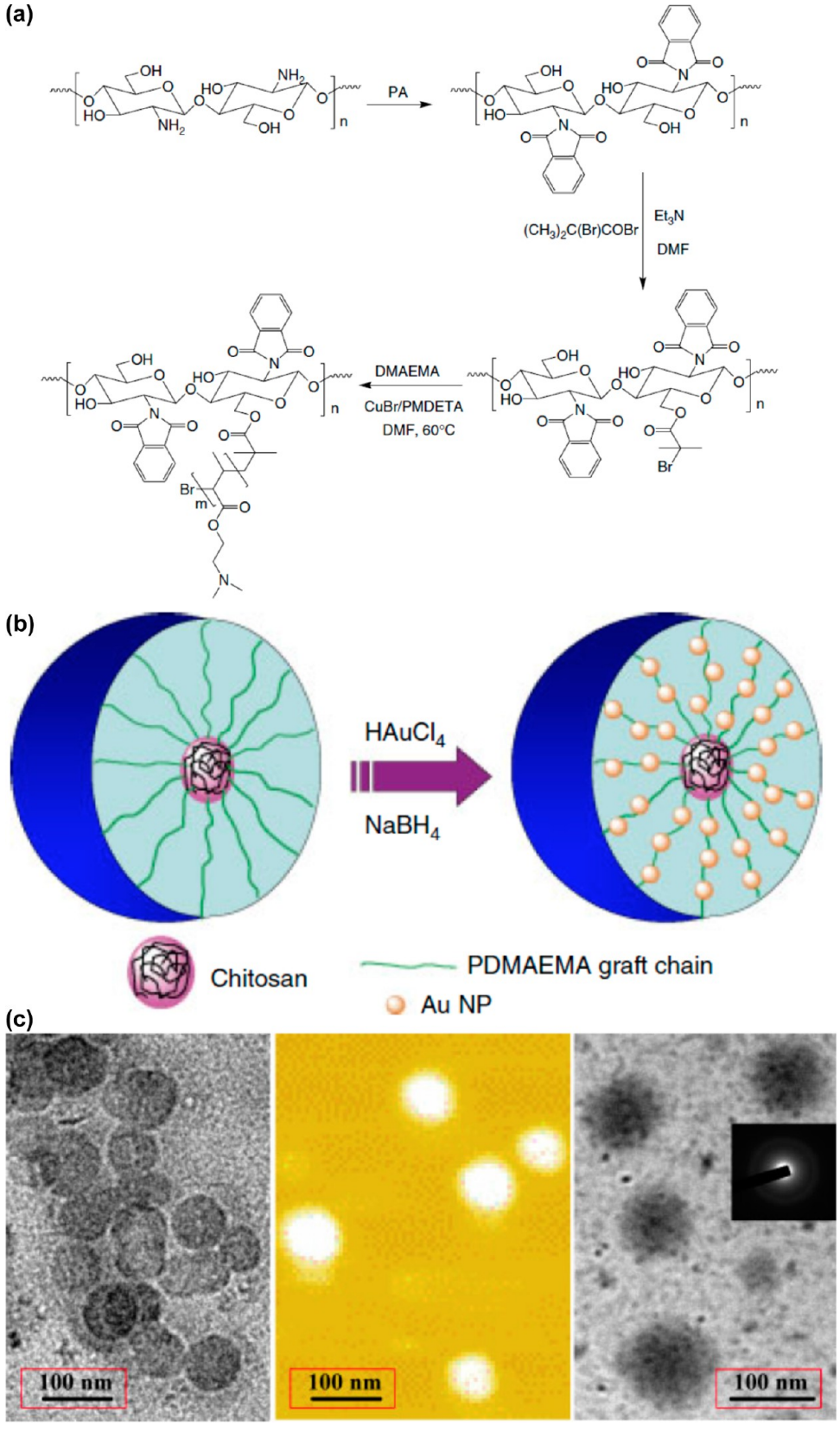

Figure 7. (a) Synthesis of chitosan-*g*-PDMAEMA graft copolymer by ATRP. (b) In situ formation and immobilization of Au NPs onto PDMAEMA chains of chitosan-*g*-PDMAEMA graft copolymer micelles. (c) TEM images of chitosan-*g*-PDMAEMA2 micelles (left) and chitosan-*g*-PDMAEMA2/Au hybrid micelles (right; inset shows the electron diffraction of Au in the hybrid micelles) and AFM image of chitosan-*g*-PDMAEMA2 micelles (center). Reprinted with permission.[56] Copyright 2011, John Wiley and Sons.

^{1}H NMR, FTIR, and GPC analyses, and subsequently, the thermoresponsive association behaviors of these synthesized copolymers in dilute aqueous solutions were studied using zeta potential, laser light scattering (LLS), and transmission electron microscopy (TEM). It was observed that the elevated temperature (>32 °C) and low pH (<4) resulted in the formation of the core−shell-structured micelles having hydrophobic PNIAPM as a core and hydrophilic chitosan (CS) as a shell. With an increase in pH at a temperature of 40 °C, these micelles were found to be converted into large aggregates and precipitated in alkaline solutions (pH > 7). Microdifferential scanning calorimetry (DSC) and UV turbidimetry were then used to study the cosolute effects on the lower critical solution temperatures (LCST) phase transition of the concentrated CS-*g*-PNIPAM solutions. It was observed from the study that the addition of salts (NaCl and NaI) and DATB resulted in shifting of the LCST to lower or higher temperatures from a pure copolymer solution, depending on the reaction mechanisms. It was also observed that mixing of cationic surfactant DTAB into copolymer solutions resulted in a reverse phenomenon.[54]

Yuan et al. have reported their study on the preparation of the bromized sodium dodecyl sulfate (SDS)−chitosan (CS) complex (Br-SCC) that was synthesized by the conjugation of 2-bromo-2-methyl propionic acid to the hydroxyl groups of SCC.[55] In this study, SCC was used as an organo-soluble precursor, and Br-SCC was used as a macromolecular ATRP initiator to carry out polymerization of methyl ether poly(ethylene glycol) methacrylate (PMPEGMA). In this reaction, copper(I) bromide/bipyridine was used as the catalyst that yielded SCC-O-PMPEGMA. Aqueous tris(hydroxymethyl) amine (Tris) medium was used to remove SDS from the product by the precipitation of a SCC-O-PMPEGMA solution. FTIR and ^{1}H NMR charactrization were used to study the chemical structure of the intermediate and CS-O-PMPEGMA. It was formulated that the substitution degree of bromobutyric acid can be modulated by the variation of the bromobutyric acid/SCC feed ratio, while by varying the PMPEGMA/Br-SCC ratio, the degree of polymerization of PMPEGMA can be easily controlled. Characterization techniques such as zeta potential analyzer, dynamic light scattering, and TEM were used to study the complexation behaviors between heparin and CS-O-PMPEGMA. It was found that with an increase in the ratio of heparin/CS molar ratio (X), the nanoparticle size increases, while the zeta potential of the particles decreases. The copolymers were found to form spherical micelles with anionic agents, while there was no effect of the grafting of PMPEGMA on the cationic density of the final product. The results demonstrated that these characteristics of CS-O-PMPEGMA make it a potential material for biomedical applications such as in tumor targeting and gene delivery.[55]

pH and temperature-responsive chitosan-*g*-PDMAEMA graft copolymers were synthesized successfully by homogeneous ATRP of [(2-(N,N-dimethylamino)ethyl methacrylate] (DMAEMA) under mild conditions.[56] The synthesis of the graft copolymers was carried out by using a chitosan macroinitiator, which was prepared by a phthaloylation of amino groups of chitosan, followed by subsequent acylation of hydroxyl groups of chitosan with 2-bromoisobutyryl bromide. Figure 7a shows the synthesis of a chitosan-*g*-PDMAEMA graft copolymer by ATRP. The graft copolymers were then obtained by ATRP of 2-(N, N-dimethylamino) ethyl methacrylate and were found to be self-assembled into stable micelles in water. Subsequently, the stable chitosan-*g*-PDMAEMA/Au hybrid micelles were prepared using in situ method via the reduction of $HAuCl_4$ with $NaBH_4$ by the embedding of Au NPs into the pH- and temperature-responsive micelles. Figure 7b depicts the in situ formation and immobilization of Au NPs onto the PDMAEMA chains of chitosan-*g*-PDMAEMA graft copolymer micelles. UV−visible spectroscopy and dynamic laser light scattering was then used to study the pH and temperature responses of the copolymer micelles and hybrid micelles. The synthesized copolymers were characterized using different techniques including TEM and atomic force microscopy (AFM). Figure 7c shows the TEM images of chitosan-*g*-PDMAEMA2 micelles and chitosan-*g*-PDMAEMA2/Au hybrid micelles and an AFM image of chitosan-*g*-PDMAEMA2 micelles. It was concluded from the study that the PDMAEMA corona acts as the "nano reactor" as well as the "anchor" during the process. The variation of temperature and pH of the external surroundings was found to tune the spatial distribution of Au NPs in the micelles. The hybrid micelles were proposed to be useful as a chemical or biomedical monitor of a specific chemical or biochemical species.[56]

Synthesis of Thermoresponsive Chitosans. ATRP reaction, combined with ring-opening polymerization (ROP) and click chemistry, was used to synthesize amphiphilic chitosan-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) copolymers with double side chains of PCL and P (MEO2MA-*co*-OEGMA).[57] Figure 8a shows the schematic for synthesis of the CS-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) copolymer via a combination of ROP, ATRP, and click chemistry. The graft ratio of PCL and P (MEO2MA-*co*-OEGMA) was adjusted via altering the feed ratio of pyrene−PCL−N3 and P (MEO2MA-*co*-OEGMA)−N3. It was formulated that due to the unique properties of click chemistry, the coupling efficiency of click chemistry was comparatively high. The synthesized chitosan-based graft copolymers were found to be assembled into micelles in water and exhibited thermosensitive properties. Characterization techniques such as TEM, DLS, and transmittance were used to study the self-assembly behavior and thermosensitive properties of chitosan-based graft copolymers. Figure 8b and c shows the plots of transmittance as a function of temperature for the CS-*g*-P (MEO2MA-*co*-OEGMA) copolymer micelle solution and temperature dependence of hydrodynamic diameters (D_h) for CS-*g*-P (MEO2MA-*co*-OEGMA) copolymers micelles as wells as CS-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) micelles solutions, respectively. It was observed from the experimental investigation that the lower critical solution temperatures (LCSTs) values of the chitosan graft copolymers micelles solutions were affected by the chitosan chains and ratio of PCL and P (MEO2MA-*co*-OEGMA) side chains. Furthermore, the temperature was also found to affect the behavior of micelles. The micelles were found to reversibly swell and shrink in response to the change in temperature. These micelles were also used to study drug release. Figure 8d shows the drug release of CS-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) micelles in PBS at 25 and 43 °C, respectively. It was found that the release rate of the model drug doxorubicin could be effectively controlled by altering the temperatures.[57]

The ATRP reaction in combination with click chemistry was also used by the same research group to prepare the amphiphilic chitosan-*g*-(PDEAEMA-*b*-PEO) copolymer.[58] The characterization of the synthesized amphiphilic copolymer was done by the ^{1}H NMR spectrum to elucidate the structure and confirmation of the synthesis. It was found that the CS-*g*-(PDEAEMA-*b*-PEO) copolymer can self-assemble into micelles in water. TEM, DLS, and transmittance were used to study the self-assembly and pH-responsive properties of the CS-*g*-

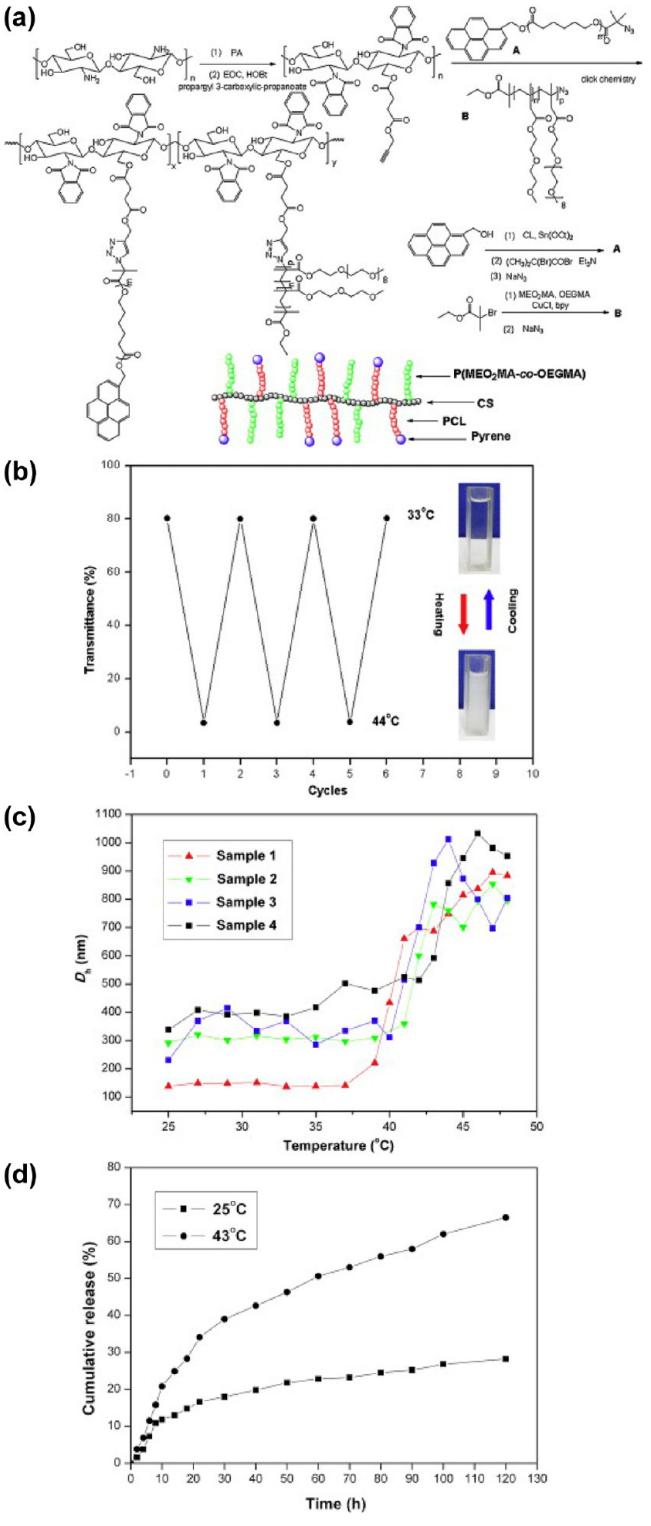

Figure 8. (a) Synthesis of CS-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) copolymer via combination of ROP, ATRP, and click chemistry. (b) Plots of transmittance as a function of temperature for CS-*g*-P (MEO_2MA-*co*-OEGMA) copolymer micelle solution (sample 2) (concentration: 2 mg/mL). (c) Temperature dependence of hydrodynamic diameters (D_h) for CS-*g*-P (MEO2MA-*co*-OEGMA) copolymers micelles (sample 1) and CS-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) (samples 2–4) micelles solutions (concentration: 1 mg/mL). (d) Drug release of CS-*g*-PCL (-*g*-P (MEO2MA-*co*-OEGMA)) micelles (sample 2) in PBS at 25 and 43 °C, respectively. Reprinted with permission.[57] Copyright 2011 Elsevier.

(PDEAEMA-*b*-PEO) copolymer. The pH values of solutions had a significant effect on the size of the micelles. By changing the value of pH, the size of micelle was found to change. The use of PEO segments during synthesis was found to improve the hydrophilicity of the copolymer and lead to avoidance of the excessive aggregation of the micelles. CS-*g*-(PDEAEMA-*b*-PEO) copolymers were suggested to be potential materials for applications in the biomedical field.[58] Chitosan/cellulose blend-based membranes were functionalized through polymer brushes via a surface-initiated atom transfer radical polymerization (ATRP) method for the chromatic warning and adsorptive removal of cadmium ions.[59] In this study, the ATRP grafting of negatively charged polymer brushes of 3-sulfopropyl methacrylate potassium salt (SMP) was first carried out on the chitosan/cellulose blend-based membrane. Subsequently 5, 10, 15, and 20 Tetrakis (1-methyl-4-pyridinio)porphyrin-tetra(p-toluene sulfonate), an optical indicator ligand, was immobilized onto the membranes utilizing the brushes grafted on the chitosan/cellulose blend-based membrane. The electrical interaction with the grafted SMP brushes was attributed to the immobilization of the ligand onto the surface of the membrane. It was found that the CS/CA-SMP-TMPyP membrane exhibited rapid and obvious colorimetric changes in an aqueous solution of cadmium ions. Furthermore, these membranes were found to exhibit significant enhancement in the performance for adsorptive removal of cadmium ions in a wide pH and concentration range. These membranes were even found to exhibit selectivity toward the cadmium ions in the presence of some other cations such as Na^+, K^+, Ca^{2+}, and Mg^{2+}. Table 3 shows the uptake and color response of the CS/CA-SMP-TMPyP membrane to Cd^{2+} in the presence of other interfering cations in the solutions. It was concluded from the study that these membranes are of a multifunctional nature and exhibit a high potential for practical applications in water and wastewater treatment.[59]

Chitosan-based thermoresponsive copolymers were also prepared using atom transfer radical polymerization of N-isopropylacrylamide.[60] To carry out the synthesis of the copolymers, first of all, the regioselective chitosan macroinitiator derivative 3,6-di-O-polyNIPAM-2-N-phthaloyl chitosan was prepared. The formation of the macroinitiator was confirmed through different characterizations. Different reaction components such as macroinitiator, ligand, NIPAM monomer, and transition metal were found to control and vary the degree of polymerization (DP) of polyNIPAM onto the chitosan derivative. Table 4 shows the reaction data for the ATRP synthesis of the copolymers. Degrees of polymerization up to 110.5 were obtained using ATRP synthesis, and it was found that the thermal properties can be significantly improved by increasing the degree of polymerization. The 3,6-di-O-polyNIPAM-2-N-phthaloyl chitosan copolymer was found to form a stable suspension and precipitate above a temperature of approximately 30 °C due to the hydrophilic-to-hydrophobic transition of the polyNIPAM graft component.[60]

The pH/temperature, multiple stimuli-responsive, chitosan-based, comb-shaped chitosan-*graft*-poly(*N*-isopropylacrylamide) copolymers were synthesized using ATRP.[61] For carrying out the synthesis, the ATRP initiating group was attached to N-phthaloyl chitosan, and after the completion of the reaction, a comb-shaped cationic copolymer containing long chitosan main chains and short PNIPAAm side chains was obtained. Polymer-pendant amino groups were then obtained by the removal of the protective groups on the N-phthaloyl chitosan-*graft*-poly(*N*-

Table 3. Uptake and Color Response of CS/CA-SMP-TMPyP Membrane to Cd^{2+} in Presence of Other Interfering Cations in the Solutions[a]

sample no.	interfering cation [X]	$[Cd^{2+}]$ ($\times 10^{-4}$ M)	[X] ($\times 10^{-4}$ M)	$[X]/[Cd^{2+}]$	Cd^{2+} uptake (mg/g)
1	–	1	–	–	3.6521
2	Na^+	1	10	10	3.6946
3	K^+	1	10	10	3.6113
4	Ca^{2+}	1	10	10	3.4062
5	Mg^{2+}	1	10	10	3.5768
6	Co^{2+}	1	1	1	3.0537
7	Ni^{2+}	1	1	1	2.9194

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Table 4. Reaction Data, Degree of Polymerization, and Monomer Conversion for ATRP of NIPAM[a]

$[M]_0:[I]_0:[CuBr]_0:[L]_0$	DMF:water (mL)	DPNIPAM[b]	conversion (%)[c]
50:1:1:1	4.5:0	2.6	5.1
50:1:1:1	4.5:1.0	3.3	6.7
50:1:1:1	4.5:2.5	4.9	9.8
50:1:3:3	4.5:2.5	14.6	29.1
100:1:1:1	4.5:0	3.7	3.7
100:1:1:1	4.5:1.0	4.9	4.9
100:1:1:1	4.5:2.5	6.6	6.6
100:1:3:3	4.5:2.5	42.9	43
200:1:1:1	4.5:0	90	45
200:1:1:1	4.5:1.0	55.5	27.8
200:1:1:1	4.5:2.5	34.3	17.2
200:1:3:3	4.5:2.5	110.5	55.3
400:1:1:1	7.0:0	80.2	20.1
400:1:3:3	7.0:2.5	70.6	17.6

[a]Reprinted with permission. Copyright 2013 Elsevier. [b]DP of polyNIPAM is determined by ^{1}H NMR data. The value was estimated by assuming that all initiators were consumed for ATRP. [c]Monomer conversion is calculated from DPNIPAM determined by NMR.

isopropylacrylamide) (PHCS-*g*-PNIPAAm) copolymer with $N_2H_4{\cdot}H_2O$. Characterization techniques such as ^{1}H NMR spectroscopy, dynamic light scattering (DLS), and transmission electron microscopy (TEM) were used to study the structural and solution behavior of the synthesized copolymers. The chitosan-*graft*-poly(*N*-isopropylacrylamide) (CS-*g*-PNIPAAm) copolymers were found to be self-assembled in an aqueous solution into stimuli-responsive core−shell micelles. The hydrodynamic diameters of these micelles were found to be about 170 nm. These copolymers were proposed to be promising materials for delivery of hydrophobic drug molecules due to their comb-shaped structure comprising hydrophobic and hydrophilic blocks.[61] Surface-initiated ATRP of cross-linked chitosan (CCS) microspheres was carried out to develop novel materials for the removal of toxic Cd (II) ions from aqueous solutions.[62] During the synthesis, pH-sensitive poly(methacrylic acid) (PMAA) was tethered on the microspheres. Table 5 shows the amount of carboxyl groups grafted onto the CCS microspheres surfaces at various polymerization times. The prepared microspheres were characterized by FTIR, XPS, SEM, and contact angle measurements to ascertain their successful synthesis. Batch adsorption studies were subsequently carried out to investigate the Cd (II) adsorption from an aqueous solution. Tables 6 and 7 show the parameters of kinetic models predicting the experimental data for Cd(II) adsorption by the CCS and CCS-*g*-PMAA microspheres and the regression parameters obtained using Langmuir isotherms on the Cd(II) adsorption data by the CCS-*g*-PMAA

Table 5. Amount of Carboxyl Groups Grafted onto CCS Microsphere Surfaces at Various Polymerization Times[a]

sample	polymerization time (h)	grafting yield (mg cm^{-2})	carboxyl concentration (μmol cm^{-2})
CCS-*g*-PMAA	1	0.263	1.563
	2	0.476	2.83
	3	0.67	3.983
	4	0.908	5.398

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microspheres, respectively. It was found that the pH of the solution has a significant impact on the Cd (II) adsorption. The maximum adsorption of the metal ions was found to be above pH 5 for the PMAA-grafted microspheres. The Langmuir fitting method was used to determine the maximum adsorption capacity, and it was found to be 1.3 mmol g^{-1} at pH 5 for the CCS-*g*-PMAA microspheres. These grafted microsphere were also investigated for the regeneration study and were found to exhibit an effective regeneration behavior with very highly stable adsorption capacity for repeated applications. The adsorption capacity of the PMAA-grafted microspheres was found to have remained almost unchanged upon a five cycle reuse. In the PMAA-grafted microspheres, Cd (II) coordination onto the PMAA chains through the complexation and electrostatic interactions between Cd (II) ions and the carboxylate groups was investigated through XPS analyses.[62]

CONCLUSIONS

Chitosan-based materials, due to their remarkable physicochemical, mechanical, and biological properties, offer a great potential for biomedical and environmental applications. A huge number of research publications for different applications employing chitosan as the core component demonstrate the commercial importance of this natural polymer. The cationic nature of chitosan makes it the most important candidate for numerous applications, which are not possible to meet by the other polysaccharides. Over the past few years, several strategies have been proposed to alter the surface characteristics of chitosan. Some of these are based on physical modifications and some on chemical modifications. Recently, the strategies to improve the existing properties of chitosan have emerged in terms of graft copolymerization techniques. These techniques are promising ways to incorporate the desired characteristics and functionalities in chitosan for the targeted applications. While this article solely focuses on the ATRP graft copolymerization of chitosan, there are also various techniques that have been used. Though chitosan offers a great alternative to other polysaccharides, it is imperative to understand the effect of incorporation of suitable functional groups on its surface for better end applications. Thus, a better

Table 6. Parameters of Kinetic Models Predicting Experimental Data for Cd (II) Adsorption by CCS and CCS-*g*-PMAA Microspheres[a]

		model parameters			
samples	models	k_f (min^{-1})	$k_s \times 10^{-3}$ (g $mmol^{-1}$ min^{-1})	q_{max} (mmol g^{-1})	χ^2
CCS	PFO[a]	0.038	–	0.15	0.76
	PSO[b]	–	6.73	0.17	0.15
CCS-*g*-PMAA	PFO	0.15	–	0.29	0.52
	PSO	–	11.71	0.32	0.24

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Table 7. Regression Parameters Obtained using Langmuir Isotherms on Cd (II) adsorption data by CCS-*g*-PMAA Microspheres[a]

		model parameters		
T (°C)	models	q_{max} (mmol g^{-1})	K_L (dm^3 $mmol^{-1}$)	χ^2
25	Langmuir	1.24	0.032	0.012
35	Langmuir	1.34	0.029	0.009
45	Langmuir	1.37	0.027	0.017

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understanding of the structure and chemistry of different polymerization reactions is most important to further develop and expand the multifunctional applications of chitosan.

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Notes

The authors declare no competing financial interest.

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REFERENCES

(1) Thakur, V. K.; Thunga, M.; Madbouly, S. A.; Kessler, M. R. PMMA-g-SOY as a sustainable novel dielectric material. *RSC Adv.* **2014**, *4*, 18240−18249.
(2) Jiang, H.; Ma, J.; Li, C. Polyaniline-MnO_2 coaxial nanofiber with hierarchical structure for high-performance supercapacitors. *J. Mater. Chem.* **2012**, *22*, 16939−16942.
(3) Jiang, H.; Lee, P. S.; Li, C. 3D carbon based nanostructures for advanced supercapacitors. *Energy Environ. Sci.* **2013**, *6*, 41−53.

(4) Thakur, V. K.; Thakur, M. K. Recent advances in green hydrogels from lignin: a review. *Int. J. Biol. Macromol.* **2015**, *72*, 834−847.
(5) Thakur, V. K.; Thakur, M. K. Recent trends in hydrogels based on psyllium polysaccharide: A review. *J. Clean Prod.* **2014**, *82*, 1−15.
(6) Agarwal, S.; Gupta, R. K.; Doraiswamy, D. A model for the collision efficiency of shear-induced agglomeration involving polymer bridging. *Colloids Surf., A* **2009**, *345*, 224−230.
(7) Drozdov, A. D.; Agarwal, S.; Gupta, R. K. The effect of temperature on the viscoelastic response of polymer melts. *Int. J. Eng. Sci.* **2005**, *43*, 304−320.
(8) Thakur, V. K.; Thakur, M. K. Processing and characterization of natural cellulose fibers/thermoset polymer composites. *Carbohydr. Polym.* **2014**, *109*, 102−117.
(9) Thakur, V. K.; Thakur, M. K.; Raghavan, P.; Kessler, M. R. Progress in green polymer composites from lignin for multifunctional applications: A review. *ACS Sustainable Chem. Eng.* **2014**, *2*, 1072−1092.
(10) Thakur, V. K.; Vennerberg, D.; Madbouly, S. A.; Kessler, M. R. Bio-inspired green surface functionalization of PMMA for multifunctional capacitors. *RSC Adv.* **2014**, *4*, 6677−6684.
(11) Thakur, V. K.; Thakur, M. K.; Gupta, R. K. Graft copolymers of natural fibers for green composites. *Carbohydr. Polym.* **2014**, *104*, 87−93.
(12) Yeh, S.-K.; Agarwal, S.; Gupta, R. K. Wood−plastic composites formulated with virgin and recycled ABS. *Compos. Sci. Technol.* **2009**, *69*, 2225−2230.
(13) Thakur, V. K.; Vennerberg, D.; Kessler, M. R. Green aqueous surface modification of polypropylene for novel polymer nanocomposites. *ACS Appl. Mater. Interfaces* **2014**, *6*, 9349−9356.
(14) Thakur, V. K.; Thakur, M. K.; Gupta, R. K. Review: Raw natural fiber-based polymer composites. *Int. J. Polym. Anal. Charact.* **2014**, *19*, 256−271.
(15) Thakur, V. K.; Thakur, M. K.; Gupta, R. K. Graft copolymers from cellulose: Synthesis, characterization and evaluation. *Carbohydr. Polym.* **2013**, *97*, 18−25.
(16) Gopalakrishnan, L.; Ramana, L. N.; Sethuraman, S.; Krishnan, U. M. Ellagic acid encapsulated chitosan nanoparticles as anti-hemorrhagic agent. *Carbohydr. Polym.* **2014**, *111*, 215−221.
(17) Ferris, C.; Casas, M.; Lucero, M. J.; de Paz, M. V.; Jimenez-Castellanos, M. R. Synthesis and characterization of a novel chitosan-N-acetyl-homocysteine thiolactone polymer using MES buffer. *Carbohydr. Polym.* **2014**, *111*, 125−132.
(18) Thakur, V. K.; Thakur, M. K.; Gupta, R. K. Rapid synthesis of graft copolymers from natural cellulose fibers. *Carbohydr. Polym.* **2013**, *98*, 820−828.
(19) Stefan, J.; Lorkowska-Zawicka, B.; Kaminski, K.; Szczubialka, K.; Nowakowska, M.; Korbut, R. The current view on biological potency of cationically modified chitosan. *J. Physiol. Pharmacol.* **2014**, *65*, 341−347.
(20) Jiang, T.; Deng, M.; James, R.; Nair, L. S.; Laurencin, C. T. Micro- and nanofabrication of chitosan structures for regenerative engineering. *Acta Biomater.* **2014**, *10*, 1632−1645.
(21) Lai, G.-J.; Shalumon, K. T.; Chen, S.-H.; Chen, J.-P. Composite chitosan/silk fibroin nanofibers for modulation of osteogenic differentiation and proliferation of human mesenchymal stem cells. *Carbohydr. Polym.* **2014**, *111*, 288−297.
(22) Lai, P.; Daear, W.; Löbenberg, R.; Prenner, E. J. Overview of the preparation of organic polymeric nanoparticles for drug delivery based on gelatine, chitosan, poly(D,L-lactide-co-glycolic acid) and polyalkylcyanoacrylate. *Colloids Surf., B* **2014**, *118*, 154−163.
(23) Jayakumar, R.; Menon, D.; Manzoor, K.; Nair, S. V.; Tamura, H. Biomedical applications of chitin and chitosan based nanomaterials—A short review. *Carbohydr. Polym.* **2010**, *82*, 227−232.
(24) Kasaai, M. R. Determination of the degree of N-acetylation for chitin and chitosan by various NMR spectroscopy techniques: A review. *Carbohydr. Polym.* **2010**, *79*, 801−810.
(25) Kasaai, M. R. A review of several reported procedures to determine the degree of N-acetylation for chitin and chitosan using infrared spectroscopy. *Carbohydr. Polym.* **2008**, *71*, 497−508.
(26) Estevinho, B. N.; Rocha, F.; Santos, L.; Alves, A. Microencapsulation with chitosan by spray drying for industry applications − A review. *Trends Food Sci. Technol.* **2013**, *31*, 138−155.
(27) Muzzarelli, R. A. A.; Greco, F.; Busilacchi, A.; Sollazzo, V.; Gigante, A. Chitosan, hyaluronan and chondroitin sulfate in tissue engineering for cartilage regeneration: A review. *Carbohydr. Polym.* **2012**, *89*, 723−739.
(28) Ragelle, H.; Vandermeulen, G.; Préat, V. Chitosan-based siRNA delivery systems. *J. Control. Release Off. J. Control. Release Soc.* **2013**, *172*, 207−218.
(29) Shukla, S. K.; Mishra, A. K.; Arotiba, O. A.; Mamba, B. B. Chitosan-based nanomaterials: A state-of-the-art review. *Int. J. Biol. Macromol.* **2013**, *59*, 46−58.
(30) Balan, V.; Verestiuc, L. Strategies to improve chitosan hemocompatibility: A review. *Eur. Polym. J.* **2014**, *53*, 171−188.
(31) Crini, G.; Badot, P.-M. Application of chitosan, a natural aminopolysaccharide, for dye removal from aqueous solutions by adsorption processes using batch studies: A review of recent literature. *Prog. Polym. Sci.* **2008**, *33*, 399−447.
(32) Croisier, F.; Jérôme, C. Chitosan-based biomaterials for tissue engineering. *Eur. Polym. J.* **2013**, *49*, 780−792.
(33) Elsabee, M. Z.; Naguib, H. F.; Morsi, R. E. Chitosan based nanofibers, review. *Mater. Sci. Eng., C* **2012**, *32*, 1711−1726.
(34) Pontoni, L.; Fabbricino, M. Use of chitosan and chitosan-derivatives to remove arsenic from aqueous solutions—a mini review. *Carbohydr. Res.* **2012**, *356*, 86−92.
(35) Muzzarelli, R. A. A. Potential of chitin/chitosan-bearing materials for uranium recovery: An interdisciplinary review. *Carbohydr. Polym.* **2011**, *84*, 54−63.
(36) Wan Ngah, W. S.; Teong, L. C.; Hanafiah, M. A. K. M. Adsorption of dyes and heavy metal ions by chitosan composites: A review. *Carbohydr. Polym.* **2011**, *83*, 1446−1456.
(37) Bhatnagar, A.; Sillanpää, M. Applications of chitin- and chitosan-derivatives for the detoxification of water and wastewater — A short review. *Adv. Colloid Interface Sci.* **2009**, *152*, 26−38.
(38) Cunha, A. G.; Gandini, A. Turning polysaccharides into hydrophobic materials: A critical review. Part 2. Hemicelluloses, chitin/chitosan, starch, pectin and alginates. *Cellulose* **2010**, *17*, 1045−1065.
(39) Prabaharan, M. Review paper: Chitosan derivatives as promising materials for controlled drug delivery. *J. Biomater. Appl.* **2008**, *23*, 5−36.
(40) Zohuriaan-Mehr, M. J. Advances in chitin and chitosan modification through graft copolymerization: a comprehensive review. *Iran Polym. J.* **2005**, *14*, 235−265.
(41) Jenkins, D. W.; Hudson, S. M. Review of vinyl graft copolymerization featuring recent advances toward controlled radical-based reactions and illustrated with chitin/chitosan trunk polymers. *Chem. Rev.* **2001**, *101*, 3245−3273.
(42) Thakur, V. K.; Thakur, M. K.; Gupta, R. K. Synthesis of lignocellulosic polymer with improved chemical resistance through free radical polymerization. *Int. J. Biol. Macromol.* **2013**, *61*, 121−126.
(43) Thakur, V. K.; Thakur, M. K.; Gupta, R. K. Development of functionalized cellulosic biopolymers by graft copolymerization. *Int. J. Biol. Macromol.* **2013**, *62*, 44−51.
(44) Xu, F. J.; Yang, W. T. Polymer vectors via controlled/living radical polymerization for gene delivery. *Prog. Polym. Sci.* **2011**, *36*, 1099−1131.
(45) Siegwart, D. J.; Oh, J. K.; Matyjaszewski, K. ATRP in the design of functional materials for biomedical applications. *Prog. Polym. Sci.* **2012**, *37*, 18−37.
(46) Zhao, Y.; Wang, L.; Xiao, A.; Yu, H. The synthesis of modified polyethylene via coordination polymerization followed by ATRP, RAFT, NMRP or ROP. *Prog. Polym. Sci.* **2010**, *35*, 1195−1216.
(47) El Tahlawy, K.; Hudson, S. M. Synthesis of a well-defined chitosan graft poly(methoxy polyethyleneglycol methacrylate) by atom transfer radical polymerization. *J. Appl. Polym. Sci.* **2003**, *89*, 901−912.
(48) Li, N.; Bai, R. B.; Liu, C. K. Enhanced and selective adsorption of mercury ions on chitosan beads grafted with polyacrylamide via surface-initiated atom transfer radical polymerization. *Langmuir* **2005**, *21*, 11780−11787.

(49) Liu, P.; Su, Z. X. Surface-initiated atom transfer radical polymerization (SI-ATRP) of styrene from chitosan particles. *Mater. Lett.* **2006**, *60*, 1137−1139.
(50) Tang, F.; Zhang, L.; Zhu, J.; Cheng, Z.; Zhu, X. Surface functionalization of chitosan nanospheres via surface-initiated AGET ATRP mediated by iron catalyst in the presence of limited amounts of air. *Ind. Eng. Chem. Res.* **2009**, *48*, 6216−6223.
(51) Munro, N. H.; Hanton, L. R.; Moratti, S. C.; Robinson, B. H. Synthesis and characterisation of chitosan-graft-poly(OEGMA) copolymers prepared by ATRP. *Carbohydr. Polym.* **2009**, *77*, 496−505.
(52) Bao, H.; Hu, J.; Gan, L. H.; Li, L. Stepped association of comb-like and stimuli-responsive graft chitosan copolymer synthesized using ATRP and active ester conjugation methods. *J. Polym. Sci., Part A: Polym. Chem.* **2009**, *47*, 6682−6692.
(53) Bao, H.; Li, L.; Gan, L. H.; Ping, Y.; Li, J.; Ravi, P. Thermo- and pH-responsive association behavior of dual hydrophilic graft chitosan terpolymer synthesized via ATRP and click chemistry. *Macromolecules* **2010**, *43*, 5679−5687.
(54) Bao, H.; Li, L.; Leong, W. C.; Gan, L. H. Thermo-responsive association of chitosan-graft-poly(N-isopropylacrylamide) in aqueous solutions. *J. Phys. Chem. B* **2010**, *114*, 10666−10673.
(55) Yuan, H.; Lin, Y.; Songwei, T.; Hongliang, J.; Kehua, T.; Liqun, W. ATRP Synthesis and Self-Assembly Behavior of Chitosan-O-Pmpegma. *Acta Polym. Sin.* **2010**, 897−902.
(56) Yuan, W.; Zhao, Z.; Yuan, J.; Gu, S.; Zhang, F.; Xie, X.; Ren, J. Synthesis of pH- and temperature-responsive chitosan-graft-poly [2-(N, N-dimethylamino) ethyl methacrylate] copolymer and gold nanoparticle stabilization by its micelles. *Polym. Int.* **2011**, *60*, 194−201.
(57) Yuan, W.; Li, X.; Gu, S.; Cao, A.; Ren, J. Amphiphilic chitosan graft copolymer via combination of ROP, ATRP and click chemistry: Synthesis, self-assembly, thermosensitivity, fluorescence, and controlled drug release. *Polymer* **2011**, *52*, 658−666.
(58) Yuan, W.; Zhao, Z.; Gu, S.; Ren, T.; Ren, J. Synthesis and self-assembly of pH-responsive chitosan graft copolymer by the combination of atom transfer radical polymerization and click chemistry. *Mater. Lett.* **2011**, *65*, 793−796.
(59) Zhang, L.; Zhao, Y.-H.; Bai, R. Development of a multifunctional membrane for chromatic warning and enhanced adsorptive removal of heavy metal ions: Application to cadmium. *J. Membr. Sci.* **2011**, *379*, 69−79.
(60) Ifuku, S.; Miwa, T.; Morimoto, M.; Saimoto, H. Thermoresponsive chitosan/N-isopropylacrylamide copolymer through atom transfer radical polymerization. *Int. J. Biol. Macromol.* **2013**, *52*, 14−19.
(61) Chen, C.; Liu, M.; Gao, C.; Lu, S.; Chen, J.; Yu, X.; Ding, E.; Yu, C.; Guo, J.; Cui, G. A convenient way to synthesize comb-shaped chitosan-graft-poly (N-isopropylacrylamide) copolymer. *Carbohydr. Polym.* **2013**, *92*, 621−628.
(62) Huang, L.; Yuan, S.; Lv, L.; Tan, G.; Liang, B.; Pehkonen, S. O. Poly(methacrylic acid)-grafted chitosan microspheres via surface-initiated ATRP for enhanced removal of Cd(II) ions from aqueous solution. *J. Colloid Interface Sci.* **2013**, *405*, 171−182.