Recent Progress of Glycopolymer Synthesis for Biomedical Applications

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Abstract

Glycopolymers are an important class of biomaterials which include carbohydrate moieties in the polymer structure. In addition to biological research of the glycopolymer's interactions with lectin-carbonate, the glycopolymer has recently been used as a new synthetic biomaterial for direct therapeutic methods, medical adhesives, and biosensors. Thus, comprehensive understanding of new advances in glycopolymer research is essential for the next level of biomaterial studies. This review article highlights commonly used glycopolymer synthesis methods and biomedical applications there of. Glycopolymers can be synthesized by modern polymerization methods that can control molecular weight, molecular weight distribution, chemical functionality, and polymer architecture. The polymerizations include free radical polymerization, atom transfer radical polymerization, reversible addition-fragmentation chain-transfer polymerization, and nitroxide-mediated polymerization. Because the carbohydrate-lectin interactions with glycopolymers involves in many biological processes, the carbohydrate containing glycopolymers are used in 1) fundamental studies to understand specificity and strength of biological bindings, 2) controllable interactions to prevent microorganism adhesion to human cells, 3) large scale bulk adhesive for medical applications, 4) biocompatible therapeutic nanoparticles, 5) direct drug delivery vehicles, and 6) precise quantitative measuring biosensor materials that can detect physiological signals.

1. Introduction

Synthetic polymers containing carbohydrate pendants, commonly referred as glycopolymer, have attracted many scientists due to their significance in biological processes. Carbohydrates contain three major components including monosaccharides, oligosaccharides, and polysaccharides. The synthetic glycopolymer generally possesses a monosaccharide and/or oligosaccharide pendant group in a repeating unit. The chemical study of carbohydrates, natural

saccharides, was first published by Emil Fischer in 1884.[1, 2] The cyclic structure of natural carbohydrates which can be seen in maltose, sucrose, lactose, and cellobiose were elucidated in 1930s by Haworth and colleagues.[3] Soon after, the macromolecule structure of saccharides and polysaccharides were discovered. Two conventional applications of polysaccharides are found in food (e.g., molasses, starch, and glycogen)[4] and structural materials (e.g., cellulose, collagen, fiber, and chitin).[5-7] In modern science research, the carbohydrate plays an important role in understanding and controlling various biological processes. For instance, heparin (Figure 1a), a sulfated polysaccharide (glycosaminoglycan), interferes with the blood clotting process in the human body by inhibiting thrombin activation during the coagulation cascades.[8] Other polysaccharide examples such as hyaluronan (Figure 1b) and chondroitin sulfate (Figure 1c) possess anti-inflammatory properties.[9]

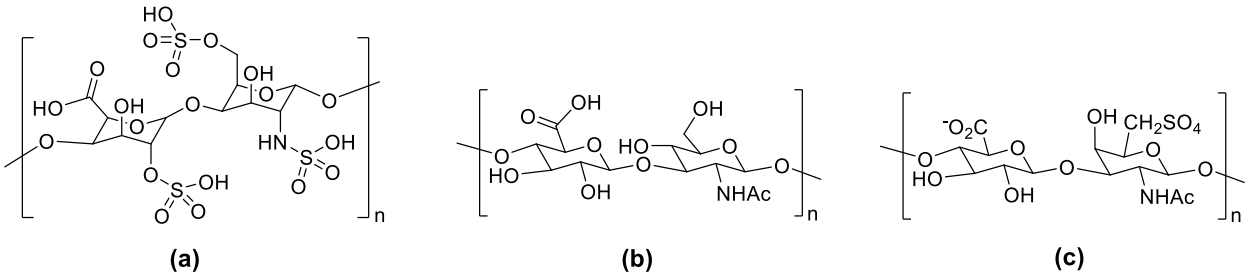


Figure 1. Structures of polysaccharide example: **(a)** heparin, **(b)** hyaluronan, and **(c)** chondroitin sulfate.

Recent advances in carbohydrate research has led to a new division of biology, glycobiology.[10] Glycobiology significantly increases understanding of sophisticated interactions of carbohydrates for biomedical applications,[11, 12] especially in selective and controlled recognition events.[13] Accurate recognition is essential to cell interactions[13] including fertilization,[14, 15] embryogenesis,[16, 17] cell migration,[18] organ formation,[19] bacterial/viral infection,[20] inflammation,[21] and cancer metastasis.[22] Precise and efficient synthesis of carbohydrates must be progressed in order to fully comprehend and utilize these important cell interactions. Synthesis of carbohydrates has been a pivotal point that resulted in crucial progress in biomedical fields such as carbohydrate-based vaccine,[23] drug carrier and delivery,[24, 25] tissue scaffold engineering,[26, 27] and HIV treatment.[28]

The specific binding interaction of a glycopolymer to carbohydrate-binding proteins (lectins)[29-31] result in cell agglutination such as hemagglutination.[32] Linear glycopolymers offer enhanced binding affinity to lectins due to the multivalent binding sites compared to a single

carbohydrate unit. This holds true as long as the functionalization of the carbohydrate moieties does not impede the recognition process.[32] In addition, linear synthetic glycopolymers are the most studied class of glycopolymers due to the simplicity of its synthesis.[33] Whitesides and co-workers have elucidated that optimum binding is achieved when the distance between two glucose pendants on polymer is equivalent to the distance between binding sites of the lectin.[34] Likewise, linear glycopolymers exhibit generally acceptable biorecognition capability. However, the application of regular linear glycopolymer is limited due to insufficient binding affinity to lectins compared to other polymer architectures. The poor binding affinity is raised from unwanted clustering which occurs often due to hydrogen bonds between hydroxyl groups of carbohydrate pendants.[35] Therefore, fields of study that require high yield of recognition, including biosensing and drug delivery, do not commonly use linear glycopolymers.

The advanced non-linear glycopolymer architecture (e.g., micelles of amphiphilic polymers, dendrimers, and nano-particle cored star-shape polymers) provides more surface area for binding between the polymer's carbohydrate and lectins.[32] In addition, new synthetic methods that enable well-defined glycopolymer chemical structures can precisely control the binding site distance that can customize the related applications. Hence, more sophisticated structures, which are non-linear and have well-defined architectures like of glycopolymers, has gained high interest in recent glycopolymer studies.

2. Polymerization methods to prepare glycopolymers

Due to excessive scientific and practical potential of glycopolymers, various architectures of glycopolymers have been prepared. Modern polymerization techniques used in preparing glycopoymers are discussed in this section.

2.1. Glycopolymer synthesis via conventional free radical polymerization (FRP)

Conventional free radical polymerization (FRP) has been the most commonly used polymerization technique to synthesize glycopolymer. Typical advantages of conventional FRP include well-established experimental methods, ample commercially available initiators, moderate tolerance to impurities, and broad range of reaction conditions in term of solvents and temperatures.[13, 32] Glycomonomers are mainly synthesized by glycosylation (a chemical reaction

between carbohydrates and other molecules) of vinyl compounds. The carbohydrate containing vinyl monomers may possess additional functionalities on a pendant group.[36]

Horejsi *et al.* reported an initial example of monomer synthesis and polymerization of glucoside acrylates *via* conventional FRP (Figure 2a).[37] The conventional FRP of glucoside acrylates was conducted in water using an ammonium persulfate initiator and a tetramethylethylenediamine (TMEDA) catalyst (Figure 2b).[37] The synthesized glycopolymer exhibits similar characteristics to natural polysaccharides in terms of lectin binding.[37]

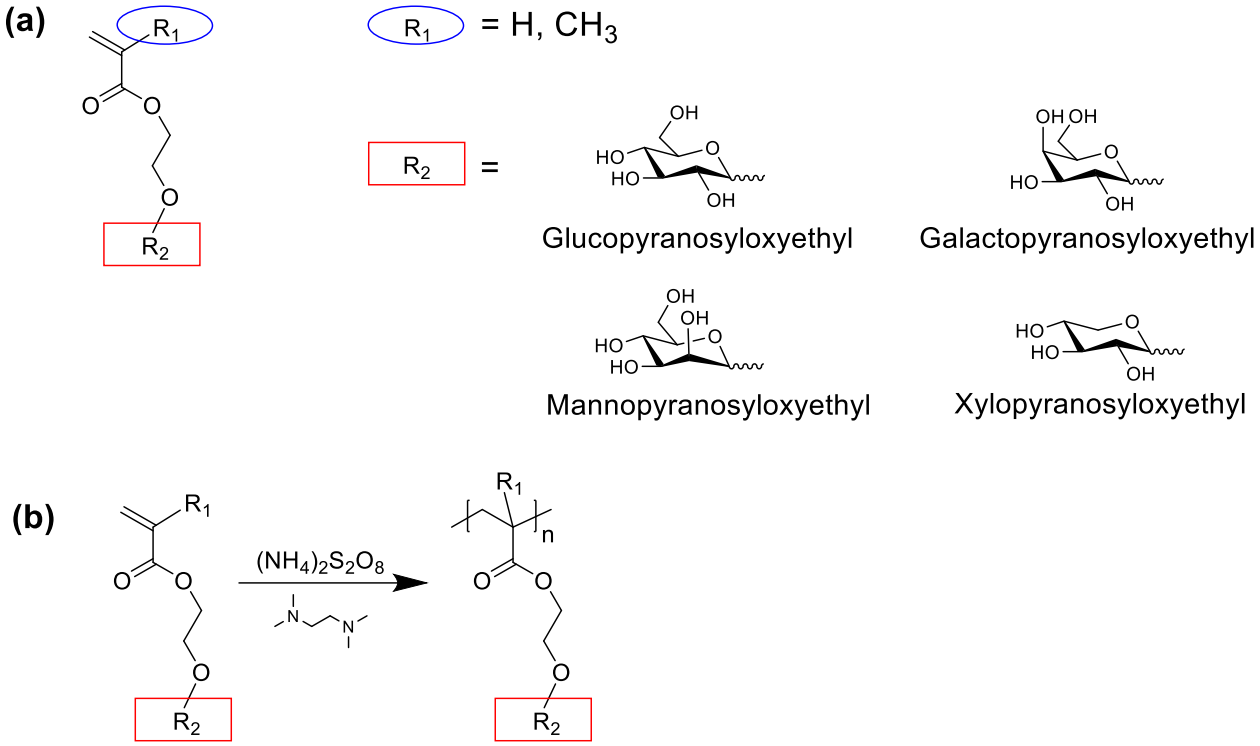


Figure 2. (a) Glucoside acrylate monomer, and **(b)** Free radical polymerization (FRP) of linear glycopolymer using ammonium persulfate as the initiator and tetramethylethylenediamine (TMEDA) as the catalyst.[37]

Coupling of oligosaccharides with *p*-vinylbenzylamine was reported by Kazukiyo *et al.*[38] Figure 3a displays the oxidation of lactose in the presence of hypoiodite to form lactone. The oxidized lactone was subsequently reacted with *p*-vinylbenzylamine (Figure 3b) by refluxing in methanol yielding a vinyl benzyl containing monomer in Figure 3b and c. This glycosylation step showed high yield without protection of hydroxyl groups on carbohydrates.[38] The glycomonomer was then polymerized *via* conventional FRP with azobisisobutyronitrile (AIBN) initiator and potassium persulfate ($K_2S_2O_8$) (Figure 3c). The prepared glycopolymer showed interaction with

Concanavalin A (Con A) which is a commonly used commercially available plant-originated lectin.

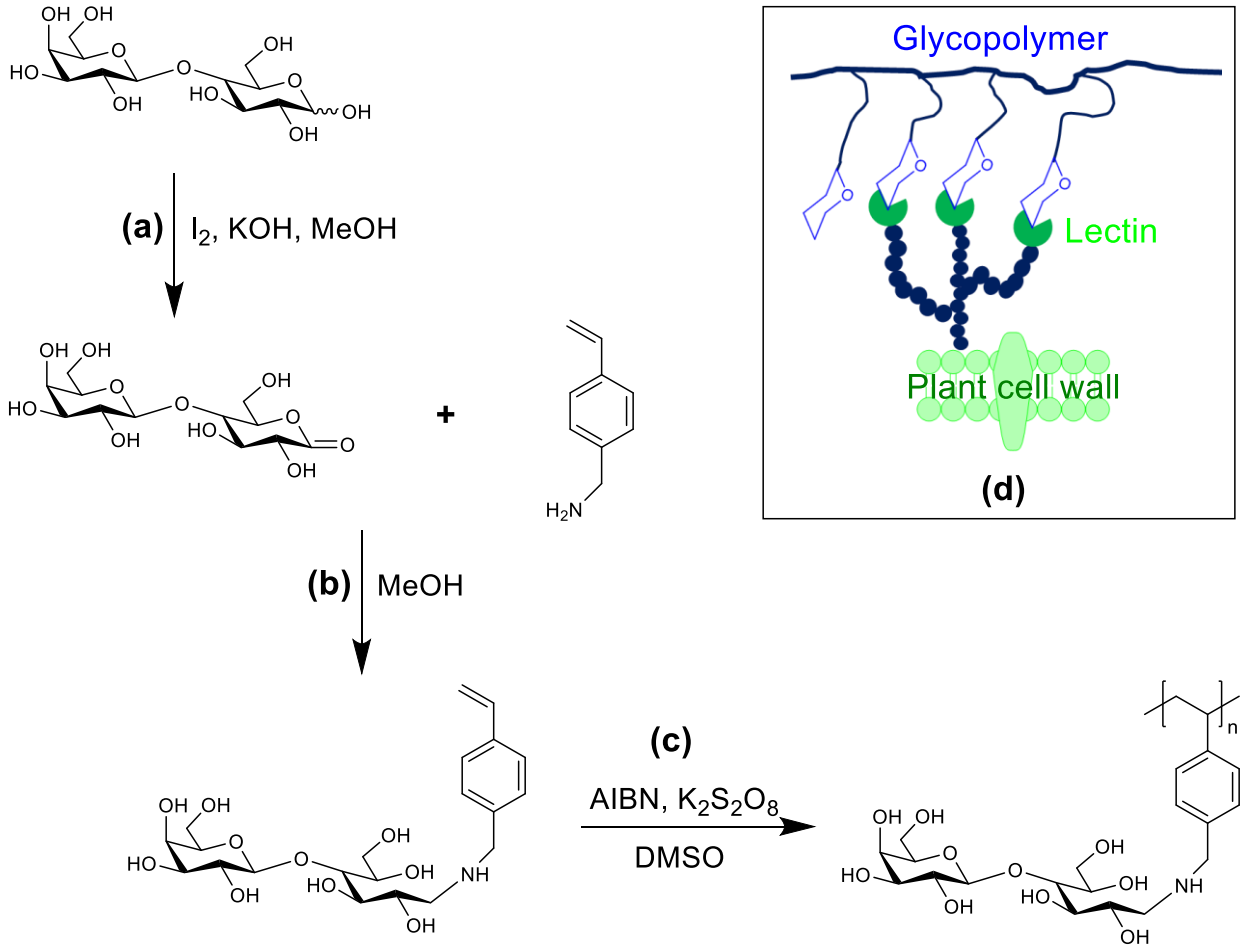


Figure 3. (a) Oxidation of lactose to lactone, **(b)** glycosylation of lactone with *p*-vinylbenzylamine, **(c)** FRP of lactose-substituted polystyrene, and **(d)** interaction between glycopolymer and *Concanavalin A* (Con A), a commonly used commercially available plant-originated lectin.[38]

Roy *et al.* reported synthesis of 4-acrylamidophenyl β-lactoside monomer from lactose as shown in Figure 4.[39] Acetyl protection of hydroxyl groups were required for the glycosylation during monomer synthesis. The protected, brominated carbohydrate was then reacted with a phenolic compounds to yield a corresponding vinyl monomers as a result of a substitution reaction (Figure 4a). The prepared 4-acrylamidophenyl β-lactoside monomer was then copolymerized with acrylamide by utilizing ammonium persulfate as an initiator and TMEDA as catalyst at 90 ^{o}C (Figure 4b).[39] The binding capability of poly(acrylamidophenyl β-lactoside) was tested with three different lectins: *A. hypogaea* (peanut) lectin, *Ricinus communis* (castor bean) agglutinin, and

wheat germ agglutinin (WGA). Lectins from peanuts and castor beans showed precipitate formation whereas wheat germ lectin did not show any interaction with the glycopolymer; hence, demonstrating the selective glycopolymer-lectin interactions.[39]

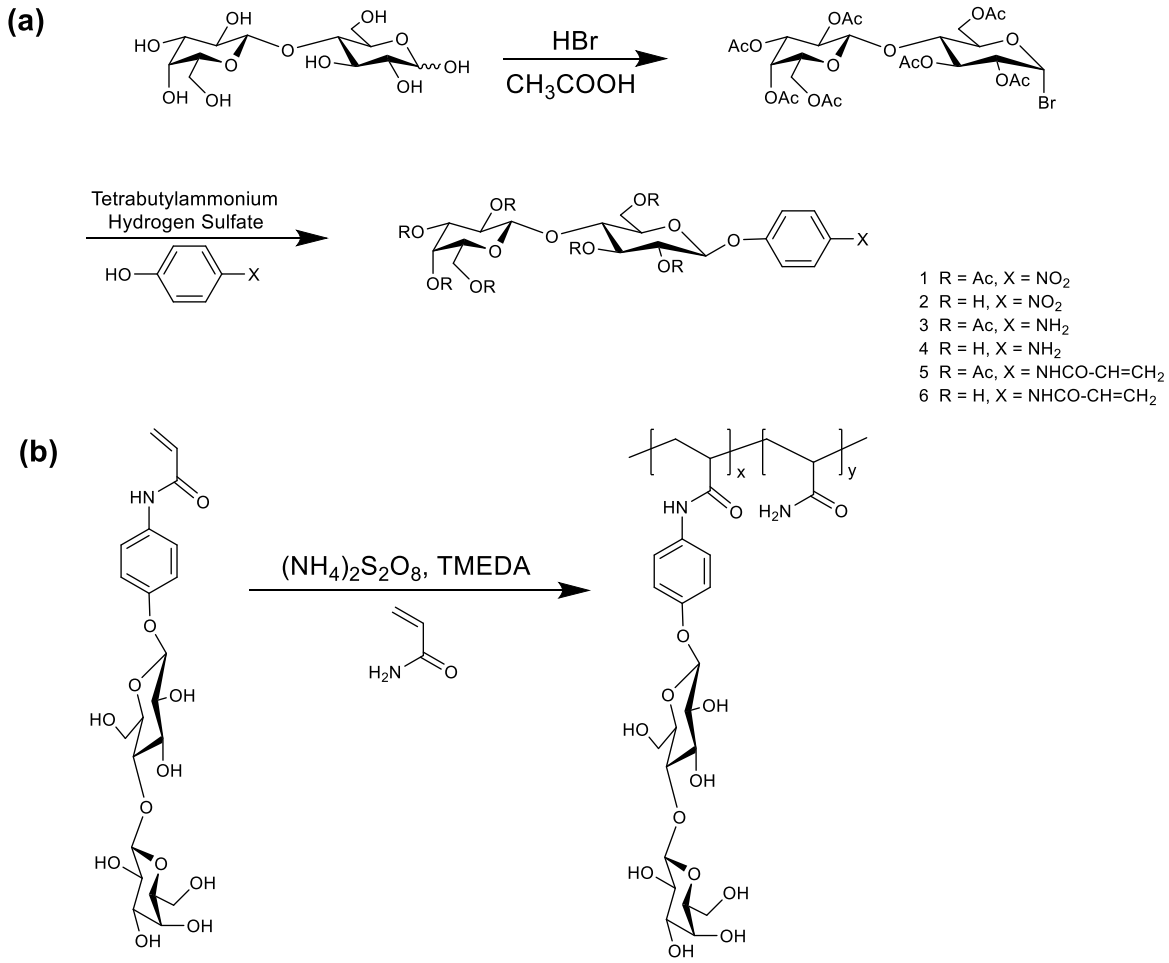


Figure 4. (a) Modification of lactose to 4-acrylamidophenyl β-lactoside to prepare monomers, and **(b)** copolymerization of 4-acrylamidophenyl β-lactoside and acrylamide *via* FRP to synthesize poly(acrylamide-*co*-acrylamidophenyl β-lactoside).[39]

Figure 5 displays other glycopolymers that are prepared from carbohydrate containing vinyl monomers through conventional FRP. As previously stated, FRP has been widely used in glycopolymer synthesis. However, conventional FRP has disadvantages including poor molecular weight control which leads high molecular weight distribution (polydispersity index, PDI > 2.0) and poor control on polymer terminals.[13]

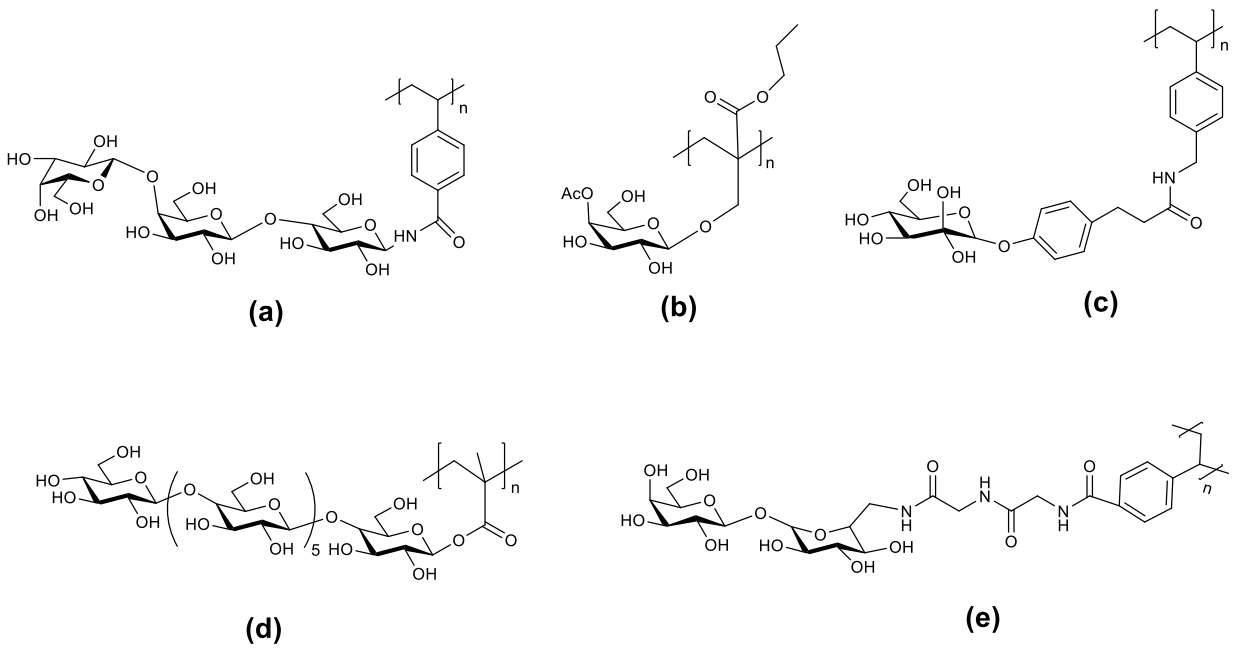


Figure 5. (a) poly(p-vinylbenzamido-β-4′-galactosyllactose),[40] **(b)** poly(ethyl α-(β-D-galactosyloxymethyl)acrylate)),[41] **(c)** poly[*p*-(2-(N-(*p-vinylbenzyl*)carbamoyl)ethyl)phenyl α-D-mannopyranoside],[42] **(d)** poly(O-methacryloyl maltoheptaoside),[43] and **(e)** poly[(galacto-trhalose)acrylate].[44]

2.2. Glycopolymer synthesis via controlled radical polymerizations (CRP)

Atom transfer radical polymerization (ATRP). ATRP is a type of controlled radical polymerization (CRP) that allows precise control of molecular weight distribution, functional terminals, molecular weights, and polymer architecture. Typical ATRP can be performed in the presence of a transition metal catalyst complex and an alkyl halide initiator for polymerization of polar group containing vinyl monomers.[45] Copper metal complexes have been most commonly used; however, diverse metal complexes, such as Ru, Fe, Mo, and Os, have also been previously reported as the transition metal catalyst complex.[46] With reference to glycopolymer, ATRP has two key advantages over FRP. ATRP can be carried out without protection of hydroxyl groups in the glycomonomer due to ATRP's excellent functional group tolerance.[47] ATRP also allows for more control over molecular weight which leads to narrow molecular distributions. Another important feature of ATRP is the halide polymer terminal which can be used for further post-polymerization modifications.[47]

Obata *et al.* reported an amphiphilic block glycopolymer, poly(2-(α-D-mannopyrano-syloxy) ethyl methacrylate)-*b*-poly(L-lactide) (PManEMA-*b*-PLLA) that is synthesized by

ATRP.[48] Two polymers, PLLA and PManEMA, were synthesized separately *via* ring opening polymerization (ROP) and ATRP as shown in Figure 6a and 6b, respectively. By selecting azide containing ATRP initiator, azide terminal PManEMA was prepared (Figure 6b). Next, the two prepared polymers were coupled by copper-catalyzed click reaction to produce a block copolymer (Figure 6).[48]

Figure 7 demonstrates surface initiated ATRP of a glycomonomer. The target surface was treated with plasma followed by the introduction of amine on the surface (Figure 7b, left). The covalently linked amine on the surface was used to integrate alkyl halide ATRP initiator. A similar surface modification was performed for gold electrodes as shown in Figure 7a and b in the right side schematic. After the successful introduction of alkyl halides on the surface, a separately prepared glycomonomer, N-acetylglucosamine (GlcNAc), was polymerized by ATRP.[49, 50] In particular, Figure 7a illustrates the application of an advanced biosensor platform that studies influence of glycopolymer graft lengths on lectin-glycan binding.[49]

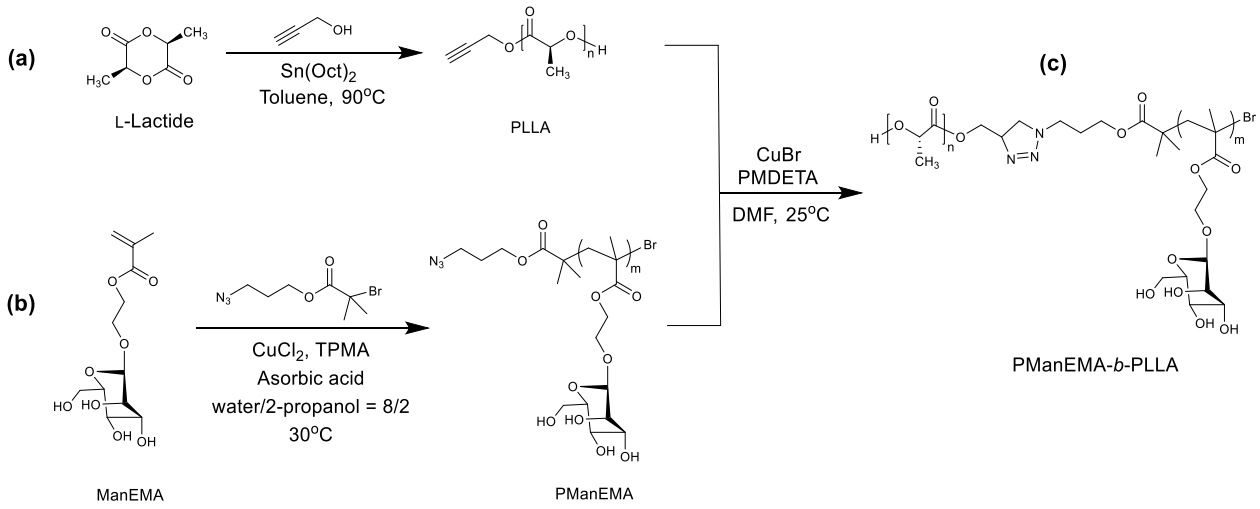


Figure 6. Synthesis of block glycopolymer, poly(2-(α-D-mannopyrano-syloxy) ethyl methacrylate)-*b*-poly(L-lactide) (PManEMA-*b*-PLLA); **(a)** synthesis of PLLA via ROP and **(b)** ATRP of ManEMA, and **(c)** PManEMA-*b*-PLLA *via* click chemistry.[48]

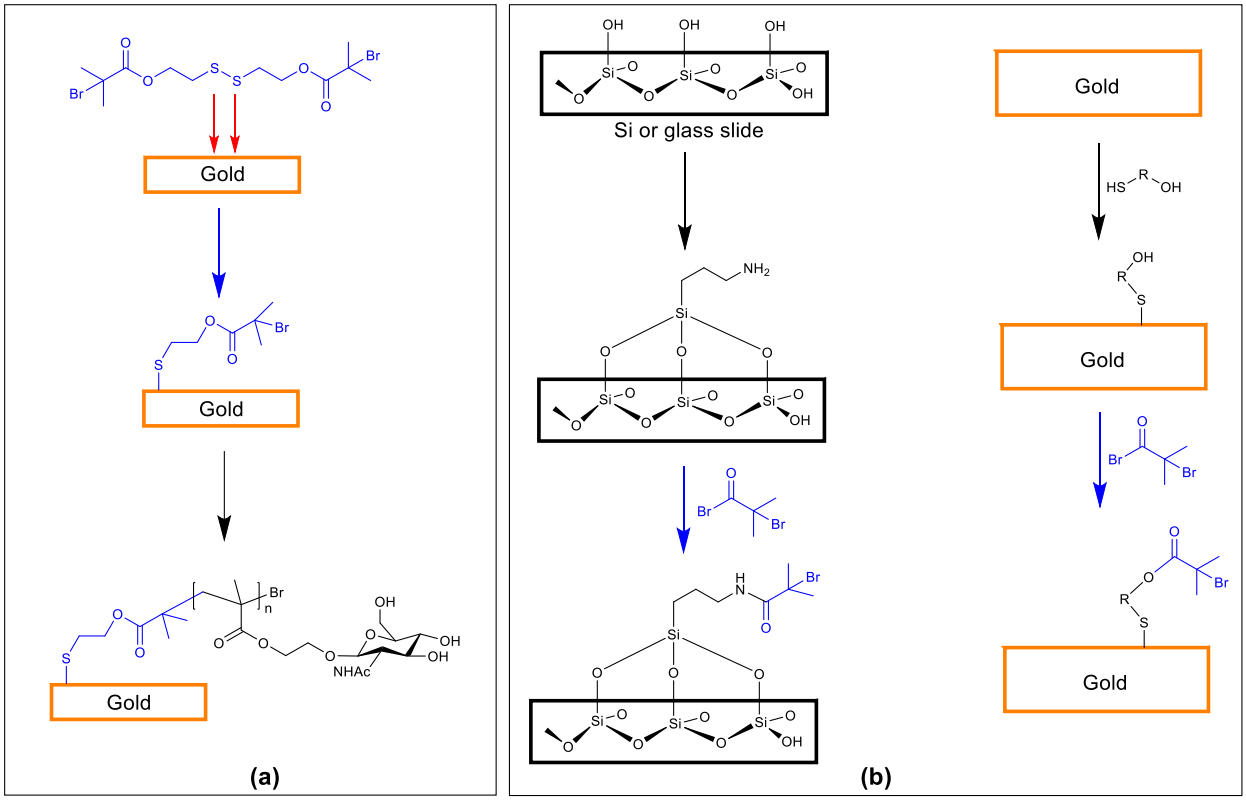


Figure 7. (a) Surface-initiated ATRP of N-acetylglucosamine (GlcNAc) monomer on plasma-treated gold electrodes,[49] and **(b)** initiator deposition on both substrate surfaces.[50]

Reversible addition-fragmentation chain-transfer polymerization (RAFT). RAFT is another efficient CRP method which was first reported in 1998 by Rizzardo *et al.* at Commonwealth Scientific and Industrial Research Organization (CSIRO).[51] Unlike ATRP, RAFT does not require a transition metal to mediate the polymerization. RAFT requires a chain transfer agent (CTA) (Figure 8a and 8c) that provides reversible deactivation of propagating radicals by degeneration of chain transfer. RAFT also requires a radical initiator such as AIBN. Commonly used CTAs of RAFT includes dithioesters, trithiocarbonates, xanthates, and dithiocarbamates, shown in Figure 8c.[52]

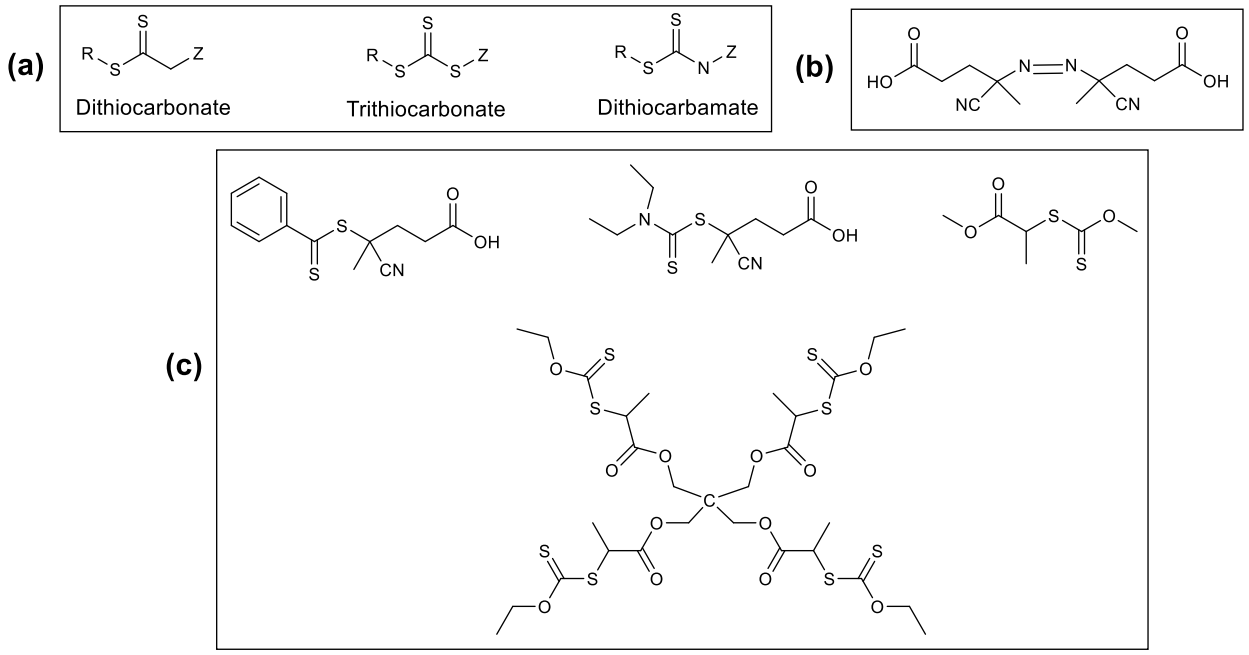


Figure 8. (a) Generalized chemical structures of commonly used chain transfer agents (CTA) in RAFT,[47] **(b)** an example of radical initiator, 4,4′-Azobis(4-cyanovaleric acid), and **(c)** representative examples of CTAs for glycopolymer synthesis *via* RAFT.[53, 54]

Well-defined poly(acryloyl glucosamine) (PAGA) with low PDI (1.1 ≤ PDI ≤ 1.3) was prepared by RAFT in aqueous media without hydroxyl protection on the glycomonomers (Figure 9).[54] The PAGA was then extended with a second monomer, N-isopropyl acrylamide (NIPAM), to form a block copolymer, PAGA-*b*-PNIPAM, as shown in Figure 9a.[54] Additionally, shown in Figure 9b, a 3-armed glycopolymer was also synthesized with the same monomer using a trifuncional RAFT CTA.[54]

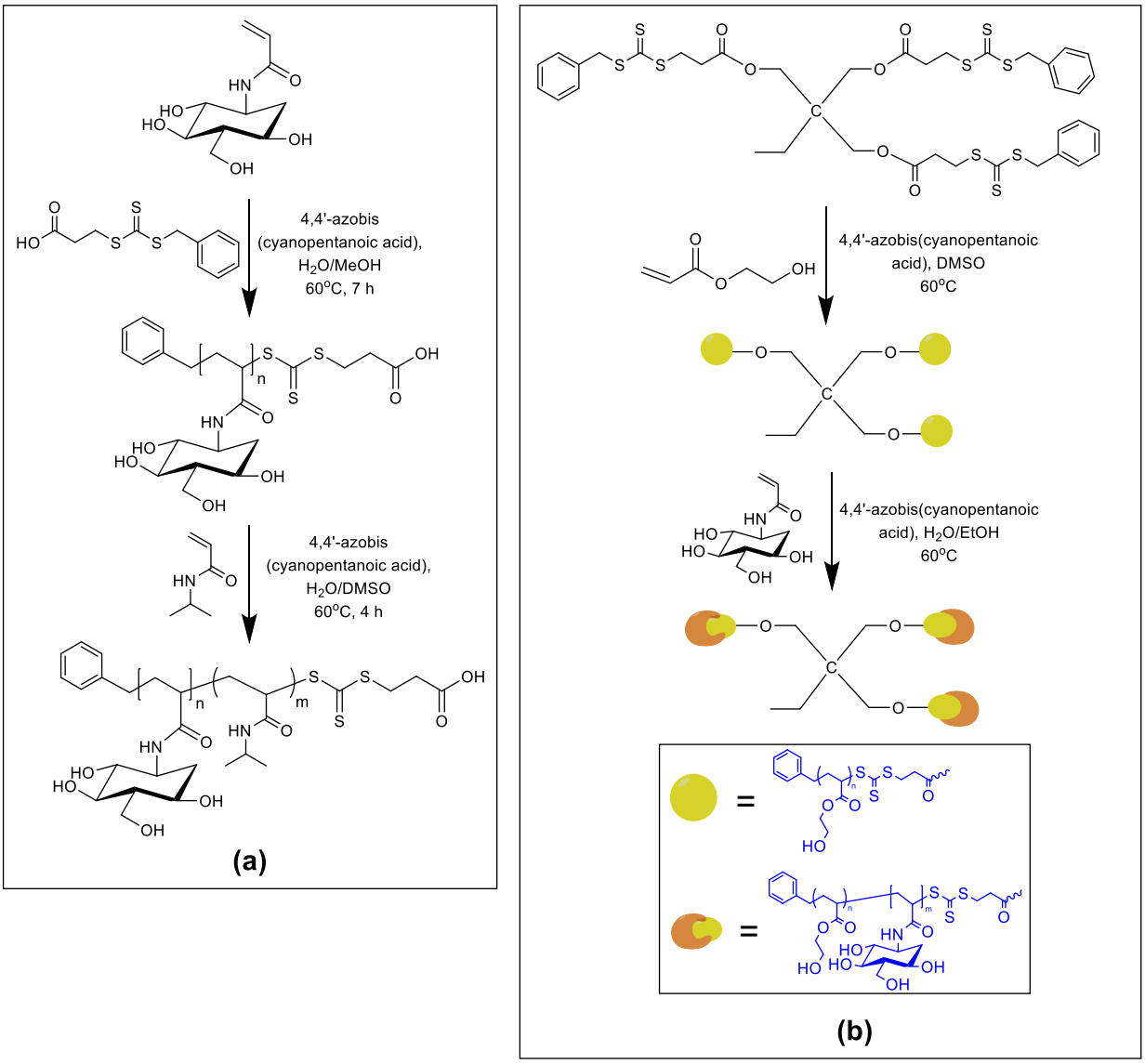


Figure 9. RAFT polymerizations of **(a)** linear glycopolymer PAGA-b-PNIPAM and **(b)** non-linear 3-armed glycopolymer.[54]

RAFT can also be used to perform the graft-from copolymerization (surface initiated polymerization) method so that the polymer can modify a target surface. Herein, the graft-from copolymerization means a monomer can be polymerized from the initiating sites on the backbone polymer which results in a high density of polymer grafts on the backbone polymer (or a surface).[55] By using a similar surface modification and following RAFT, Figure 10 illustrates the graft-from of glycopolymer, poly(acryloyl glucosamine) (PAGA), on the CTA integrated silicon wafer using RAFT polymerization *via* Z-group approach.[56]

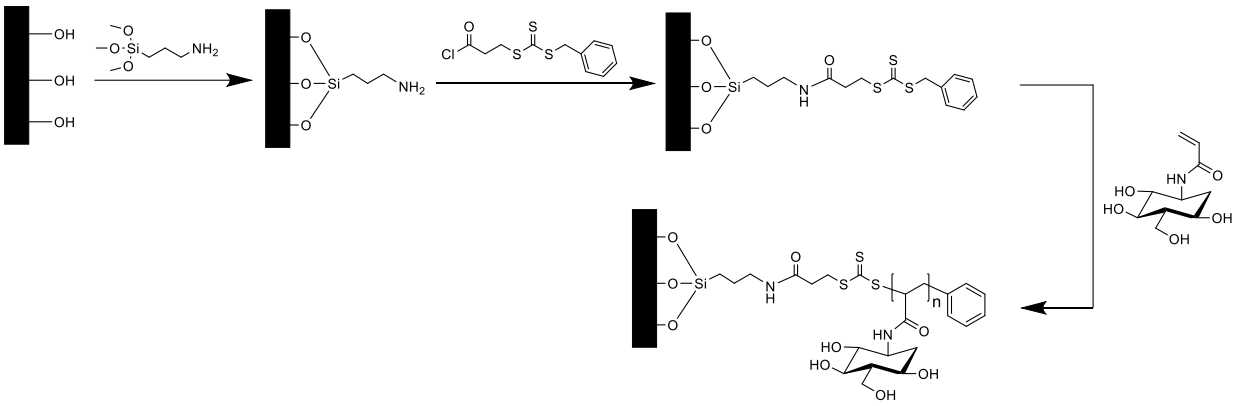


Figure 10. Synthesis of poly(acryloyl glucosamine) (PAGA) grafts on a silicon wafer surface using RAFT *via* Z-group approach.[54]

Nitroxide-mediated polymerization (NMP). NMP employs nitroxides or their associated alkylated derivatives, such as alkoxyamines as an initiator.[57] The use of NMP for polymerization of glycopolymers is not common, because other modern CRP techniques, such as ATRP and RAFT, are much more convenient to conduct and well-studied in general. In this section, we discussed few examples of glycopolymers preparation *via* NMP.

In 1998, Fukuda *et al*. synthesized the first glycopolymer *via* NMP from protected and unprotected monomers, to form poly(N-(*p*-vinylbenzyl)-[*O*-*β*-D-galactopyranosyl-(1→4)]-D-gluconamide), shown in Figure 11.[58] The acetyl protected glycomonomer displayed higher control during polymerization than an unprotected glycomonomer.

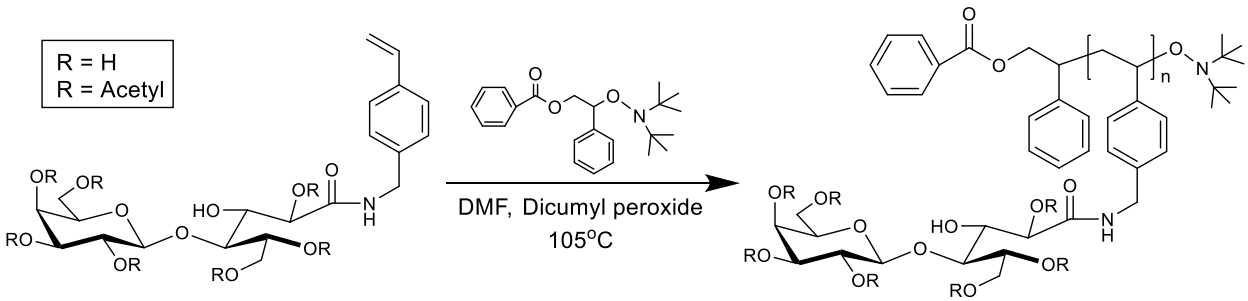


Figure 11. Synthesis of protected (and unprotected) poly(N-(*p*-vinylbenzyl)-[*O*-*β*-D-galactopyranosyl-(1→4)]-D-gluconamide) *via* NMP.[58]

Additionally, a well-defined glycopolymer, poly(2-(2′,3′,4′,6′-tetra-*O*-acetyl-*β*-D-galactosyloxy)ethyl methacrylate-*co*-styrene) (poly(AcGalEMA-*co*-S)), was successfully prepared by NMP using N-tert-butyl-N-(1-diethylphosphono-2,2-dimethylpropyl) (BlocBuilder^{TM}) as the NMP agent (Figure 12).[59] The introduction of a hydrophobic styrene block on the glycopolymer produced an amphiphilic polymer which can undergo self-assembly into

micelles.[59] It is also proven that the polymerization conditions and deacetylation (deprotection) of poly(AcGalEMA-*co*-S) did not have a negative affect the biofunctionality of the copolymer when it tested with a lectin, peanut agglutinin (PNA).[59]

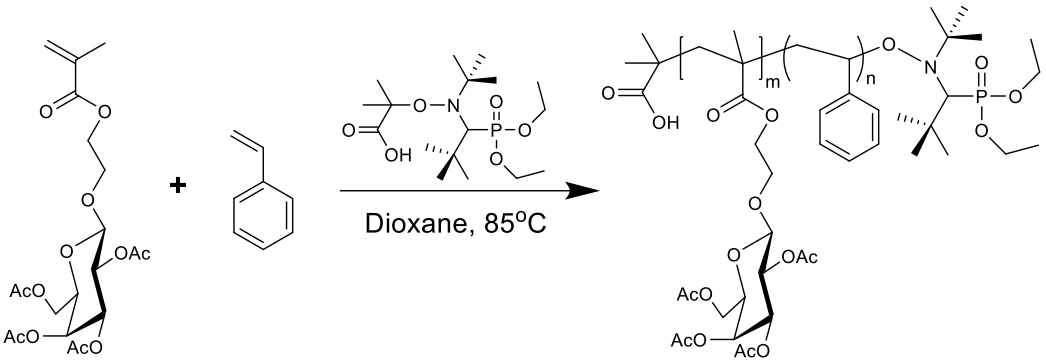


Figure 12. Synthesis of glycopolymer, poly(2-(2′,3′,4′,6′-tetra-*O*-acetyl-*β*-D-galactosyloxy)ethyl methacrylate-*co*-styrene) (poly(AcGalEMA-*co*-S)) *via* NMP.[59]

2.3. Glycopolymer synthesis via post-polymerization modification

Besides direct polymerization of carbohydrate containing monomers, glycopolymers have also been synthesized through post-polymerization modification technique.[60-66] The post-polymerization modification can convert a polymer to possess a broad range of side-chain functionalities *via* modification reactions.[67] In particular, the post-polymerization modification is an efficient method to prepare functional polymers that cannot be directly polymerized from monomers due to poor functional group tolerance of the polymerization.

Post-polymerization modification through azide-alkyne "click chemistry". A α-mannopyranoside-containing glycopolymer was prepared by post-polymerization modification approach via copper-catalyzed azide-alkyne cycloaddition (Figure 13a).[60] The maleimide-terminated glycopolymer is selectively bound to cysteine (Cys34) protein of bovine serum albumin (BSA) (Figure 13b). The conjugate of protein-glycopolymer offers an easy pathway of defined biological molecules which is promising in the development of new generation of therapeutic agents.[60]

Chen and coworkers reported a one pot synthesis, which performs RAFT polymerization and "click reaction" simultaneously, of glycopolymer (Figure 14a). The glycopolymer was then reduced to form thiol-terminated glycopolymer (Figure 14b) which was subsequently grafted on the surface of gold nanorods by taking advantage of Au-S interaction (Figure 14c).[62] The glycopolymer-coated gold nanorods showed robust and selective recognition with lectin. There was no evidence that triazole moiety negatively impact the lectin-binding property.[62]

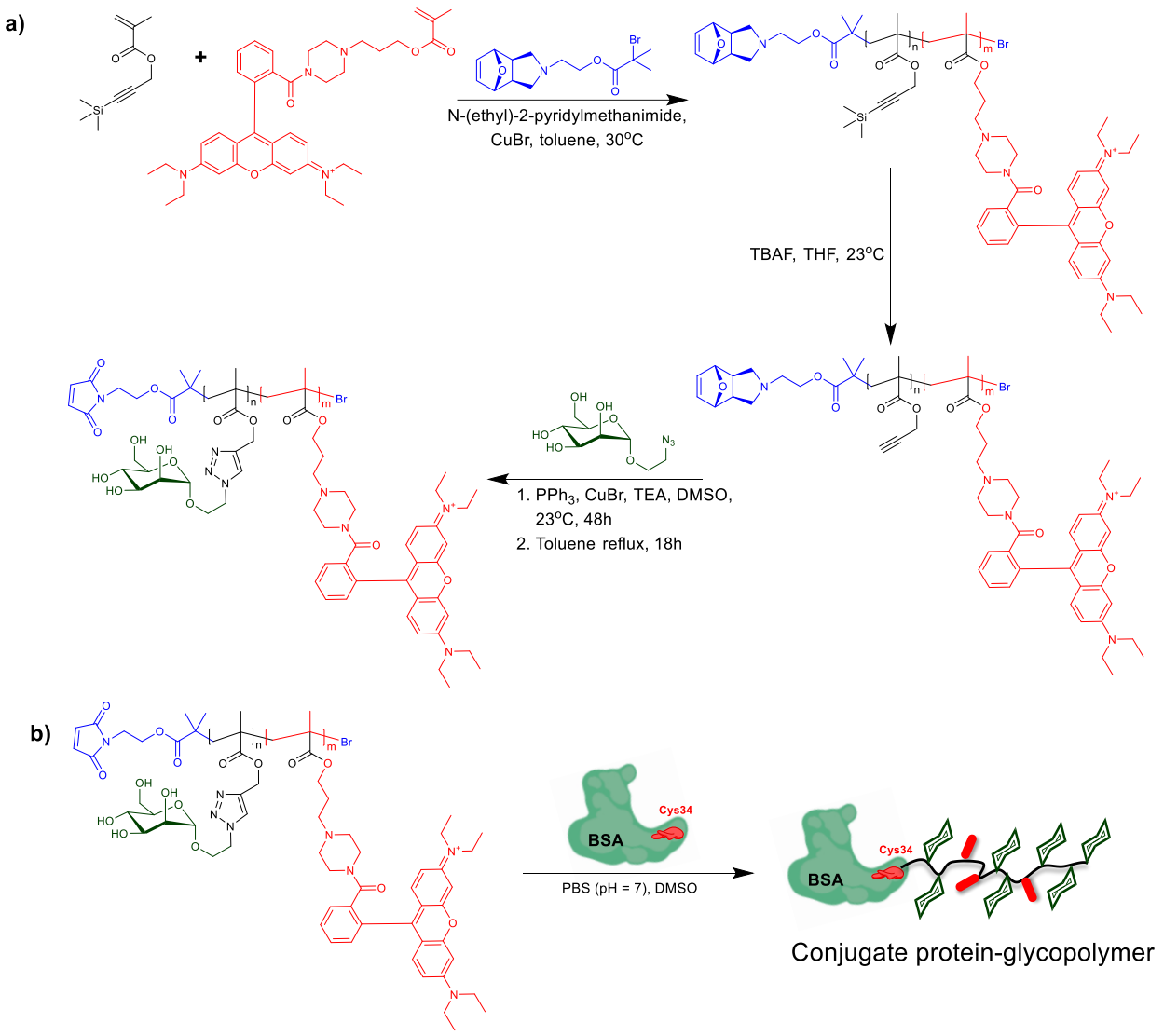


Figure 13. Synthesis of **(a)** α-mannopyranoside-containing glycopolymer *via* post-polymerization modification using copper catalyzed click chemistry, and **(b)** facile and selective formation of protein-glycopolymer conjugate through maleimide and cys34 protein of BSA.[60]

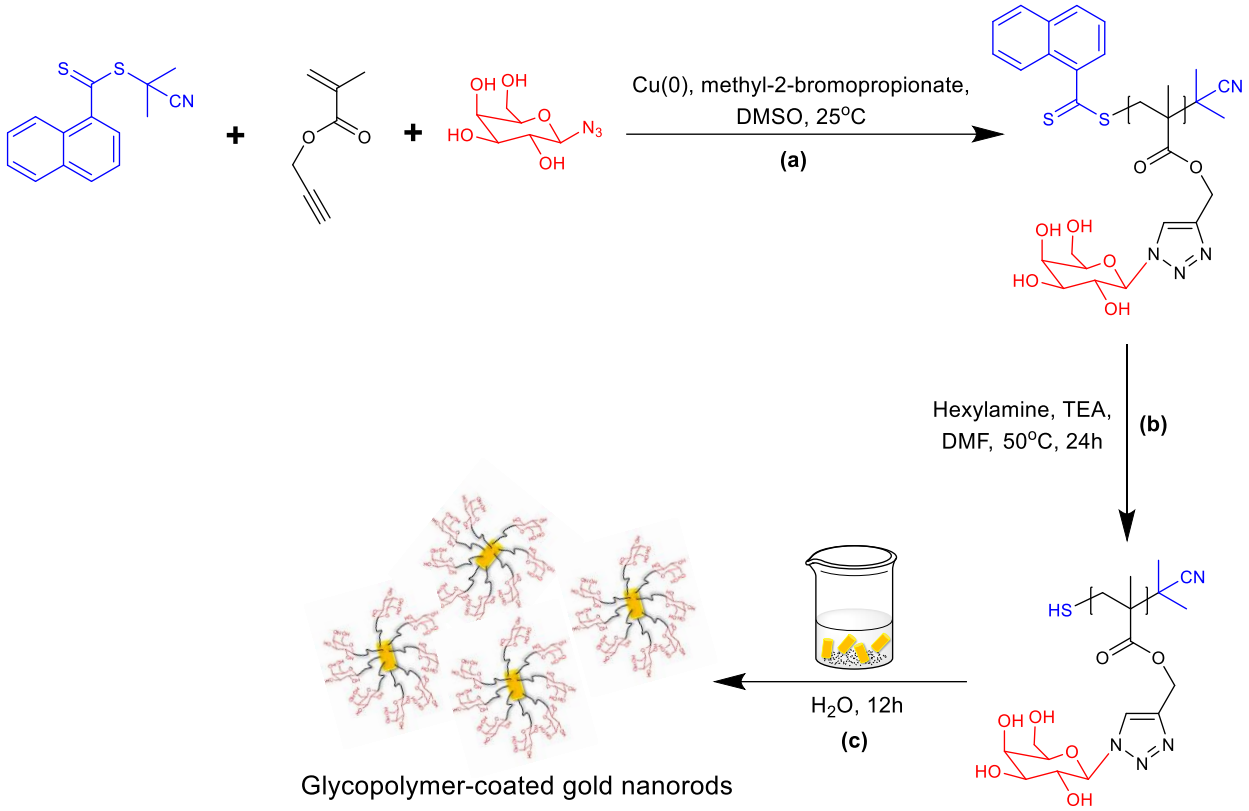


Figure 14. (a) One pot reaction combining RAFT polymerization and "click chemistry", **(b)** post-polymerization reduction to form thiol-terminated glycopolymer, and (c) glycopolymer-coated gold nanorods.[62]

Glycopolymer can also be prepared by ring opening of glycidyl functional group followed by copper-catalyzed azide-alkyne cycloaddition. Jiang *et al*. successfully synthesized various glucose pendant containing polymers which self-assembled in an aqueous solution with polystyrene (PS) as the inert hydrophobic core and glycopolymer (PR) as the shell.[63] The synthesis consisted of multi-step reactions (Figure 15a). The first step was RAFT to create an amphiphilic block copolymer, polystyrene-*block*-poly(glycidyl methacrylate), followed by reduction to form thiol chain terminal for attachment of fluorescein isothiocyanate (FITC). Azide-containing glycopolymer was prepared by simultaneous ring opening of glycidyl group and azide substitution. The azide of the synthesized polymer underwent copper-catalyzed azide-alkyne cycloaddition with alkynated-glucose molecules to form the desired block copolymer (Figure 15a).[63] In an aqueous solution, the glycopolymer self-assembled with a diameter of 34-36 nm as shown in Figure 15b. The self-assembled glycopolymers were observed at macrophages both in vitro and in vivo. Based

upon this behavior, the new glycopolymer is expected to be used in targeting therapeutic application.[63]

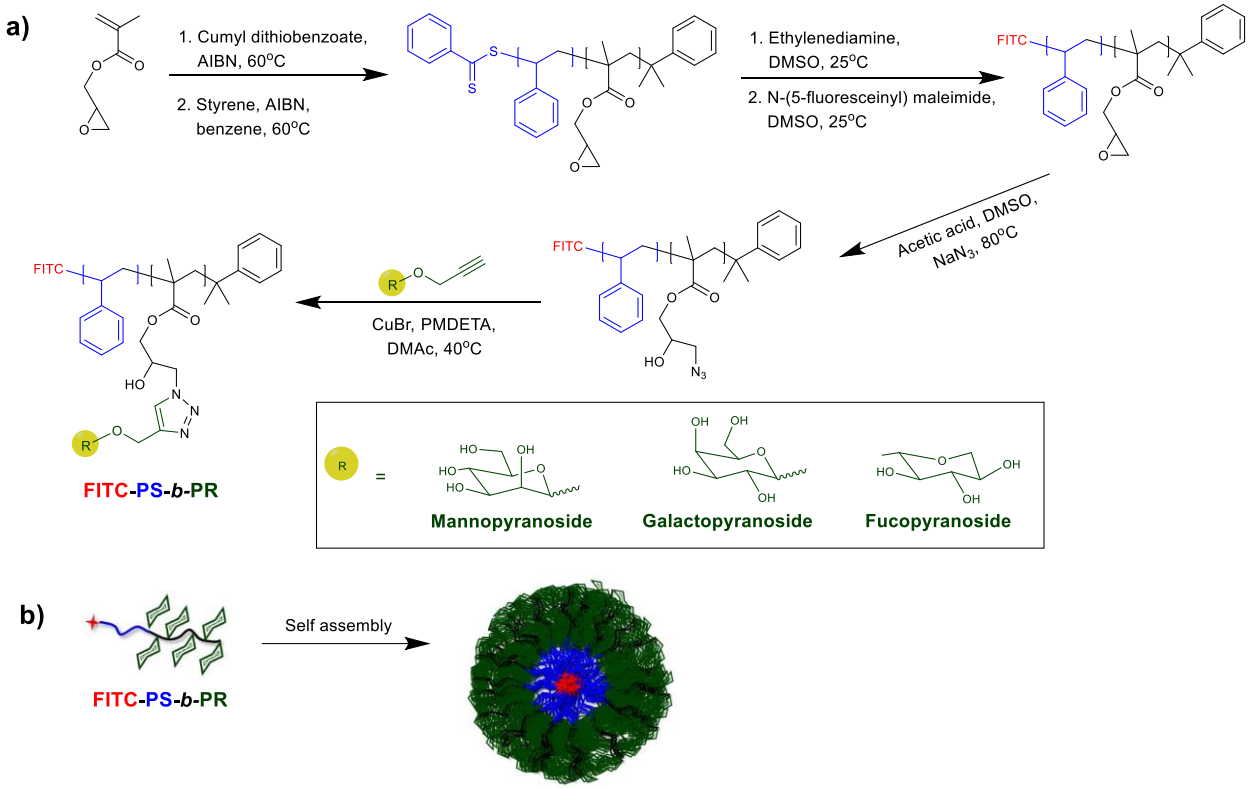


Figure 15. (a) Synthesis of fluorescent-labelled amphiphilic glycopolymers, FITC-PS-*b*-PR, and **(b)** Self-assembly of glycopolymer containing block copolymer in an aqueous solution.[63]

Post-polymerization modification through thiol-ene "click chemistry". In addition to the frequently used "click chemistry", copper-catalyzed azide-alkyne cycloaddition, thiol-ene reaction has also been widely reported as a post-polymerization modification to prepare functional glycopolymers.[61, 65, 66] Thiol-ene reaction is highly efficient reaction and has high tolerance against various functional groups. Moreover, the coupling reaction occurs in metal-free condition which is an attractive feature for biomedical applications.[68] Finally, sulfide linkage (thioether linkage, R-S-R′) in protein-sulfide-oligosaccharide has been reported to have high flexibility.[69]

Stenzel *et al.* have synthesized a block copolymer containing glucose pendant *via* thiol-ene click chemistry (Figure 16).[61] The polymer showed an unique thermo-responsive behavior with a lower critical solution temperature (LCST) of 29 °C.[61] This route opens a new non-toxic (metal-free) pathway of developing sophisticated glycosylated macromolecular architecture which can be beneficial for advancing targeted drug delivery system.[61]

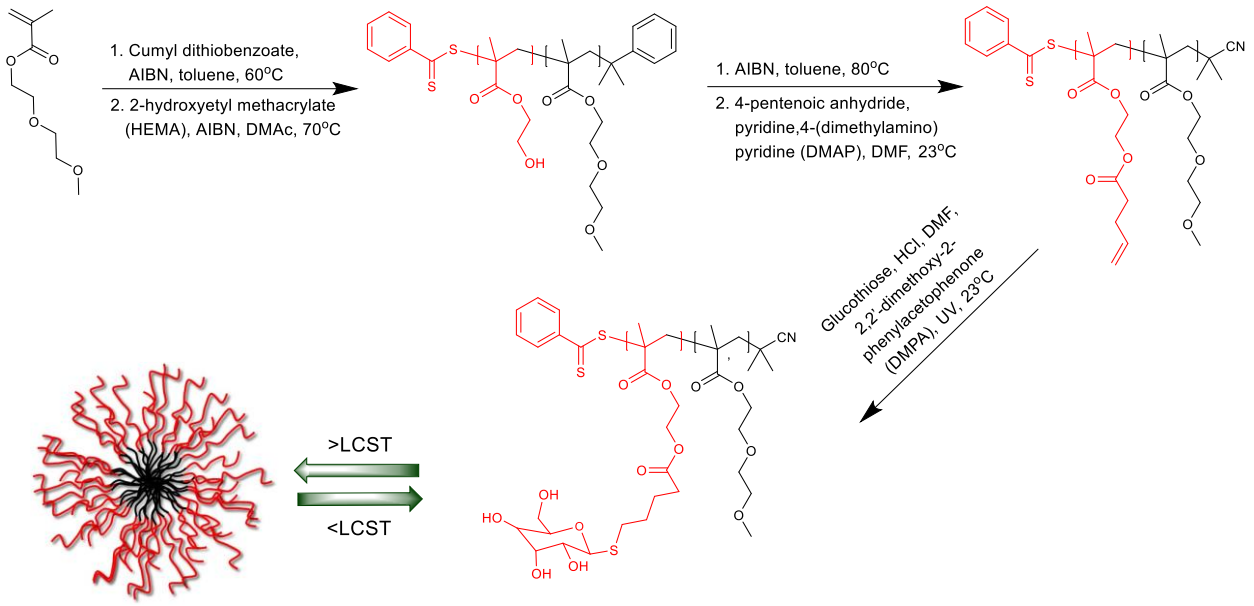


Figure 16. Synthesis of poly(di(ethylene glycol) methyl ether methacrylate)-block-poly(2-hydroxyethyl methacrylate) and post-polymerization modification of thermo-responsive glycopolymer *via* thiol-ene "click chemistry".[61]

Microarray technology plays an important role for inspecting carbohydrate-protein interactions due to its large number of glucose-pendants generated with variability in size and composition.[70, 71] Microarrays using an inject fabrication technique from well-defined glycopolymer achieved by post-polymerization modification of poly(allyl glycidyl ether) (PAGE) was reported.[66] The fabrication technique produced a highly dense and broad range of glucose epitopes in a rapid manner on a single glass chip.[66] In this work, PAGE was used as a main structure which is suitable for various post-polymerization reactions through pendant alkene moieties. Additionally, the poly(ethylene glycol) backbone of PAGE shows high hydrophilicity that enables introduction of highly dense glucose-pendants on a polymer backbone.[66, 72, 73]

A surface modification of glass surface (substrate) with isocyanate was performed to allow covalent immobilization of PAGE on the substrate. The surface was further modified by thiol-ene reaction with thiolated-glucose molecules, such as α-D-mannose, β-D-galactose, and β- D-glucose (Figure 17).[66] This microarray was used for investigating selective glucose binding to lectin using fluorescence technique. The high-throughput analysis is essential for elaboration of cell-surface interactions, adhesion mechanisms, as well as multivalent inhibitors development.[66]

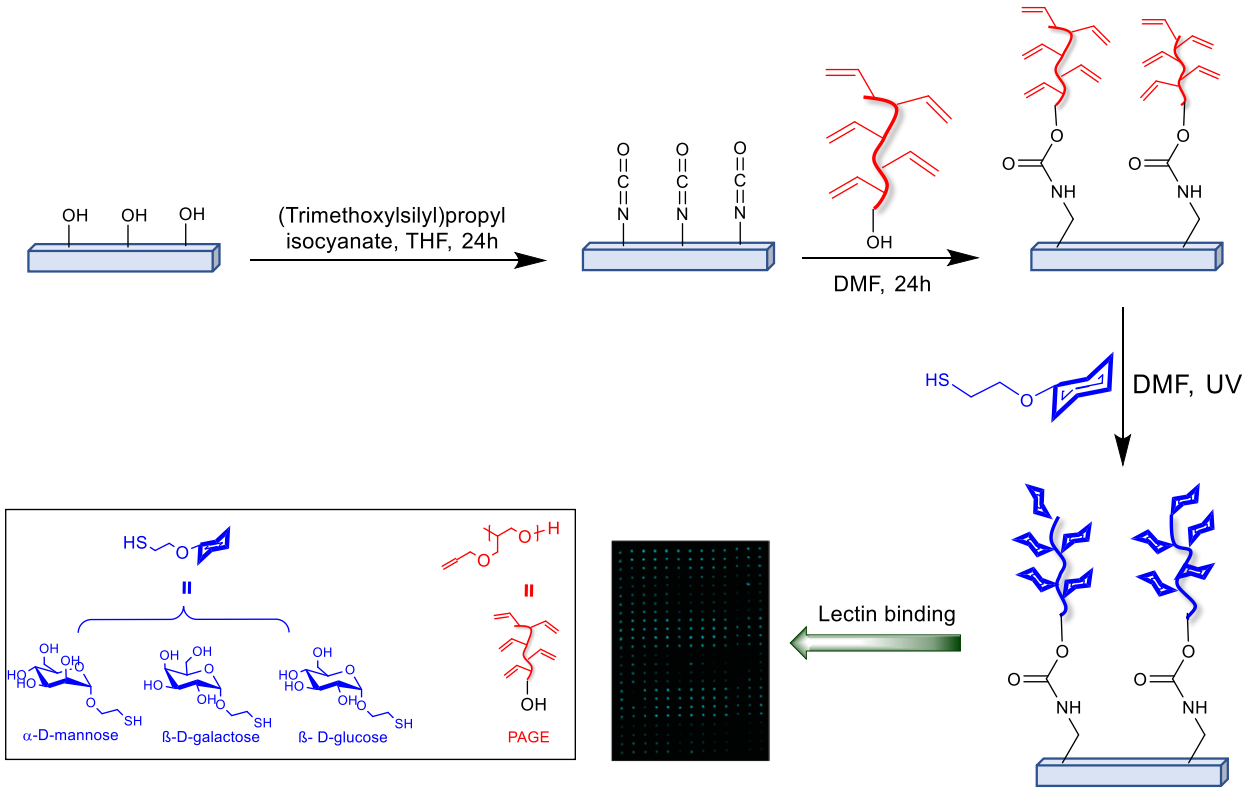


Figure 17. Preparation of glycopolymer-microarray utilizing thiol-ene "click reaction" for high-throughput analysis of carbohydrate-lectin binding properties.[66] Copyright 2017, American Chemical Society.

Another example of thiol-ene "click chemistry" in glycopolymer synthesis was reported by Maynard *et al.*[65] A copolymer, poly(5,6-benzo-2-methylene-1,3-dioxepane-*co*-but-3-enyl methacrylate) [poly(BMDO-*co*-bMA)], was first prepared by RAFT with a trithiocarbonate CTA. The protected thiolated-trehalose underwent thiol-ene reaction to yield poly(BMDO-*co*-bMA) followed by deprotection side chain of glycopolymer, poly(BMDO-*co*-bMA-trehalose) (Figure 18).[65]

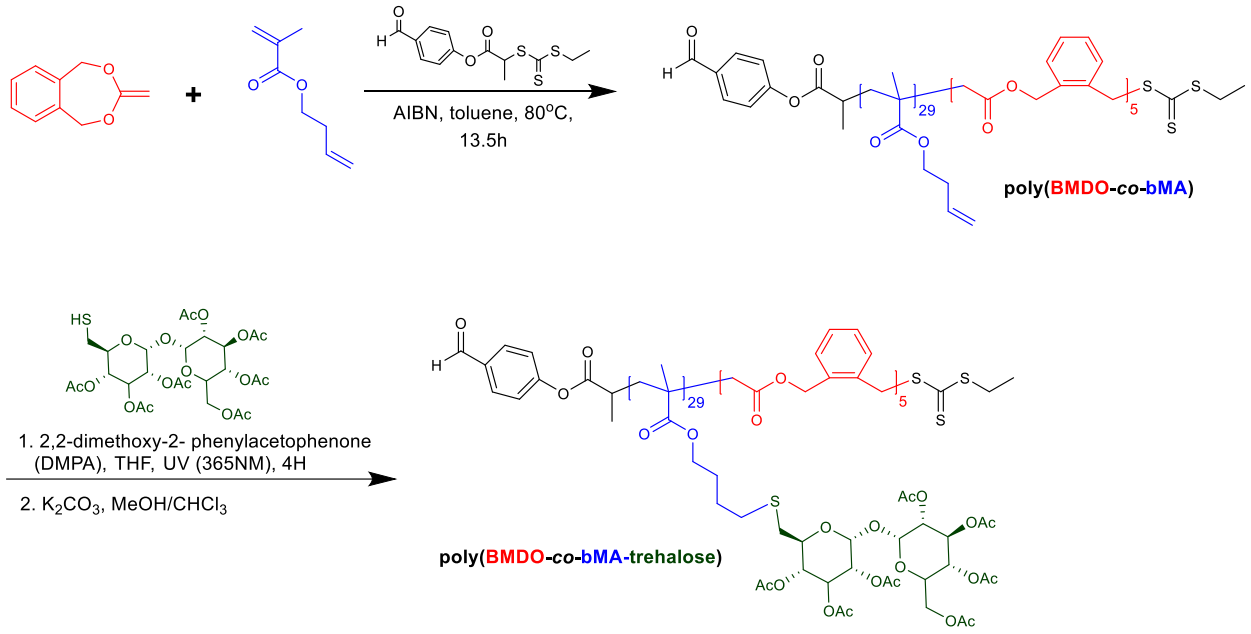


Figure 18. Preparation of degradable trehalose-side chain glycopolymer, poly(BMDO-*co*-bMA-trehalose), *via* RAFT followed by thiol-ene "click" reaction.

3. Applications of glycopolymers

3.1. Fundamental study of carbohydrate-lectin interactions

Carbohydrate-lectin interaction is the most studied field of application for synthetic glycopolymers. By definition, lectins are proteins that bind to carbohydrates with high affinity and specificity.[74] Because glycopolymers are synthetic polymers that contain carbohydrate pendants, the glycopolymer is known to have the ability to mimic natural polysaccharides during biological recognition events[75-80] due to their binding affinity to lectins (Figure 19).

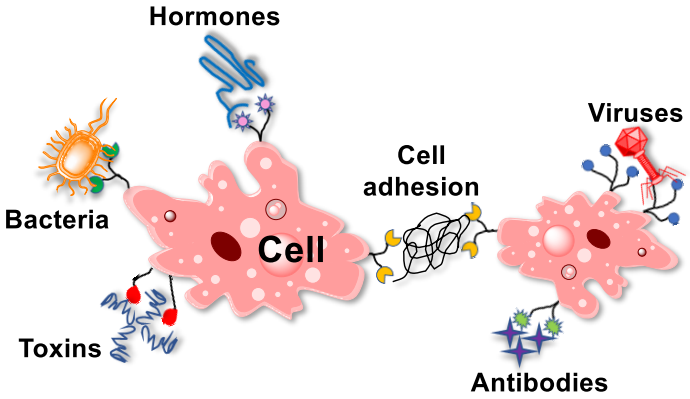


Figure 19. Illustration of various biological recognitions that use lectin's high and specific affinity for carbohydrates; examples include affinities in bacteria, toxins, hormones, antibodies, viruses, and cell-cell adhesion.[75-80] The figure is imaginary to describe multiple affinities in a single illustration, not a real.

Concanavalin A or Con A is the most widely used plant lectin found in *Canavalia ensiformis* or commonly known as jack bean. Con A has been used to study multivalent binding of glycopolymers that contains mannose and glucose pendants.[61] Another lectin from the plant legume family is PNA which is extracted from *Arachis hypogaea*. PNA specifically binds to galactose pendants on glycopolymers.[61] Additionally, WGA from *Triticum vulgare* is also commonly used to study lectin binding to glycopolymers. WGA binds selectively to *N*-acetylglucosamine and *N*-acetylneuraminic acid (a sialic acid).[31] Unlike plant lectins, animal lectins are more complex structurally, and they can be generally classified in five main types: C-type, I-type, P-type, S-type (galectins) lectins, and pentraxins according to their structural features.[31] Animal lectins are crucial to biological functions such as recognition molecules within the immune system, regulation of cellular growth, cell-cell interactions, and extracellular molecular bridging.[32] Because of the selective binding affinity of Con A to glucose molecules, Con A is very crucial for the investigation of carbohydrate-protein interactions that occurs on cell surface.[32, 61]

Unique interactions (lock-and-key metaphors) between monosaccharide and lectin are normally weak. The simplest strategy to enhance overall binding strength is to increase the number of binding sites.[81] Because glycopolymers contains many carbohydrate pendants, the study of lectin binding affinity to glycopolymers is an important field to research to further biological applications. Kiessling and co-workers reported 7-oxanorbornene pendant which comprised of linear glycopolymers that are prepared by ring opening metathesis polymerization (ROMP). The glycopolymer was investigated by utilizing agglutination inhibition assay with regard to their multivalent effects on Con A binding efficiency.[82] In this report, it was revealed that the density of saccharides is an important control factor for carbohydrate-lectin interactions. By decreasing the density of carbohydrate pendants on an aliphatic backbone, the binding activity of a glycopolymer towards bulky Con A increased due to less steric hindrance (Figure 20).[83]

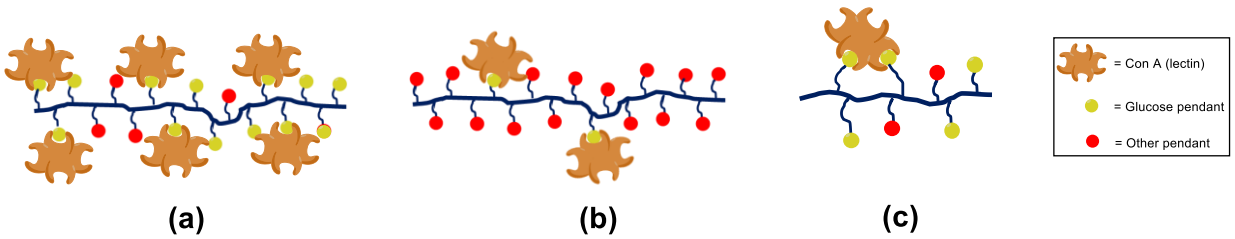


Figure 20. Illustration of Con A binding on multivalent ligands **(a)** High density carbohydrate pendants shown in the high density glycopolymer which allows many receptors to attach to a single molecule. However, the attachment is limited by the steric hindrance which results in unused molecules, **(b)** Low density carbohydrate pendants; low density glycopolymer allows fewer total receptors per molecule. The space between carbohydrates on the glycopolymer is too large to bind efficiently.[83] **(c)** Optimum binding activity occurs when the distance between two glucose pendants is equivalent with the distance between two binding sites of a lectin.[34]

Generally, glycopolymers offer enhanced binding activity compared to single glucose ligand due to increase in available binding sites. Therefore, optimum distance-dependent binding can be achieved if the distance between two carbohydrates is equivalent to the distance between two binding sites on a lectin.[34] For the optimum distance-dependent binding, the glycopolymer must be flexible. If the glycopolymer is stiff, the exact binding does not occur.[34] Other parameters of efficient lectin-glycopolymer binding are molecular weights and spacers between the glucose pendant and the polymer backbone. Kiessling *et al.* discovered that an increase in molecular weight of the glycopolymer provides an increase in the binding capability to Con A.[84] Additionally, flexible spacers between glucose pendants and the polymer backbone enhances multivalent binding because the distance between binding sites can be adequately adjusted for an optimal binding.[85]

In addition to the linear homoglycopolymer, more complex architectures of glycopolymers have been reported. Figure 21 exhibits an example of an amphiphilic block copolymer possessing a hydrophilic glycopolymer block and a hydrophobic polymer block. The amphiphilic feature allows for structural diversities such as micelles,[59, 86-88] vesicles,[86, 87] helix, and worm-like aggregates.[87] The non-linear polymer architecture results in an increase of surface area of glycopolymers thus, enhancing the lectin-glycopolymer binding probability.[32]

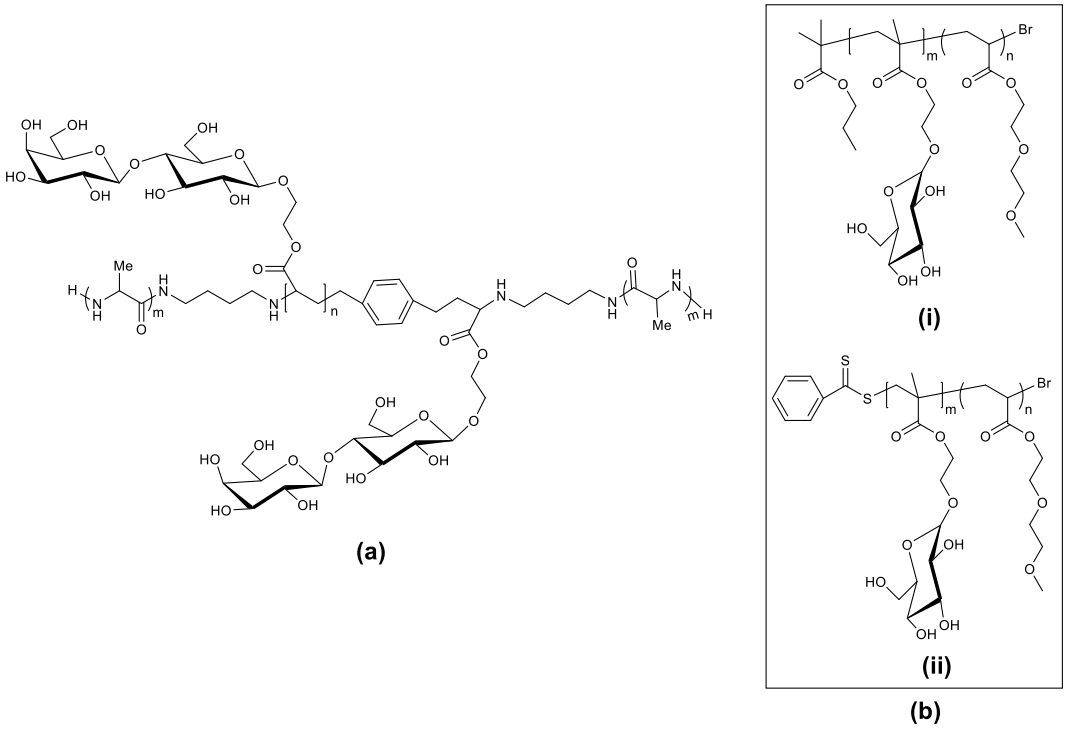


Figure 21. (a) poly[(L-alanine)-*b*-poly(2-acryloyloxyethyllactoside)-*b*-poly(L-alanine)] which exhibits micelle-like structure in solution,[88] and **(b)** poly[(2-glucosyloxyethyl methacrylate)-*b*-poly(diethyleneglycolmethacrylate)],which exhibits a vesicle-like structure in solution. Those glycopolymers are prepared by ATRP and RAFT.[89]

Glycopolymers have also been integrated to metallic and organic nanoparticles prior to the glycopolymer-lectin interaction. Gaojian *et al.* reported two synthetic approaches of producing glucose modified nanoparticles. Figure 22a shows introduction of monosaccharides on a nanoparticle *via* click-chemistry. The second example is surface initiated ATRP, conducted to have glycopolymers on a nanoparticle (Figure 122b).[90] Both modified nanoparticles show efficient recognition to Con A lectin (Figure 22c).[90]

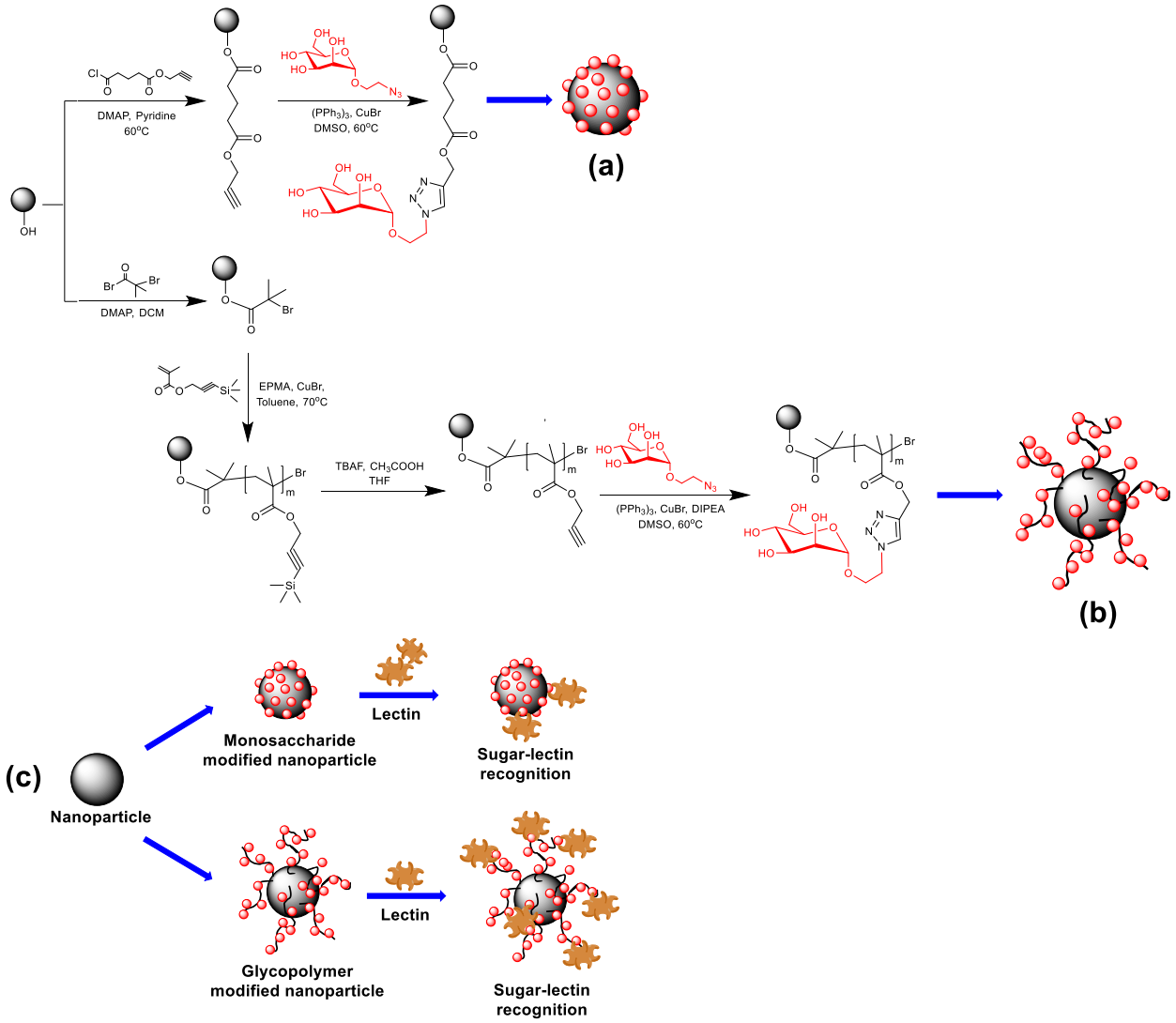


Figure 22. (a) monosaccharide-modified nanoparticle *via* click-chemistry, **(b)** glycopolymer-modified nanoparticle *via* ATRP, and **(c)** sugar-lectin recognition on the modified nanoparticles.[90]

3.2. Carbohydrate to cell, bacteria, and virus interactions

Based upon strong and specific affinity of carbohydrates to lectins, the interaction of glycopolymers to lectin containing cells and microorganisms (bacteria and virus) have been studied. The study of the glycopolymer interaction has led to a precise understanding of microorganisms' infection mechanisms to the host. The first stage of infection is physical attachment of microorganisms on a cell.[34] For example, the influenza virus attaches to the surface of a bronchial epithelial cell as illustrated in Figure 23.[76, 91] The influenza virus membrane contains a highly dense (2-4/100 nm^2) glycoprotein, *i.e.* hemagglutinin.[76, 92] The counterpart, bronchial epithelial cell membrane consists of densely (50-200/nm^2) packed of glycoprotein with a N-

acetylneuraminic acid (sialic acid) terminal sugar. Multiple attachments of hemagglutinin-sialic acid initiate the infection of the cells.[76] Therefore, precise control of microorganisms-human cell interaction can be important in decreasing human cells' susceptibility to infectious diseases.

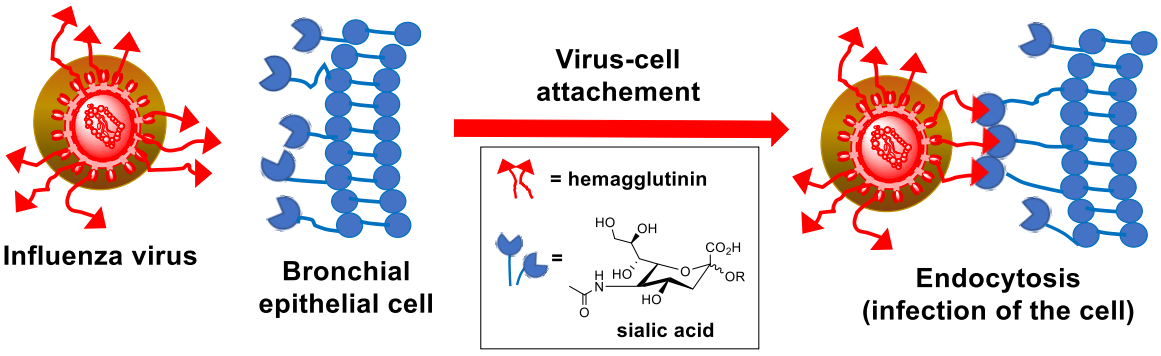


Figure 23. Attachment of influenza virus to bronchial epithelial cell *via* interaction of hemagglutinin of the virus to sialic acid on the cell;[76, 91] the described bronchial epithelial cell is a simplified figure showing part of outer surface of the cell.

Hardy *et al.* reported glycopolymer functionalized spider silk mimics enhance cell adhesion.[77] Alkyne-capped-poly(-6-O-methacryloyl-D-galactopyranose) (PMAGal) was synthesized *via* ATRP (Figure 24a).[77] The engineered spider silk (eADF4(C16)), in Figure 24b, was modified with alkyne-capped-PMAGal at various degree of polymerizations (Degree of polymerization = 31, 64, and 97). Fibroblast cell line (M-MSV-BALB/3T3, mouse embryo fibroblast) was used to study cell adhesion. Based on this study, functionalization of spider silk substrates with PMAGal enhanced the attachment of fibroblast on the substrates (cell surface area of 310-390 μm^2) compared to unmodified spider silk substrate (cell surface area of 210 μm^2) (Figure 24c).[77]

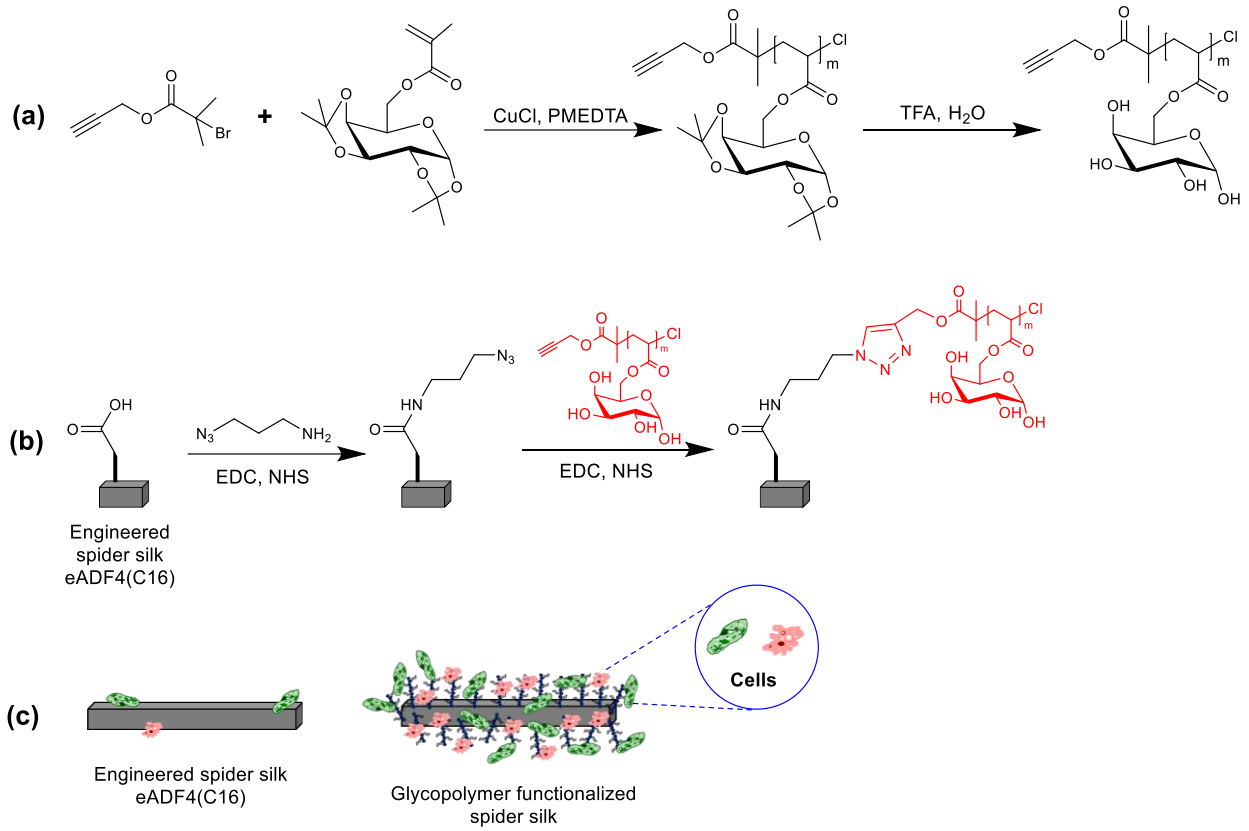


Figure 24. (a) Synthesis of alkyne-capped PMAGal *via* ATRP, **(b)** PMAGal surface-functionalized spider silk eADF4(C16) *via* click chemistry, and **(c)** cell attachment comparison between spider silk (control) and glycopolymer-modified spider silk.[77]

Lees *et al*. reported the interaction between a virus and the glycoprotein of cells can be suppressed by three approaches: (i) utilizing ligand to compete with sialic acid on the cell wall so the hemagglutinin binding sites on the virion are sterically blocked, (ii) modification of sialic acid groups so the cells binding sites are not accessible for the hemagglutinin of the virus, and (iii) blocking hemagglutinin of the virus which only allows them to access the sialic acid on a cell.[76] However, blocking all sialic acid moieties of the cells is impractical due to possibility of interfering with normal cell functions. In this work, a copolymer, poly(acrylamide-*co*-*O*-glycoside) (Figure 25a), was investigated to understand the function of hemagglutinin inhibition through cell binding site blocking *via* capping and/or steric inhibition (Figure 25b).[76]

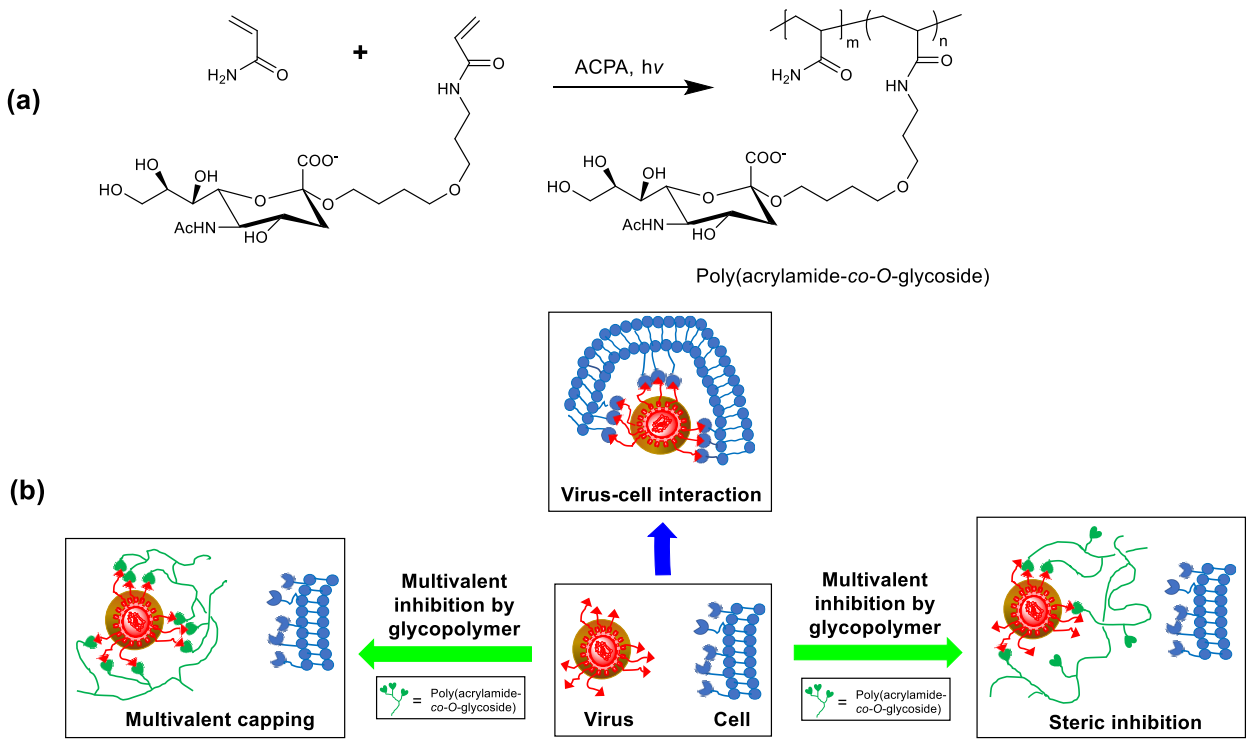


Figure 25. (a) Synthesis of poly(acrylamide-*co*-*O*-glycoside), and **(b)** multivalent inhibition of hemagglutinin of virion by glycopolymer.[76]

Glycopolymers can be an anti-bacteria-adhesion agent. Similar to that of a viral infection, bacteria adhesion to a cell is the initial stage of a bacterial infection. For instance, an overgrowth population of *Escherichia coli (E. coli)* causes many infections such as urinary tract and bladder infection, meningitis, and bowel diseases which often require antibiotic treatment. *E. coli* carries hair-like organelles, type 1 piliated fimbriae, on their cell wall.[93] The type 1 pili contains fibrillar short-tip structure bearing FimG and FimH adhesion which is responsible for binding to mannose units of oligosaccharides.[93] A n-Heptyl α-D-mannose (HM)-based glycopolymer was studied as a FimH adhesion inhibitor to reduce a virulence factor of *E. coli* (Figure 26).[79] Based on an *in vitro* study, the HM-based glycopolymer showed inhibition of *E. coli* adhesion to intestinal epithelial cells at a low density of mannose in the glycopolymer.[79]

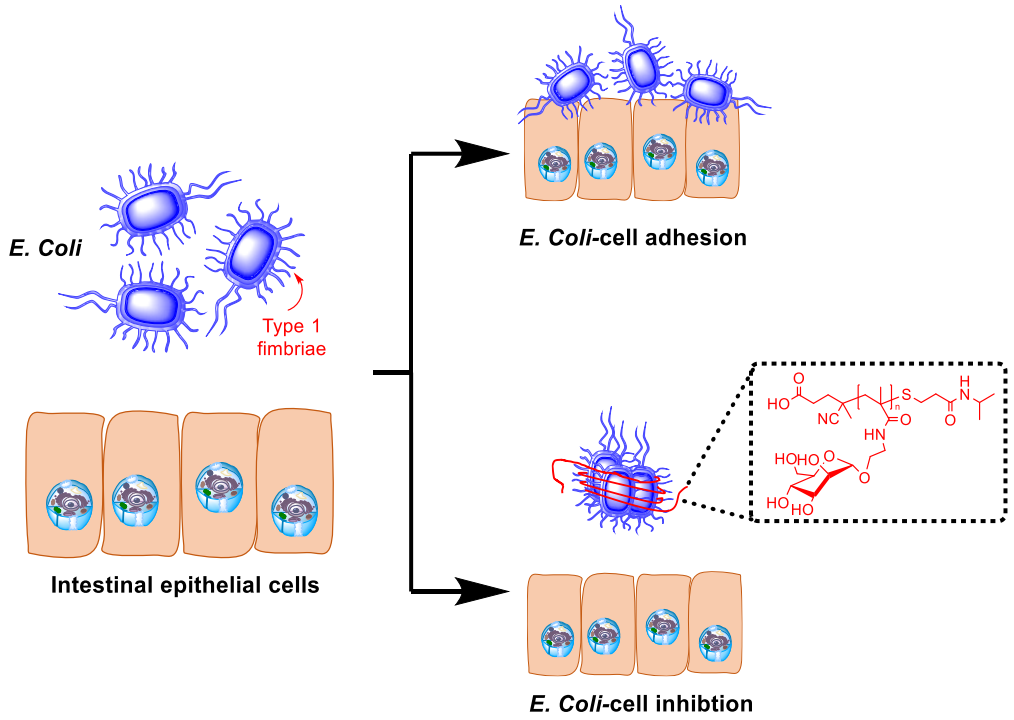


Figure 26. Principle of n-Heptyl α-D-mannose (HM)-based glycopolymer as an anti-bacteria-adhesion agent against *E. Coli* bacteria.[79]

In relatively new study, glycopolymers have also been reported to have a crucial role in immunology.[63, 94] In particular, cancer immunotherapy uses immune system to eliminate cancer cells. This process is designed to allow immune cells attacking target cancer cells specifically.[95, 96] Jiang *et al.* developed an artificial glycocalyx based on self-assembled glyco-nanoparticles (glyco-NPs) that is discussed in Figure 15.[61] Herein, glycocalyx is a highly dense carbohydrate-coating on the surface of cells.[97, 98] The glyco-NPs was able to reverse the immunosuppressive phenotype which impairs antitumor immune response.[61] Then, the enhanced immune-function of macrophase can perform tumor immunotherapy. The reversal of the immunosuppresive phenotype is controlled by macrophage polarization, which is occurred by the synthesized glycopolymer.[99 61]

As an additional example of anti-cancer immunotherapy, two synthetic glycopolymers, poly(N-methacryloylglucosamine) (pMAG) and poly(N-methacryloylmannosamine) (pMAM), have been used to create engineered tumor cell membranes as shwon in Figure 27.[94] In this example, tumor cells membrane were engineered with pMAG and pMAM. pMAG and pMAM functions as binding sites allowing selective recognition to macrophage lectins, such as mannose receptor (MR), and complement receptor three (CR3). The binding, between pMAG-MR and

pMAM-CR3, triggers immune responses which lead elimination of the tumor cells (Fig. 27).[94, 100] These examples of glycopolymer application in immunology would offer strong potential in thev development of immunotherapy for cancer treament.

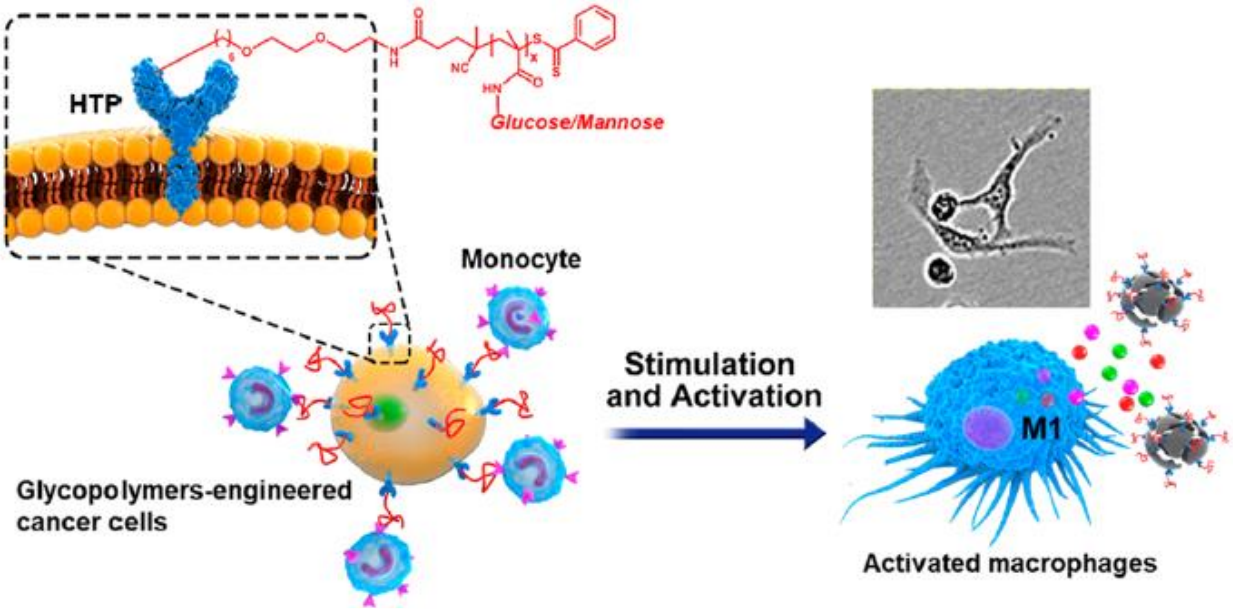


Figure 27. Gycopolymers for cancer immunotherapy: Glycopolymer-bound tumor cells enhanceds tumor cell recognition of macrophages to trigger immune response against tumor cells.[94] Copyright 2019, American Chemical Society.

3.3. Glycopolymer-based adhesives and materials

In previous sections, direct biological interactions of glycopolymers have been described to demonstrate various applications. Glycopolymers have also been recently reported to be utilized in tissue adhesives, sealants[101] and pressure sensitive adhesives which exhibit mechanically elastomeric behavior.[102, 103] The advantage of glycopolymers-based adhesive material include their abundance and sustainability of the natural resources.[103]

The glycopolymer, poly(6-methacryloyl-α-D-galactopyranose) (polyMG), has been introduced into collagen to form an interpenetrated polymer network hydrogel.[101] The hydrogel was prepared by photo-crosslinking of MG with poly(ethylene glycol) diacrylate in the presence of photo-initiator, Irgacure-2959. Herein, the crosslinking and polymerization occurs simultaneously with collagen (Figure 28).[101] The hydrogel was used in corneal application due to its exceptional transparency which resembles human cornea.[101]

For the corneal substitute application, mechanical properties of the hydrogel are very important. By introducing small amount of MG to collagen (MG to collagen of 1:16, 1:8, 1:4, 1:2),

the tensile strength of hydrogel increased significantly from 520 kPa (pure collagen) to 530 kPa (ratio of 1:16), 540 kPa (ratio of 1:8), 570 kPa (ratio of 1:4), and peaking at 720 kPa (ratio of 1:2).[101] However; at ratio of 1:1 (MG to collagen), the tensile strength started decreasing presumably due to over-crosslinking between MG and collagen in hydrogel.[101] Additionally, modulus of hydrogel contained MG also shows similar trend with tensile strength, which was higher with introduction of MG to the collagen (1200 kPa, 1230 kPa, and 1300 kPa for 1:8, 1:4, and 1:2 ratio of MG to collagen) than pure collagen modulus at 700 kPa.[101] No significant change of elongation at break (±35-53%) reported when MG was introduced to collagen which is close to the value of human corneal tissue, 45-75% elongation at break.[101, 104] *In vitro* testing also revealed that corneal epithelial cells count favored the glycopolymer hydrogel over the pure collagen hydrogel. In addition to the excellent optical and biological properties, the synthesized glycopolymer hydrogel prevents bacteria adhesion.[101]

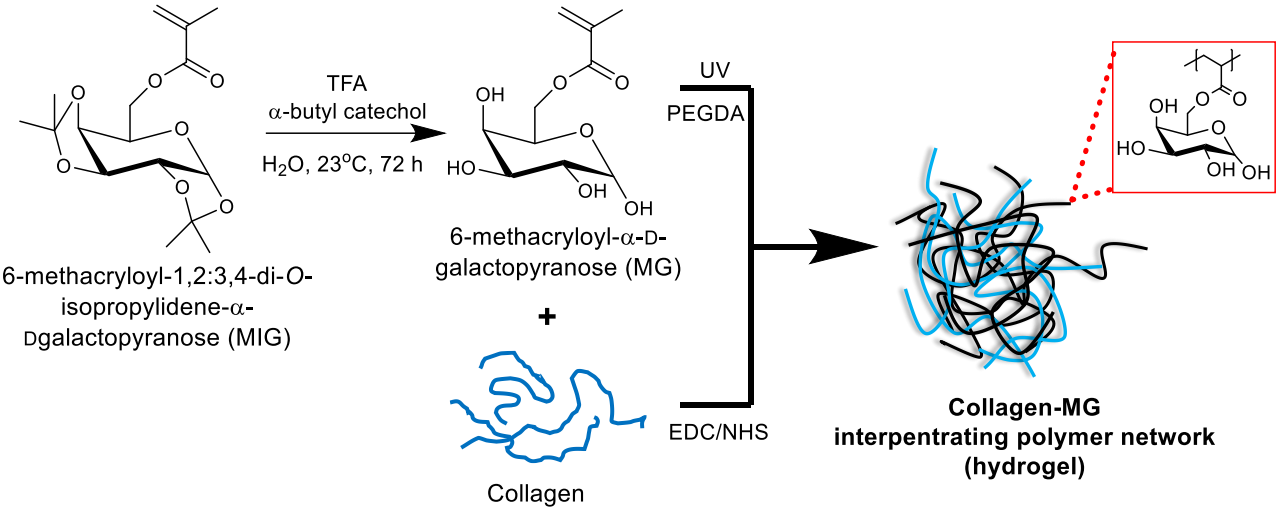
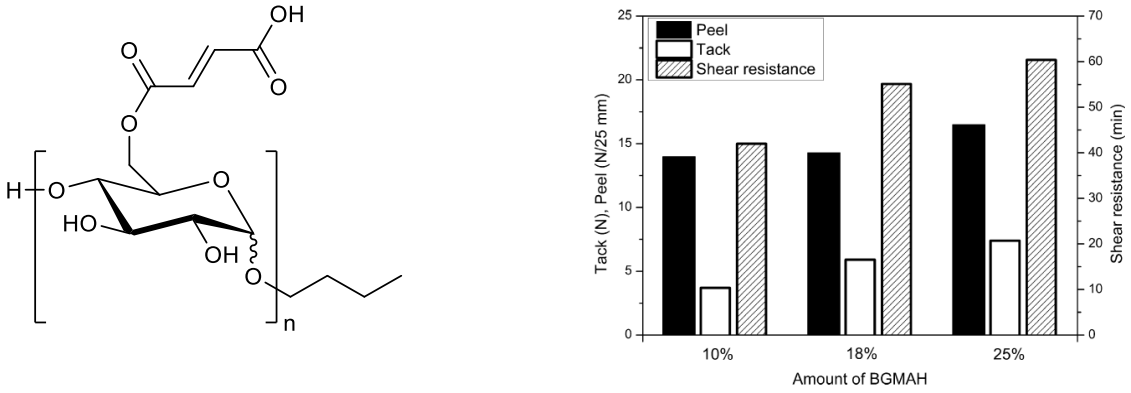


Figure 28. Synthesis of collagen-poly(6-methacryloyl-α-D-galactopyranose) (polyMG) interpenetrating polymer network for hydrogel application.[101]

In 2015, Pokeržnik and Krajnc used a glucose-based surfmer (surfactant and monomer) and butyl polyglucoside maleic acid ester (BGMAH) to prepare poly(n-butyl acrylate) (PnBA) copolymer *via* emulsion polymerization. Due to the -COOH and C=C bonds in the chemical structure (Figure 29a), BGMAH behaves not only as a anionic surfactant but also as a vinyl monomer.[103] Various amounts of BGMAH in an acrylate-based pressure sensitive adhesive was investigated. Increasing the amount of BMGAH up to 25% did not change the elastic moduli (G')

of the polymer. The higher percentage of BMGAH in the copolymer showed the higher adhesion properties (Figure 29b).[103]



(a) **(b)**

Figure 29. (a) Butyl polyglucoside maleic acid ester (BGMAH) structure, and **(b)** Effect of BGMAH amount in the copolymer on peel and tack strength.[103]

An important design principle of thermoplastic elastomers is ABA-type triblock copolymer that contains both soft/rubbery segment (B block) and glassy segment (A block) in ABA-type triblock copolymers. The two glassy segments A allows the entire polymer to resist the flow resulting in an elastomeric property.[105] To achieve this elastomeric behavior, Nasiri and Reineke developed an ABA-type triblock copolymer. In the new triblock copolymer, poly(glucose-6-acrylate-1,2,3,4-tetraacetate), PGATA is the outer glassy segment A, and PnBA is the soft/rubbery middle segment B (Figure 30a).[102] The triblock copolymer, containing 14% GATA, was mixed with 30% weight of tackifier. The mixture exhibited 2.31 N/m^2 peel adhesion strength[102] which is comparable to many commercial pressure sensitive adhesives such as duct tape, electrical tape, and paper tape (peel adhesion strength range of 1.9-4.2 N/m^2).[106]

Due to the importance of glycopolymers in biological processes, a study of glycopolymers as a bulk tissue adhesive is a new and attractive field to be explored. The glycopolymer is an excellent candidate as a tissue adhesive base material because of its high water compatibility and high flexibility as a hydrogel. During the adhesive bond formation, the polymer should be flexible for easy access to the substrate surface, meaning the adhesive polymer should wet the substrate surface quickly. Therefore, high flexibility is essential for high quality adhesives.[107-111]

In addition, water compatibility in biomedical application is always important because the human body is 60% water and most of the human body is wet, save for the outer skin.[112] However, the glycopolymer has not been studied for tissue adhesives applications in spite of suitable properties as a tissue adhesive. An initial example of glycopolymer-based adhesive includes spider silk protein ((eADF4(C16))) containing glycopolymer. This new glycopolymer-based material showed improved cell (BALB/3T3 mouse fibroblasts) adhesion compared to non-glycopolymer samples.[77]

Recently, we have developed a glucosamine pendant containing glycopolymer-based tissue adhesive that shows strong and controllable bulk adhesion (not cell adhesion) between large biological surfaces. The new glycopolymer adhesive, poly(2-methacrylamido glucopyranose-*co*-N-methacryloyl-3,4-dihydroxyl-L-phenylalanine-*co*-8-azidooctyl methacrylate) [poly(MG-*co*-MDOPA-*co*-AOM)], was prepared by free radical polymerization of three methacrylate monomers (a hydrophilic glycopolymer segment, a mussel-inspired catechol segment, and a crosslinking azide segment) as shown in Figure 30b.[113] Even without crosslinking, the new terpolymer adhesive demonstrated 20-fold higher adhesion strength (115 kPa) compared to a commercial rubber cement (5.8 kPa). The bulk adhesion properties of the terpolymer were enhanced by covalent bond forming crosslinking via strain-promoted azide–alkyne cycloaddition (SPAAC).[113]

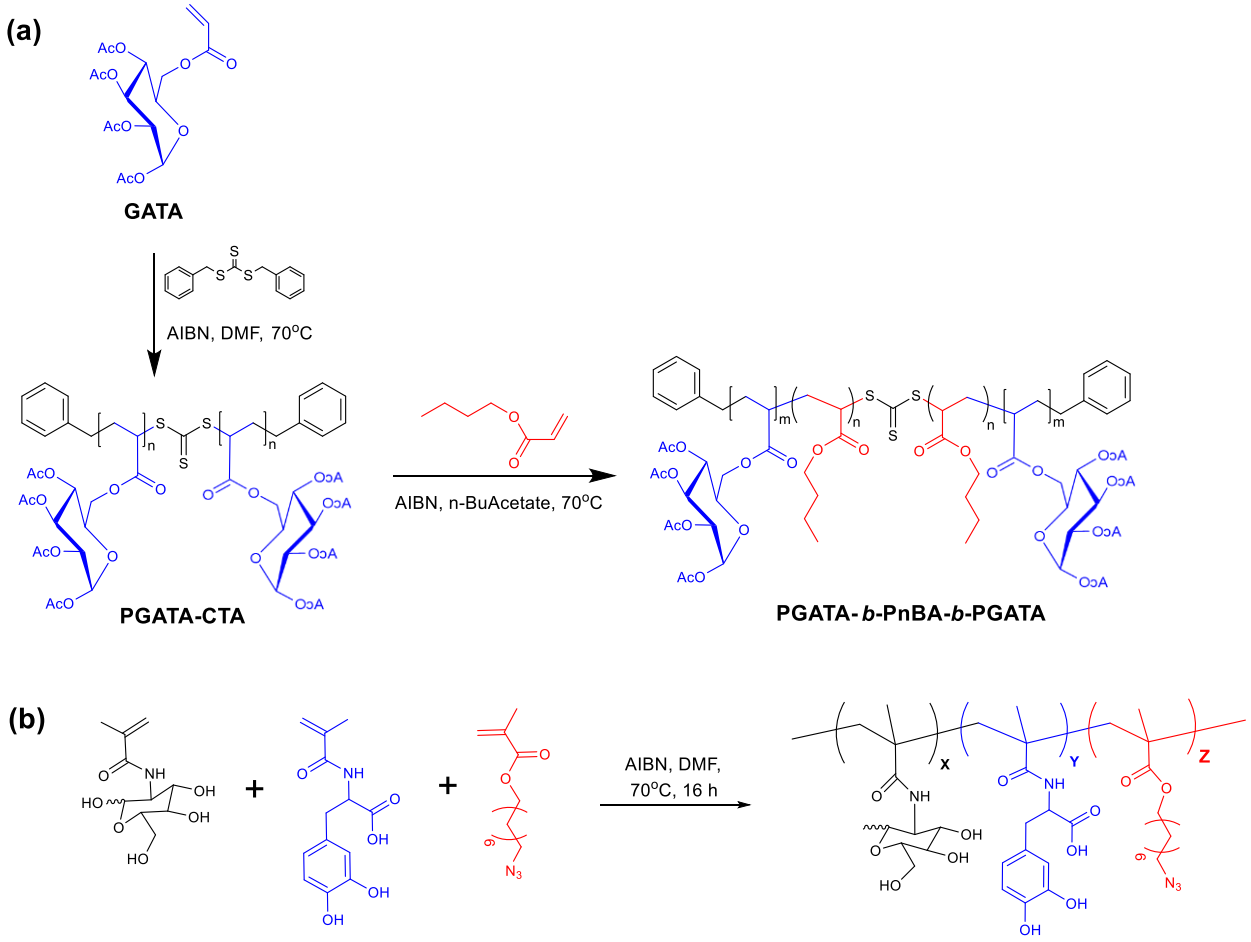


Figure 30. (a) Synthesis of ABA triblock copolymer, poly(GATA-*b*-nBA-*b*-GATA), copolymer *via* RAFT,[102] and **(b)** synthesis of a controllable terpolymer bioadhesive, poly(MG-*co*-MDOPA-*co*-AOM).[113]

3.4. Glycopolymer Nanoparticles

Glucose-coated nanoparticles (glyco-nanoparticles, GNPs) integrate multivalent glycopolymers on a nano-size inorganic core, such as iron oxide,[114, 115] silver,[116] and gold.[117, 118] The GNP have significantly advanced molecular imaging technology.[119] The most significant challenge is synthesis of well-defined nanoparticles that possess high density of multifunctional polymers on the nanoparticle surface.[120-124] The well-defined glycopolymer nanoparticle is particularly important for a cell targeted treatment.

Gold nanoparticles have been used in many biomedical applications including fluorescent materials, cell imaging, and radiolabeling substrates due to their inertness, photo-stability, simplicity of preparation and facile conjugation with biological molecules.[117, 118] However, there are still multiple challenges that involve stability of nanoparticle materials in physiological

condition, poor permeability to various biological membranes, rapid renal elimination, and insufficient target cell recognitions.[125] There has been a lot of effort to overcome those limitations by designing and synthesizing new glycopolymers for nanoparticles. A glycopolymer functionalized gold nanoparticles, which forms a core-shell architecture, was reported by Gokhan *et al.* The glycopolymer was prepared by RAFT polymerization of mannose-methacrylic acid (Figure 31).[126]

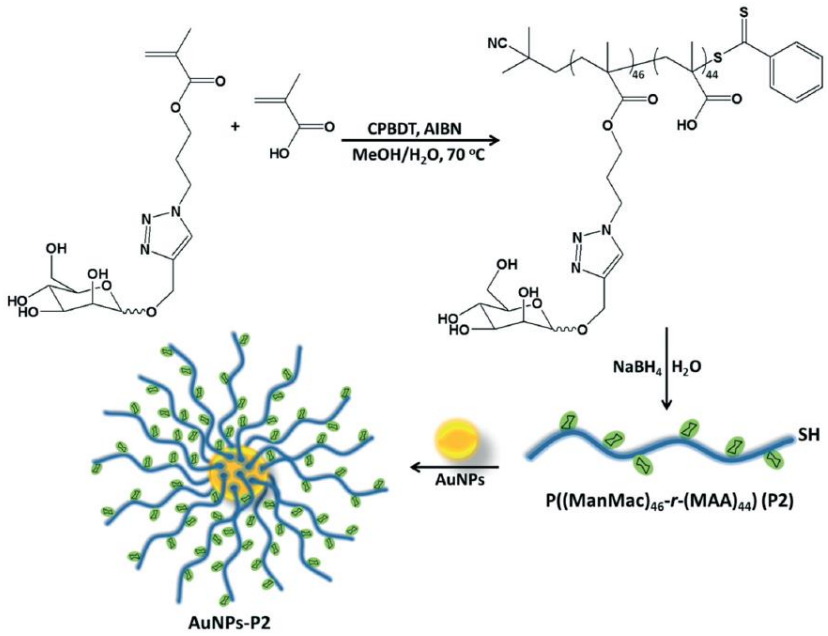


Figure 31. Glycopolymer-funcationalized gold nanoparticles (GNPs) for preparation of pH-sensitive anti-cancer agent delivery. Thiol terminals linked the glycopolymer to gold nanoparticle surface. Reproduced with permission.[126] Copyright 2018, Royal Society of Chemistry.

GNPs has also been reported as a photocaustic imaging agent and photothermal therapy agents of tumors.[127, 128] Those features are enabled by high-density functional groups on the nano-particle.[121] The gylcopolymer for the GNPs was prepared by self-assembly of amphiphilic poly(lactose)-modified perylenediimide (PLAC−PDI) that was synthesized by ATRP (Figure 32). The selective binding of lactose on GNPs to asialoglycoprotein receptors at HepG2 cells (human liver cancer cells) enabled treatment of hepatocellular carcinoma (liver cancer).[129-131] The self-assembling amphiphilic glycopolymer offers high selectivity for a photothermal therapy agent to a hepatic tumor.

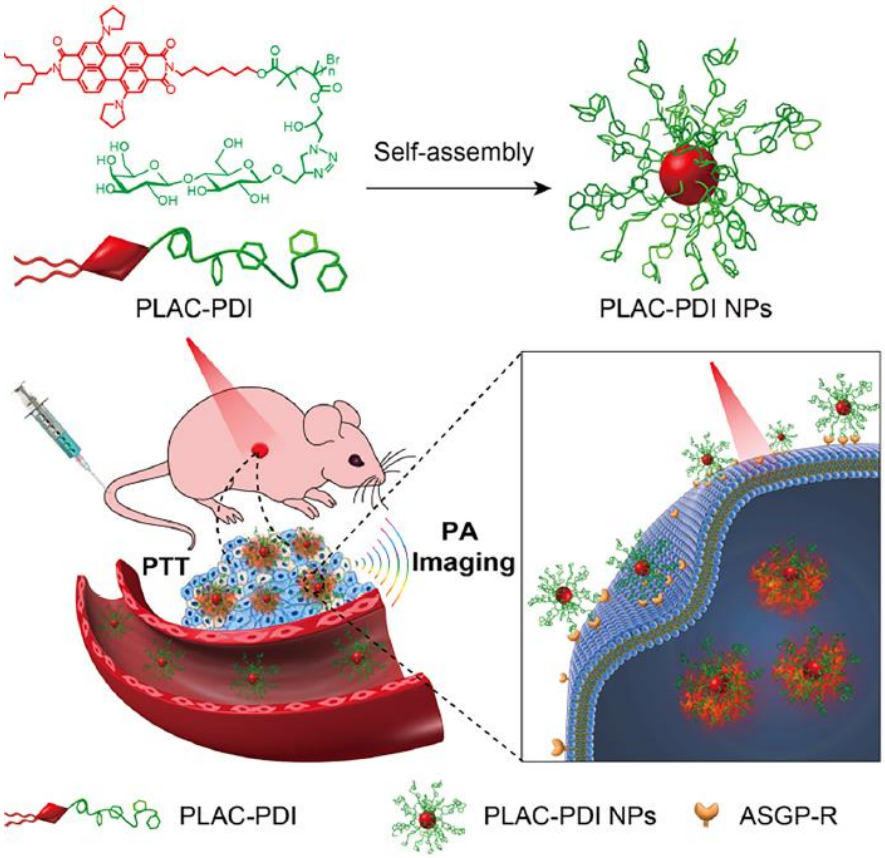


Figure 32. Preparation of GNP, poly(lactose)-modified perylenediimide (PLAC−PDI) nanoparticles, for hepatic tumor targeted photocaustic imaging and photothermal therapy application.[121] Copyright 2017, American Chemical Society.

Glycopolymers have been incorporated into biologically applicable quantum dots (QDs) which are water soluble, biocompatible, and enabled to emit near infrared.[132, 133] Guo *et al.* developed a polyvalent glycan-quantum dot (Figure 33) which is an excellent tool used to investigate multivalent protein-glycan interactions via multi-modal readout strategies (e.g., Förster resonance energy transfer (FRET), hydrodynamic size measurement, and transmission electron microscopy imaging).[134] The new glycan-quantum dot enables the dissection of multivalent protein-ligand which leads inhibition of viral infection at a cellular level.

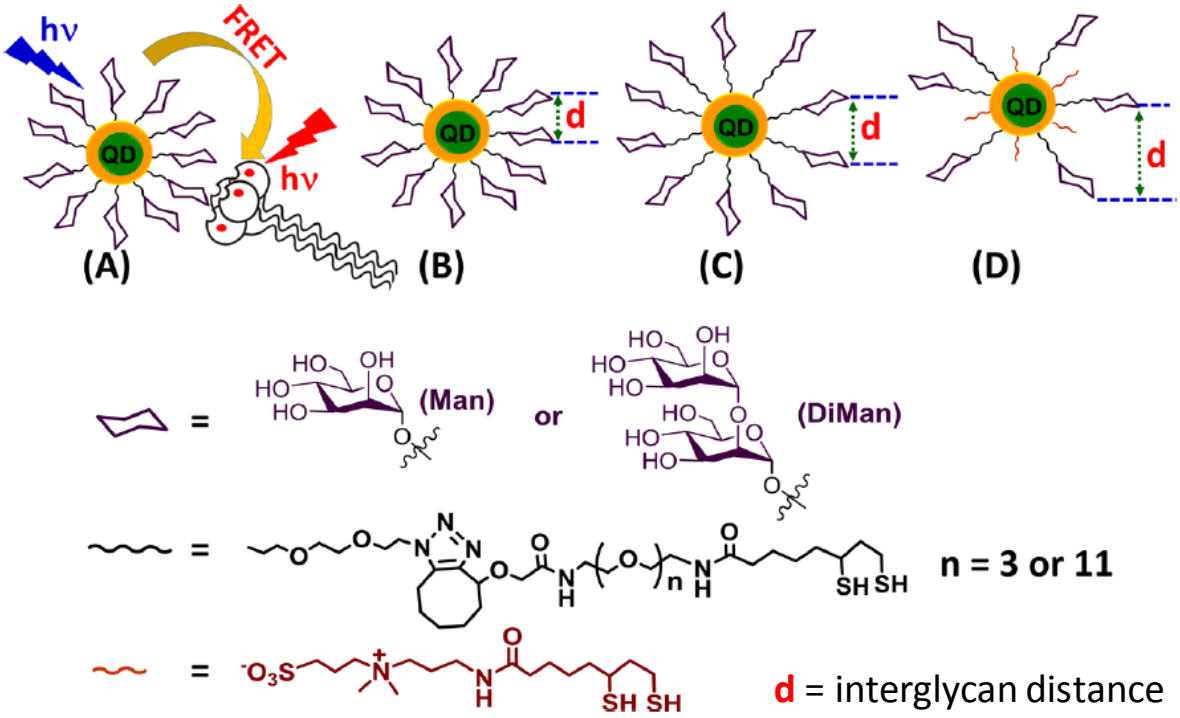


Figure 33. **(a)** Polyvalent glycan-quantum dots by QD-sensitized dye FRET mechanism, **(b-d)** tuning mechanism of QD surface glycan valency and interglycan distance (d) *via* ethylene glycol linker length (n=3 for b, and n = 11 for d; d =) and glycan dilution with an inert dihydrolipoic acid-zwitterion spacer ligand.[134] Copyright 2017, American Chemical Society.

3.5. Drug delivery and biosensor

Glycopolymer has been used for drug delivery systems in a wide variety of ways and it has been well-reviewed in multiple review papers.[135-137] Thus, the present report will discuss a few selected examples of glycopolymer-based drug delivery systems. A photo-responsive block copolymer, poly(methoxyphenyl azo phenoxy ethyl methacrylate-*b*-β-d-galactopyranosyl ethyl methacrylate) (poly(AzOMA-*b*-βGalEtMa), was synthesized via RAFT polymerization (Figure 34a) for delivery of hydrophobic small molecule drugs.[138] The synthesized amphiphilic polymer, (poly(AzOMA-*b*-βGalEtMa), can undergo self-assembly in an aqueous solution to form micelles (Figure 34b).[138] A drug mimic, Nile red, loaded glycopolymer micelles showed high cellular uptake in human melanoma A375 cells (Figure 34c).[138] The safe drug delivery to melanoma cancer cells are most likely due to the structure of the cancer cells which contains galectin-3 receptors. The galectin-3 is known to have strong interaction with galactose; therefore, promoting cell uptake by endocytosis.[139]

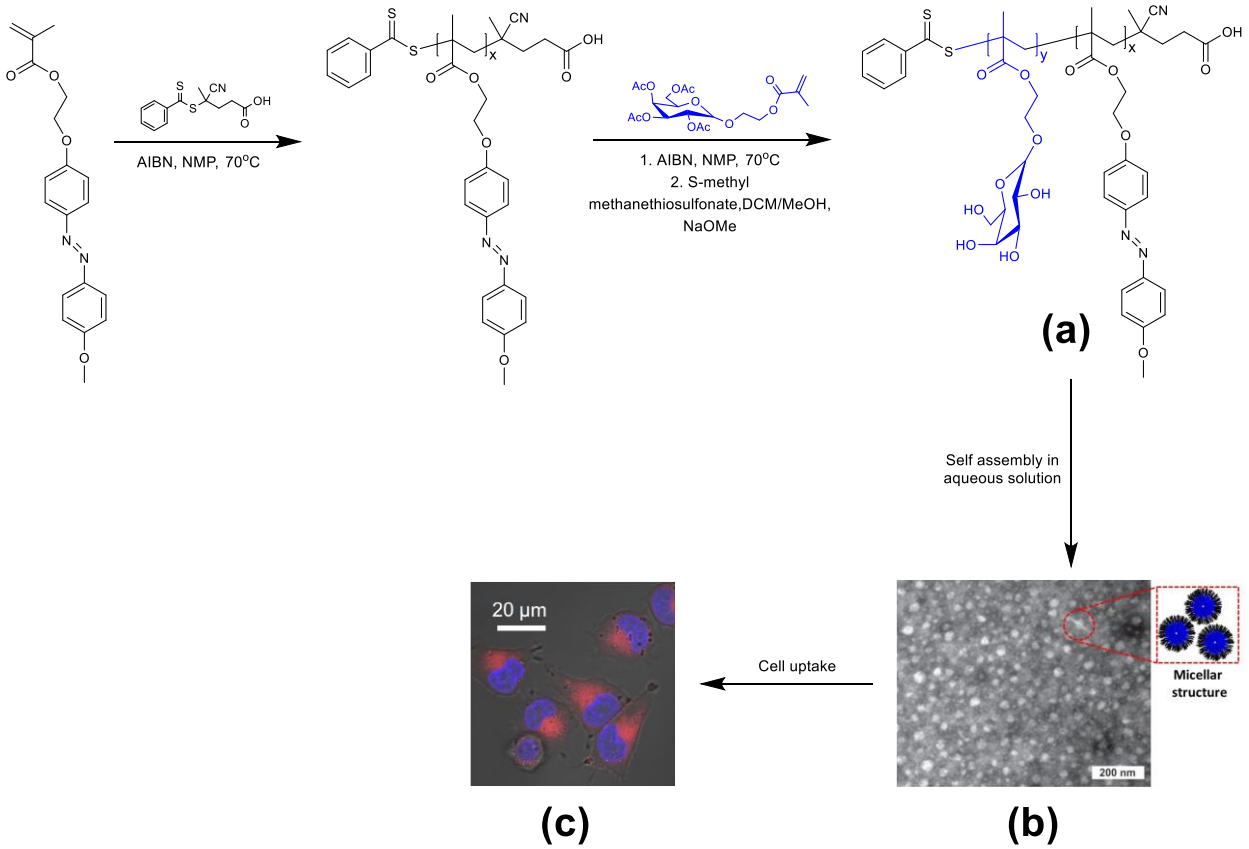


Figure 34. (a) Synthesis route of photo-responsive block copolymer, poly(AzOMA-*b*-βGalEtMa), via RAFT polymerization, **(b)** Micelle formation was observed by TEM, and **(c)** Confocal image of A375 melanoma cells incubating with Nile red dye loaded polymer.[138] Copyright 2015, Elsevier.

In diabetes treatment, glycopolymer has an important role due to its selective interaction to lectins. Mees *et al.* introduced the above discussed selective glucose-Con A interaction to glycopolymers. As shown in Figure 35a, terminal -OH of poly(2-ethyl-2-oxazoline) was modified to add a glycopolymer by reductive amination. The copolymer underwent agglutination upon addition of Con A forming turbid solution (aggregate sizes: 200-1000 nm). After addition of large excess glucose, the agglutinated polymers (glycopolymer + Con A) were disrupted and then it formed linear polymers (Figure 35b).[140]

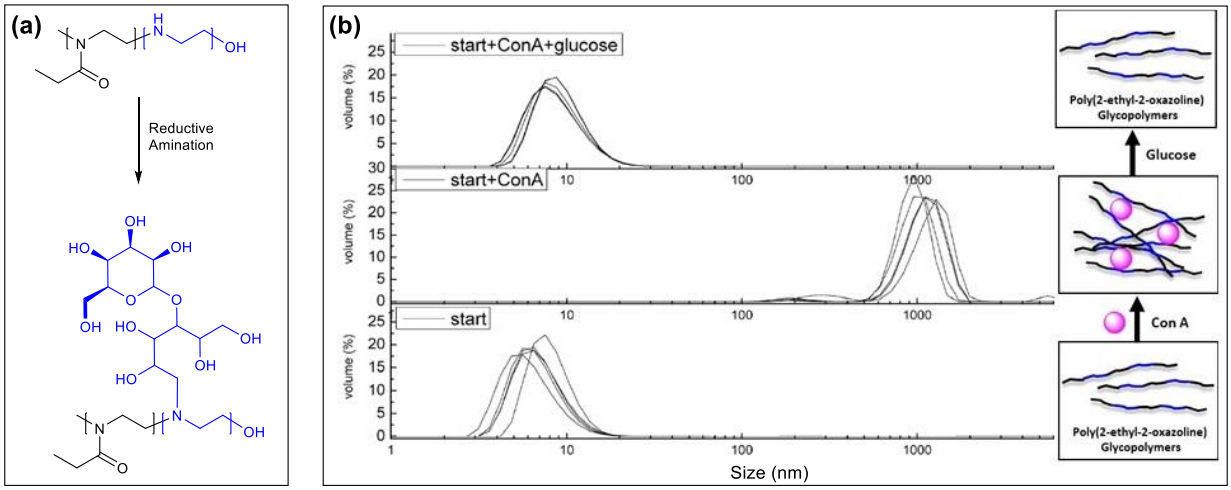


Figure 35. **(a)** Synthesis of poly(2-ethyl-2-oxazoline)-based modified glycopolymers, and **(b)** DLS volume plots of poly(2-ethyl-2-oxazoline)-based modified glycopolymers (glycopolymer only, bottom plot), after introduction of Con A (glycopolymer + Con A, middle plot), and after addition of excess glucose (glycopolymer + Con A + excess glucose, top plot).[140] Copyright 2016, American Chemical Society.

In a recent study, a glucose sensor, poly(N-isopropylacrylamide-co-glycosyloxyethyl methacrylate) microgels were synthesized in the presence of crosslinker, N,N′-methylenebisacrylamide (BIS), by free radical polymerization (Figure 36a).[141] A large number of glucose pendants allows the glycopolymer to have multiple interactions with Con A to form contracted microgels. The contracted microgel immediately expands upon exposure to glucose. By using this phenomena, concentration of glucose can be quantitatively monitored by surface plasmon resonance (SPR) (Figure 36b). When the glycopolymer microgels are swollen due to glucose, the SPR's reflected light intensity decreases because of the lowered refractive index of the swollen microgel and vice versa.[141]

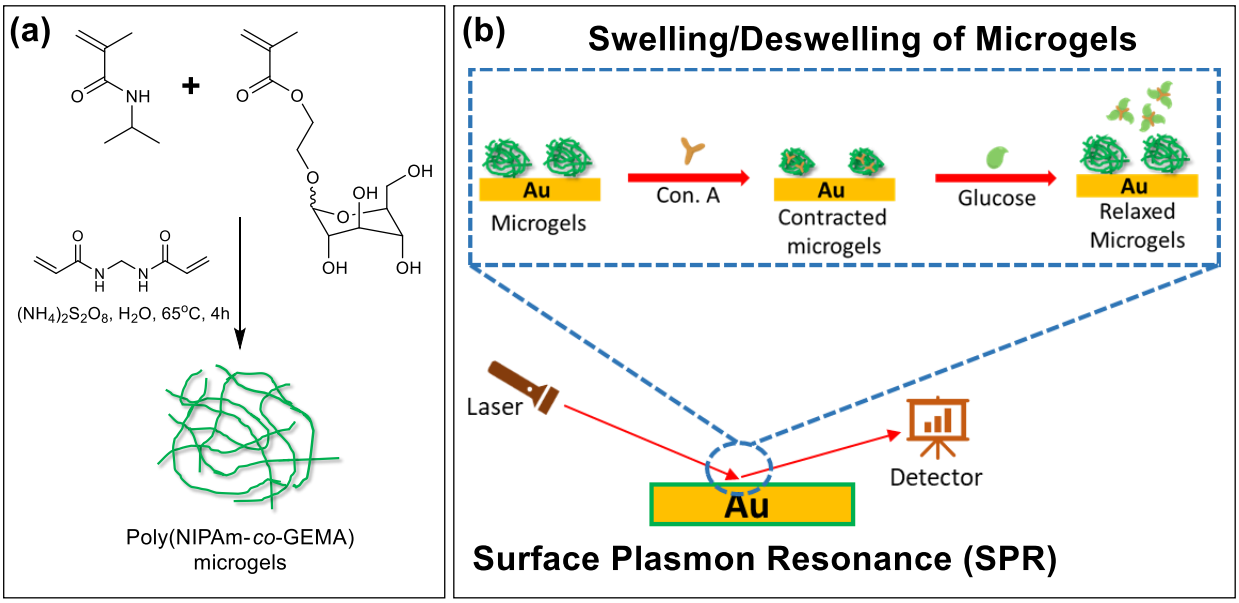


Figure 36. (a) Synthesis of poly(NIPAm-co-GEMA) microgels, and **(b)** microgel-based sensing mechanism for glucose detection.[141]

4. Conclusions

In glycopolymers research, synthesis of a well-defined polymer that includes a variety of multiple polar functionalities is an important first step in biomedical applications. Because of the accuracy of polymerization and the convenience, ATRP and RAFT are two major synthetic tools used to prepare well-defined functional glycopolymers as stated in recent literature. Furthermore, those synthetic approaches will continue to advance due to the requirement of new chemical functionalities for individualized application. Significant challenges include synthesis of the more precisely defined glycopolymer molecular structure as well as the new polar functional group tolerance. Because main chemical driving forces of carbohydrate-lectin interaction are hydrogen bonding, supramolecular interaction (e.g., van der Waals interactions), and salt bridges, precisely defining glycopolymer's chemical structures will increase understanding and control of carbohydrate-lectin interaction. This has resulted in a high demand for various synthetic tools used to prepare 3-dimensional glycopolymer structures. The new 3-dimensional glycopolymers may include helical structured glycopolymers, isotactic/syndiotactic glycopolymer, and crystalized glycopolymers. Finally, the development of new synthetic approaches to combine multiple distinctive properties will direct glycopolymer research in the future. As briefly discussed in the review, a recent example of multifunctional glycopolymer is [poly(MG-*co*-MDOPA-*co*-AOM)]

that can be used for 1) glycopolymer-based tissue adhesives, and 2) selective biological recognition (lectin-glycopolymer interaction).

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