Polymeric Materials Containing Bile Acids

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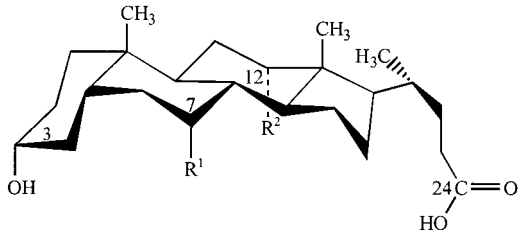
ABSTRACT

Bile acids are biological compounds in the body that have interesting properties and have been used to make special chemical structures in molecular recognition. Various polymers have been synthesized from bile acids. The materials should preserve some of the properties of bile acids, such as biocompatibility, high stability of the steroid nucleus, reactivity of the side groups, optical activity and self-assembling capacity. The synthesis and applications proposed for such polymers are discussed.

Introduction

Bile acids are natural compounds that are synthesized from cholesterol in the liver, stored in the gall bladder, and released in the small intestine for the digestion of fats and lipids. They return to the liver by absorption through the ileum membrane. This enterohepatic circulation of the bile acids is rigorously maintained in the human body and is the key for biological processes, such as emulsification and membrane transport of cholesterol, vitamins, retinol, β-caroten, etc.[1,2]

All bile acids possess a steroid skeleton known for its rigidity (Figure 1). In contrast to the cholestane steroids (trans isomers), bile acids belong to the coprostane family, where the cis connection between ring A and ring B imparts a curvature to the steroid skeleton, making possible the formation of a cavity.[3] The two faces of the cavity have very different properties, with an α face where several hydroxyl groups are directed convergently to the concavity, forming the hydrophilic part together with the carboxylic side chain, and a completely hydrophobic β face where three methyl groups are present. Because of this facial amphiphilicity, bile acids form micelles and other supramolecular structures in a stepwise manner. The organization is induced by the back-to-back hydrophobic interactions, but hydrogen bonds also play an important role.[4,5] The biological activity of bile acids is mainly based on their surfactant properties in their salt form. They can form mixed micelles with water-insoluble compounds. Bile acids also possess several chiral centers. Their carboxylic and hydroxyl groups can be easily modified, for which the chemistry has been thoroughly studied.[3,6] Therefore, bile acids are molecules of choice for many applications.



	R^1	R^2
Cholic acid	OH	OH
Deoxycholic acid	H	OH
Chenodeoxycholic acid	OH	H
Lithocholic acid	H	H
Ursodeoxycholic acid	β-OH	H

FIGURE 1. Chemical structure of bile acids.

The natural enterohepatic circulation of the bile acids has been considered useful in the synthesis of new prodrugs with liver-specific drug targeting or with improved intestinal absorption for peptides and hydrophilic drugs.[7] Amino derivatives of bile acids obtained by the modification of carboxylic side chain or replacement of OH groups with amine moieties were found useful for binding inorganic ions,[8] for enantioselective recognition of amino acid derivatives,[9] or as DNA receptors.[10−12] Unlike the more rigid cholestanes, such as cholesterol and its derivatives, the cis A/B junction of coprostanes allows ring A of bile acids to bend inward and produce a concave curvature where all of the OH groups line on the inside of the molecules. Modification of the OH groups at positions 3 and 12 with pyrene-1-carbonyl chloride produced "molecular tweezers" capable of forming complexes with aromatic guests through $\pi-\pi$ interactions.[13] Recently, bile acid dimers were used in the design of molecular architecture as enzyme models that can catalyze reactions by an enzyme-like mechanism[14] or as "molecular umbrellas" in which two bile acid molecules attached to an active agent can protect the agent from the environment.[15,16] Cyclic dimers and oligomers called cyclocholates (without spacers) and cholaphanes (with spacers) were proposed as enzyme mimics or for molecular recognition.[17−20]

The interesting properties of bile acids and their derivatives have prompted researchers to use these compounds in the design of new polymeric materials. These polymers could preserve some properties of bile acids, such as facial amphiphilicity, chirality, capacity of self-assembling, and high chemical stability of the steroid

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nucleus combined with the reactivity of the side groups, and could impart new properties by the higher molecular weights. We have used bile acids in the synthesis of new hydrogels or degradable polymers in an effort to make new polymers with improved biocompatibility for potential biomedical applications.

We will review the synthesis of these new polymeric materials based on bile acids as well as the properties and applications of the materials. In addition to the proposed biomedical uses, other applications such as chromatographic supports and photoresists, will be also discussed.

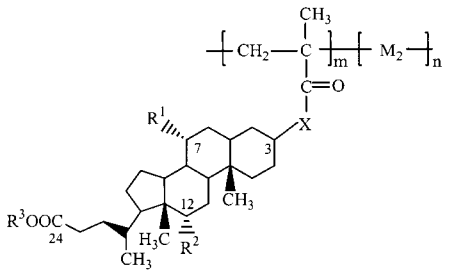
Polymerization of Monomers Containing Bile Acids

Bile acids can be chemically modified on their hydroxyl groups or carboxylic side chain to introduce functional groups capable of free radical polymerization or polycondensation. These polymers are designed as new materials with good mechanical properties or with a high degree of water uptake (hydrogels), and some can be degraded under physiological conditions.

Radical Polymerization of Acrylic Monomers. Monomers in which a polymerizable group (usually a methacryloyl group) is attached to the position C3 of a bile acid, directly or via a spacer, have been synthesized.[21−29] Cholic acid was often conveniently selected,[21−28] but lithocholic acid,[21] or deoxycholic acid[29] were also used. The higher reactivity of OH at position C3 allowed it to react easily with methacryloyl chloride without the protection of the other OH groups at positions C7 and C12. Esterification with less hindered acid chlorides, for instance, 4-(11-methacryloyloxy-undecyloxy)cynamoyl chloride, provided a mixture of isomers modified at C3 and C7 or C12.[22] In the last case, the selective acylation of the C3 OH group could be achieved by Mitsunobu reaction, with 4-(11-methacryloyloxy-undecyloxy)cynamic acid as a reagent, in the presence of azodicarboxydiethylester and triphenylphosphine.[22] The reaction proceeded with the inversion of the configuration at C3. In all cases, the carboxylic groups at position C24 needed protection to avoid side reactions and to improve the solubility of the products in organic solvents. The protection was achieved by esterification with methanol,[21,23−27] 2-trimethylsilylethanol,[22] or *tert*-butyl alcohol,[29] depending on the application for which the polymeric material was intended. The bond at position C3 can be in either α or β configuration, depending on the derivatization procedure.[22,24,29]

Methacrylate esters and methacrylamides of bile acids could easily undergo homopolymerization and copolymerization. The general chemical structure of the polymers is shown in Figure 2.

The properties of homopolymers depend on the configuration at C3 and the nature of the spacer group (X). Thus, the homopolymers obtained from 3β-methacrylamides and 3β-methacrylates of cholic acid had higher degrees of conversion, higher molecular weights, narrower molecular weight distributions, and better solubilities than the 3α homopolymers.[23,24] The higher reactivity of 3β



I

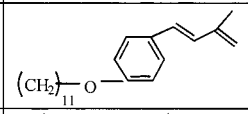
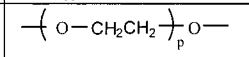
	R^3	X	M_2	Ref.
Ia	Me	O	-	21, 24
Ib	Na	O	-	22
Ic	Me	NH	-	23, 24
Id	Me	O	styrene	21
Ie	Me	O	MMA	21
If	Me	O, NH	MAA	25,26
		O, NH	HEMA	25
Ig	t-Bu	O	t-BuA	29
Ih	$(CH_2)_2Si(CH_3)_3$		-	22
Ij	Na		-	22
Ik	Me		-	27
Im	Me	p = 0, 1, 2, 4, 6	NIPAM	28

FIGURE 2. General chemical structure of homo- and copolymers obtained by radical polymerization of 3-methacryloyl derivatives of bile acids. The bond at position 3 can be in α- or β-configuration.

monomers was attributed to their stereochemistry. Molecular modeling and X-ray crystallography showed that the rigid steroid skeleton is in a linear position with methacryloyl moiety in the case of 3β monomers but forms a sharp angle in the case of 3α monomers.[24,30] Consequently, the propagation of the polymer chain can proceed more easily for the less sterically hindered 3β monomers. Therefore, stereochemically controlled procedures for preparing pure 3β-methacrylate and 3β-methacrylamide of cholic acid have been successfully developed.[31] Even though the β configuration favors the polymerization, Zhu and co-workers succeeded in polymerizing α monomers to polymers of relatively high molecular weights in high yields. However, failure of such a polymerization was reported by others.[29]

Copolymerization of the methacrylic monomers containing bile acids has also been successfully performed. The copolymerization ratios were determined for the pair 3β-(methacryloyloxy) cholic acid methyl ester and styrene (r_1 = 0.80, r_2 = 0.91).[21] In most cases, the chemical composition of the copolymers with acrylic or vinylic monomers, as determined by ^{1}H NMR or solid state ^{13}C NMR analysis, was very close to the ratio of the comonomers in the copolymerization mixture. [25,26] This was considered to be an indication of a similar reactivity of the comonomers. DSC analysis of the copolymers indicated the presence of a single T_g value located between the T_g's of the corresponding homolpolymers,[25,26,28] showing that the copolymers were random.

The hydrophilicity of the homopolymers depends on the nature of the X group (NH > O),[24] the configuration

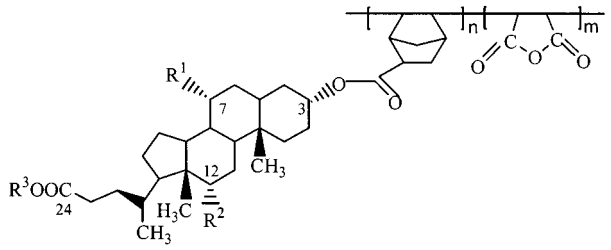
at the C3 (3β-polymers were more hydrophilic than their 3α-analogues),[24] and the form of the COOH group (ester, free acid, or salt). The homopolymers with free carboxylic groups were obtained by selective hydrolysis of the protecting ester group, and their water uptake was twice that determined before hydrolysis.[24] The hydrophilicity of the bile acid-containing polymers can be significantly improved either by introducing more polar comonomers, such as methacrylic acid (MAA) or hydroxyethyl methacrylate (HEMA), or by using a more flexible and hydrophilic spacer, as in the case of polymers where X was ethylene glycol or its oligomers (structure **Ik**).[27] The water uptake of these polymers increased with the increasing length of the polar spacer, and for $n = 6$, it reached 50 and 90 wt % before and after ester group hydrolysis, respectively.

Copolymerization of 3α-[methacryloyloxy(oligoethoxy)]-cholic acid methyl esters with *N*-isopropylacrylamide (NIPAM) led to copolymers with both thermosensitive and pH-sensitive properties (structure **Im**).[28] In comparison with NIPAM homopolymer, the lower critical solution temperature (LCST) of copolymers decreased with the increasing content of the cholic acid-containing monomer and increased with the increasing length of the hydrophilic spacer and increasing pH (in the case of polymers with free carboxylic groups). These polymers formed self-aggregates below LCST as a result of the hydrophobic interactions among pendant steroid moieties.

Ahlheim and co-workers have investigated the possibility to obtain polymers with organized structures (vesicles, micelles) as those formed by bile acids themselves.[22] The monomers with carboxyl groups as a sodium salt could form organized structures, but the type of the structure was dependent on the flexibility of the monomer. In the absence of a spacer between the methacryloyl moiety and the bile acid nucleus, the monomers were too rigid to form vesicles, but they formed micelles. Monomers with an 11-undecyl cinnamoyl spacer formed vesicles. However, the polymers obtained from these organized monomers displayed a very low degree of ordered structures.[22]

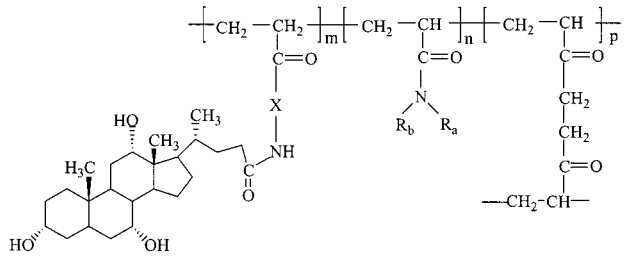
In addition to the methacrylic monomers and the corresponding polymers with the structure **I**, some other bile acid-based monomers have been synthesized in order to obtain polymers with special properties. For instance, cholic and deoxycholic acids were introduced as pendant groups of norbornene moieties, and the monomers were copolymerized with maleic anhydride to prepare new photoresists. Polymers (structure **II**, Figure 3) had a molecular weight of ~3500, a good thermal stability (T_d about 255 °C), and an excellent transmittance at 193 nm.[32,33]

There have been only two attempts to obtain polymers from monomers in which the bile acid is attached through its carboxyl group to a polymerizable double bond. Cholic acid was coupled to a (meth)acryloyl group via a spacer.[34,35] The obtained monomers were copolymerized with different *N*-alkyl acrylamides, with or without a cross-linker (structure **III**, Figure 4). Polymers with the structure **IIIa**



II

FIGURE 3. Chemical structure of copolymers based on norbornyl deivatives of bile acids. R^3 = *tert*-butyl.



III

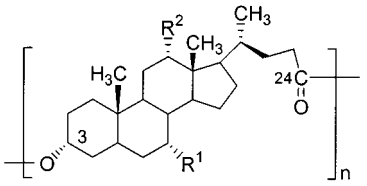
	X	R_a	R_b	m/n/p	Ref.
IIIa	O-$(CH_2)_6$	H, methyl, ethyl, isopropyl	H or R_a	1/10/2	34
IIIb	NH-$(CH_2)_2$	Isopropyl	H	1/99 – 7/93 p = 0	35

FIGURE 4. Chemical structure of polymers obtained from C24 derivatives of bile acids.

were designed as imprinting materials. Therefore, the removal rate of the pendant bile acid by aminolysis with aqueous *N*,*N*-dimethylethylenediamine was found to be dependent on the type of alkylacrylamide comonomer used.[34] Polymers with the structure **IIIb** displayed thermosensitive properties. Their LCST values decreased with increasing content of bile acid-bearing monomers.[35]

Polycondensation of Bile Acids. The first attempt to perform bile acid polycondensation was reported by Ahlheim and Hallensleben,[36] who used *p*-toluenesulfonic acid as a catalyst and high reaction temperatures. Under these conditions, they obtained branched polymers from bile acids containing more than one hydroxyl group (deoxycholic and cholic acids). The resulting polymers had low molecular weights and low solubility in organic solvents.

Linear polycondensation of bile acids, without branching or cross-linking (structure **IV**, Figure 5) has been achieved either with an enzyme (lipase) as a catalyst[37] or by the use of a combination of a coupling reagent (*N*,*N*-diisopropylcarbodiimide) and a catalyst (1:1 complex of *N*,*N*-(dimethylamino)pyridine and *p*-toluenesulfonic acid) at room temperature.[38] In the last case, poly(lithocholic acid) and poly(deoxycholic acid) with relatively high molecular weights (20−60 kD) were obtained, but poly(cholic acid) had an M_w of only 2.3 kD. The polymers had a good thermal stability and displayed a partial crystallinity (15−30%, depending on the type of bile acid used);



IV

FIGURE 5. Chemical structure of polymers obtained by polycondensation of bile acids.

however, these polymers had a very rigid structure and reduced solubility in common organic solvents.

Another example of polycondensation is the polyanhydride prepared by Gouin et al. from dimers of bile acids obtained by linking the C3−OH groups via alkylene diester or diether spacers.[39,40] The dimers of lithocholic acid were used in preparation of homo- or copolyanhydrides with the structure **V** (Figure 6).[41] The polyanhydrides had a molecular weight of ~18 kD, and their T_g values decreased from 85 °C for homopolymer to 13 °C for copolymer containing 90% of sebacic acid. The degradation studies performed on pellets immersed in phosphate buffer of pH 7.4 showed near zero-order kinetics profiles for the erosion of the polymer matrix without bulk degradation. To accelerate the degradation of the polymers, smaller diacids, such as sebacic acid, were used as comonomers. The rate of degradation can be varied by adjusting the comonomer content. It increased with increasing content of the more hydrophilic component, sebacic acid.[41]

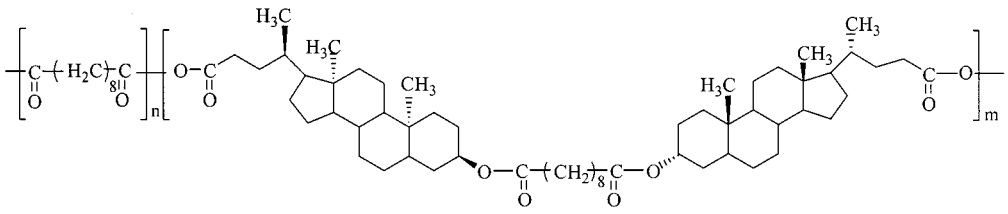
Chemical Attachment of Bile Acids to Polymers

Polymers bearing reactive groups such as OH or NH_2 were chemically modified with bile acids, mainly through the COOH groups at position C24 (Figure 7). Modification of the OH groups of polymers was performed with the help of coupling reagents, such as carbodiimides[42] or carbonyldiimidazol.[44] Coupling of the bile acid to the NH_2 groups of polymers was realized also in the presence of carbodiimides[44−50] or by using activated esters of bile acid, for instance, 24-hydroxysuccinimide esters.[51] Coupling procedures specific to other polymer supports have also been used, as described below.

Many modified polymers are hydrophilic, and the aim of such a modification was the synthesis of new amphiphilic polymers capable of self-aggregation as a result of the hydrophobic interactions between bile acid moieties. According to the position of the reactive groups along the polymer backbone, one can obtain polymers with pendant bile acid moieties or polymers end-capped with bile acids.

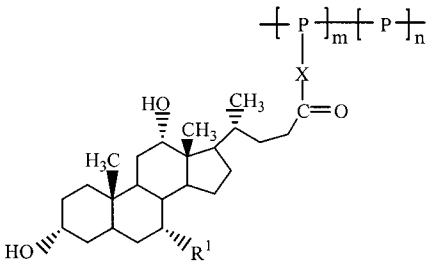
Polymers with Bile Acid Moieties as Pendants. Bile acids have been covalently attached to polymers containing reactive groups, such as polysaccharides (dextran,[42] aminohexylamino agarose,[44] chitosan,[45] partially N-desulfated heparin[51]), proteins (bovine serum albumin, BSA[46]), and poly(*p*-aminomethylstyrene).[47,48] The degree of substitution depended on the application foreseen for the modified polymers and was rather low (5−15 mol %) if the polymers should be water-soluble. In the last case, the obtained amphiphilic polymers can self-aggregate. Self-aggregation in water solutions has been investigated for chitosan,[45] dextran,[53] or heparin[51] modified with cholic or deoxycholic acids. These hydrophobically modified polysaccharides aggregated in water solutions and formed nanoparticles with a mean diameter of 20−200 nm, but the properties of the aggregates were different for the various polymeric supports.

The critical aggregation concentration (CAC) of chitosan modified with deoxycholic acid (structure **VId**) was 0.01−0.05 mg/mL, depending on the degree of substitution with bile acid. The aggregates were very compact (fluorescence anisotropy, $r \approx 0.3$, where r is proportional to the viscosity inside the aggregates) and had a mean diameter of 160−180 nm.[45] The same qualitative behavior has been noted for water solutions of dextran modified with cholic or deoxycholic acid (structure **VIa**).[53] However, the quantitative data were different. The CACs (0.2−0.4 mg/mL) were 1 order of magnitude higher than for chitosan derivatives and the compactness (or viscosity) ($r \approx 0.12$) of the aggregates was lower. The difference can be assigned to the higher hydrophilicity and flexibility of dextran backbone, but can also be the result of different experimental conditions used for the analysis. In the case of a water solution of dextran derivatives, the study was extended over a large range of polymer concentrations (0.001−2 wt %). Fluorescence measurements correlated with dynamic and static light scattering studies showed that the aggregates were formed over the whole concentration range, but the size and shape of the aggregates were strongly dependent on the polymer concentration. Below the CAC, the polymers formed large and loose aggregates (average diameter, 150−200 nm) built from 10 to 20 polymer chains connected by weak intermolecular hydrophobic interactions. They were swollen by water, which was a good solvent (the second virial coefficient was positive). Above the CAC, the polymer coils became



V

FIGURE 6. Chemical structure of polyanhydrides obtained from a dimer of lithocholic acid and sebacic acid.



VI

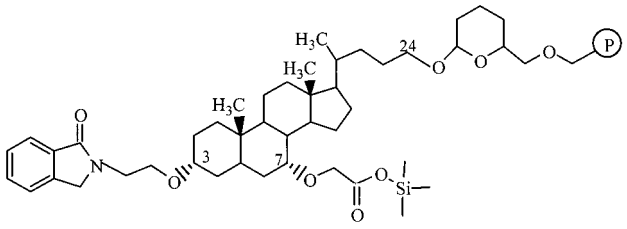
	Polymeric Support (P)	X	Ref.
VIa	Dextran	O	42
VIb		$NHCH_2COO$	42
VIc		$NH(CH_2)_5COO$	42
VId	Chitosan	NH	45
VIe	Desulfated heparin	NH	51
VIf	Agarose	$NH\text{-}(CH_2)_6\text{-}NH$	44
VIg	BSA	NH	46
VIh	Polystyrene	CH_2NH	47, 48

FIGURE 7. Structure of polymers obtained by chemical modification of polymers with bile acids via their C24-COOH group.

more compact, and this transition was accompanied by both the disruption of intermolecular associations and the change in the quality of the solvent (the second virial coefficient became negative). In the concentrated solutions (above 1 mg/mL), the aggregates included only 2−3 polymer chains, and they were very small in size (20 nm in diameter).[53]

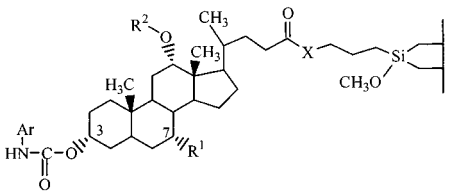
The amino groups of lysine moieties in bovine serum albumin (BSA) have been modified with deoxycholic acid, and the modified polymer was cross-linked with 1,8-diaminooctane to obtain microparticles with a mean diameter of 20 μm.[46] The modified BSA microparticles showed a lower swelling in water and a higher sorption of hydrophobic compounds in comparison with the unmodified protein.

Cross-linked polystyrene beads were used as polymeric supports for the binding of different bile acids[47,48,54] in an effort to prepare new synthetic receptors for combinatorial chemistry. Bile acids were bound to the polymeric support through their carboxylic groups. The difference in the reactivity of the OH groups was used for attaching different peptides. Chenodeoxycholic acid was bound to poly(*p*-aminomethylstyrene) (structure **VIh**), and then the OH groups at positions C3 and C7 were modified in a stepwise manner with different sequences of tripeptides.[47] In another attempt, the carboxyl group of chenodeoxycholic acid was reduced to an OH group, and after that, the OH groups at positions 3 and 7 were modified to obtain a precursor for a tetrapeptide mimic. This bile acid derivative was bound through the OH group at position 24 to the polystyrene beads via a dihydropyran acid labile spacer,[55] yielding a polymer with the structure **VII** (Figure 8). After the desired modifications at the functionalities



VII

FIGURE 8. Chemical structure of polystyrene resin modified with an amino acid derivative of chenodeoxycholic acid. p represents the crosslinked polystyrene bead.



VIII

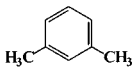
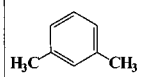
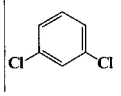
	R^1	Ar	R^2	X	Ref.
VIIIa	OH	Phenyl	H	-	56
VIIIb	H		H, Ar	$NH(CH_2)_3S$	57
VIIIc				$NH(CH_2)_3S$	57

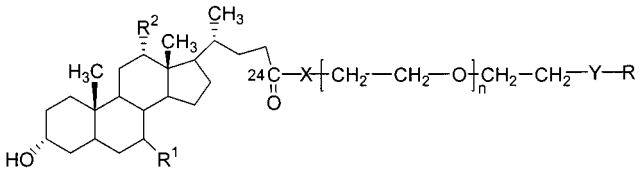
FIGURE 9. Structure of chromatographic supports obtained by binding of bile acid derivatives to silica gel.

at positions 3 and 7, the bile acid skeleton was cleaved from the resin and purified.[54]

The chirality of bile acids has been used for the synthesis of new chiral chromatographic stationary phases. The cholic and deoxycholic acid derivatives containing different aryl groups at positions C3 or C12 and an allyl moiety at C24 were bound to a silica gel support in the presence of trialkoxysilanes. The polymers with the structure **VIII** (Figure 9) were obtained. A variety of chiral supports were prepared by varying the type and number of the aryl substituents.[56,57]

Polymers End-Capped with Bile Acids. Polymers having reactive end groups, such as poly(ethylene glycol) (PEG), have been chemically modified with bile acids (Figure 10). One of the first reports dealt with binding of ursodeoxycholic acid (3α,7β-dihydroxy-5β-cholan-24-oic acid) to both ends of PEG, in an attempt to prepare polymeric prodrugs for treatment of gallbladder diseases.[43] The authors proved that the OH groups of bile acid remained unmodified.

End amino-group containing PEGs were chemically modified with deoxycholic or cholic acid at only one chain end, the other end being a methoxy group (**IXb**) or a sugar moiety (**IXc**). The resulting amphiphilic polymers were water-soluble and formed self-aggregates in water solutions. The CACs were 0.04−0.05 mg/mL,[49,52] much lower than the critical micellar concentration (CMC) of the corresponding bile salts. The aggregates had a spherical



IX

	X	Y	R	Ref.
IXa	O	O	bile acid	43
IXb	NH	O	CH_3	52
IXc	NH	NH	galactose	49

FIGURE 10. Chemical structure of poly(ethylene glycol) end-capped with bile acids.

form and a size of 120−180 nm in water (determined by dynamic light scattering) and 10−30 nm in dry form, as determined by transmission electron microscopy.[52] A PEG modified with 3 deoxycholic acid molecules at one end formed smaller aggregates (~95 nm in water) at lower CAC (0.007−0.01 mg/mL).[58]

Finally, an amino-terminated poly(*N*-isopropylacrylamide) (PNIPAM) was end-capped with cholic acid, and a polymer with a molecular weight of 8 000 displayed a temperature sensitivity similar to that of unmodified PNIPAM (LCST about 31.5 °C) and formed self-aggregates with a CAC of 0.089 mg/mL, again much lower than the CMC of sodium cholate.[50]

Applications of Bile Acid-Containing Polymers

Biomedical Applications. Polymers prepared from natural compounds, such as bile acids, can be used for biological purposes as a result of their biocompatibility and nontoxicity. Their amphiphilicity can be exploited for entrapping and delivery of both hydrophilic and hydrophobic bioactive compounds.

Drug Delivery Systems. Polymers prepared from bile acids were tested as potential drug delivery systems, either in the form of physical mixtures with the drug, or as drug−polymer conjugates. Chitosan and heparin modified with bile acids (structures **VId** and **VIe**), which can form nanoparticles in water, were proposed as colloidal delivery systems of drugs.[51,59] These systems have the advantage of a longer circulation time, because they are not taken up by the reticuloendothelial system. In vitro experiments with a system obtained from chitosan modified with deoxycholic acid (**VId**) and a very hydrophobic cytostatic drug, adriamycin (ADR), have shown that the loading of ADR in chitosan self-aggregated nanoparticles was very efficient, and the drug was physically entrapped in the hydrophobic core of the nanoparticles. The in vitro drug release in PBS solutions at pH 7.2 was very slow.[59]

Cross-linked BSA microparticles with pendant deoxycholic acid moieties (structure **VIg**) were tested as controlled-release systems for indomethacin.[46] The in vitro release experiments indicated initial zero-order release kinetics that lasted a longer time than in the case of unmodified BSA, but after ~100 h, the release rate decreased. On the contrary, polyanhydrides with structure **V** showed a near-zero-order profile of erosion rate for a very long period (100 days), and the release of a model compound, such as *p*-nitroaniline had also a quasi-zero-order rate and was almost parallel to the erosion profile. The findings suggest that these polyanhydrides can be used as implants from which the active drug can be released at a constant rate over a long period of time, avoiding the saw-tooth patterns of conventional administration or the first-burst pattern of other matrix systems undergoing bulk erosion.[41]

The systems presented above can release the drug by diffusion or erosion mechanisms. The ursodeoxycholic acid−PEG conjugates (structure **IXa**) can release the drug by a stepwise hydrolysis of the ester bond connecting the drug to the polymer. After the oral administration of one of these conjugates to human volunteers, it was found that the half-life of the drug in the blood was longer, and the concentration in the blood was more constant than after administration of an equivalent dose of free drugs.[43] This indicates that the ursodeoxycholic acid−PEG conjugate might have a better therapeutic efficacy than the free drug.

New Receptors for Combinatorial Chemistry. Combinatorial synthesis is an important tool in medicinal chemistry in the search for new bioactive compounds. Bile acids have been used as new nonpeptide scaffolds that can enlarge the library of the compounds. The polymers with structure **VIh** were modified with peptide sequences at positions 3 and 7 and were tested as receptors for opioid peptides[47] or for enkephalin.[48] The tests allowed the accumulation of data about the precise peptide structures, imparting their selectivity for the chosen substrate. In another approach, the design of the bile acid-containing polymer with structure **VIII** included the bile acid moiety into the active peptide substrate. It was assumed that the compact hydrophobic surface of bile acid could have beneficial effects on the pharmacokinetics of these peptides.[54]

Separation Techniques. Polymers containing bile acids are quite appropriate for different separation techniques as a result of their amphiphilic character and the presence of chiral centers.

Affinity chromatography performed on the columns filled with aminohexylamino sepharose modified with deoxycholic acid (structure **VIf**) allowed the separation of high-purity albumin from human plasma.[44] The same polymer proved to be useful in the isolation of receptor/carrier proteins involved in the enterohepatic transport of bile acids.

The binding of bile acid derivatives to silica gels provided chiral stationary phases with improved column lifetime, more reproducible chromatography, and good enantiomer separation.[56,57] The cholic acid-based chromatographic support with structure **VIIIa** had a great enantioselectivity for binaphthyl racemic mixtures.[56] The efficacy of the stationary phase obtained by binding deoxycholic acid derivatives with different aryl substituents at positions C3 and C12 (structure **VIIIc**) was dependent on the relative positions of the substituents.

When these substituents were in "matched positions", the bile acid acted as a biselector system and was able to separate racemic mixtures of both π-donor and π-acceptor analytes.[57]

Photoresist Materials at 193 nm. Photoresist materials are radiation-sensitive polymers that can help to yield positive or negative images on semiconductors and other surfaces at resolutions of micro- or even nanometer scales. Shorter-wavelength laser sources at 193 and 157 nm allow higher resolution and higher density of storage capacities, but also require the use of nonaromatic compounds.

Bile acids have an alicyclic structure transparent at 193 nm, a high C/H ratio for etching resistance,[33] and hydrophilic groups (COOH, OH), ensuring adhesion to a substrate. The introduction of bile acids as pendant groups of acrylic monomers (structure **If**)[29] or norbornyl monomers (structure **IV**)[32,33] has yielded polymers with good dry-etching resistance, good transmittance at 193 nm, and good lithographic performances. All of this indicates that derivatives of bile acids are among the best potential materials for 193 nm photoresists, and with some modifications, they can be designed also for 157 nm photoresists.

Conclusions

Recent research shows that bile acids are interesting and useful starting materials in the preparation of different polymers for various applications. The study of their physicochemical properties and their interactions with bioactive compounds helps in the understanding of many biological processes. Their natural origin, amphiphilic and acid−base properties, and ease of chemical modification ensure the versatility of their uses, especially in the biomedical and pharmaceutical fields, including controlled drug delivery, stimuli-responsive systems, liquid crystals, or dental filling and bone-repair materials.

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