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REVIEW

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Bulk and surface modifications of polylactide

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Abstract This article reviews various methods of modifying the bulk and surface properties of poly(lactic acid) (PLA) so that the polymer may be used as a drug carrier in a drug delivery system (DDS) and as a cell scaffold in tissue engineering. Copolymerization of lactide with other lactone-type monomers or monomers with functional groups such as malic acid, copolymerization of lactide with macromolecular monomer such as poly(ethylene glycol) (PEG) or dextran, as well as blending polylactide and natural derivatives and other methods of bulk modification are discussed. Surface modifications of PLA-type copolymers, such as surface coating, chemical modification, and plasma treatment are described. Cell culture technology proves the efficiency of bulk and surface modification and the potential application of PLA in tissue engineering.

Keywords Polylactide · Bulk modification · Surface modification · Hydrophilicity · Cell affinity · Tissue engineering

Introduction

Biomaterials represent one of the most interesting areas of science, where both chemical and medical scientists are contributing to human health care and improving the quality of life. A great number of polymers have been evaluated as potential biomaterials, due to their various compositions, special structures and excellent properties that cover a wide range of applications [1].

Biodegradable polymers are widely used in the medical field, in drug carriers, wound dressing [2], medical devices [3] and scaffolds in tissue engineering [4, 5]. Because aliphatic polyesters contain flexible ester bonds, and they degrade into non-toxic matter in solutions of various pH, they appear to be the most promising biodegradable polymers for clinical applications. With their outstanding biocompatibility and variable degradability [6–8], polylactones such as polylactide (PLA), polyglycolide (PGA) and polycaprolactone (PCL) as well as their copolymers are becoming one of the most commonly used synthetic biodegradable polymers in medical field. For example, PLA is manufactured with lactic acid, which is derived from renewable resources [9] such as potato, corn and sugar beet, and it can be metabolized via the tricarboxylic acid cycle in vivo. It has many advantageous physical properties, such as good mechanical properties, transparency, thermal stability, oil resistance and gas impermeability, as well as easy processing. It was approved by the US FDA as far back as the 1970s, and has since been widely utilized in sutures, clips, plates and screws, ultrasound contrast agents [10], nerve guides [11] and in drug delivery devices in clinical [12] applications. However, if we consider the practical requirements of tissue engineering and drug delivery systems (DDS), it is apparent that homo-PLA also has many obvious disadvantages: the degradation rate of the PLA cannot meet the different requirements of various tissue engineering scaffolds; the poor hydrophilicity of the PLA greatly affects cell adhesion onto the surface and penetration into the scaffolds. Furthermore, there are no cell recognition sites on the surfaces of PLA scaffolds, which leads to poor cell affinity and failed tissue engineering. On the other hand, in the case of using PLA directly as a drug system carrier, burst release or biphasic release has often occurred [13]. Therefore, in order to meet the various clinical requirements, we need to improve the PLA properties by modifying either the bulk or the surface. In this paper, we chart our attempts to modify the hydrophilicity, degradability and cell affinity of PLA. To do this, we modified the bulk and surface of PLA by introducing hydrophilic and biocompatible components and by adjusting the surface energy, surface charge and surface roughness.

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Bulk modification

Critical factors that influence the biodegradability of polymers include chemical components, compositions and morphological structure. In our bulk modification of PLA, we focus on the variety and amount of hydrolytic groups, the flexibility and crystallinity of molecular chains, and the hydrophilic groups. The bulk modification of PLA includes copolymerizing the lactide with other lactone-type monomers, a hydrophilic macromonomer (poly(ethylene glycol), PEG), other monomers with functional groups (amino and carboxylic groups), and blending the PLA with other materials.

Copolymerization of lactide with other lactone-type monomers

Polyglycolide has high crystallinity, a high melting point, and very poor solubility. If we copolymerize glycolide (GA) and lactide (LA), the product—PLGA—should exhibit better properties than PLA and PGA. PLGAs usually exhibit lower crystallinities and T_m values. The degradation characteristics of the PLGA could be adjusted by controlling the ratio of LA to GA in the feeding dose. The PLGA composed of 50:50 (LA/GA) is entirely amorphous. The degradation rate of PLGA is more rapid than that of PLA and can be increased by increasing GA content [14]. However, the solubility and toughness of the copolymer is limited by the composition.

The ABA block copolymer of L-lactide and ε-caprolactone (PLA–PCL–PLA) was synthesized by bulk copolymerization of LA and CL in the presence of stannous octoate (catalyst) at 120 °C [15]. The copolymer showed lower crystallinity and moved more freely when stretched due to the introduction of the flexible chain of PCL.

In order to further adjust the degradation rate and the flexibility of the polymeric chain, a third component—caprolactone (CL)—was introduced into the PLGA. The poly(glycolide-co-lactide-co-caprolactone) tri-component copolymer (PGLC) was synthesized by ring-opening copolymerization of glycolide, L-lactide and ε-caprolactone in the presence of stannous octoate (catalyst) [16]. By adjusting the component ratio of the three monomers, copolymers with much faster degradation rates than PLGA could be obtained. On the other hand, because of the lower T_g of PCL, the T_g of PGLC decreased as the CL and GA content increased. With a special molar ratio of GA/LA/CL = 30/70/4, a PGLC with its T_g equal to body temperature could be obtained, which could potentially be used as a thermally-sensitive drug carrier (Fig. 1).

The mechanical property measurement indicated that increasing the CL content reduces the toughness but enhances the flexibility of the copolymers. The elongation of the PGLC at break increased significantly with the introduction of the CL content. Because the T_g of the PGLC could be lower than the body temperature, the rubbery state also contributes to a fast degradation of the polymer. By means of bulk modification, a series of copolylactides with half-lives of degradation ranging from several weeks to more than two years, as well as various physical appearances and mechanical properties, could be obtained by adjusting the component ratio of lactide to the other lactone-type monomers and the molecular weights of the copolymers (Table 1) [17]. However, we could not significantly improve the hydrophilicity of the copolymer using this method.

Copolymerization of the lactide with poly(ethylene glycol)

Poly(ethylene glycol) is a highly biocompatible, nontoxic material with excellent hydrophilicity. It can make

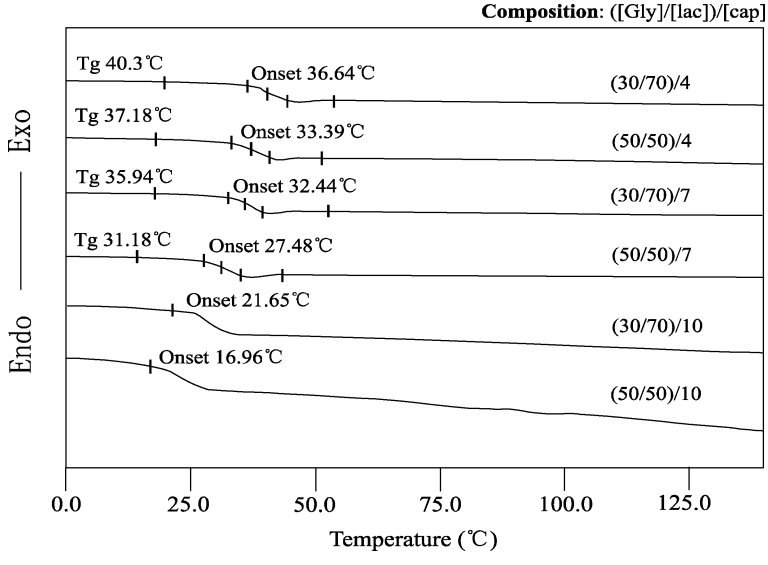


Fig. 1 Dependence of T_g on the composition of PGLC

Table 1 In vitro degradation rates of various copolylactides [17]

Polymers	$[\eta]_0$ (dl/g)	T_{50}[a] (%wt) weeks	Tensile strength (MPa)	Physical appearance (room temperature)
PLLA	13.0	~110	–	Solid
r-PGLC(10/10/80)[b, c]	2.14	> 60	31.5	Solid, waxy
PLLGA(90/10)	1.57	> 50	33.9	Solid
PDLLGA(90/10)	1.16	~18	24.1	Solid
b-PGLC(35/35/30)[d]	0.51	~11	11.7	Solid
PLLGA(70/30)	0.95	~10	28.9	Solid
PDLLA	6.5	~10	–	Solid
r-PGLC(27/63/10)	1.53	~9	25.6	Slightly tacky, rubbery
r-PGLC(27.9/65.1/7)	1.08	8~9	–	Solid
PDLLGA(70/30)	1.10	~8	16.7	Solid
PLLGA(70/30)	0.42	~6	–	Solid
r-PGLC(45/45/10)	1.12	~5	24.8	Slightly tacky, rubbery
b-PGLC(45/45/10)	0.61	4~5	22.4	Solid
r-PGLC(63/27/10)	0.46	3~4	19.7	Tacky, soft rubbery
PLLGA(50/50)	0.63	~3	20.9	Solid

[a] T_{50} is the time taken for 50% of the weight of the sample to be lost
[b] The number refers to molar ratio of glycotyl (G), lactyl (L), and caproxyl (C)
[c] *r* refers to random copolymer
[d] *b* refers to block copolymer

a surface highly resistant to biological fouling, and can reduce protein adsorption and resistance to bacterial and animal cell adhesion [18]. It is also apparently not readily recognized by the immune system. Since it is very soluble in water and many organic solvents, it can also be readily cleared from the body, and it has two hydroxy groups with reactive ends, we chose to use PEG as the macromonomer to improve the hydrophilicity and the biocompatibility of the copolymer. Modifying proteins with PEG has been shown to reduce the immunogenicities and antigenicities of these proteins and to increase circulation times [19]. PEG also has the ability to lower the toxicity and to enhance the in vivo antitumor activity of adrimycin [20].

Di- and tri-block PLA–PEG copolymers

In order to modulate the biodegradation rate, the hydrophilicity, the mechanical properties and the drug delivery behavior of PLA, poly(lactide-co-PEG) block copolymer (PLE) with a multicomponent nature was synthesized via polyesterification in the presence of PEG, using antimony trioxide (Sb_2O_3) as the catalyst [21]. Block polymers of D, L-lactide and PEG were prepared in the presence of a ring-opening catalyst [22]. The PEG was also treated with phosgene and then reacted with PLGA oligomers by polycondensation. High molecular weight block copolymers containing both PLGA and PEG segments could be synthesized [23, 24], and PLLA-b-PEG copolymer could be synthesized by using a metal oxide or stannous octoate catalyst. When potassium tert-butoxide was used as the catalyst, the copolymers were synthesized in toluene at 80 °C [25, 26].

PLA–PEG–PLA triblock copolymer (tri-PLE) was prepared in toluene at 70 °C by ring opening polymerization of lactide using PEG as macroinitiator in the presence of $Sn(Oct)_2$ as catalyst [27]. It was also synthesized by LA and PEG at 120–140 °C via bulk ring opening polymerization in the absence of a catalyst [28], or by a coupling reaction of prepolymers of PLA and PEO [29]. The properties of copolymers containing PLA of various molecular weights showed that phase-separation of the PLA and PEG segments occurred in the copolymers, and that the crystallization of the PLA segment was greatly affected by the PEO segment [30]. On the other hand, the PDLLA-b-PEG-b-PDLLA copolymer could be synthesized via anionic polymerization using potassium poly(ethylene glycol)ate as a microinitiator [31].

Although di-PLE and tri-PLE were synthesized, and the hydrophilicities of the copolymers were obviously improved by the introduction of the PEG component, the hydrophilicities and molecular weights of these copolymers were influenced by the content and molecular weight of PEG. The poor miscibility between PEG and PLA segments often lead to phase separation in the copolymers. The T_g value of the copolymer was also high. Therefore, the di-PLE and tri-PLE copolymers did not have good mechanical strength and flexibility. Further modification was necessary.

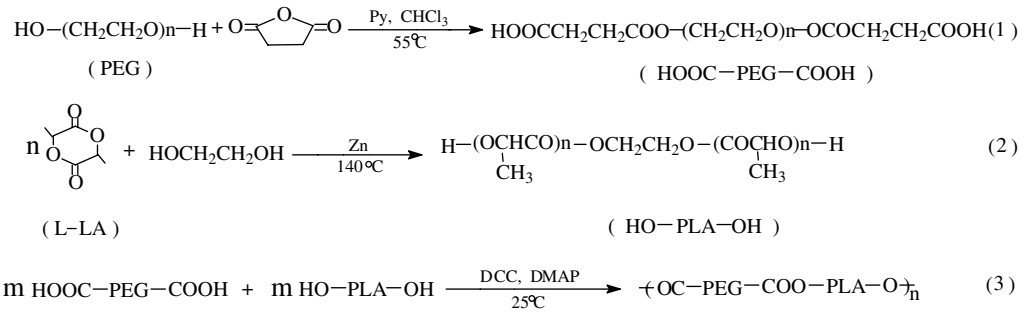
PLA–PEG multi-block copolymers

In order to overcome the phase separation that existed in the di- and tri-PLE, a PLA–PEG multiblock copolymer (m-PLE) with predetermined block lengths was synthesized by copolycondensating PLA-diols and PEG-diacids using dicyclohexylcarbodiimide as a coupling agent and dimethylaminopyridines as catalyst (Scheme 1) [32].

A series of m-PLEs with different PEG segment lengths and various compositions were synthesized. Compared with tri-PLE, it could be seen that, with the same segment length of PEG and component ratio of PLA–PEG, the inherent viscosity of m-PLE was much higher than that of tri-PLE (Table 2). The results demonstrated that the molecular weight of m-PLE could be greatly increased by coupling prepolymers of PLA-diols and PEG-diacids [32, 33].

DSC measurements showed that all of the m-PLE copolymers had only one glass transition. This means that the miscibility of the two components was enhanced in all cases, and phase separation did not happen in the copolymer. The crystallinity of both the PEG and the PLA segments decreased after copolymerization. It is thought that this m-PLE is composed of many short PLA and PEG segments. The miscibility between the short segments of PLA and PEG was improved and the hydrophilicities of the copolymers were increased

Scheme 1 Synthetic procedure for PLA–PEG multiblock copolymers



without needing to account for the segment length of PEG. Therefore, phase separation was avoided, and the molecular weight and mechanical properties of the m-PLE were also enhanced.

Polycaprolactone/polylactide/poly(ethylene oxide) (PCEL) copolymers

In order to improve the properties of the PLE copolymers further, a third component—caprolactone—was introduced, and a copolylactone based on PLA/PEG/PCL (PCEL) was developed [34]. Porous PCEL microspheres were easily prepared by an emulsion–solvent evaporation method in the absence of porogen. The introduction of the PEG content led to the morphologies of the microspheres changing from smooth to porous. The pore sizes of the microspheres increased as the content and molecular weight of the PEG component increased. The relationship between the chemical composition and the morphological structures of the microspheres was studied. It was proposed that during the solidification of the microspheres, the hydrophilic PEG segments had a tendency to orientate and swell in the aqueous phase, resulting in the formation of larger microspheres. After the microspheres were dried by freeze-drying under vacuum, the porous structure was obtained [35]. It was found that different compositions of the PCEL caused morphological variations among the microspheres. The hydrophilicity and degradation rate of the PCEL increased as the PEG content was increased [36]. Therefore, porous film- and microsphere-like scaffolds with various degradation rates could be easily prepared from the PCEL copolymers without the need for pore-forming agents. A novel material for application in tissue engineering scaffold has been found.

Table 2 Comparison of the inherent viscosities of tri- and multi-PLA-PEG block copolymers

Polymers[a]	[η] (dl/g)
Tri-PLE1/1(2000)	0.20
Tri-PLE2/1(2000)	0.30
Tri-PLE4/1(2000)	0.43
Tri-PLE1/1(4000)	0.36
Tri-PLE2/1(4000)	0.46
Tri-PLE4/1(4000)	0.66
Multi-PLE1/1(2000)	1.20
Multi-PLE2/1(2000)	1.57
Multi-PLE4/1(2000)	1.60
Multi-PLE1/1(4000)	1.33
Multi-PLE2/1(4000)	1.47
Multi-PLE4/1(4000)	1.31
PLLA	1.62

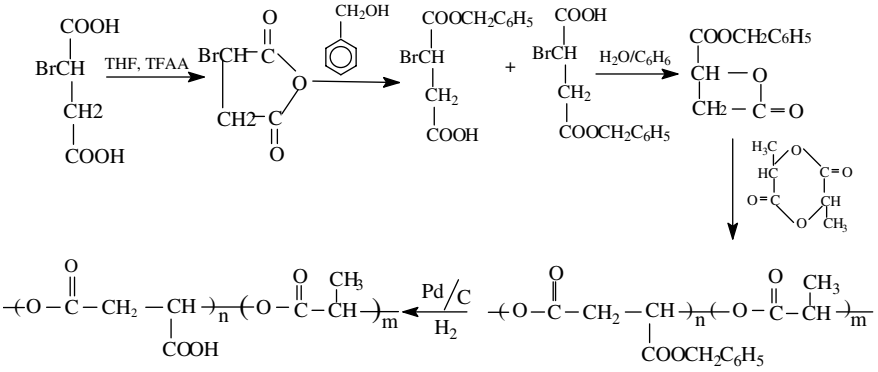
[a]Measured in chloroform at 30 °C

Star- and dendrimer-like copolylactides

Star-like copolymers possess more and shorter side chains than linear block copolymers in the similar molecular weight range. The steric architecture of the star-block copolymers contributes to a reduction in T_g, T_m, and crystallinity which also affects the formation of microspheres. Due to the widespread application of PLA in pharmaceutical and other medical fields, incorporating star- or dendrimer-like macromolecules with PLA will give a well-defined polymer structure that may have some valuable potential applications. Compared with linear polymers, dendrimers possess high surface area-to-volume ratios, exhibit numerous end groups for functionalization, and have small polydispersity indices (PDI) with well-defined interior and exterior regions. Divergent syntheses and characterizations of novel dendrimers composed of glycerol and lactic acid have been reported [37]. A third generation [G3] poly(glycerol-lactic acid) dendrimer ([G3]-PGLLA) with a tetrafunctional core was prepared in three steps. Briefly, an efficient divergent procedure for synthesizing novel aliphatic biodendrimers composed of glycerol and lactic acid was described. The key constituents of the polymer are combined in reiterative reaction steps from simple and abundant starting materials.

New types of star-branched AB block copolymers consisting of A blocks (PLLA or PLGA) and star-branched poly(ethylene oxide) (PEO) B blocks were synthesized by solution polymerization in toluene catalyzed by aluminum triethylene at 110 °C. The potential of these star-branched AB copolymers, as opposed to linear copolymers consisting of A block (PLGA) attached to central B blocks (PEO), as a drug carrier for sustained release has been investigated [38, 39]. For the recombinant human erythropoietin (EPO) loaded micr-

Scheme 2 Synthetic route for poly(L-lactide-co-RS-β-malic acid)



ospheres prepared from the linear block polymers, the reduction of lactic acid content resulted in a decrease in particle size and encapsulation efficiency, and an increase in the initial drug release. Due to the presence of more reactive OH groups in the star-shaped PEO, the incorporation of star-branched PEO domains instead of linear PEO into the polymeric matrix reduced the possibility of PEO loss in the surrounding buffer solution.

Poly(amidoamine) (PAMAM) dendrimers are good biomaterials known for their non-immunogenic properties and low mammalian toxicities, especially when their surfaces contain anionic or neutral groups, such as carboxylic or hydroxy groups [40]. Star-shaped PLA was synthesized using PAMAM with terminal hydroxy groups as an initiator [41]. Highly branched polylactide initiated by amine-terminated PAMAM dendrimer has also been synthesized [42]. The reaction was performed at a higher temperature compared to that of the lactide because of the weak reactivities of the amine or amide groups. The inherent viscosity of PAMAM-g-PLA was much smaller than that of the corresponding linear PLA with a similar M_n, which demonstrated that PAMAM-g-PLA should have a highly branched structure. Because PLLA chains are attached to a PAMAM core and the chain movements were hindered, the crystallizability of PAMAM was weakened. The degradation behaviors of PAMAM-g-PLA and PLA microspheres were compared. The weight loss of PAMAM-g-PLA was higher than that of the corresponding linear PLA for the much shorter PLA chains in the star polymer.

Poly(L-lactide-co-*RS*-β-malic acid)

Using to the methods above, it is possible to greatly improve the hydrophilicity and degradation rates of copolymers of PLA. However, another disadvantage of PLA is its lack of natural recognition sites for cells, and this problem still occurs in the bulk modified PLA copolymers. Other methods (such as the introduction of cell-recognizable groups) are needed to further improve the cell affinities of the polymers. Because using a polymer with pendant carboxyl groups or amino groups would help to immobilize bioactive agents such as amino acids, peptides and proteins on the surface, biodegradable poly(malic acid)-type polymers, with pendant carboxyl groups, are of interest to researchers [43–45]. A copolymer of malic acid and α-hydroxyl acids appeared to be a good substitute for the poly(α-hydroxyl acid)s. A functional biodegradable copolymer of L-lactide and RS-β-benzyl malate (MA) was synthesized by an improved ring-opening copolymerization method [46] in the presence of stannous octoate as catalyst, as shown in Scheme 2 [47].

The molecular weight of the copolymer decreased with increasing MA content, and was lower than that of the PLA homopolymer prepared under the same conditions. The compositions of the functional carboxy groups in the poly(L-lactide-co-β-malic acid) could be controlled by adjusting the MA content in the feeding dose. The hydrophilicity and degradation rate of the copolymers increased with increasing malic acid content.

Copolymers of PLA and other materials

Natural polymers are usually biodegradable and offer excellent biocompatibilities and cell affinities, but suffer from batch-to-batch variation because of their sources. Naturally-derived materials isolated from plants, animals, or human tissues are also expensive. It is therefore logical to study the merits of natural polymers and try to build these into synthetic polymers.

Polylactide-g-dextran

Polysaccharides such as starch and dextran are important natural biodegradable hydrophilic polymers with enzymatic degradation behaviors and good biocompatibilities. Starch-based polymers and their composites have been investigated as potential biomedical materials, especially in orthopaedic fields [48, 49]. These starch-based polymers have already been shown to be non-cytotoxic and they do not induce harmful tissue response [50, 51].

Brush-like biodegradable polylactide-grafted dextran copolymer (PLA-g-dextran) was synthesized by bulk polymerization with trimethylsilyl (TMS)-protected

Table 3 Main properties of PLA-g-dextran copolymers compared with pure linear PLA

Sample	[η] (dl/g)	Glucose units (mol%)	Contact angle (°)		Water uptake (%)	σ (MPa)
			Before de-protection	After de-protection		
PLA-g-dextran-1	2.50	0.8	84.5 ± 1.8	72.5 ± 1.1	4.2 ± 0.08	32.6 ± 2.7
PLA-g-dextran-2	1.57	1.5	85.0 ± 1.3	72.1 ± 0.9	6.3 ± 0.11	29.2 ± 1.7
PLA-g-dextran-3	0.82	3.1	89.8 ± 2.2	69.5 ± 2.1	7.7 ± 0.20	25.5 ± 2.1
Linear-PLA	2.41	–	–	80.1 ± 1.4	0.6 ± 0.05	39.2 ± 1.6

dextran as a macroinitiator [52]. With the introduction of hydrophilic glucose units, the hydrophilicity of the copolymer was obviously enhanced, compared with pure PLA, as shown in Table 3. This can be explained due to the increase in the hydrophilicity of the non-linear structure of the brush-like grafted PLA as the number of terminal polar groups increases. However, the tensile strength of the PLA-g-dextran decreased as the glucose unit content increased, because it had a brush-like structure.

Because dextran possesses a good cell affinity, the cell attachment on the PLA-g-dextran film was better than that on the PLA film, as shown in Fig. 2; in other words, the cell affinity of PLA-g-dextran was improved by introducing the dextran component.

Polylactide-dextran blend

In addition to the copolymerization technique, the bulk properties of PLA could also be modified by blending with other materials. By blending PLA with naturally-derived dextran, a kind of new biodegradable material could be obtained. Moreover, using solvent-casting and particle-leaching techniques, a sponge-like scaffold could be fabricated [53]. In order to obtain a uniform blend of PLA and dextran, hydroxyls of dextran were protected via TMS groups in order to make dextran soluble. Using a mixed solvent of dichloroform and benzene, a homogeneous solution of PLA and TMS-protected dextran could be obtained. As shown in Fig. 3, the hydrophilicity and cell affinity of the PLA-dextran blend was improved significantly compared with the pure PLA.

Polylactide-g-chitosan

Chitosan is a promising natural polymer with good biocompatibility, non-toxicity, and biodegradability, and it is widely used for wound healing and dressing, as a drug delivery carrier, a blood anti-coagulant, and an anti-tumor agent in the medical world, but the mechanical properties of chitosan are poor. So, we might hope to improve the properties of a synthetic polymer through copolymerization with chitosan. Chitosan derivatives with various side chains can be a source of manipulation for specific drug-delivery applications [54]. A pH-sensitive physically crosslinked hydrogel was synthesized by grafting D, L-lactic acid onto the amino groups in chitosan in the absence of a catalyst [55]. This method brings together the good properties of synthetic and natural polymers. Because the side chains substitute the $-NH_2$ groups of the chitosan randomly along the chain and destroy the regularity of packing between chitosan chains, the crystallinity of chitosan gradually decreases after grafting. The water uptake behavior of the hydrogels was related to the side chain length and the degree of substitution. A brush-like copolymer of poly (D, L-lactide) grafted onto chitosan by graft copolymerization was prepared with

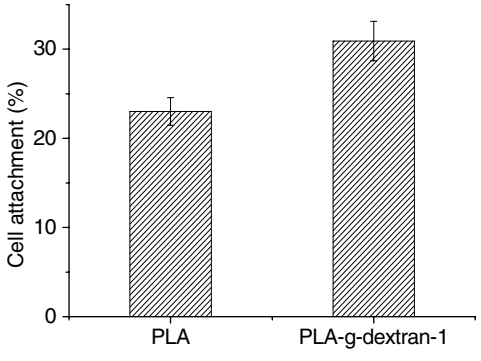


Fig. 2 Attaching efficiencies of mouse 3T3 fibroblasts on films fabricated from pure PLA and PLA-g-dextran-1 after being cultured for 2 h. The statistical difference between the two sets of data was below 0.05 ($p < 0.05$)

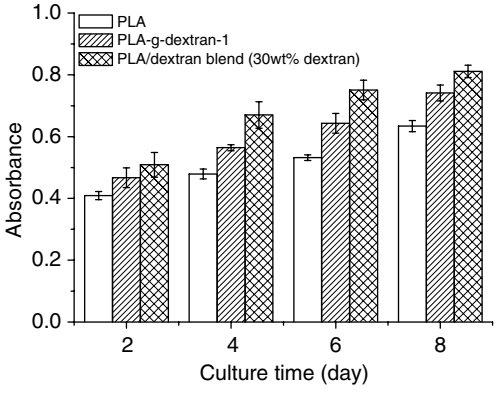


Fig. 3 MTT-tetrazolium assays after mouse 3T3 fibroblasts were cultured on PLA, PLA-g-dextran-1 and PLA/dextran blend (containing 30 wt% dextran) scaffolds. Formazan absorbance is expressed as a function of culture time. The statistical difference between the three sets of data, determined every day, was below 0.05 ($p < 0.05$)

Table 4 Factors that affect cell affinity and their influence

Affecting factors	Effects
Hydrophilicity/hydrophobicity	Hydrophilic surfaces favor cell adhesion and growth
Surface free energy	High surface energy favors cell adhesion and spreading
Surface charge properties	Static effects between a surface (with positive charge) and cells (negative charge) favor cell adhesion
Surface chemical structures	Amino-, hydroxy-, carboxyl-, sulfonic-, acylamino- groups favor cell adhesion and growth
Surface morphology	A rough surface favors cell adhesion and regeneration of biological membrane

triethylaluminum as catalyst at 70 °C. Increasing the lactide content in the feed ratio can enhance the grafting percentage [56].

Surface modification

Surface characteristics are critical to cell affinity and cell adhesion on the surface of the materials. Surface characteristics such as hydrophilicity, surface energy, charge properties, smoothness govern the biocompatibility of the material surface with tissues and cells. The main factors affecting cell affinity, and their influences, are summarized in Table 4.

In some special cases, the optimum conditions for cell attachment were found to be a surface with intermediate wettability [57], a positive charge [58], high surface energy, and a cell growth rate independent of surface chemistry [59]. Because their excellent mechanical strengths, adjustable degradation rates and the optimal (micro) stress environment they create for the growing tissue, and their easy manipulation into desired shapes, a lot of attention has been paid to modifying the bulk properties of PLA-type polymers so that they can be used in tissue engineering. However, like a lot of synthetic polymers, even after bulk modification, there are no cell recognition sites on the surface of PLA. The cell affinities of such polymers cannot satisfy the special requirements of tissue engineering and some special biomedical applications. Further surface modification is necessary. In order to attach cell-recognition ligands onto the polymer surface for modifying cell adhesion and proliferation on the polymer, surface modification methods such as surface coating, surface chemical modification, plasma treatment and hybrid modification (see Table 5) were applied.

Surface coating

Coating the surface with extracellular matrix (ECM) proteins such as fibronectin, vitronectin, and collagen, provides an adhesive interface between the polymer scaffold surface and cells that resemble the native cellular milieu. It is one of the simplest surface modification methods. Cell attachment, migration and growth on the polymer surface were mediated by proteins, adsorbed from the culture medium or secreted by the cultured cells. ECM proteins, including fibronectin, collagen, vitronectin, thrombospondin, tenascin, laminin, and entactin, have the ability to prompt cell adhesion. Therefore, if the polymer surfaces are immobilized with these bioactive factors before further treatment, the biocompatibilities of the surfaces can be significantly improved [60]. However, surface coating is often time-consuming and expensive. Moreover, passive adsorption could induce the competitive adsorption of other materials in the system and change the configuration of the adsorbed fibronectin molecules. As a result, its cell-binding activity would be influenced and reduced. Further investigations found that certain short amino acid sequences appear to bind to receptors on cell surfaces and mediate cell adhesion. The cell-binding domain of fibronectin, vetronectin and collagen contains the tripeptide RGD (Arg-Gly-Asp). As RGD is critical to cell adhesion, many researchers have investigated the addition of this sequence to synthetic polymer substrates [61–63]. The RGD was directly coated onto synthetic surfaces, and it significantly enhanced cell adhesion and growth. When the PLA surface was coated with a reactive block copolymer of α-acetal-PEG-PLA [64], the acetal groups were converted into aldehyde groups, which are highly stable in water and highly reactive with primary amino groups.

Table 5 Surface modification methods

Modification	Mechanism	Method used
Surface coating	Immobilizing growth factors and attachment factors to improve cell affinity and biocompatibility	Surface coating
Chemical modification	Modifying chemical compositions and components of the materials by copolymerization of different monomers	Random, graft and block copolymerization
Plasma treatment	Inducing cell recognition sites onto the surfaces of the biomaterials by introducing various functional groups	Low temperature plasma treatment
Hybrid modification	Obtaining biomaterials with complementary properties by blending multiple materials	Blending, hybrid compositions

Surface chemical modification

Surface alkali hydrolysis treatment is a simple and convenient chemical modification method. After surface hydrolysis of aliphatic polyester, hydrophilic carboxyl and hydroxyl can be produced by cleaving the ester bond. The resulting groups can also be used to conjugate the bioactive molecules, such as L-lysine, collagen and Arg-Gly-Asp peptide, which can be recognized by the cell adhesion receptors. However, strong alkali treatment is accompanied with extended bulk degradation of the polyester, and the residual alkali is not easily removed except by rinsing. It was shown that a mild alkali treatment could not break the ester bonds effectively in a short time. An improved method for enhancing the cell affinity of a macroporous PLLA scaffold surface is to use an improved surface-treating medium, a mixture of aqueous 0.25 M NaOH/ethanol. The ethanol was applied as a co-treating medium to wet the polylactone and assist the hydroxide nucleophilic attack on the ester bond. Therefore, a low concentration of NaOH could be applied to avoid severe bulk degradation, and the residual alkali was easy to remove. After treatment under optimal conditions, the surface hydrophilicity and the surface energy of PLLA were improved significantly, and the surface roughness was also changed. Mouse 3T3 fibroblast culture results indicated that the improved surface-treating medium was effective and convenient for enhancing the cell affinity of the PLLA cell scaffold [65].

The synthetic RGD-containing peptides could also be immobilized to PLA [66]. This method supports suitable recognition molecules for cells containing cell adhesion receptors. The attachment of hydrophilic PEG chains to the PLA surface can regulate the cell adhesion behavior on the surface, as well as control the protein and peptide adsorption [67].

Plasma treatment

Plasma treatment was performed in a vacuum, and the plasma tended to be pervasive [68]. It was demonstrated to be an efficient and unique method for modifying the surface property of scaffolds with complex shapes without changing the bulk properties. The plasma technique easily induces the desired groups or chains onto the surface of the polymer [69–74], so it is especially suited to improving the cell affinities of cell scaffolds. The surface hydrophilicity of the polymer can be improved by adjusting the kind of gas used and the treatment parameters. After ammonia plasma treatment, the surface energy of PDLLA samples increased and the polar groups were incorporated into the PDLLA surface. The hydrophilicity of the surface was also improved (Table 6). Plasma treatment can also reduce cell loss during cell seeding and can avoid the negative effects of the trace ethanol on the cell culture. Because the plasma treatment effect is difficult to maintain due to the thermal motions of surface molecules, a low-temperature treatment was suggested to efficiently maintain plasma-modified surface properties [75].

Table 6 Effects of different methods of modification on the surface energies of modified samples and a control

Modification methods	Contact angle (°)		Surface energy[a] (mJ m^{-2})			
	θH_2O	θCH_2I_2	γ_s	γ_s^d	γ_s^p	X^p
None	78.0	37.0	43.2	32.5	10.7	0.25
NH_3 plasma	21.5	40.0	69.1	26.7	42.4	0.61
N_2 plasma	25.5	33.5	68.2	29.2	39.0	0.57
O_2 plasma	45.0	26.5	60.1	32.4	27.7	0.46
($O_2 + NH_3$ plasma)[b]	17.0	32.0	71.5	29.5	42.0	0.59
Collagen coating	61.9	38.3	49.1	29.5	19.6	0.39
(Collagen anchorage)[c]	54.3	42.8	51.8	27.0	24.8	0.48

[a] γ_s surface energy, γ_s^d dispersive components, γ_s^p polar components $X^p = \gamma_s^p/\gamma_s$. Plasma modification parameters were 50 w, 20 Pa, and 120 s except for additional descriptions

[b] 60 s for O_2 and then 60 s for NH_3, 50 w, 20 Pa

[c] NH_3 plasma pre-treatment conditions: 50 w, 20 Pa, 300 s

To further improve the effect of plasma treatment, a method combining plasma treatment and collagen anchorage was proposed [76]. XPS analysis revealed that the polar O-containing groups and N-containing groups as well as the positive charges could be incorporated into the modified sample surface, while the collagen was anchored on the sample surface. Additionally, the surface of the sample turned rough after the plasma treatment (Fig. 4), which suggested plasma etching or removal of the surface layer. After plasma pre-treatments, more collagen could be easily anchored on the surface of the samples. The enriched polar groups on the surface provided many sites to attach the collagen by polar interaction and hydrogen bonding.

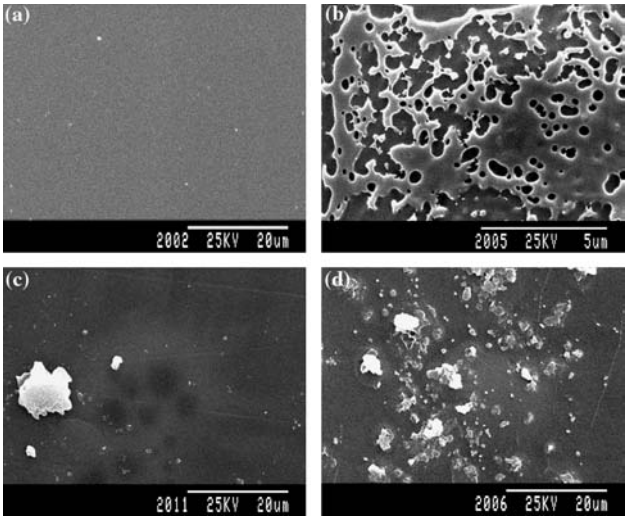


Fig. 4a–d SEM observations of the surface morphologies of PDLLA samples. **a** Control (×2,000); **b** modified by NH_3 plasma (50 w, 20 Pa, 300 s) (×10,000); **c** modified by collagen coating (×2,000); **d** modified by ammonia plasma pre-treatment then collagen anchorage (×2,000)

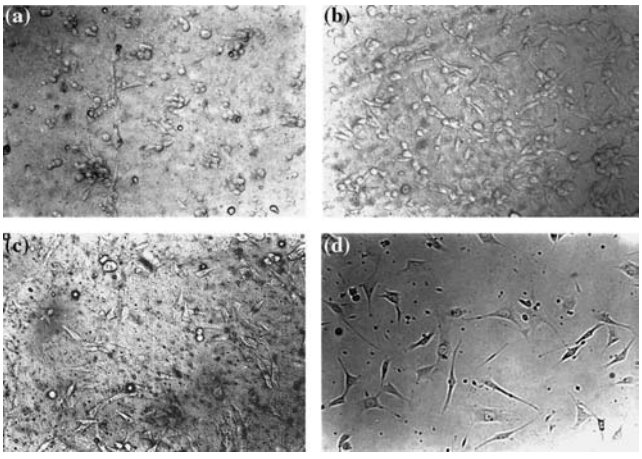


Fig. 5a–d Photomicrograph (×150) of 3T3 fibroblasts cultured on various PDLLA samples for 9 h.**a** Control;**b** modified by NH_3 plasma (50 w, 20 Pa, 300 sec);**c** modified by collagen coating;**d** modified by ammonia plasma pre-treatment then collagen anchorage

The results of cell culture experiments showed that the cell affinity of the PDLLA surface after a combination of plasma treatment and collagen anchorage was greatly improved. After being cultured for a short time, the cells stretched very well on the collagen-anchored sample, which was much better than the individual plasma-modified or collagen-coated samples, as shown in Fig. 5.

The depth that could be modified by NH_3 plasma treatment was measured. It was found that the plasma modifying depth could reach as deep as 4.0 mm for a three-dimensional PLLA scaffold [77]. Under shear conditions, the cells were almost completely removed from the untreated PLLA in a short time, but for the PLLA modified by combining plasma treatment and collagen anchorage, the cells detached very slowly. After 90 min under shear stress, a large number of cells still remained on the PLLA films.

Conclusions

Although PLA-type polymers have many good properties, their hydrophilicities, biocompatibilities, and cell affinities are still not good enough for some tissue engineering and other biotechnology uses. Focusing on the application of PLA-type polymers for medical use, we have reviewed methods of modifying PLA-type polymers by bulk modification and surface modification. Using bulk modification, such as copolymerization with other lactone-type monomers, PEG, monomers with functional groups, and blending with other materials, the degradation rates, hydrophilicities, mechanical properties and surface properties of PLA-type polymers can be significantly improved. Moreover, surface modifications of the polymers, such as surface coating, chemical modification, plasma treatment, and a combination of plasma treatment with collagen anchorage can improve the cell affinities of PLA-type polymers. This ability to improve the properties of PLA-type polymers, giving them excellent biocompatibilities, biodegradabilities and cell affinities, points to a promising future for them in medical science and particularly in tissue engineering, DDS and other human health care fields.

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