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One-Pot Synthesis of Brush-Like Polymers via Integrated Ring-Opening Metathesis Polymerization and Polymerization of Amino Acid *N*-Carboxyanhydrides

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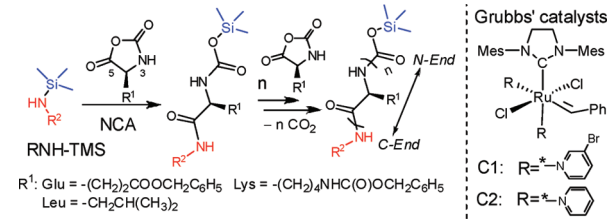
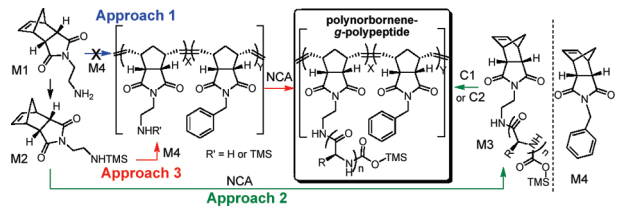
The preparation of brush-like polymers using controlled polymerization techniques, such as anionic, radical, and ring-opening metathesis polymerization (ROMP), has attracted much attention in the past decade.[1] Brush-like polymers derived from these methods contain flexible side-chain polymers, such as poly(methyl methacrylate), poly(styrene), and poly(ethylene glycol).[2] Incorporating polypeptides that have intrinsic secondary structures into the brush side chains could significantly expand the horizon of brush-like macromolecules by providing materials with unprecedented properties. However, reports on brush-like macromolecules bearing polypeptide side chains are scarce, and most are focused on grafting oligo- or polypeptides to the backbone polymers. These works primarily utilize the so-called "grafting to" or "grafting through" strategy, which has no control over the site of grafting.[3] There have also been reports of grafting polypeptides through polymerization of amino acid *N*-carboxyanhydrides (NCAs) onto a backbone polymer bearing amine functional groups.[4] However, amine-initiated NCA polymerizations proceed through a complex mechanism and give polypeptides with poorly controlled molecular weights (MWs). We recently reported a new type of controlled NCA polymerization initiated by the *N*-trimethylsilyl (*N*-TMS) group (Scheme 1).[5] We report here the integration of this controlled NCA polymerization with ROMP, which allows for an unprecedented one-pot synthesis of brush-like polymers bearing polypeptides as the brush side chains.

Three NCAs, γ-benzyl-L-glutamate NCA (Glu-NCA), ε-CBZ-L-lysine NCA (Lys-NCA), and L-leucine NCA (Leu-NCA) (Scheme 1 left), two Grubbs' catalysts (C1 and C2, Scheme 1 right),[6] and three norbornene compounds, 5-norbornene-endo-2,3-dicarboximide (M1) and its derivatives (M2 and M4) (Scheme 2), were used in this study. To facilitate controlled NCA polymerization, the polymer backbone derived from ROMP should have the desired *N*-TMS group.[5b] We first attempted to prepare such polymers by ROMP of an amine-containing norbornene (M1) or a monomer mixture (M1 and M4) (Approach 1, Scheme 2). No ROMP of M1 was observed in the presence of either C1 or C2. ^{1}H NMR analysis indicated that M1 remained largely intact (>97%), presumably due to the deactivation of C1 or C2 by the amine of M1 (P1, Table 1).[7] We also attempted to use the "grafting through" strategy (Approach 2, Scheme 2). M2 was first utilized to polymerize Glu-NCA to make a norbornene-terminated polypeptide macromonomer (M3) that contained the desired *N*-TMS group.[8] However, when this macromonomer was utilized for ROMP using a variety of monomer/initiator (M/I) ratios, polymers with uncontrolled and broad molecular weight distributions (MWDs) were obtained (data not shown).

Scheme 1



Scheme 2



TMS can be used as an amine-protecting group since the *N*-TMS amine has reduced nucleophilicity compared to the parent amine. We reasoned that M2, a norbornene containing the *N*-TMS group, would likely make a successful ROMP monomer and that the polymers resulting from the ROMP of M2 might function as macromolecular initiators for subsequent NCA polymerization (Approach 3, Scheme 2). As expected, poly(norbornene dimide)s with well-controlled MWs and narrow MWDs were obtained in C1-catalyzed ROMP of M2 at M/I ratios ranging from 11 to 100 (P2−P5, Table 1). C1 displayed remarkable activity—the polymerization was completed within 30 min. To control the polypeptide grafting density of the brush-like polymers,

Table 1. Poly(norbornene diimide)s Prepared by ROMP

polymer	catalyst	monomer(s)	M_{tot}/I	M_n (M_n*)[a] (× 10^3 g/mol)	MWD (M_w/M_n)
P1	C1	M1	60	no reaction	/
P2	C1	M2	11	2.0 (2.3)[b]	1.13
P3	C1	M2	20	3.5 (4.1)[b]	1.27
P4	C1	M2	50	12.5 (15.3)[c]	1.20
P5	C1	M2	100	27.6 (30.6)[c]	1.13
P6	C1	M2+M4[d]	80	19.3 (19.2)	1.07
P7	C2	M2+M4[d]	60	12.5 (14.9)	1.02
P8	C1	M2+M4[d]	130	29.9 (31.4)	1.11
P9	C1	M2+M4[d]	70	16.2 (16.7)	1.04
P10	C1	M2+M4[d]	90	23.4 (21.2)	1.04

[a] M_n = the obtained M_n; M_n* = the expected M_n. [b] Determined by MALDI-TOF MS. [c] Measured by GPC after converting the P-NHTMS to P-NHBoc (P = P4 or P5). [d] Random copolymerization, M4/M2 (molar ratio) = 60:20 (P6), 50:10 (P7), 100:30 (P8), 50:20 (P9), and 60:30 (P10).

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Table 2. Synthesis of Poly(norbornene)-*g*-Polypeptides

entry	polymer	cat.	x (x*)[a]	y (y*)[b]	NCA	n (n*)[c]	M_n (M_n*)[d] ($\times 10^4$ g/mol)	MWD (M_w/M_n)
1	P2-*g*-Glu_{50}	C1	9(11)[e]	0	Glu	50(50)	10.0(12.3)	1.05
2	P2-*g*-Glu_{100}	C1	9(11)[e]	0	Glu	113(100)	22.5(24.3)	1.10
3	P2-*g*-Glu_{200}	C1	9(11)[e]	0	Glu	185(200)	38.7(48.4)	1.14
4	P3-*g*-Glu_{50}	C1	17(20)[e]	0	Glu	50(50)	19.1(22.3)	1.15
5	P6-*g*-Glu_{30}	C1	23(20)	58(60)	Glu	34(30)	19.0(15.1)	1.03
6	P7-*g*-Glu_{30}	C2	6(10)	42(50)	Glu	32(30)	5.5(8.0)	1.05
7	P7-*g*-Lys_{30}	C2	6(10)	42(50)	Lys	37(30)	7.2(9.3)	1.13
8	P8-*g*-Glu_{100}	C1	28(30)	95(100)	Glu	81(100)	52.5(57.0)	1.04
9	P10-*g*-($_{10}Glu_{30}$-*b*-Leu_{10})	C1	28(30)	70(60)	Glu +Leu	25/7 (30/10)[f]	19.7(25.2)	1.13

[a] x = the obtained degree of polymerization (DP) of poly(M2), x* = the expected DP of poly(M2). [b] y = the obtained DP of poly(M4), y* = the expected DP of poly(M4). [c] n = the obtained DP of polypeptides, n* = the expected DP of polypeptides. [d] M_n = the obtained M_n; M_n* = the expected M_n. [e] Determined by MALDI-TOF MS. [f] Obtained DP of PBLG/poly(Leu) (the expected DP of PBLG/poly(Leu)).

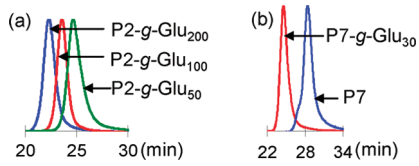

Figure 1. (a) GPC overlay of the P2-*g*-Glu_{50}, P2-*g*-Glu_{100}, and P2-*g*-Glu_{200}; (b) GPC overlay of P7 and P7-*g*-Glu_{30}.

we evaluated the random copolymerization of M2 with comonomer M4 (P6−P10, Table 1). Well-controlled polymerizations were also observed at various M2/M4 ratios. The resulting polymers all have well-controlled MWs and low MWDs (M_w/M_n = 1.02−1.14). ^{1}H NMR studies confirmed that the TMS groups were quantitatively preserved in these polymers.[8]

We then tested whether these polymers could be used as initiators for controlled NCA polymerization. Polynorbornenes containing *N*-TMS amine groups were prepared via ROMP in THF followed by the removal of the solvent and the pyridine ligand of C1 or C2 under vacuum. Anhydrous DMF was added to dissolve the resulting polymers, followed by the addition of Glu-NCA at various NCA/M2 ratios (entries 1−3, Table 2). The NCA polymerizations initiated by the pendant amine-TMS group of the ROMP polymers proceeded in a similar fashion to those initiated by *N*-TMS amine containing small molecules,[5] and they gave polynorbornene-*g*-poly(γ-benzyl-L-glutamate) (PBLG) with the expected MW and narrow MWD. Polynorbornene-*g*-PBLG is denoted as Pn-*g*-Glu_m, where "Pn" corresponds to the poly(norbornene) shown in Table 1 and "m" is the Glu-NCA/M2 ratio. Monomodal GPC distribution patterns were observed with all three brush-like polymers derived from P2 containing PBLG brush side chains of different lengths (Figure 1a). PBLG side chains as short as 20 repeating units could be readily obtained (data not shown). Controlled NCA polymerizations were also observed with the use of random copolymers as macroinitiators. P7, a copolymer that has an M2/M4/C2 ratio of 6:42:1 (M2/M4/C2 feeding ratio of 10:50:1), was used as the macroinitiator for Glu-NCA polymerizations. At a Glu-NCA/M2 ratio of 30, P7-*g*-Glu_{30} with an M_n of 5.5 $\times$ 10^4 g/mol was obtained, which corresponded to a degree of polymerization of 32 (entry 6, Table 2). GPC analyses of P7 and P7-*g*-Glu_{30} demonstrated the formation of a brush-like polymer (Figure 1b). No homo-PBLG was detectable in the polymerization solution.[8]

This technique can be expanded to other types of NCAs. When the polymerization of Lys-NCA was mediated by P7 at a Lys-NCA/M2 ratio of 30, P7-*g*-Lys_{30} with the expected MW and a very narrow MWD was obtained (entry 7, Table 2). Brush-like polymers containing block copolypeptide arms could also be readily prepared through this one-pot polymerization strategy. Sequential addition of Glu- and Leu-NCA to P10 at a Glu-NCA/M2 ratio of 30 and a Leu-NCA/M2 ratio of 10 resulted in P10-*g*-(Glu_{30}-*b*-Leu_{10}) with the anticipated composition, MW, and narrow MWD (entry 9, Table 2). Kinetic studies have shown that P8-mediated Glu-NCA polymerization is only marginally slower than the polymerization mediated by M2.[8] The M_n values of the P8-*g*-PBLG that were collected by terminating the P8-mediated Glu-NCA polymerization at selected time intervals were linearly correlated with the conversion of NCA,[8] demonstrating that the amine-TMS groups of P8 mediated living NCA polymerizations.[5b]

To investigate the secondary structure of the brush polymers, circular dichroism (CD) spectrometry was performed using P6-*g*-Glu_{30} and P3-*g*-Glu_{50}. CD analyses showed that the PBLG brush side chains of both polymers indeed adopt an α-helical conformation.[8] It is known that brush-like hybrid polymers may form nanoaggregates, such as micelles.[9] When methanol was added to a DMF solution of P7-*g*-Glu_{30}, the colorless solution turned slightly blue, and strong light scattering was observed, suggesting the formation of aggregates. Analysis of these aggregates using TEM revealed that they adopted spherical structures with sizes around 60−150 nm. Exploration of detailed assembly mechanisms is underway.

In conclusion, we have demonstrated an unprecedented strategy of integrating ROMP and ROP of NCAs to make brush-like polymers containing polypeptide side chains. Given that numerous ROMP and NCA monomers are widely available, we believe that this novel polymerization technique will allow easy access to numerous hybrid materials with broad applications.

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Supporting Information Available: Experimental procedures and NMR spectra of polymers. This material is available free of charge via the Internet at http://pubs.acs.org.

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