Tuning the Mechanical Properties of Bioinspired Catechol Polymers by Incorporating Dual Coordination Bonds

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Coordination bonds between catechol-functionalized proteins and metal ions serve as sacrificial bonds that play essential roles in the expression of good mechanical performance and the self-healing properties of mussel byssal threads, which has inspired researchers to develop a range of catechol-based functional polymers. Herein, we investigated the mechanical properties of catechol-functionalized network polymers crosslinked by dual coordination bonds, namely catechol–Ca and catechol–B. As the bonding strengths are different, the viscoelastic properties of these materials can be controlled by changing the relative concentrations of these two bonds. The dual-coordination polymer with a 75:25 ratio of catechol–B:catechol–Ca (P(B75:Ca25)) exhibited a higher Young’s modulus and break strength than the corresponding single-coordination polymer (PCa100 or PB100). The chemical structure of the polymer itself remained unchanged; hence this is a facile experimental system for solely investigating the effects of dynamic bonds on the mechanical properties of network polymers.

Keywords: Supramolecular network, Dynamic bonding, Coordination, Microphase separation, Ionic cluster

1. Introduction

Marine mussels can firmly attach themselves to almost any surface using byssal threads, even in wet environments, while most synthetic glues fail to do so [1,2]. Studies have revealed the presence of peculiar catechol-functionalized proteins in the threads synthesized by the post-translational modification of tyrosine residues. These proteins play pivotal roles in universal adhesion [3]. Furthermore, in order to accommodate large strains applied repeatedly in a wave-swept seacoast without detaching itself from the rock surface, the mussel cleverly overcomes the high hardness that is usually lost at the expense of extensibility (i.e., a tradeoff relationship). This is achieved by reinforcing the thread with a protective protein coating (cuticle) that is five times harder (~0.1 GPa) than the thread core [4]. The cuticle has a multiphase sea-island structure in which the densities of the catechol–Fe crosslinks are higher in the island phases than in the sea phases [5]. As the coordination bonds between metal ions and the catechol moieties are reversible [6], they can function as sacrificial crosslinks that dissipate energy and limit crack propagation.

Inspired by this sophisticated catechol-functionalized biopolymer, researchers have recently developed a range of functional materials that include versatile coatings [7,8], wet adhesives [9,10], pH-responsive capsules [11,12], self-healing gels [13–15], and elastomers [16,17]. Of particular importance, Holten-Andersen et al. reported a self-healing gel prepared from a catechol-terminated tetra-PEG complexed with Fe3+ [13]; the use of Ti3+ instead of Fe3+ resulted in gels with different viscoelastic properties. Birkedal synthesized catechol-functionalized polyallylamine and varied the coordinating metals in group 13 from Al3+ to Ga3+ and In3+ [15]. The modulus was observed to decrease in the same order, which is
consistent with the acidic hardness of the cations based on the theory of hard and soft acids and bases (HSAB) [15]. Complexation stability also followed this order, as catechol is a hard basic ligand. This systematic study showed that the mechanical properties of catechol-functionalized gels can be finely tuned through the judicious choice of the metal ion.

In 2016, we reported the seawater assisted self-healing of catechol-functionalized elastomers complexed with Ca\(^{2+}\) [16]; the Young’s modulus and break strength increased with increasing amounts of added Ca\(^{2+}\), suggestive of the formation of crosslinks. The polymer self-healed with high efficiency (78% based on toughness) in seawater owing to the shuffling of catechol–Ca\(^{2+}\) bonds. However, severe water uptake and swelling occurred (~20 wt%) when the polymer was fully immersed in seawater. The use of benzene-1,4-diboronic acid (BDBA) instead of Ca\(^{2+}\) as the cross-linker solved this issue and resulted in a higher self-healing efficiency (91% based on toughness) and long-term stability in seawater (at least 1 month) [17]. Herein, we combine these two coordination motifs (catechol–Ca and catechol–B) in one network polymer. The mechanical properties of network polymers with varying catechol–Ca/catechol–B ratios were systematically investigated in order to understand the effects of these crosslinks on the bulk mechanical properties of the resulting material.

Fig. 1. The chemical structure of the P(DA-co-BA) catechol-functionalized polymer.

2. Experimental
2.1. Materials
All solvents, including methanol, chloroform, ethanol (super dehydrated), and tetrahydrofuran (super dehydrated, stabilizer free), as well as triethylamine, butyl acrylate, 2,2’-azobis(isobutyronitrile) (AIBN), calcium chloride, and benzene-1,4-diboronic acid (BDBA) were obtained from Wako Pure Chemical Industries. Lithium chloride was purchased from Sigma-Aldrich, while pyrocatechol was purchased from TCI. Butyl acrylate was purified by passing it through basic alumina before use. AIBN was purified by recrystallization from diethyl ether.

2.2. Synthesis of P(DA-co-BA)
Full synthesis details for P(DA-co-BA) (Fig. 1) have been reported previously [16]. Briefly, to a nitrogen-purged 200 mL Schlenk flask were added dopamine acrylamide (DA, 3.73 g, 18.0 mmol), AIBN (148 mg, 0.643 mmol), n-butyl acrylate (BA, 23.3 mL, 21.0 g, 163 mmol), and anhydrous ethanol (90 mL). The solution was degassed by three freeze-pump-thaw cycles and backfilled with nitrogen. The solution was then placed in an oil bath at 75 °C and stirred for 3 h. The solution was then quenched in liquid nitrogen and its volume was reduced to 50 mL by rotary evaporation under reduced pressure. Tetrahydrofuran (30 mL) was then added, and the resulting solution was precipitated from cold water (5 °C) in a polytetrafluoroethylene beaker. The precipitate was collected and dried under vacuum to yield a light-yellow viscous oil (11.8 g, yield 48%). 1H NMR (400 MHz, CDCl\(_3\), \(\delta\): 2.28–0.92 (93H, aliphatic region), 2.69 (2H, CONH-CH\(_2\)-CH\(_2\)), 3.55 (2H, CONH-CH\(_2\)-CH\(_2\)), 4.04 (18H, COO-CH\(_2\)-CH\(_2\)-CH\(_3\)), 6.81–6.59 (3H, Ar). Size-exclusion chromatography (SEC, 10 mM LiCl in DMF as eluent, polystyrene standards): \(M_n = 92,000\), \(M_w/M_n = 2.0\). The molar ratio of DA:BA in P(DA-co-BA) was calculated to be 10:90 by integrating the 1H-NMR signals at 6.81–6.59 ppm (3H), and 4.04 ppm (2H).

2.3. Film preparation
The preparation the PB\(_{50}\)Ca\(_{50}\) network polymer, with a 50:50 ratio of catechol–B:catechol–Ca, serves as a typical example. To that end, P(DA-co-BA) (1.80 g, containing 1.34 mmol catechol) was dissolved in chloroform (30 mL). A solution of calcium chloride (0.335 mmol), BDBA (0.335 mmol), and trimethylamine (0.135 g, 1.34 mmol) in methanol (1.5 mL) was then added. The solution was stirred for 3 h, poured into a Teflon dish, and allowed to evaporate for 24 h at room temperature. The sample was placed in a vacuum oven at 50 °C for another 3 h to remove residual solvent. A film with an average thickness of 0.3 mm was prepared by compression molding (12.5 MPa) between two Teflon disks at 60 °C for 30 min. For the tensile testing, dumbbell-shaped specimens (Fig. 2) were punched from the films using a stainless-steel mold.
2.4. Characterization

SEC was performed on a TOSOH HLC-8220 chromatograph using 10 mM LiCl in DMF as the eluent. A calibration curve was prepared using polystyrene standards. $^1$H NMR spectra were recorded on a JEOL JNM-AL 400 spectrometer in deuterated solvents at 20 °C. Equilibrium constants were determined by mixing pyrocatechol (25 mM) and BDBA (62.5 mM) in the presence of trimethylamine (50 mM) in methanol-d$_4$. After incubating for 24 h, the $^1$H NMR spectrum was acquired. Peaks corresponding to free pyrocatechol (6.63–6.67 and 6.73–6.76 ppm), the mono-complex (6.49–6.52 and 6.57–6.60 ppm), and the bis-complex (6.46–6.49 and 6.54–6.56 ppm) were used to calculate the equilibrium constants. Tensile testing was performed on a SHIMADZU TCE-N300 tensile tester; the specimens were extended at rate of 5 mm min$^{-1}$ at room temperature. Each experiment was conducted at least three times. Films were cut into 20 × 10 mm rectangular shapes for dynamic mechanical analysis (DMA). Data were recorded using a SEIKO DMS6100 instrument in tensile mode over the –40 to 180 °C range at a heating rate of 3 °C min$^{-1}$ and a frequency of 1 Hz with 1% strain. Atomic force microscopy (AFM) was performed using a Nanocute microscope (SII Nano Technology, Inc.) fitted with a self-sensitive PRC-DF40P micro-cantilever in dynamic force mode.

3. Results and discussion

3.1. Catechol-functionalized network polymers crosslinked with single-coordination bonds

We first prepared network polymers crosslinked with single coordination bonds (PCa$_{100}$ and PB$_{100}$). In these polymers, the amount of added crosslinker (Ca$^{2+}$ or BDBA) was half the molar amount of the catechol moieties in P(DA-co-BA), since we assumed that two catechol moieties bind to one crosslinker (Fig. 3).

Figure 4 displays representative tensile-test results for PCa$_{100}$ and PB$_{100}$. The maximum tensile strength recorded for PCa$_{100}$ was 4.26 ± 0.07 MPa, with a strain at break of 243 ± 20%, while PB$_{100}$ exhibited a maximum tensile strength of 2.08 ± 0.39 MPa, with a strain at break of 1085 ± 132%. The Young’s moduli of PCa$_{100}$ and PB$_{100}$ were 3.96 ± 0.29 and 2.12 ± 0.79 MPa, respectively. PCa$_{100}$ was stiffer and less extensible than PB$_{100}$, suggesting that the catechol–Ca bond is stronger than the catechol–B bond.

To understand this difference, we evaluated the equilibrium constants $K_1$ and $K_2$ (Fig. 3) using pyrocatechol as a model compound (Table 1); $K$ is the overall equilibrium constant ($K = K_1 \times K_2$). The values for pyrocatechol–Ca are taken from the literature [16], while the values for pyrocatechol–B were determined in this study (see section 2.4). The overall equilibrium constant ($K$) for pyrocatechol–Ca ($10^{8.70}$ M$^{-2}$) is three orders of magnitude larger than that of pyrocatechol–B ($10^{5.82}$ M$^{-2}$), which confirms that the catechol–Ca interaction is substantially stronger than the catechol–B interaction. Stronger interactions result in a higher number of apparent crosslinks in the equilibrium state. As a consequence, PCa$_{100}$ is mechanically stiffer than PB$_{100}$.

|                        | log $K_1$ | log $K_2$ | log $K$  |
|------------------------|-----------|-----------|----------|
| Pyrocatechol–Ca        | 5.45      | 3.25      | 8.70     |
| Pyrocatechol–B         | 3.26      | 2.56      | 5.82     |
3.2. Catechol-functionalized network polymers crosslinked through dual coordination bonds

A series of network polymers crosslinked with dual coordination bonds were prepared and their mechanical properties examined, the results of which are summarized in Table 2. Here “PBXCa100-X” refers to a polymer in which X mol% of Ca$^{2+}$ crosslinks in PCa100 have been replaced with BDBA crosslinks. Interestingly, PB75Ca25 exhibited the highest Young’s modulus (5.70 ± 0.70 MPa) and maximum tensile stress (4.47 ± 0.13 MPa) among the tested polymers. These values are higher than PB100 and PCa100, suggesting that catechol–Ca and catechol–B enhance mechanical properties through synergism.

Table 2. Physical properties of the PBXCa100-X network polymers.

|                  | Young’s modulus (MPa) | Elongation at break (%) | Maximum tensile stress (MPa) | Toughness (MJ m$^{-3}$) |
|------------------|-----------------------|-------------------------|------------------------------|-------------------------|
| PCa100           | 3.96 ± 0.29           | 243 ± 20                | 4.26 ± 0.07                  | 6.69 ± 0.79             |
| PB5Ca95          | 1.72 ± 0.13           | 313 ± 48                | 4.01 ± 0.69                  | 6.59 ± 1.77             |
| PB25Ca75         | 4.96 ± 0.36           | 83 ± 5                  | 3.16 ± 0.22                  | 1.38 ± 0.13             |
| PB50Ca50         | 3.81 ± 0.26           | 105 ± 16                | 2.95 ± 0.66                  | 1.69 ± 0.51             |
| PB75Ca25         | 5.70 ± 0.70           | 159 ± 16                | 4.47 ± 0.13                  | 4.12 ± 0.40             |
| PB95Ca5          | 2.49 ± 0.07           | 786 ± 127               | 2.21 ± 0.20                  | 13.1 ± 2.11             |
| PB100            | 2.12 ± 0.79           | 1085 ± 132              | 2.08 ± 0.39                  | 14.3 ± 0.32             |

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The dual crosslinking strategy has been previously exploited to toughen hydrogels [18–23]; in these cases, the weaker crosslinks act as sacrificial bonds. When an external loading is applied, the sacrificial bonds rupture and dissipate energy. As these sacrificial bonds can reform and rupture multiple times, the amount of energy per unit volume that a material can absorb before fracturing (toughness) increases. However, in this study, the toughness of each network polymer crosslinked with dual coordination bonds (PB5Ca95, PB25Ca75, PB50Ca50, PB75Ca25, and PB95Ca5) was lower than that of PB100 (Table 2). High toughness is usually achieved by a trade-off between stiffness and yield strength. As these polymers have different stiffnesses, we cannot directly compare their toughness values.

The thermal properties of PB75Ca25 were investigated by dynamic mechanical analysis (DMA) in tensile mode and compared to those of PB100 and PCa100 (Fig. 5). All three samples exhibited decreases in storage modulus (Fig. 5a) and a tan $\delta$ peak at around –10 °C (Fig. 5b, inset), which corresponds to the glass-transition temperature of P(DA-co-BA). Above this glass-transition temperature, the storage modulus of each sample gradually decreased in the elastic rubber state, followed by rapid decreases at ~80°C (PB100) and ~130°C (PCa100), which are ascribed to dissociation of the coordination bonds. At tan $\delta = 1$ (Fig. 5b), the polymers are in transition between solid-elastic behavior and fluid-flow-like behavior due to the dissociation of the coordination crosslinks. Catechol–Ca dissociated at higher temperature than catechol–B, which is consistent with the aforementioned equilibrium constants. In contrast, the decrease in modulus due to the dissociation of coordination bonds was not observed for PB75Ca25 over the tested temperature range (< 180 °C). PB75Ca25 not only exhibited superior mechanical properties but also better thermal stability than PB100 or PCa100.

The nanostructures of thin film samples of P(DA-co-BA), PB100, PB75Ca25, and PCa100 were investigated by AFM (Fig. 6). The AFM phase images of P(DA-co-BA) and PB100 did not show
notable microphase-separated morphologies, which is consistent with expectation, since P(DA-co-BA) is a random copolymer rather than a block copolymer. The AFM image of PB\textsubscript{100} strongly suggests that catechol–B crosslinks are finely dispersed in this copolymer. In contrast, PCA\textsubscript{100} showed a microphase-separated morphology with granular domains (< 100 nm), which indicates that the catechol–Ca crosslinks form aggregates, consistent with that reported previously [17]. Catechol–Ca crosslinks are charged and hydrophilic, consequently they can become segregated from the hydrophobic polymer matrix.

Fig. 6. AFM phase images of (a) P(DA-co-BA), (b) PB\textsubscript{100}, (c) PB\textsubscript{75}Ca\textsubscript{25}, and (d) PCA\textsubscript{100}.

The granular domains enriched by catechol–Ca crosslinks are still clearly visible in PB\textsubscript{75}Ca\textsubscript{25}, in which two types of coordination bonds operate in one network (Fig. 6c). Therefore, the network structure consists of hard spherical domains (rich in catechol–Ca crosslinks) surrounded by the softer matrix (rich in catechol–B crosslinks). This nanostructure, illustrated in Fig. 7, resembles the granular microstructure found in mussel byssus cuticles [5]. Conceivably, this multi-phase nanostructure, with optimum contrast of crosslinking densities (dense in island phases and sparse in sea phases), led to the good mechanical performance of PB\textsubscript{75}Ca\textsubscript{25}.

4. Conclusion
In this study, we successfully tuned the viscoelastic properties of a material by varying the relative concentrations of two types of coordination crosslinks incorporated in a network structure. We found the PB\textsubscript{75}Ca\textsubscript{25} exhibited good mechanical performance. AFM revealed the multiphase sea-island structure formed through self-organization resembles that found in mussel byssus cuticles. It is likely that biomimicking this multi-phase structure, with optimum contrast between crosslinking densities, led to the good mechanical performance of PB\textsubscript{75}Ca\textsubscript{25}. This study also introduced a facile experimental system for investigating the effects of dynamic crosslinks on the mechanical properties of network polymers, since there is no requirement to modify the chemical structure of the polymer itself.

Acknowledgement
This work was partly supported by a research grant from the JGC-S Scholarship Foundation.

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