Tailored Modification of Thioacrylates in a Versatile, Sequence-Defined Procedure

Joshua O. Holloway, Suzan Aksakal, Filip E. Du Prez,* and C. Remzi Becer*

A strategy for the synthesis of sequence-defined oligomers using a selective side-group insertion approach making use of thiophenol-catalyzed amidation reactions is herein reported. In this context, a new thiolactone-based, multi-step, iterative protocol is designed, utilizing thioacrylates in combination with solid-phase synthesis for step-by-step growth, resulting in sequence-defined oligomers. Sequence definition and structure variation are introduced by substituting the thioacrylate side groups with a wide variety of amines. The step-by-step growth of the oligomers is followed by liquid chromatography–mass spectrometry and high-resolution mass spectrometry to determine both conversion and purity.

1. Introduction

The field of polymer chemistry has grown exponentially in recent decades, yielding macromolecules of unprecedented structure and design for wide-ranging applications, most notably through recent advances in controlled polymerizations and click chemistry approaches. [1–5] However, when it comes to complete structural control over the sequenced order in which the monomers are placed, Nature is way ahead of current synthetic materials. [5,6] However, scientists are now experiencing an evolution in synthetic macromolecules that takes inspiration from Nature, resulting in tailor-made, complex architectures, with precise control over the monomeric sequence, as was previously only witnessed in biopolymers such as DNA. [7–11] This has led to the development of the field of sequence-defined macromolecular synthesis. This tailoring of macromolecules on a molecular level is enabled through a series of highly efficient and selective reactions, [12] mostly based on unique, biologically relevant, organic chemical reactions such as amid bond formation, [13] thiol–thioester exchange, [14] and disulfide formation. [15] There are numerous recent examples of biomimetic, synthetic approaches such as molecular knots, which can be found in DNA strands or the synthesis of molecular machines, which resulted in the 2016 Nobel Prize in chemistry, that mimic ribosome. [16,17]

Since its introduction in 2001, [18] “click” chemistry has become a powerful tool across many branches of synthetic chemistry and while it has become one of the most established reactions in polymer science, [4,5,19,20] it is through this technique that many sequence-defined, synthetic approaches have arisen. [9–11,21–26] One of the methods toward sequence-defined synthesis and tailored functionalization of monodisperse oligomers is a thiolactone-based chemical approach [20,21] using a solid-phase, Merrifield-inspired synthesis. [27] The promising versatility of the aforementioned method is achieved by introduction of highly efficient, iterative reactions to create a multistep, protecting group-free, synthetic cycle. First, aminolysis of the thiolactone moiety and a subsequent Michael addition of the in situ generated thiol to the double bond of an acrylic compound occur in a one-pot fashion. Then, re-introduction of the thiolactone group was, after a series of observations, the most effective and precise method to proceed the synthetic cycle. Previously reported approaches resulted in an elegant incorporation of a diverse range of side groups with impressive oligomeric length and purity of structures. [10]

However, the previous use of acrylics in this synthetic cycle was limited to acrylates and a small number of acrylamides, so our first aim was the expansion of the monomeric toolbox with the use of promising sulfur analogues, also known as thioacrylates, to unleash new potential applications. [28] Reversible addition fragmentation chain transfer polymerization (RAFT) of thioacrylates has already been described and the advantage of this monomer class is that it enables various postpolymerization functionalizations, [28] while these reactions were previously restricted to only acrylates with activated esters. By using thioacrylates and adapting the iterative, thiolactone-based protocol, we are herein able to report a redesigned synthesis strategy incorporating thioacrylates for increased side-chain functionalization and further future applications. Ring opening of the thiolactone by aminolysis and subsequent thiol–ene reaction with ethyl thioacrylate allowed the introduction of thioacrylate moieties. Therefore, through a thiophenol-catalyzed amidation reaction, we were able to substitute the thioacrylate side group with any chosen primary amine, resulting in the corresponding amide structure. The amidation reaction of the thioester moiety

J. O. Holloway, Prof. F. E. Du Prez
Polymer Chemistry Research Group
Centre of Macromolecular Chemistry (CMaC)
Department of Organic and Macromolecular Chemistry
Faculty of Science
Ghent University
Krijgslaan 281 S4-bis, Ghent B-9000, Belgium
E-mail: Filip.DuPrez@UGent.be
S. Aksakal, Dr. C. Remzi Becer
Polymer Chemistry Laboratory
School of Engineering and Materials Science
Queen Mary
University of London E1 4NS, London, UK
E-mail: cbecer@qmul.ac.uk

The ORCID identification number(s) for the author(s) of this article can be found under https://doi.org/10.1002/marc.201700500.

DOI: 10.1002/marc.201700500
has opened the possibility to introduce several side-chain functionalities. The limited acrylamide library that can be utilized to also result in such amide structure can, therefore, be overcome and mimicked by using the respective primary amine, which are already available in a broad variety, to achieve complete sequence-control of the monomeric order.

The four-step protocol, which is designed for each monomeric addition to the sequenced chain, is illustrated in Figure 1. Through this iterative approach, multifunctional, oligomeric molecules to the length of an octamer were synthesized from a thiolactone moiety that is immobilized on a 2-chlorotrityl chloride functionalized solid-phase resin. The new approach gives a broader diversity of side-chain functionality and better opportunity for potential new applications, for example, with use of biofunctional/biocompatible amines, while demonstrating postfunctionalization on a sequence-defined level under mild conditions.

2. Results and Discussion

The use of thioacrylates required a new protocol to be designed, somewhat combining previously reported approaches to enable us to harness the advantages of using these acrylics as a Michael acceptor in a thiolactone-based, sequence-defined protocol. The first step of the four-step protocol consists of the reaction between the thiolactone group, attached to a solid phase, with ethanolamine. While the amine function is known to react in a chemoselective way with a thiolactone unit, the alcohol function serves in the last, fourth step as a functional handle to react with α-thiolactone-γ-isocyanate (Figure 1). In order to confirm if the aminolysis (step 1) and Michael-addition reaction (step 2) could be merged into one step with the thioacrylates, as was done for an earlier reported protocol, the reactivity of the acrylic group of the thioacrylate toward an amine has first been compared with that of a regular acrylate (Figure 2). While a slow reaction does occur between the ethyl acrylate and the ethanolamine, as witnessed by a 23% reduction in the integrals of the acrylic proton resonances over 2 h and a shift in the other proton resonances, this reaction is sufficiently slow, so as not to interfere with the relatively fast thiolactone aminolysis.

On the other hand, reaction between the ethanolamine and ethyl thioacrylate was very rapid and was complete within 18 min (as demonstrated by the rapid disappearance of the acrylic protons in Figure 2b), thus illustrating why thiolactone aminolysis was ineffective when both molecules were present.
A synthetic route was found as an alternative way to overcome this side reaction, in which each of the steps was carried out separately. In order to prevent disulfide bond formation from the newly released thiol moieties of thiolactone aminolysis, two equivalents of dimethylphenylphosphine (DMPP) were added to reduce the disulfide bonds as they formed. As it was observed that DMPP also catalyzed the reaction between the ethyl thioacrylate and alcohol group of the applied ethanolamine, it was not possible to first carry out aminolysis, followed by addition of ethyl thioacrylate and DMPP to both reduce the disulfide bonds and catalyze the thiol–Michael addition. On the other hand, the addition of DMPP with the aminolysis caused

Figure 2. $^1$H NMR spectra recorded every 6 min to compare the reactivity of ethanolamine with a) ethyl acrylate and b) ethyl thioacrylate. Measurement at $t = 0$ was also taken for reaction (b) because of the expected fast reaction between ethyl thioacrylate and ethanolamine. Full assignment is detailed in the Supporting Information.
no further problems, in terms of undesired side products or side reactions, which were a concern when compared with another previously reported approach.\textsuperscript{[21]} As such, the free thiol was able to undergo subsequent Michael addition in the following step with ethyl thioacrylate. N,N′-Dimethylformamide (DMF) was selected as the reaction solvent as it provided a low-viscosity reaction medium that for further use within potential applications, purification by preparative high-performance liquid chromatography (HPLC), as typically performed in peptide solid-phase chemistry, would be possible with the final oligomers, as all of the remaining resin had to be cleaved from the oligomer to yield sufficient compound to be fully analyzed by NMR spectroscopy.

A range of functional, primary amines were then selected (Figure S6, Supporting Information) to introduce a wider scope of side groups to the protocol in order to further increase the monomeric alphabet and demonstrate the versatility of this approach. A monomeric cycle was prepared with each amine, to check their compatibility within the protocol. These monomers were then cleaved from the solid-phase resin and fully analyzed, both by LCMS and high resolution mass spectrometry (HRMS) to evaluate the conversion and purity and by 1D and 2D NMR techniques where the various functional side groups were identifiable and the structures were fully assigned. A final oligomer was then prepared, incorporating all of these amines into one, multifunctional, sequence-defined hexamer (Figure 4c). LCMS and HRMS were again measured after each complete cycle and the final oligomer was also analyzed by NMR spectroscopy.

It should be noted that the molecules synthesized on the resin are cleaved from this support using 1% trifluoroacetic acid (TFA). The use of a TFA solution can potentially lead to a slight oxidation of one of the thioether bonds in the desired structure, resulting in a small side peak to the left of the main peak in the LCMS spectrum (as seen below in Figure 4 and demonstrated in Figure S12, Supporting Information). While the LCMS trace of each cycle in the multifunctional hexamer clearly shows that the protocol works (Figure 4c), indeed the purity of this oligomer in the last step appears not to be as high as that of the benzyl-butyl octamers from Figure 4a,b, as evidenced by their LCMS traces in the same figure. A possible explanation for the reduced purity can be that the combination of these varying functional groups into one oligomer introduces unforeseen folding interactions that may limit the efficiency of the synthesis. Folding and structural arrangement is an area that needs more investigation in this field as it is not currently clear how precisely sequence control can affect this. Individually, the primary amines used reacted completely, resulting in pure monomers using this protocol as reported in the Supporting Information. For instance, the tetramer step in Figure 4c appears to be in high purity in comparison to the trimer and pentamer steps, suggesting that the present impurities do not directly affect the subsequent step.

To improve reproducibility and scientific accuracy in such a repetitive, synthetic process, further optimization of this protocol, including automation, would most likely further improve the purity of the resulting oligomers. However, it is worth noting that for further use within potential applications, purification by preparative high-performance liquid chromatography (HPLC), as typically performed in peptide solid-phase chemistry, would be required in a similar way as an organic compound would be purified by, for example, column chromatography before further use.

3. Conclusions

A new chemical protocol for the synthesis of multifunctional and post-modified sequence-defined oligomers has been successfully
demonstrated. The protocol was constructed following $^1$H NMR model studies and careful study of the kinetics and the compatibility of certain steps and reagents to ensure efficient reaction and to eliminate the need for protecting groups. Through this, several sequence-defined oligomers have been synthesized, to demonstrate the versatility of this approach and its potential in terms of side-chain functionalization. We have also shown how advantageous solid-phase chemistry is in such iterative protocols as used in the synthesis of sequence-defined oligomers, as they allow for small-scale reactions, easy handling, and facile purification by extensive washing. In future studies, this protocol will be expanded for the incorporation of more specific functional groups, thus resulting in unique applications for such sequence-defined oligomers, further confirming the need for the advancement of this specific branch of polymer chemistry. Finally, the successful coupling of thiolactone and thioacrylate chemistry has been achieved, thus demonstrating the compatibility of these two classes of molecules in click-inspired reactions and within sequence-defined chemistry.

Supporting Information
Supporting Information is available from the Wiley Online Library or from the author.
Acknowledgements

J.O.H. and S.A. contributed equally to this work. J.O.H. and S.A. would like to acknowledge funding for their Ph.D. scholarships from the European Union’s Horizon 2020 research and innovation program under the Marie Skłodowska-Curie Grant Agreement No. 642083. The authors would like to thank Timothee Courtin (Ghent University) for his assistance with NMR spectroscopy measurements and Jan Goeman (Ghent University) for LCMS and HRMS measurements.

Conflict of Interest

The authors declare no conflict of interest.

Keywords

amidation, sequence-defined polymers, thioacrylate, thiolactone

Received: July 24, 2017
Revised: August 28, 2017
Published online: