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# Exploring Strategies To Bias Sequence in Natural and Synthetic Oligomers and Polymers

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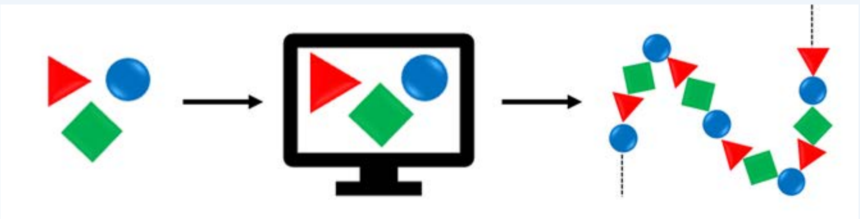


**CONSPECTUS:** Millions of years of biological evolution have driven the development of highly sophisticated molecular machinery found within living systems. These systems produce polymers such as proteins and nucleic acids with incredible fidelity and function. In nature, the precise molecular sequence is the factor that determines the function of these macromolecules. Given that the ability to precisely define sequence emerges naturally, the fact that biology achieves unprecedented control over the unit sequence of the monomers through evolved enzymatic catalysis is incredible. Indeed, the ability to engineer systems that allow polymer synthesis with precise sequence control is a feat that technology is yet to replicate in artificial synthetic systems. This is the case because, without access to evolutionary control for finely tuned biological catalysts, the inability to correct errors or harness multiple competing processes means that the prospects for digital control of polymerization have been firmly bootstrapped to biological systems or limited to stepwise synthetic protocols.

In this Account, we give an overview of strategies that have been used over the last 5 years in efforts to program polymer synthesis with sequence control in the laboratory. We also briefly explore how the use of robotics, algorithms, and stochastic chemical processes might lead to new understanding, mechanisms, and strategies to achieve full digital control. The aim is to see whether it is possible to go beyond bootstrapping to biological polymers or stepwise chemical synthesis. We start by describing nonenzymatic techniques used to obtain sequence-controlled natural polymers, a field that lends itself to direct application of insights gleaned from biology. We discuss major advances, such as the use of rotaxane-based molecular machines and templated approaches, including the utilization of biological polymers as templates for purely synthetic chains. We then discuss synthetic polymer chemistry, whose array of techniques allows the production of polymers with enormous structural and functional diversity, but so far with only limited control over the unit sequence itself.

Synthetic polymers can be subdivided into multiple classes depending on the nature of processes used to synthesize them, such as by addition or condensation. Consequently, varied approaches for sequence control have been demonstrated in the area, including but not limited to click reactions, iterative solid-phase chemistry, and exploiting the chemical affinity of the monomers themselves. In addition to those, we highlight the importance of environmental bias in possible control of polymerization at the single-unit level, such as using catalyst switching or external stimuli.

Even the most successful experimental sequence control approach needs appropriate tools to verify its scope and validity; therefore, we devote part of the present Account to possible analytical approaches to sequence readout, starting with well-established tandem mass spectrometry techniques and touching on those more applicable to specific classes of processes, such as diffusion-ordered NMR spectroscopy. Finally, we discuss progress in modeling and automation of sequence-controlled polymers. We postulate that developments in analytical chemistry, bioinformatics, and computer modeling will lead to new ways of exploring the development of new strategies for the realization of sequence control by means of sequence bias. This is the case because treating the assembly of polymers as a network of chemical reactions will enable the development of control strategies that can bias the outcome of the polymer assembly. The grand aim would be the synthesis of complex polymers in one step with a precisely defined digital sequence.

## 1. INTRODUCTION

Modern biology owes its extraordinary chemical complexity to functional oligomers and polymers, including sugars, proteins, and nucleic acids, which have been designed, refined, and adapted by the process of evolution. Their utility is set by the primary linear sequence of units in the chains, for instance amino acids in proteins and nucleobases in nucleic acids. From this primary sequence, additional secondary, tertiary and even quaternary structural features emerge. The fact that the

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pathophysiology of genetic disorders is manifested by defects in sequence attests to the immense importance carried by the sequence, down to single units whose replacement with seemingly related ones leads to widespread disruption of metabolic activities. Over the years, synthetic polymer chemists have been trying to establish an analogous relationship between structure and function of human-made macromolecules. To do this, they have been trying to devise efficient ways of gaining control of the polymer sequence, but full control of every monomer identity remains a challenging if not impossible prospect. Recent reviews describe progress from the past decade,[1,2] but in this Account we focus on progress since 2013 and show how this ambition and grand vision of sequence control at the molecular level might be achieved.

## 2. SEQUENCE IN NATURAL POLYMERS

Biological systems have almost complete sequence control of polymerization, converting genetic code into defined protein sequences involving three steps: DNA replication, DNA to RNA transcription, and RNA to protein translation. Nature has always performed these complex processes in a precise way, including ways to both limit and correct errors. The ability to implement sequence-controlled polymerization with the precision and efficiency exhibited by biological systems would pave the way toward the development of new types of materials, those of sequence-controlled "inorganic" or non-biological matter. To achieve this aim, many different biological approaches to sequence control have been studied and explored. These include nonenzymatic templating to steer the coupling of simple monomers mainly through Watson−Crick base pairing.[3] In a more complex approach, enzymes have been used to catalyze oligomerization in vitro. A famous example is the polymerase chain reaction (PCR), a process that involves copying and amplification of a certain DNA sequence. Similar methods have also been studied for non-natural nucleic acid polymerization. The most complex yet effective approach to develop sequence-controlled polymerization is the use of proxies in the form of living organisms, usually bacteria, through the introduction of artificial genes. This method is advantageous because it tolerates both natural and non-natural monomers. Chemistry-based systems have also been attempted as an alternative to biological approaches. The concept of the artificial ribosome based upon supramolecular molecular machines was recently developed.[4,5] This approach is based on a rotaxane-based machine that travels along a track of amino acids, coupling them in a sequence-controlled manner according to the movement direction of a thiolated ring (as illustrated in Figure 1).[5]

### Natural Polymers as Templates for Sequence-Specific Polymerization

Sequence-controlled polymerization in natural polymers such as proteins is perfectly controlled by template (macro)-molecules bearing sequence information through which monomers can be selectively recognized and coupled. Inspired by that, natural-template-assisted constrained peptide sequence synthesis and selection were recently achieved for an enzymatically catalyzed mixture utilizing electrostatic interactions between charged amino acids and oppositely charged polysaccharide templates.[6] In the absence of any template, peptide sequence selection could be also achieved under programmable reaction conditions. These are enzyme-assisted

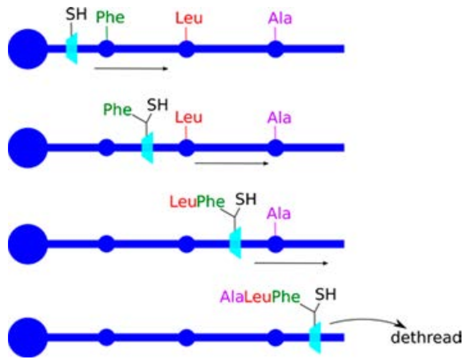


**Figure 1.** Concept of a rotaxane-based molecular machine for sequence-controlled peptide synthesis. Adapted with permission from ref 5. Copyright 2013 AAAS.

dipeptide polymerizations where the sequence of the most thermodynamically stable peptide was selected by the system.[7]

In a recent study, Liu and co-workers developed an enzyme-free, DNA-templated translation system[8] that enabled translation of DNA into sequence-defined synthetic polymers (Figure 2). In this approach, macrocyclic substrates hybridize

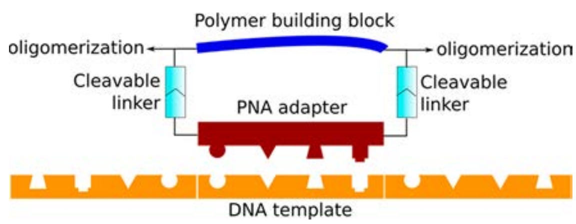


**Figure 2.** Schematic representation of sequence-defined polymerization using enzyme-free, DNA-templated synthesis of non-nucleic acid polymers. Adapted with permission from ref 8. Copyright 2013 Macmillan Publishers Ltd.

with codons on a DNA analogue, peptide nucleic acid (PNA), allowing for polymer building block organization along the template, coupling, and oligomerization. This is followed by linker cleavage, releasing the PNA adapters and liberating the product. By this approach, 16 monomers were successfully coupled in a defined manner to form synthetic polymers with molecular weights of 26 kDa and 90 residues of densely functionalized $\beta$-amino acids.

## 3. SEQUENCE CONTROL IN SYNTHETIC POLYMERS

Sequence-controlled polymerization controls all of biology, but despite many years of developments in polymer science, molecular-level sequence control has not been achieved in any large-scale technology. A key development in the future of polymer science could be the precise sequence control of polymeric materials. In this section, we discuss some promising research directions leading to sequence-controlled synthetic polymers.

### 3.1. Step-Growth and Multistep-Growth Polymers

The common feature of polymerization processes described in this section is the sequential addition of monomers to a growing chain, typically involving functional group coupling and formation of byproducts. There are significant mechanistic differences among members of this group, and they can be accordingly divided into step-growth and multistep-growth processes.


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**Step-Growth Polymers.** Polyesters and polyamides, used in many practical applications in daily life, are leading examples of this type of polymer. Since unit addition takes place by reactions between functional groups on chain ends, sequence control has recently been demonstrated using click chemistry or multicomponent reactions. The click chemistry processes were envisioned by Sharpless[9] as reactions that are fast, highly specific, and high-yielding, furnishing well-defined products under mild conditions. Their utility in polymer science is well-known,[10] and they have also found uses in control of chain sequences. In this regard, the thiol−yne coupling was used by Han and co-workers[11] to produce a sequence-controlled polymer by exploiting successive additions of a thiol group to a carbon−carbon triple bond (Figure 3, top panel). This allowed random, periodic, and linear copolymers to be obtained in a controlled fashion.

The development of multicomponent reactions for polymer science takes advantage of their inherent selectivity and atom economy, in some cases involving multiple substrates. They are useful for polymer chemistry, as well-defined monomers can undergo reactions to furnish a polymer with a specific sequence. In one example, a fast Biginelli reaction between a keto ester, an aldehyde, and urea was used to produce sequence-controlled polymers under mild conditions (Figure 4, left).[12] Another multicomponent process, the Passerini reaction between isocyanides, aldehydes, and carboxylic acids, has also been developed (Figure 4, right).[13] In another example, amine−thiol−ene conjugation followed by alkyne−azide−amine coupling was employed in an elegant sequence that resulted in a well-defined polymer.[14]

**Multistep-Growth Polymers.** This term refers to polymers produced using solid-phase iterative chemistry.[15] In contrast to step-growth polymers, chains grow only on one end, with the other tethered to a support. Unit sequence in such polymers has chiefly been controlled in a similar manner as in solid-phase peptide synthesis, using the same cycles of binding and release.[16,17] One attractive aspect of the fact that the macromolecules used are synthetic is that they can be functionalized with groups allowing for orthogonal chemistry, thereby dispensing with the costly binding and cleavage steps. Peptoids (N-substituted glycine polymers) sit on the boundary between natural and synthetic polymers. Since their first synthesis using a stepwise method,[18] their use has substantially increased, leading to various applications. There is an excellent review on this topic by Zuckermann and co-workers.[19] This method can be also modified to use liquid-phase-based supports, such as native polystyrene chains[20] or fluorinated hydrocarbon chains (Figure 5).[21]

Phosphoramidite coupling,[16,22] historically used in oligonucleotide synthesis, has also been applied in the synthesis of sequence-controlled polymers (Figure 3, bottom panel), with the nature of the phosphoramidite bond allowing for easy sequence readout using mass spectrometry.

**Outlook.** Sequence control in iteratively synthesized polymers is an extremely dynamic field, with many more examples to be found in recent comprehensive reviews.[23] Many of the step-growth processes described above lead to polymers that cannot adequately be termed "sequence-controlled" but rather are "sequence-defined". The subtle but important difference lies in the fact that the experimentalist defines the sequence by setting up the materials to take part in a multicomponent reaction, or in other words, the sequence is defined by the functional groups and the characteristics of the

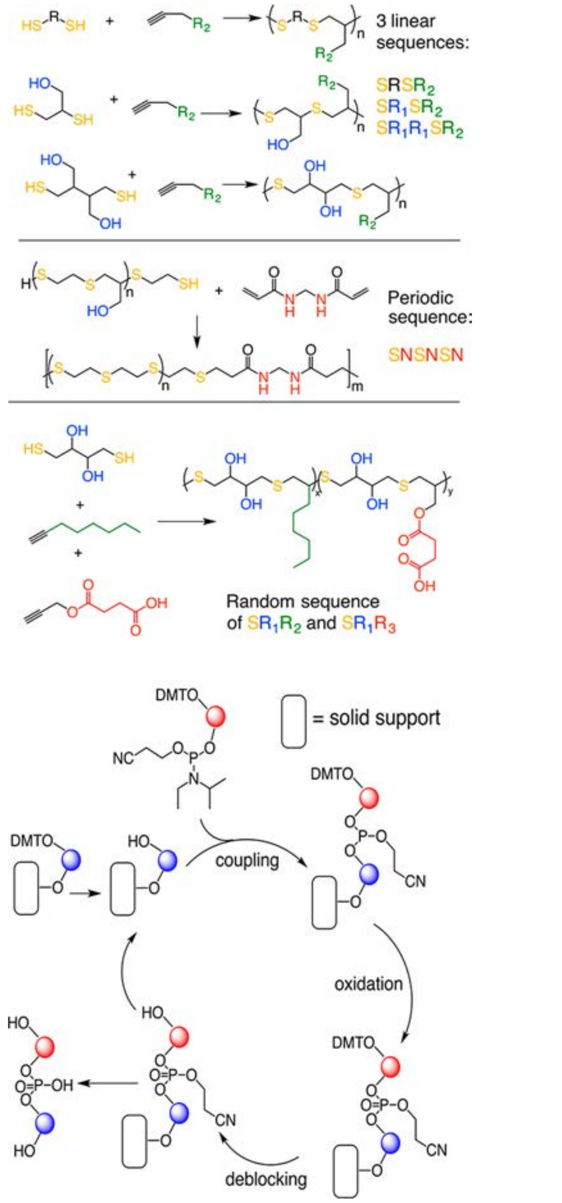


**Figure 3.** Click approaches used in producing sequence-controlled polymers. (top) Thiol−yne coupling in step-growth polymerization. Adapted with permission from ref 11. Copyright 2014 Macmillan Publishers Ltd. (bottom) Phosphoramidite chemistry in a multistep-growth process. Adapted from ref 22. Copyright 2016 American Chemical Society.

reaction. On the other hand, in a true "sequence-controlled" process, the sequence emerges as a result of, e.g., external factors acting in a sequential manner or sequential addition of monomers. Perhaps the closest to that aim are results presented by Du Prez, Madder, and co-workers,[24] who utilized successive thiol−ene couplings to build up a polymer chain in a sequence determined by the order in which the monomers were added.

Sequential addition is at the core of multistep-growth processes, with recent reports indicating that polymers up to 100 units long can be obtained by harnessing DNA synthesizers in a solid-phase protocol.[25] Further advances are undoubtedly to be made in liquid-only systems. For instance, liquid-handling robots that can produce well-defined volumes of reactants could be used to control reactions at liquid−liquid interfaces between droplets (see Figure 6).[26] A promising extension to


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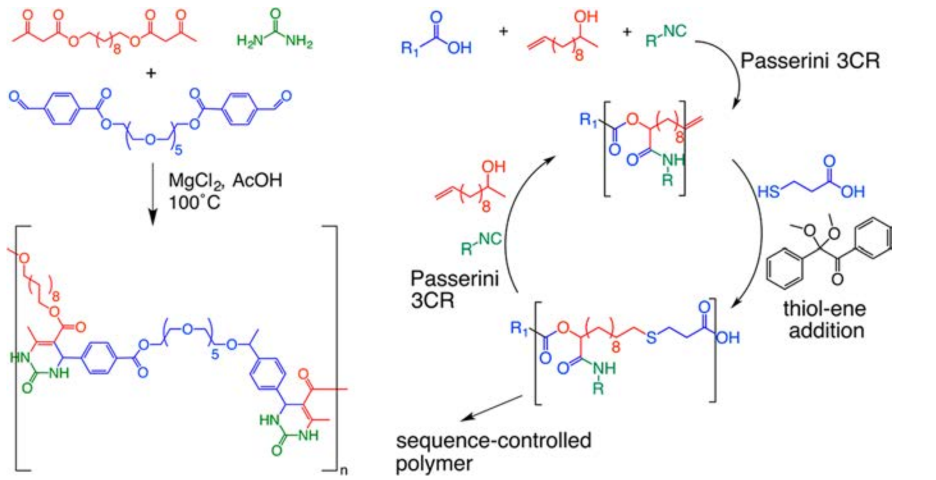


**Figure 4.** Three-component (left) Biginelli (adapted from ref 12; copyright 2016 American Chemical Society) and (right) Passerini (adapted with permission from ref 13; copyright 2014 Wiley) reactions and their utility for controlling sequence in step-growth polymerization.

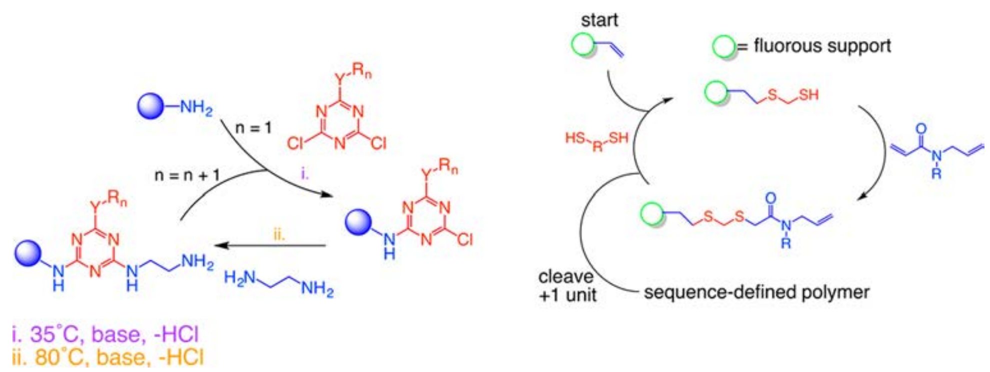


**Figure 5.** Selected iterative approaches to sequence control in multistep-growth polymers based on (left) solid (adapted with permission from ref 17; copyright 2016 the authors, published by Wiley-VCH) and (right) liquid (adapted from ref 21; copyright 2014 American Chemical Society) supports.

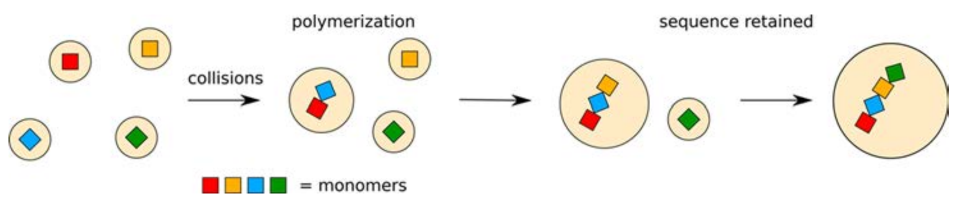


**Figure 6.** Collisions of monomer-containing droplets in a specific sequence leading to retention of that sequence in the resulting polymer.

the realm of sequence control could feature functionalized monomers dissolved in individual droplets and buildup of chains upon individual droplet collisions. This would necessarily have to take into account the time-scale disparity between the collisions and the relevant reaction rates. As a result, the coupling would have to be relatively fast. One promising candidate could be triazolinedione chemistry.[27]

### 3.2. Chain-Growth Polymers

In contrast to step-growth polymers, the propagation of chain-growth polymers is relatively fast because of the presence of reactive intermediates such as carbocations, carbanions, or free radicals. Ionic and radical variants both lend themselves to sequence control, typically by chemical stimuli.

**Free Radical Polymerization.** There has been immense progress regarding molecular weight control in free radical polymerization processes over the last 20 years, and as a result, most of these reactions can now be run in a controlled fashion. This is because of the emergence of "living" free radical polymerization approaches, such as atom transfer radical polymerization (ATRP), reversible activation-fragmentation chain transfer polymerization (RAFT), and single electron transfer living radical polymerization (SET-LRP). Further control of sequence in these reactions can be imposed in multiple ways. As an example, sequence-controlled multiblock polyacrylates were obtained by Haddleton and co-workers by simple addition of the monomers in a desired sequence to a UV-light-controlled SET-LRP medium using a copper complex as a catalyst.[28] On the other hand, using the RAFT process, Perrier and co-workers demonstrated the efficient sequence-controlled synthesis of multiblock copolymers, with each block being up to 100 units long.[29]

**Ionic Polymerization.** In ionic polymerization, carbocations or carbanions (as opposed to free radicals) are the active species. The chain length distribution of the resulting polymers is much more tightly controlled, but the active species are much


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more sensitive to impurities such as oxygen. Nevertheless, these reactions have been used in sequence-controlled polymerizations. In one example, Kanazawa and Aoshima[30] demonstrated cationic terpolymerization of vinyl ethers, oxiranes, and ketones with strict selectivity of the units, resulting in repeating vinyl ether–oxirane–ketone sequences. Wurm and co-workers[31] reported the polymerization of a mixture of up to five different aziridine-based monomers with substituents characterized by varied electron-withdrawing strengths. The result was sequence control stemming from reactivity differences: the most reactive monomers were completely consumed in the amount of time in which the least reactive reached only 20% conversion. In another recent example, diphenylethylene derivatives were copolymerized with either styrene or butadiene to provide perfectly alternating or telechelic copolymers.[32] This strategy was based on steric hindrance provided by diphenylethylene, which is unable to polymerize on its own.

**Ring-Opening Polymerization.** The characteristic feature of ring-opening polymerizations is the fact that the monomers are cyclic and chain growth takes place through successive ring-opening and addition of the resulting segments to the active center. Lactones, lactams, and cyclic carbonates are examples of typical monomers; sequence control is typically achieved by modifying the properties of polymerization catalysts in order to influence their affinity for a particular monomer class. Li, Guo, and co-workers implemented this approach by switching the catalyst between Brønsted acidic (optimized for cyclic lactones) and basic/conjugate acidic (efficient in L-lactide polymerization) through addition of a Brønsted base.[33] Another approach involved encoding a sequence in a macrocycle, which was then polymerized in an entropy-driven fashion.[34]

**Outlook.** Other than the approaches described above, the sequence in polymer chains has been imposed by exploiting differences in reactivity between specific functional groups in these chains, as demonstrated by Kamigaito in the polymerization of maleimide and styrene (or limonene) units, whereby the maleimide units were further functionalized with sequence-defined side chains,[35] and by Sawamoto, whose group demonstrated individual addition of bulky methacrylate units followed by their transesterification with different alcohols to obtain sequence-defined polymers.[36]

Examples of external stimuli that could conceivably be used to control processes of this type are illumination and oxidation–reduction potential (Figure 7). Redox-responsive catalysts for ring-opening polymerizations were first described by Diaconescu and co-workers.[37] Briefly, by changing the oxidation state of the catalytic metal center, the catalyst not only can be switched between active and inactive states but also can be made active toward different classes of monomers. One can thus envision a mixture of monomers exposed to changing redox states of a catalyst that could give rise to chemoselective emergence of sequence-controlled polymers. Indeed, this was recently achieved by two groups: Williams and co-workers[38] were able to show that mixtures of monomers selected from four different classes can be selectively polymerized by switching the dizinc catalyst between different oxidation states (Figure 8), whereas Byers et al.[39] demonstrated similar behavior for mixtures of lactides and epoxides in the case of an iron-based catalyst.

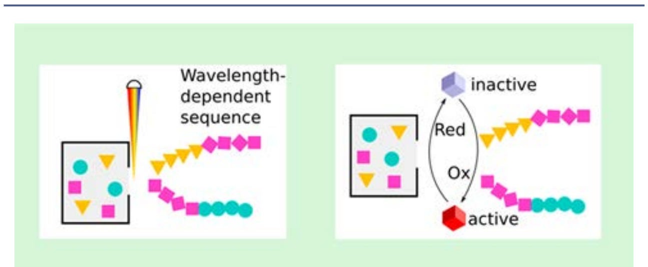


Figure 7. Different aspects of the external environment as factors controlling sequence distribution: (left) light and (right) oxidation–reduction potential.

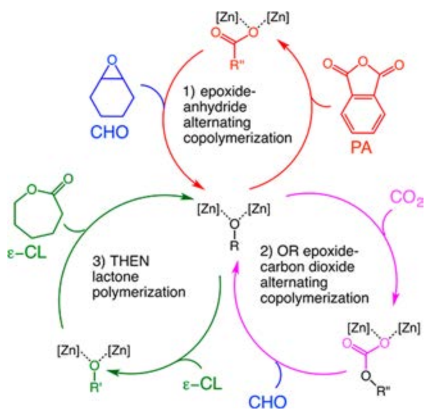


Figure 8. Example of chemoselective emergence of a sequence-controlled copolymer from a mixture of competing monomers determined by the electron-withdrawing strengths of substituents in ring-opening polymerization. Adapted from ref 38. Copyright 2016 American Chemical Society.

The Boyer group has hinted at a major step toward illumination-controlled oligomer sequence emergence by showing that illumination can impose sequence control on the level of individual monomer units.[40] A related study by the same group[41] further showed that the light wavelength used is able to effectively select a species from a mixture of monomers. This suggests a possible generalization wherein the wavelength of light acts as an environmental factor controlling the sequence emerging from a mixture of monomers.

### 3.3. Multistep Flow Synthesis and Iterative Exponential Growth (Flow-IEG)

Iterative exponential growth (Flow-IEG) combines multistep continuous flow chemistry and polymer synthesis for semi-automated synthesis of polymers, as shown by Jamison and co-workers.[42,43] They have chosen a copper-catalyzed azide–alkyne cycloaddition reaction to polymerize an ester monomer functionalized with a triisopropylsilyl (TIPS)-protected alkyne and an alkyl bromide, as illustrated in Figure 9. This approach was successfully validated for achieving sequence-controlled polymerization by targeting an alternating sequence $(ABAB)_n$ and a sequence with an $(AABB)_n$ repeating unit. As a result, pure high-molecular-weight polymers were obtained. The user-friendly nature, scalability, and modularity of Flow-IEG provides a general strategy for the automated synthesis of sequence- and architecture-defined uniform macromolecules.

## 4. HYBRIDS

Polymer hybridization is of great interest, especially when it comes to developing materials with new properties. However, achieving hybrid polymerization by combining natural and non-natural building blocks in a sequence-defined manner is still a


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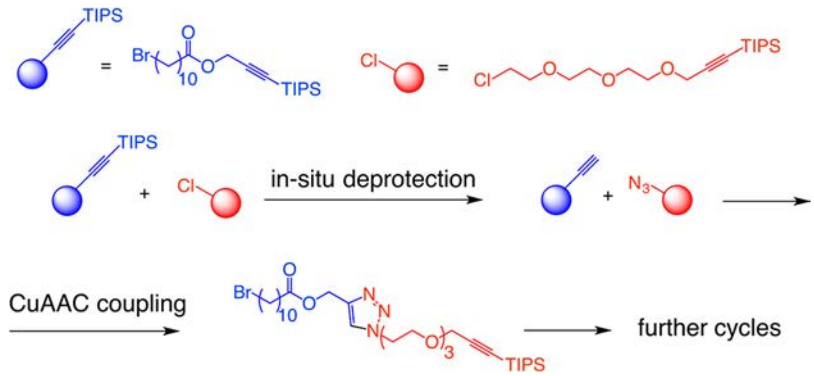


**Figure 9.** Example implementation of sequence-controlled polymerization based on Huisgen copper-catalyzed azide−alkyne cycloaddition in a Flow-IEG process. Adapted with permission from ref 43. Copyright 2015 PNAS.

challenge. To achieve this, Sleiman and co-workers recently coupled perfluorocarbons with nucleic acids and other non-natural polymers. This was accomplished by means of an automated approach using phosphoramidite chemistry, as illustrated in Figure 10.[44] As a result of the introduced

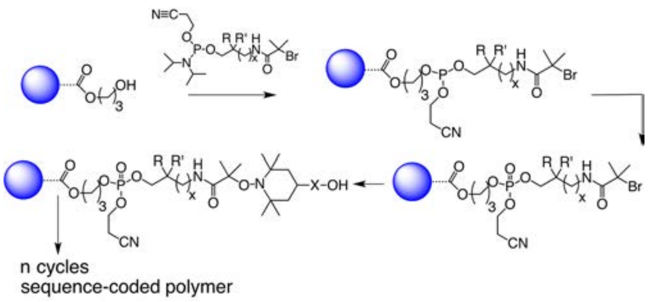


**Figure 10.** Sequence-controlled oligomer synthesis with phosphoramidites grafted sequentially onto a growing chain attached to a solid support. Adapted with permission from ref 44. Copyright 2016 Royal Society of Chemistry.

perfluorocarbon chains, the thermal stability and nuclease resistance of the DNA strands was significantly improved (by up to 20 °C), which resulted in self-assembly of monodisperse micellar nanoparticles.

## 5. ANALYSIS, AUTOMATION, AND MODELING

### 5.1. Analytical and Sequencing Methods

Natural, enzymatically controlled poly- and oligo-merization processes have been invented by evolution with numerous safety checks and correction steps that ensure that sequence fidelity is preserved. However, in the case of synthetic polymers, every novel sequence control protocol necessarily needs reliable analytical tools to confirm that the attempted sequential polymerization was indeed successful. Tandem mass spectrometry (MS/MS) has historically been the technique of choice for investigating sequences of easily fragmented polymers such as poly(alkoxyamine amide)s or poly(triazole amide)s. In recent reports,[45,46] Lutz, Charles, and co-workers described sequence readout using MS/MS with electrospray ionization (ESI) as a means of retrieving binary information that was earlier encoded in the chains. These techniques are best suited to polymers containing easily cleavable ether or amide functions. In contrast, the main chains of common vinyl polymers are built up entirely of carbon atoms and are thus nowhere near as amenable to fragmentation. For such polymers, information from several different techniques must be collected to obtain reliable unit sequence details. As a starting point, molecular weight, and thus the degree of polymerization, must be obtained. This is typically provided by gel permeation chromatography/size exclusion chromatography but requires polymer purification and extensive calibration. In situ molecular weight measurements on complex monomer/polymer mixtures can be more conveniently conducted using diffusion-ordered NMR spectroscopy (DOSY).[47]

Matrix-assisted laser desorption ionization mass spectrometry (MALDI-MS) is a technique that has long been used to analyze molecular weight distributions of synthetic polymers. These include copolymers produced from substrates with different reactivities and sequence analysis in alternating copolymers.[32,48] This technique has limitations that preclude its usefulness for detailed readout of sequence in high-molecular-weight polymers, but for other cases, the combination of unit ratios obtained from NMR measurements with molecular weights measured by MALDI-MS is potentially the most powerful. If the respective monomer propagation rates, as well as addition and sequence times, are known, a descriptive model could give unambiguous averaged sequences for the produced polymers and therefore confirmation of the sequence-controlled nature of the reaction.

### 5.2. Automated Synthesis of Sequence-Controlled Polymers

All of the methodologies described above could be easily extended by introducing a degree of automation. This would result in shifting the burden of overseeing the sequence of events leading to polymers from chemists to computer-controlled reaction setups, thus avoiding potential reproducibility issues and ensuring that variation between experiments is minimal.

Reports concerning automation of polymer synthesis have been relatively scarce, but there exist several examples of automation, with both commercial and in-house setups. The former approach was taken by Matyjaszewski and co-workers, who used a commercially available DNA synthesizer to conduct photocontrolled ATRP by programming a specific sequence of monomer additions to be performed by the machine.[49] This allowed the production of well-defined homopolymers, block copolymers, and DNA−polymer hybrids, but the nature of the equipment necessarily limited broader applications. A more flexible implementation, presented by Du Prez, Espeel and co-workers,[50] involved an automated peptide synthesizer adapted


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to conduct sequential thiolactone ring openings and acrylate couplings (Figure 11), which led to strictly sequence-defined oligomers with diverse functional groups.

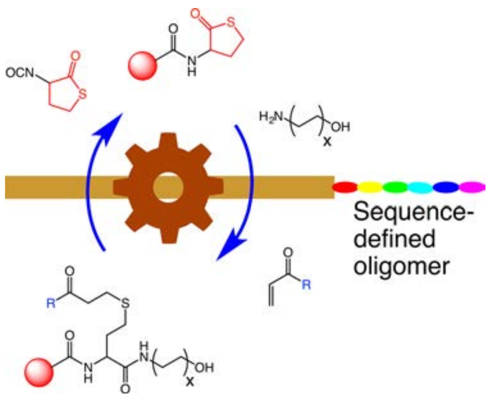


**Figure 11.** Operational principle of an automated system inspired by peptide synthesizers and used to produce sequence-defined oligomers. Adapted from ref 50. Copyright 2016 American Chemical Society.

### 5.3. Modeling Approaches

Theoretical modeling is well-established in sequence studies of natural polymers such as proteins, primarily because of the existence of the Protein Data Bank, which can be used to train computational procedures such as neural networks.[51] However, the synthetic polymer space is nowhere near as deeply explored, and the analogous approach would be prohibitively expensive. As a result, alternative theoretical frameworks are being developed, sometimes with very specific optimization targets in mind.[52,53] An important strand of stochastic approaches to polymerization modeling relates to processes conducted in a continuous fashion, for example in flow reactors. Numerous additional parameters such as monomer residence time and mixing rate come into play here, but models have nevertheless been developed to simulate these conditions.[54,55]

## 6. PROGRAMMING SEQUENCE AND FUTURE CHALLENGES

In this Account, we have attempted to give an overview of recent advances in polymer sequence control, along with outlooks to future challenges and directions. These can be more broadly put in a context of two main strands: selection of sequence from a random mixture and improving our understanding of the relationships between sequence and polymer properties. By analogy to natural polymers, synthetic macromolecules with the same chemical nature but possessing different sequences can be expected to exhibit different properties. Recent examples of this tendency include optoelectronic property differences between polyvinylenes[56] and divergent properties of random versus alternating polyesters, including hydrolytic susceptibility, fluoride ion affinity, ductility, and Young's modulus.[57]

The solid-phase-based processes are being developed with an outlook to increase coupling yields and rates, since the sequential addition of units means that there is typically only one type of monomer present in the reaction medium at any given moment. An example recently reported by us[58] is based on rehydration/dehydration cycles, which allowed efficient uncatalyzed formation of oligopeptides in unprecedented yields. Importantly, the system was fully controllable digitally: parameters such as cycle number and duration, monomer concentration, temperature, and pH could be set and controlled, allowing for the straightforward exploration of all the different environmental variables.

In the case of chain-growth polymers, we have discussed major advances made recently in the synthesis of multiblock polymers. We also recognize that there are additional ambitious prospects related to the importance of bias in polymerization on the level of individual units. Here the understanding of how a molecular constructor might be designed from scratch to build complex self-replicating architectures might seem outlandish, but this is perhaps part of the key problem and is not limited just to chemistry but is also of relevance to computer science and technology.[59] For biology to emerge, such a problem had to be solved without an explicit constructor.[60] Indeed, oligomers that can self-replicate must be able to emerge naturally (for biology even to exist),[61] and these will produce molecules and systems that are more complex than would be expected if the process forming them were random.[62] To that end, in earlier sections we have given numerous examples in which the desired sequence arises from a mixture of monomers through control by factors as diverse as steric hindrance, chemical reactivity, the presence of a template, and catalyst affinity. These are important because any complex mixture of many monomers has to be intrinsically biased to produce well-defined polymer sequences. Otherwise, when all of the monomers are equally likely to polymerize, the product will consist of perfectly random chains only. Therefore, the stochastic modeling techniques and analytical approaches summarized above can be utilized to better understand the influence of different environmental factors on complex monomer mixtures and the possibilities of "pushing" the mixtures in the direction of increasing function. An ultimate breakthrough, bringing the field closer to the molecular machinery of biology,[63] might come from incorporating the one-pot, one-instruction nature of chain-growth polymerization and sequential characteristics of solid-phase processes into one system (Figure 12).

In the former case, radical-based kinetics enables rapid chain growth, but the incorporation of individual monomer units is

| PROCESS | Chain-growth | Step-growth |
| --- | --- | --- |
| maximum no. of units | millions | hundreds |
| units per operation | millions | 1 |
| initial operations | 1 | hundreds |
| programming feasibility | – | + |

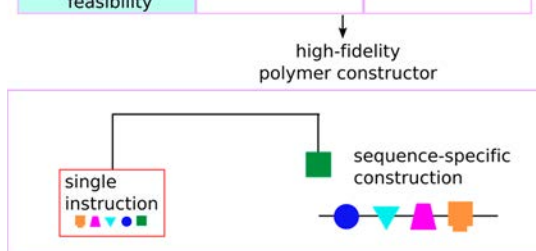


**Figure 12.** To truly mimic nature, high-fidelity synthesis of long polymer chains will need to be implemented in a manner that requires only a single instruction (i.e., what might be called a "one-pot" setup). This approach stands in contrast to chain-growth and step-growth classes of processes but incorporates elements of both.


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harder to control. In the latter, the chain elongation kinetics is slower, but sequential addition is easier to implement. The high-fidelity synthesis of long, sequence-controlled chains determined by a unique set of initial conditions would then pave the way for efficient exploration of the sequence space, with potential goals as diverse as (auto)catalytic activity, material properties, control of microstructure, and self-assembly. It is our wish that this Account will be used to define a new goal by which polymer science and molecular synthesis come together to aim for high-precision assembly of millions of bonds in just one programmable operation. Even small steps toward such a feat, while currently far from reach, would show that the dream of molecular "hard" nanotechnology might best be solved using sequence-controlled polymer systems. These, like those found in biology, would be "engines of creation".[64]

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### Notes

The authors declare no competing financial interest.

### Biographies

**Jan K. Szymański** obtained his M.Sc. in chemistry at Adam Mickiewicz University in Poznan, Poland (2007), followed by a doctorate at the Institute of Physical Chemistry of the Polish Academy of Sciences in Warsaw (2012), working on complex reactions in small volumes and their applications for information processing. He then moved to Harvard University to study nonequilibrium formation of polymer self-asssemblies in an origins-of-life context as a member of the Harvard Origins of Life Initiative. He has been a member of the Cronin group since February 2016, and his work in the group involves designing complex reactions in oil droplets, with an outlook to sequence control in macromolecules and evolving the physical behavior of the droplets. His other research interests include controlling chemical reactions with external stimuli such as light and oxidation−reduction potential.

**Yousef M. Abul-Haija** is a postdoctoral research associate in the Cronin research group at the University of Glasgow, currently working on exploring the emergence of peptide assemblies without biological constraints to investigate how alternative biologies might be created. He earned his Ph.D. in soft supramolecular materials in the research group of Prof. Rein Ulijn at the University of Strathclyde in 2015, which was followed by a one-year postdoctoral position in the same group. He also worked in industry (Hikma Pharmaceuticals in Jordan) for two years. He completed his B.Sc. (2006) and M.Sc. (2009) in Applied Chemistry at Jordan University of Science and Technology, where he worked on developing polymer-based materials through copolymerization and cross-linking. He is interested in the design of supramolecular materials, complex chemical systems, peptide nanotechnology, and structural and functional control of chemical networks.

**Leroy Cronin** is the Regius Professor of Chemistry at the School of Chemistry, University of Glasgow. He was an undergraduate and D.Phil. student at the University of York and a research fellow at the University of Edinburgh and the University of Bielefeld. Starting in 2000 he was a lecturer at the University of Birmingham before moving to the University of Glasgow in 2002. There he was promoted to Professor (2006), Gardiner Professor (2009), and most recently to the Regius Chair (2013). He has received several awards, including the RSC Bob Hay Lectureship, the RSC Corday Morgan Medal and Prize, and the RSE/BP Hutton Prize in Energy Innovation. His research spans a range of fields under the umbrella of "complex chemical systems", focusing on understanding and controlling self-assembly and self-organization in chemistry to develop functional molecular and nanomolecular chemical systems, along with linking of architectural design with function and engineering of system-level functions.

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