Combinatorial Methods, Automated Synthesis and High-Throughput Screening in Polymer Research: Past and Present

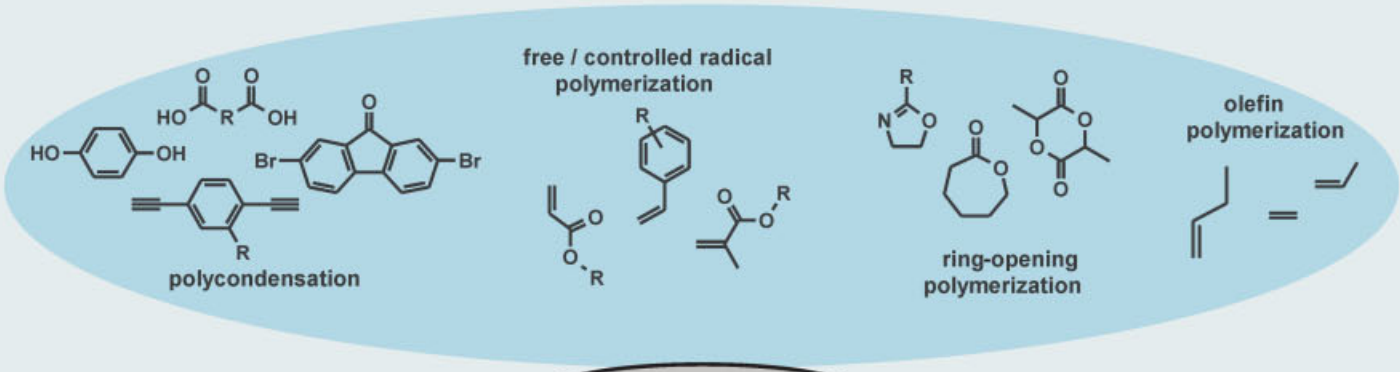
ARTICLE *in* MACROMOLECULAR RAPID COMMUNICATIONS · JANUARY 2003

Impact Factor: 4.94 · DOI: 10.1002/marc.200390013

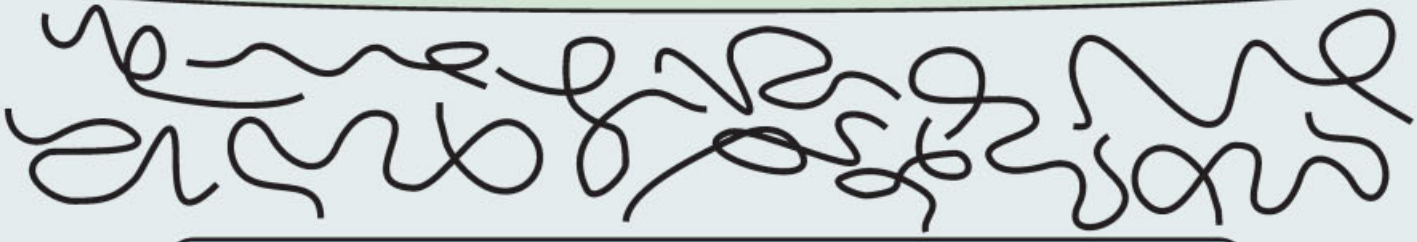
3 AUTHORS, INCLUDING:

Hoogenboom Richard
Ghent University
413 PUBLICATIONS **10,345** CITATIONS
SEE PROFILE

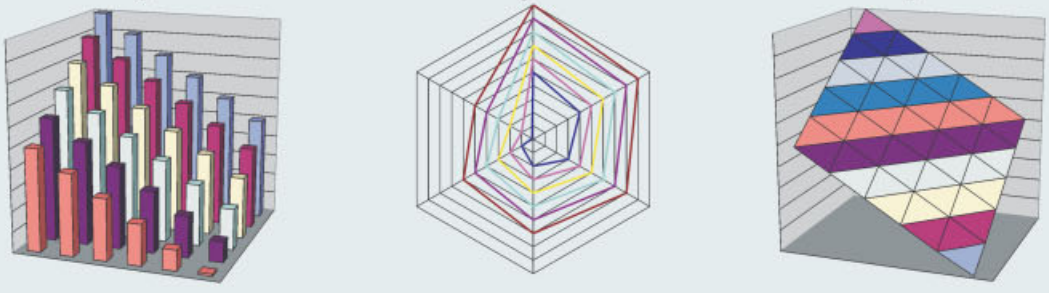
Michael A R Meier
Karlsruhe Institute of Technology
129 PUBLICATIONS **4,421** CITATIONS
SEE PROFILE



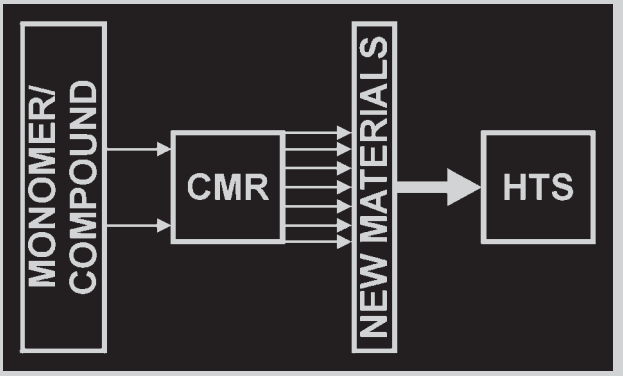
Automated and Combinatorial Polymer Synthesis



High-Throughput Screening and Property Testing



Review: Combinatorial techniques, parallel experimentation and high-throughput methods represent a very promising approach in order to speed up the preparation and investigation of new polymeric materials: a large variety of parameters can be screened simultaneously resulting in new structure/property relationships. The field of polymer research seems to be perfectly suited for parallel and combinatorial methods due to the fact that many parameters can be varied during synthesis, processing, blending as well as compounding. In addition, numerous important parameters have to be investigated, such as molecular weight, polydispersity, viscosity, hardness, stiffness and other application-specific properties. A number of corresponding high-throughput techniques have been developed in the last few years and their introduction into the commercial market further boosted the development. These combinatorial approaches can reduce the time-to-market for new polymeric materials drastically compared to traditional approaches and allow a much more detailed understanding of polymers from the macroscopic to the nanoscopic scale. Here we provide an overview of the present status of combinatorial and parallel polymer synthesis and high-throughput screening.



Combinatorial Methods, Automated Synthesis and High-Throughput Screening in Polymer Research: Past and Present

*Richard Hoogenboom, Michael A. R. Meier, Ulrich S. Schubert**

Laboratory of Macromolecular Chemistry and Nanoscience, Eindhoven University of Technology and Dutch Polymer Institute (DPI), PO Box 513, 5600 MB Eindhoven, The Netherlands
E-mail: u.s.schubert@tue.nl

Keywords: automated synthesis; combinatorial chemistry; high-throughput screening; materials; polymers

1 Introduction

The development and design of new (polymeric) materials is based on the synthesis of new compounds and the optimization of existing materials. Therefore, researchers have to synthesize and screen many reaction conditions and different compounds, e.g., monomer, catalyst, molecular weight, reaction time, reaction temperature and so on, which represents a very time-consuming process. In order to speed up this research, combinatorial techniques, parallel experimentation, as well as high-throughput methods represent a very promising approach: many different parameters can be screened (simultaneously or in fast serial mode) and the results can be easily compared, which may result in new structure/property relationships. However, the capability of performing a large amount of experiments does not necessarily speed up research if the experiments are not properly selected. Therefore, design of experiments (DoE) is often utilized.[1–5] DoE methods utilize probability and statistics to define the minimum number of experiments required to determine significant relationships between input and output parameters.

Combinatorial and high-throughput methods in pharmaceuticals research were very successful.[6,7] This success resulted in an increased attention to parallel and combinatorial approaches for the synthesis and discovery of new inorganic materials, catalysts, and organic polymers. We would like to provide a short overview of the history and development of combinatorial techniques, which led to the emergence of combinatorial and high-throughput research in the field of polymer chemistry.

The first examples of combinatorial approaches in material research can be addressed to Edison[8] and Ciamician.[9] Thomas A. Edison already applied parallel and combinatorial methods in materials research as early as 1878.[8] To discover suitable filament materials for the incandescent

  1022-1336/2003/0101–15$17.50+.50/0

lamp, he tested over 1600 different earths, minerals and ores. He finally discovered that carbonized cotton thread in a vacuum light bulb was the optimal material. This discovery led to electrical lighting, as it is known in present days. In 1912, the Italian photochemist Ciamician investigated the possibility of using a photochemical process for batteries by exposing hundreds of flasks with potentially photoactive materials to the sun at the roof of the university of Bologna (Figure 1).[9] In these first parallel approaches, the experiments were set up manually and screened in parallel. The first example of automated preparation and screening of inorganic materials was reported by Hanak in 1970.[10,11] The use of a radio-frequency co-sputtering technique allowed the synthesis of nearly complete binary or ternary solid alloy systems in one experiment. For rapid screening, 50 gold contacts were evaporated along the library so that superconducting transition temperature and resistivity could be investigated at regular composition intervals of 2%.

The extensive use of combinatorial techniques was first adapted in pharmaceutical research.[12–17] The time-to-market for new drugs can in principle be decreased tremendously, because hundred to thousand times more compounds can be synthesized and screened compared to traditional approaches. The success of combinatorial methods in pharmaceutical research is closely related to the fact that it is relatively easy to rapidly screen new libraries of compounds on purity (liquid chromatography/mass spectrometry) and to identify bioactive materials by standard binding assays.[18]

In materials research, parallel and combinatorial techniques have only been used intensively during the last decade,[19] since only then the first high-throughput screening techniques for materials became available.[18] A

Richard Hoogenboom was born in 1978 in Rotterdam (The Netherlands). In 2002 he completed his undergraduate research on quadruple hydrogen bonding of the 2-ureido-4-[1H]-pyrimidinone unit in water in the group of Bert Meijer (Eindhoven University of Technology (TU/e), The Netherlands). He is currently working on his Ph.D. thesis at the TU/e under the supervision of Ulrich Schubert, focusing on supramolecular initiators for controlled polymerization techniques and automated high-throughput synthesis of well-defined polymers.

Michael A. R. Meier was born in Ingolstadt (Germany) in 1975. He studied chemistry at the University of Regensburg. His diploma thesis dealt with the fluorosensing of ammonium ions via molecular recognition in polymeric emulsion membranes and was carried out at the Institute of Analytical Chemistry, Chemo- & Biosensors at the University of Regensburg with Professor O. Wolfbeis. In 2002 he started his Ph.D. work with Ulrich Schubert at the TU/e in the field of combinatorial polymer research and supramolecular polymers.

Ulrich S. Schubert was born in Tübingen in 1969. He studied chemistry at the Universities of Frankfurt and Bayreuth (both Germany) and the Virginia Commonwealth University, Richmond (USA). His Ph.D. work was performed under the supervision of Professor Eisenbach (Bayreuth, Germany) and Professor Newkome (Florida, USA). In 1995 he obtained his doctorate degree with Prof. Eisenbach. After a postdoctoral stay with Professor Lehn at the Université Strasbourg (France) he moved to the Technische Universität München (Germany) to obtain his habilitation in 1999 (with Professor Nuyken). From 1999 to 2000 he held a temporal position as a professor at the Center for NanoScience at the Universität München (Germany). In Summer 2000 he has been appointed a Full Professor at the TU/e (Chair for Macromolecular Chemistry and Nanoscience). He received the Bayerische Habilitations-Förderpreis, the Habilitandenpreis of the GDCh (Makromolekulare Chemie), a Heisenberg-Stipendium of the DFG and a Dozenten-Stipendium of the Fonds der Chemischen Industrie. His research interests are focused on organic heterocyclic chemistry, supramolecular materials, combinatorial materials research, nanoscience, and tailor-made macromolecules.

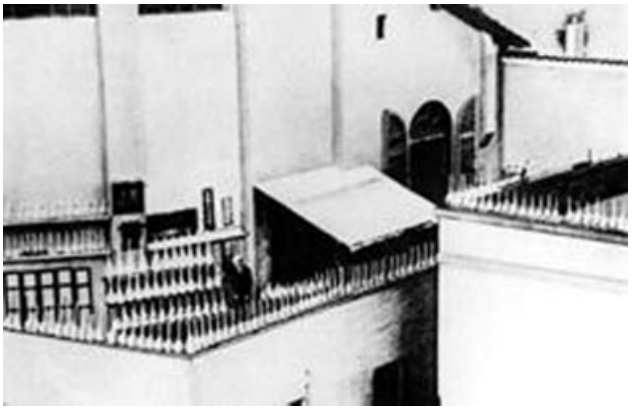


Figure 1. Ciamician tested hundreds of samples in parallel on the roof of his laboratory at the University of Bologna. (Photo courtesy of the University of Bologna).

combination of thin-film deposition and physical masking steps allowed the synthesis of libraries of inorganic compounds. Novel superconducting materials[20] and inorganic phosphorus compounds[21–24] have been discovered using this technique. Systematic variation of composition and processing conditions is particularly well-suited to ternary and higher-order inorganic materials, for which predictions of basic properties have been unsuccessful.[21] In the area of catalysis, parallel combinatorial techniques have also been introduced.[25–32] High-throughput screening of catalyst activities were reported by utilizing, e.g., laser-induced resonance-enhanced multiphoton ionization,[25] thermographic techniques,[26] and fluorescence.[29] Furthermore, parallel synthesis and high-throughput screening have proven to be successful for faster discovery of novel catalysts.[25,31]

The field of polymer research seems to be perfectly suited for parallel and combinatorial approaches due to the fact that many parameters can be varied during synthesis, like monomers, catalysts, initiators or end-cappers as well as many reaction, processing, blending and compounding conditions. In addition, numerous important parameters have to be investigated, such as molecular weight, polydispersity, viscosity, hardness, stiffness and other application-specific properties. However, until the 1990s,[33] combinatorial (bio)polymer research was nearly completely unexplored because of the absence of suitable high-throughput screening techniques. In the last few years, several Reviews and Feature Articles on combinatorial materials research were published,[5,17,19,34–39] mainly covering inorganic materials and catalysis. Therefore, we would like to provide here an overview of the present status of combinatorial and parallel polymer synthesis and the available high-throughput screening techniques. This Review only covers the open literature (journals and patents), but we are totally aware of the fact that many more results may have been obtained already within companies.

2 Parallel, Automated and Combinatorial Polymer Synthesis

The goals in parallel polymer synthesis are mainly related to the design of new materials with special properties or the study of structure/property relationships. Several approaches have been applied to achieve these goals, ranging from a rather simple parallel manual synthesis to completely automated workflows with synthetic robots. Furthermore, different synthetic polymerization methods have been used, ranging from polycondensation to living polymerization techniques. This chapter will provide detailed information concerning the synthetic parallel and combinatorial approaches that have been used in polymer science so far.

2.1 Polycondensation

A manual combinatorial approach in polymer synthesis was first reported by Kohn and coworkers in 1997.[40,41] Polycondensation reactions of diphenols and diacids with small but systematical structural variations were reacted in a parallel fashion to obtain a library of 112 polyarylates. A maximum of 32 parallel reactions were manually carried out in separate reaction vessels in a water shaker bath on a 200 mg scale. The library was screened using standard techniques (gel permeation chromatography (GPC), differential scanning calorimetry (DSC), air/water contact angle), whereby predictable changes in surface wettability (Figure 2a), glass transition temperature (Figure 2b), and cellular response (measured by in-vitro studies) were found. Another degradable polymer library (140 members) consisting of diacrylate and amine monomers was synthesized and screened by Langer et al. in a non-automated way by performing small-scale reactions in parallel and applying routine analysis techniques afterwards.[42] The time required in order to prepare and screen all polymers was approximately 2 weeks, which could be significantly decreased by the incorporation of robotic systems, as mentioned by the authors. Utilizing this combinatorial approach, an unpredicted new synthetic transvection vector was discovered.[42] A higher degree of automation is reported for the synthesis of conjugated polymers from dihalogenated and diethynyl monomers by a Pd-catalyzed carbon-carbon coupling reaction (Figure 3).[43] For this approach 96 vials (1 mL), dispatched in the standard 12×8 format, were filled in a glovebox with different combinations of monomers and the catalyst by a multichannel pipette system. Polymerizations were performed for 24 h at 60 °C and the polymers were subsequently screened for optimal fluorescent behavior utilizing a hand-held UV lamp. The ''hits'' of this first screening were then analyzed by means of GPC and synthesized in larger scale in a conventional approach. The comparison of the fluorescent properties of the polymers obtained from the combinatorial and the classical

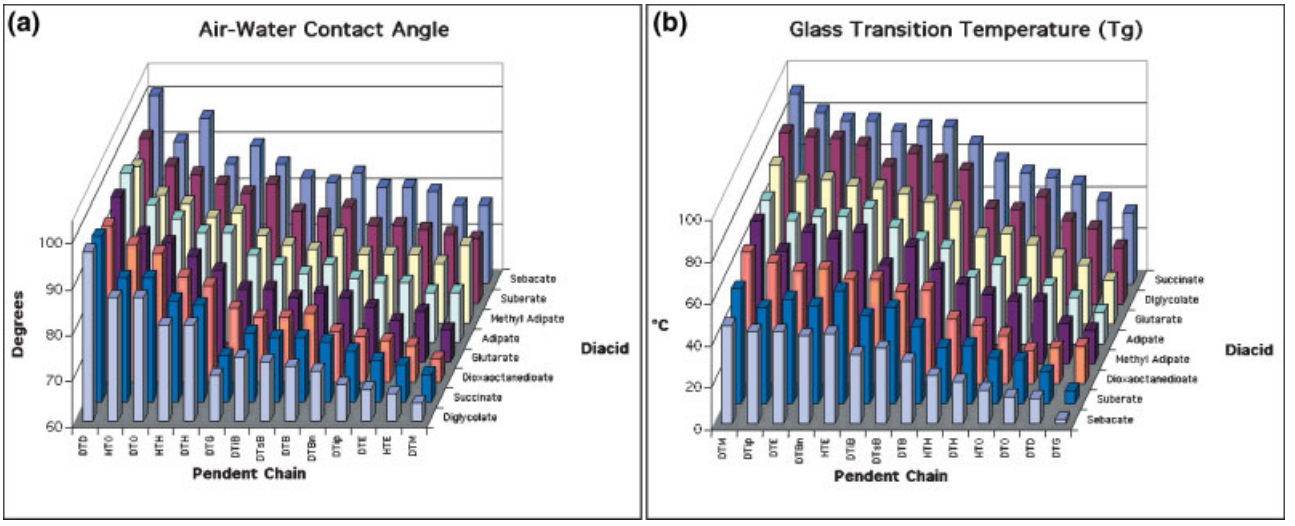


Figure 2. (a) Glass transition temperatures and (b) air/water contact angles for a 112-polymer library as a function of the polymer pendant chain (*x* axis) and backbone structure (*y* axis). (Laboratory of Professor Kohn, Rutgers University.[40,41])

approach revealed that the combinatorial approach indeed reflected the real fluorescence properties of the polymers. Therefore, the authors concluded that the high-throughput method applied is suitable for the fast qualitative screening of new compounds for light-emitting diodes. A parallel approach for melt polymerizations was described by GE.[44] Solutions of bisphenol A (BPA), diphenylcarbonate (DPC) and catalyst were delivered to approximately 4 mL vials resulting in 100 mg reagent per vial. The solvent was allowed to evaporate at 80 °C and the reaction vials were placed in a heating block and polymerized by increasing the temperature stepwise to 240 °C. The resulting polymer films (0.5 mm thick) were dissolved in chloroform and characterized utilizing standard GPC. These parallel melt polymerizations appeared to be reproducible and could be used, e.g., for catalyst optimization. In addition, automated parallel melt polymerizations of BPA and DPC with various catalysts in a 96-well glass microtiter plate were described by the same authors.[44]

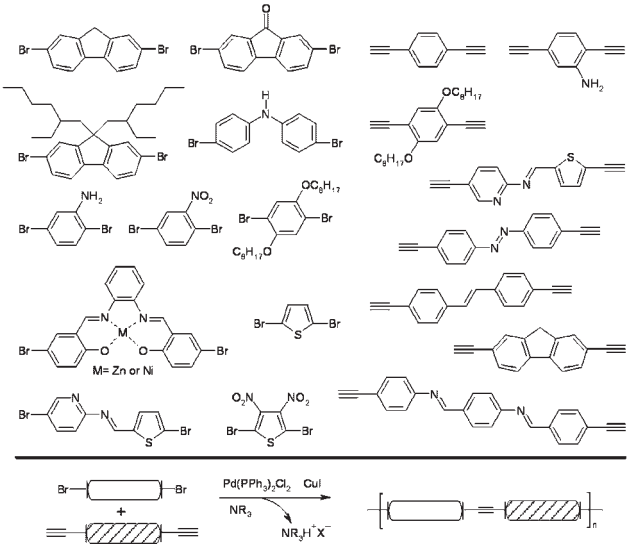


Figure 3. Library of monomers used by Lavastre et al. for the parallel Pd-catalyzed polymerization of conjugated polymers.

2.2 Radical Polymerization

2.2.1 Free-Radical Polymerization

Suspension polymerization was carried out successfully utilizing a multiparallel polymerization system as reported by Bradley (Figure 4).[45–47] This system was used to perform the parallel synthesis of a set of polystyrene resins. Polymerizations with varying ratios of divinylbenzene (DVB), styrene and vinylbenzyl chloride were thermally initiated with 2,2′-azoisobutyronitrile (AIBN). The resulting beads were washed, Soxhlet-extracted, dried in vacuo and sieved to afford resins in five size ranges. The resulting size distribution of the polymer beads was reproducible and

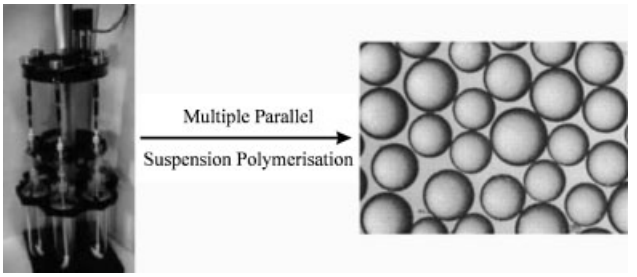


Figure 4. Apparatus for parallel suspension polymerization (left), and an example of 1% DVB/PS resin beads obtained in DMF. (Taken from the literature.[45])

showed that this parallel approach is useful for the rapid design of polymeric bead supports and to optimize polymerization conditions.

A manual parallel free-radical polymerization approach towards new soluble supports for organic synthesis has been described by Janda et al.[48] Bifunctional initiators with both an α-nitrilediazene (–N=N–) and a 2,2,6,6-tetramethylpiperidine-*N*-oxyl (TEMPO) group were utilized to synthesize block copolymers, whereby five different monomers were utilized. The α-nitrilediazene was first polymerized at 70 °C with different monomers. After work-up, the polymers were split and used as macroinitiators to polymerize the different second blocks with the TEMPO groups at 130 °C. Furthermore, a TEMPO/methacrylate initiator was synthesized and polymerized first with different monomers using AIBN at 60 °C. These polymers were again split and further polymerized with different monomers at 130 °C to give different graft copolymers.

Sellergren described a manual parallel approach for the photoinitiated free-radical polymerization of molecularly imprinted polymers.[49] The reagents were manually pipetted into 1.5 mL vials and then symmetrically placed around a UV lamp for polymerization (Figure 5). Later on, a fully automated procedure in order to synthesize and evaluate molecularly imprinted polymers was reported, whereby thermal initiation was used since this technique was considered easier to implement and more widely applicable than photoinitiation.[50] Mixtures of different monomer solutions were dispensed automatically into 1.5 mL glass vials. Subsequently, the mixtures were thermally initiated at 45 °C and allowed to polymerize for 24 h. Approximately 60 polymers could be prepared in parallel applying this procedure. The polymers were automatically evaluated by means of their binding constants to the imprinted analytes. Therefore, the analytes were extracted with porogen and, after complete extraction, a template solution in porogen was added. During this procedure the supernatants were analyzed utilizing online high-performance liquid chromatography (HPLC)/UV. Long et al. described another combinatorial approach for free-radical polymerizations.[3,51]

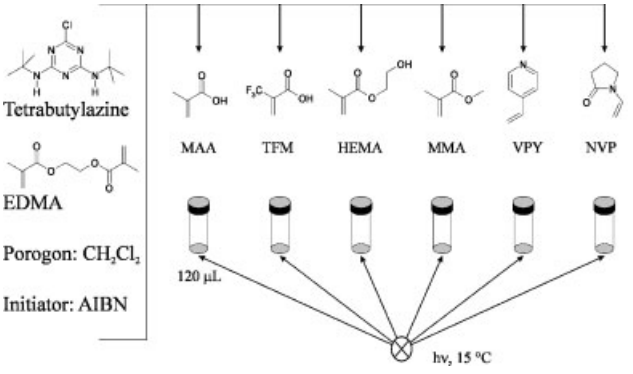


Figure 5. Manual parallel photoinitiated free-radical polymerization of molecularly imprinted polymers.

Poly[styrene-*co*-(methyl methacrylate)]s and poly[styrene-*co*-(butyl methacrylate)]s were synthesized by systematically varying the monomer ratios in a robotic system. The products were precipitated automatically and standard analytics (NMR, GPC, DSC) were used to characterize the resulting polymer library. Symyx described a fully automated method for the parallel free-radical polymerization of styrene with acrylonitrile as a terminating agent.[52] Different ratios of monomer and terminator solutions were delivered by an ink-jet dispenser to 16 predefined regions (45 µL each) on a 3×3 cm^2 pyrex substrate. After the addition of benzoyl peroxide solutions polymerizations were performed at 60 °C and, upon completion of the polymerizations, the solvent was removed under reduced pressure.

2.2.2 Controlled Radical Polymerization

Controlled radical polymerizations were also performed successfully utilizing automated synthesis robots, including reversible addition/fragmentation transfer (RAFT),[53] atom-transfer radical (ATRP),[54–57] and nitroxide-mediated polymerizations.[58,59]

The RAFT polymerization technique was used to prepare graft copolymers with controlled length and spacing of the grafted chains in a 96-well parallel batch reactor equipped with a liquid dispensing robot.[53] Backbones of varying molecular weights were chemically modified in order to attach RAFT control agents with different degrees of modification. These modified polymers were polymerized with different monomers to generate a library of graft copolymers. Fluorescence labeling of the polymers was utilized to quantify the absorption to a substrate (Figure 6). This

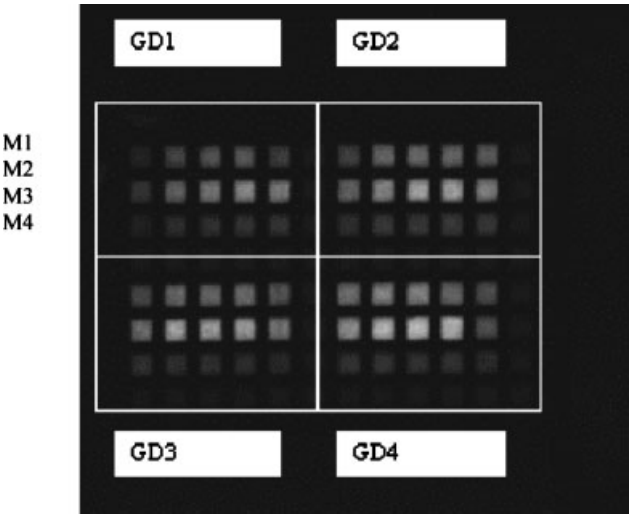


Figure 6. Fluorescence imaging of an 80-member library of graft copolymers adsorbed to a substrate. Each polymer of a quarter has the same grafting density (GD1–GD4), while the length of the grafts increases from left to right and the monomer changes from top to bottom (M1–M4). (Taken from the literature.[53])

procedure is capable of producing 200 to 300 materials per day. A similar automated controlled radical polymerization (MADIX) was reported by Chapon and coworkers.[60] MADIX differs from RAFT in the nature of the chain-transfer agent: MADIX is performed with xanthates (RS(C=S)OZ), whereas RAFT may be performed with all kinds of thiocarbonylthio (RS(C=S)Z) compounds. Both homopolymers and diblock copolymers were automatically synthesized with two different xanthates in a reproducible way. GPC characterization demonstrated that the automatically synthesized polymers were highly comparable with polymers obtained from classical polymerizations.

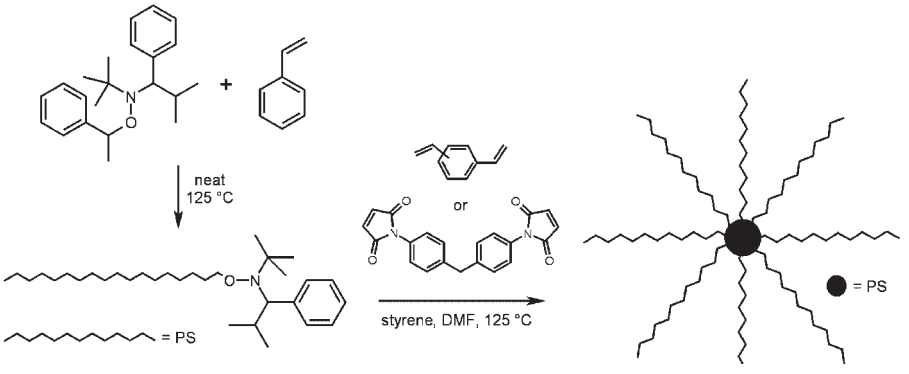
Symyx demonstrated that it is possible to polymerize styrene and butyl acrylate in parallel and fully automated by ATRP.[54,55] In this way libraries with 48 to 140 members with volumes of 0.1 to 20 mL per reaction vessel could be created. Robotic systems were used to dispense all reagents and to prepare samples for high-throughput characterization. Schubert and coworkers have also reported the utilization of an automated synthesizer for ATRP.[56,57] Both reproducibility and comparability with classical ATRP were demonstrated. In addition, a method for automated GPC sample preparation by means of online column chromatography (to remove the catalyst) was reported.

Hawker et al. evaluated the usage of macroinitiators for nitroxide-mediated polymerization by setting up a library of 96 members with different ratios of macroinitiator to styrene monomer and different ratios of macroinitiator to 1,1′-(methylenebis(4,1-phenylene)bismaleimide or divinylbenzene (Scheme 1).[58,59] As a result, optimized conditions for the formation of star polymers were obtained. The next step was the generation of a 168-member library based on the hits of the initial library, and the investigation of seven different macroinitiators within this library. This provided further insight into the reaction conditions that are required in order to form well-defined star polymers.

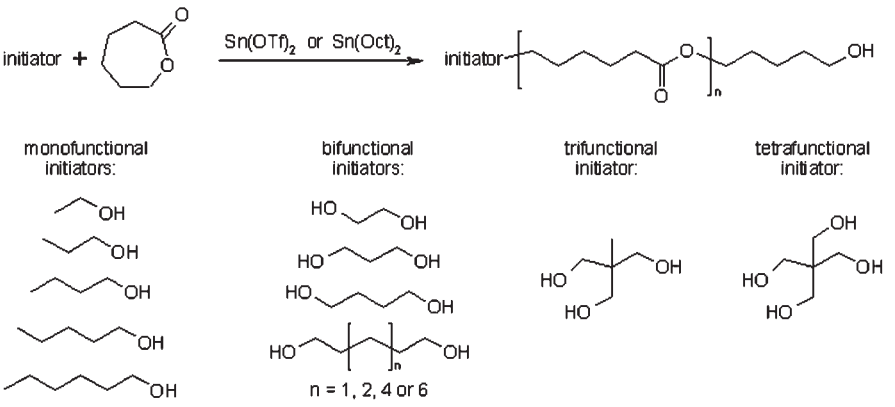
2.3 Ring-Opening Polymerization (ROP)

Hedrick and coworkers reported parallel (up to 20) controlled ROPs of lactides and lactones.[61,62] The catalytic behavior of 4-(*N,N*-dimethylamino)pyridine (DMAP) and 4-pyrrolidinopyridine (PPY) in the polymerization of lactides was studied using ethanol as an initiator with 0.1 to 4 equivalents of amine with respect to initiating alcohol and varying monomer-to-initiator ratios ([M]/[I]).[61] DMAP and PPY showed comparable catalytic activity, whereby polymers with narrow polydispersity and molecular weights closely tacking the [M]/[I] ratios were obtained. The automated synthesizer was also utilized for rapid screening and optimization of both catalyst/initiator systems and the associated polymerization conditions as shown in Scheme 2. Polymerizations were initiated with mono- to tetrafunctional alcohols and both tin(II) triflate and tin(II) ethylhexanoate were examined as catalysts.[62]

The living cationic ROP of 2-ethyl-2-oxazoline was performed in a fully automated fashion by Schubert et al.[63,64] Stock solutions of initiator and monomer were dispensed in different ratios into the reaction vessels (up to 40) and vortexed for 24 h at 80 °C. Piperidine was subsequently added to terminate polymerization, and the resulting polymers were automatically precipitated and transferred into sample vials. Both the reproducibility and living character of the polymerizations were demonstrated by online GPC, offline ^{1}H NMR spectroscopy and matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS). Temperature optimization of the cationic ROP of 2-ethyl-2-oxazoline in *N,N*-dimethylacetamide utilizing an automated synthesizer equipped with an individually heatable reactor block was also reported.[65] Parallel polymerizations were performed at 9 different temperatures, whereby samples were automatically taken at suitable time periods. These samples were characterized with both online GPC and offline gas chromatography (GC). The results revealed an optimal



Scheme 1. Presentation of the general synthesis of star-shaped polystyrene. The first step is done utilizing a classical set-up, and star-synthesis was carried out utilizing an automated synthesizer.



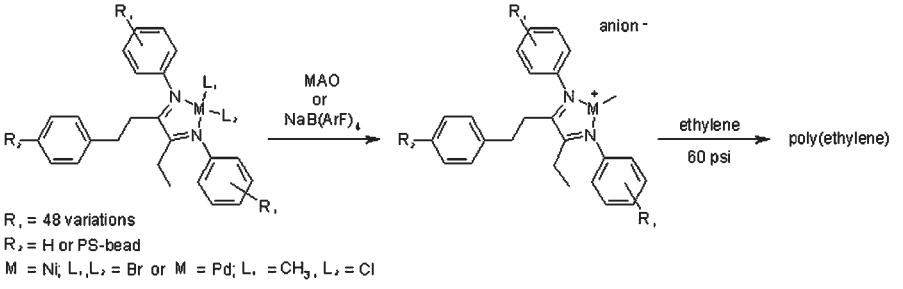
Scheme 2. Initiators and catalysts screened for the ring-opening polymerization of ε-caprolactone.

polymerization temperature for this system of 100 °C. The activation energy for the polymerization of 2-ethyl-2-oxazoline was determined from GC characterization.

2.4 Polyolefins

The discovery and optimization of olefin polymerization catalysts is an area of great interest, since the chemical industry produces approximately 46 million metric tons of polyolefins annually.[66] In order to speed up the catalyst research in this area, Symyx has synthesized a library of both solid-supported and free palladium and nickel catalysts utilizing an automated parallel approach.[67] The resulting catalysts were fully automatically (up to 48 parallel reactions) screened for ethylene polymerization (Scheme 3). The reactors were loaded inside a glovebox utilizing a 3-axis liquid handling robot. The pressure of each reaction chamber was individually controlled and monitored, and the reaction mixtures were mechanically stirred with Teflon-coated stirring blades. The results revealed that those complexes with the highest steric hindrance in the ortho positions of the aryl rings produced the highest-molecular-weight polyethylene. Electronic factors did not seem to have much influence on yield or molecular weight. Mülhaupt described the usage of an automated synthesizer for the parallel synthesis of ligands, organometallic compounds, supported catalysts, as well as for the automated polymerization of olefins.[68] Polymerizations were performed in minireactors (13 mL to 100 mL), resulting in a significantly improved quality as compared to high-throughput polymerizations utilizing simple arrays (up to 1 mL vials or wells).

A completely different non-automated combinatorial approach to polyolefin research was reported by Müllen.[69] A mixture of (two) catalysts labeled with fluorescent dyes were mixed and used for ethylene polymerization. During polymerization each catalyst particle formed a product granule, which can be considered as a microreactor. By exposing the obtained granules to UV light the different catalysts showed different fluorescence behavior as shown in Figure 7. Particles of each catalyst could thus be separated and characterized with standard techniques. It was shown that no exchange of labels occurred and that the granules resulting from the mixed experiment revealed the same properties as polymers obtained from single runs with the catalysts. This approach can easily be expanded due to the great variety of dyes available that cover the whole UV-vis spectrum. Coates also described a split-and-pool strategy for the development of new polyolefin catalysts.[70] First a pool of 12 ligands was synthesized by condensation



Scheme 3. Ethylene polymerization procedure in combination with automatically screened variations.

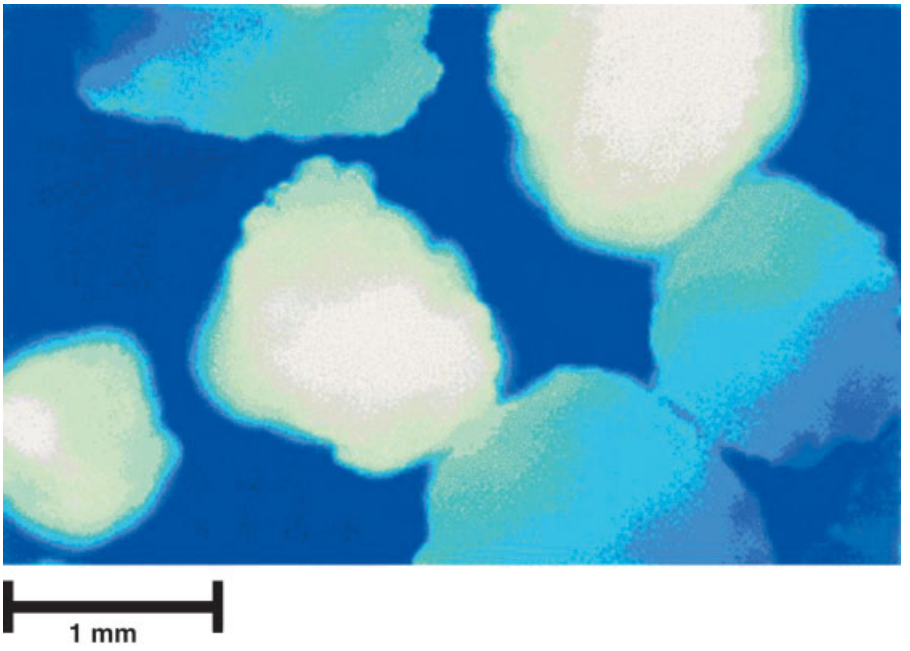


Figure 7. Fluorescence microscopy image of the polymer products obtained from simultaneous polymerization of mixed labeled catalysts. (Taken from the literature.[69])

of three salicylaldehydes with four amines. This pool was subsequently complexed with titanium tetrachloride resulting in a 78-member library of complexes. The complete library was activated with methylaluminoxane in toluene and exposed to propylene. 10% of the resulting polymer were insoluble in refluxing diethyl ether and this was unexpectedly found to be syndiotactic poly(propylene). The catalyst responsible for the formation of stereoregular poly(propylene) was identified by utilizing four sub-libraries for the polymerization of propylene.

2.5 Supramolecular Polymerization

In contrast to conventional polymers, which are based on covalent bonds, supramolecular polymers are formed via both covalent bonds and non-covalent interactions, such as hydrogen bonds, metal-coordination, or ionic interactions.[71,72] Schubert and coworkers reported the fully automated synthesis of main-chain supramolecular coordination polymers.[73,74] Complexation reactions of bis(2,2′:6′,2″-terpyridine)-functionalized poly(ethylene oxide) with four different metal(II) acetates were performed automatically, and the resulting supramolecular polymers were characterized by means of UV-vis spectroscopy revealing that the classical laboratory approach could be transferred to the automated system. Viscosity measurements in solution demonstrated a strong dependence on the metal ions and counterions applied. It was therefore suggested that the parallel and combinatorial synthetic approaches could lead to a broad variety of new supramolecular materials.

3 Combinatorial Polymer Libraries

New methods for the preparation of gradient libraries of polymers have become available in the last few years.[75–81] Special flow-coating devices have been designed by Meredith, Karim, and Amis et al. in order to prepare thickness-gradient libraries of thin films. The set-up of this system was based on a velocity-gradient knife-edge coating device. With this equipment, drops of polymer solutions can be spread over the substrate at constant acceleration resulting in polymer films with a gradient in thickness (see, e.g., Figure 8).[77,78] Gradients in polymer composition are also accessible by automated premixing of solutions of different polymers, whereby samples of the different polymer mixtures are automatically aspirated into a syringe. These samples are deposited as a thin stripe on the substrate, which is then spread as a film orthogonal to the composition gradient using a computer-controlled knife-edge coater as described above.[75,76] Another method for the preparation of polymer libraries is by applying a linear temperature gradient over a thin polymer film that is placed on an aluminium heating stage.[76,77] In this way it is possible to prepare thin films of polymers as composition/thickness, composition/temperature or thickness/temperature two-dimensional combinatorial libraries. The information that may be derived from these libraries will be discussed in Section 4.3.

A fourth method to obtain a gradient polymer film is described by Dickinson and Walt.[81] A fiber bundle, consisting of approximately 6 000 single optical fibers, was attached to a micropositioner at one end, and the other end was immersed in 1 mL of a stirred solution containing

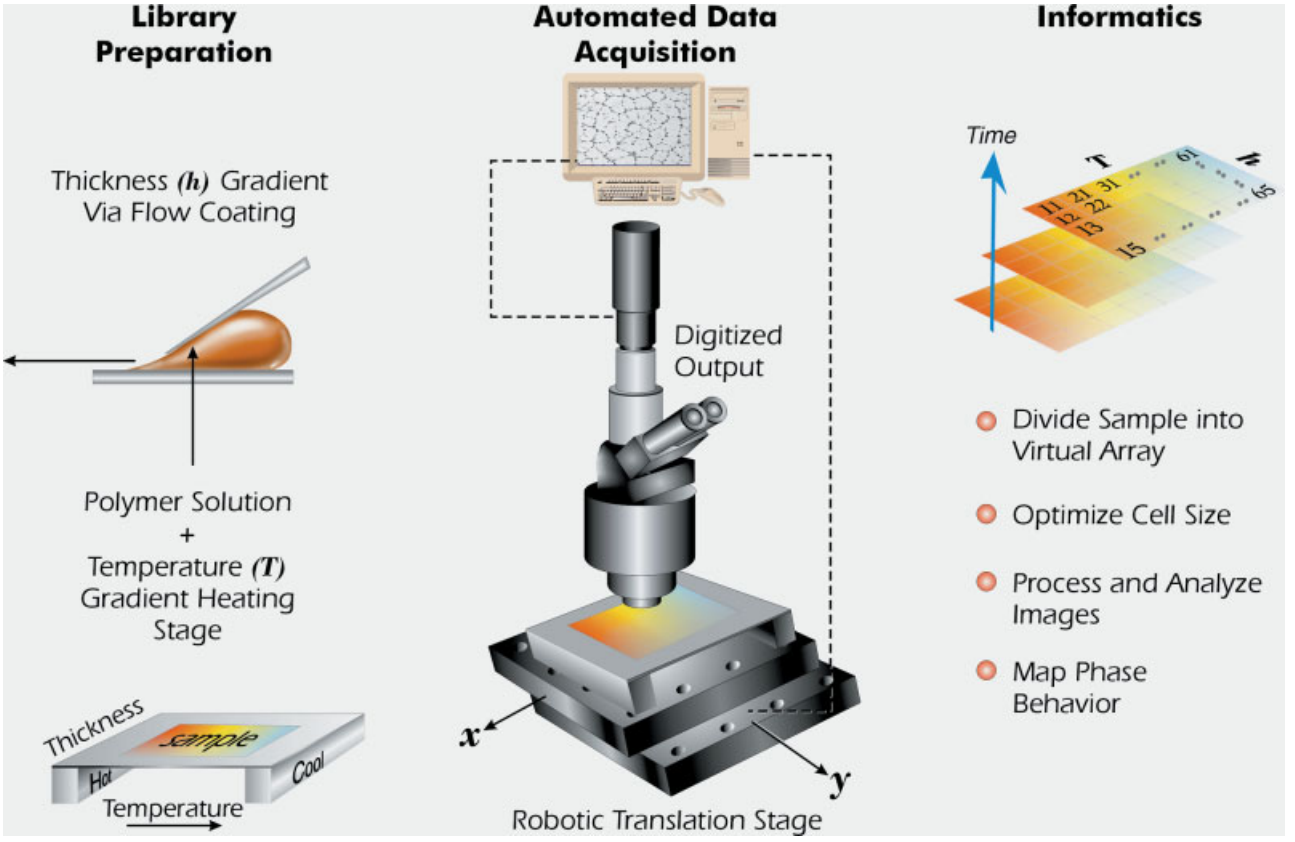


Figure 8. Schematic representation of the three steps involved in combinatorial characterization of thin polymer films: (1) preparation of thickness- and temperature-gradient libraries, (2) automated optical microscope, and (3) data reduction. (Taken from the literature.[77])

monomer (acryloxypropyl)methylsiloxane (PS901) and photoinitiator (BEE) in chloroform. By focusing UV light on the fiber, simultaneously scanning the light beam across the fiber, and adding a second monomer (PS802) to the polymerization solution, a gradient polymer stripe was obtained on the distal face of the fiber. After soaking in a solution of Nile Red to entrap the dye in the polymer matrix, the sensor shows shifts in emission wavelength depending on the polarity of the local environment. The polymer was screened for sensor response across the gradient. Similarly, discrete polymer sensing regions could be fabricated at designated regions across the distal face of the fiber as shown in Figure 9.

Recently, Potyrailo and coworkers described the use of a microextruder equipped with two feeders to make one-dimensional libraries of polymeric compositions.[82] As little as 2–10 g of polymer were required, and step or gradient changes could be introduced to the composition within 1 min per formulation. Weathering tests were performed on the libraries by exposing the samples to artificial sunlight. An automatically scanning fluorescence spectroscopic system was utilized to determine the weathering of the polymers by excitation of the degradation products.

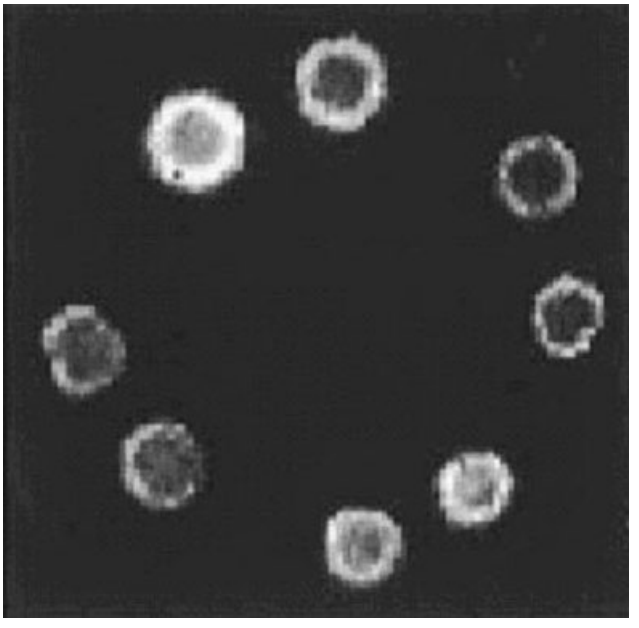


Figure 9. Fluorescence image of an optical array, showing eight discrete polymer sensing regions, each 40–50 μm in diameter. (Taken from the literature.[81])

4 High-Throughput Characterization of Polymers

As described in Section 2, the parallel synthesis of new polymeric materials has gained more and more attention during the last few years. Unfortunately, the corresponding screening techniques are less developed up to now. The concept of high-throughput screening (HTS) can be dated back to the 1950s and was developed due to the need of fast and automated analyses in clinical testing and medicine.[35] Since then HTS has mainly stayed a field of research applied to and developed for parallel catalyst design[83,84] and drug discovery.[85,86] There are only a few examples where HTS is applied to synthetic polymers in order to obtain information about molecular weight, optical properties, morphology, etc.[17,39] The existing techniques will be the subject of discussion in the following paragraphs.

4.1 Screening of Molecular Weights

GPC represents the standard and most widely used method to determine molecular weights and the corresponding molecular weight distributions of polymers in solution.[87,88] The major drawback of this technique considering HTS strategies is the relatively long analysis time of up to 30 min for standard systems, which would represent a serious bottleneck. Therefore, much faster GPC systems have been developed in recent years, applying one or more of the following features: parallelization, shorter columns, flow-injection analysis (FIA), and high-speed GPC columns.[87–89] Figure 10 shows a conventional GPC calibration in comparison with a high speed approach.[90] This example demonstrates the possible time-saving effects very impressively. The resolution of the chromatographic system on the other hand will be lowered when using high-speed instead of conventional columns. Nevertheless, high-speed GPC is the method of choice for the fast and easy determination of molecular weights and molecular weight distributions of polymer libraries. Short screening columns with analysis times of less than 3 min that are now available from different manufacturers will allow faster GPC measurements.[90,91] Due to the progress in developing monolithic materials, the availability of new types of columns can be expected in the near future.[92–94] Almost all parallel synthetic approaches described up to now utilize conventional GPC systems for screening purposes (see, e.g., ref.[40–43]), thus facing a significant bottleneck regarding the most important parameters in polymer characteristics: the molecular weight and its distribution. Ultrafast GPC screening techniques however are commercially available. Symyx, for example, has introduced Rapid GPC™ systems, which are capable of analyzing GPC samples in the range 40 s to 2 min,[95] and Dow adopted these systems for their purposes.[96] Symyx compared the data obtained by ultrafast screening of weight-average molecular weights with the results obtained by conventional GPC measurements (Figure 11),[97] which clearly shows that fast GPC is practicable for screening polymer libraries in terms of molecular weights.

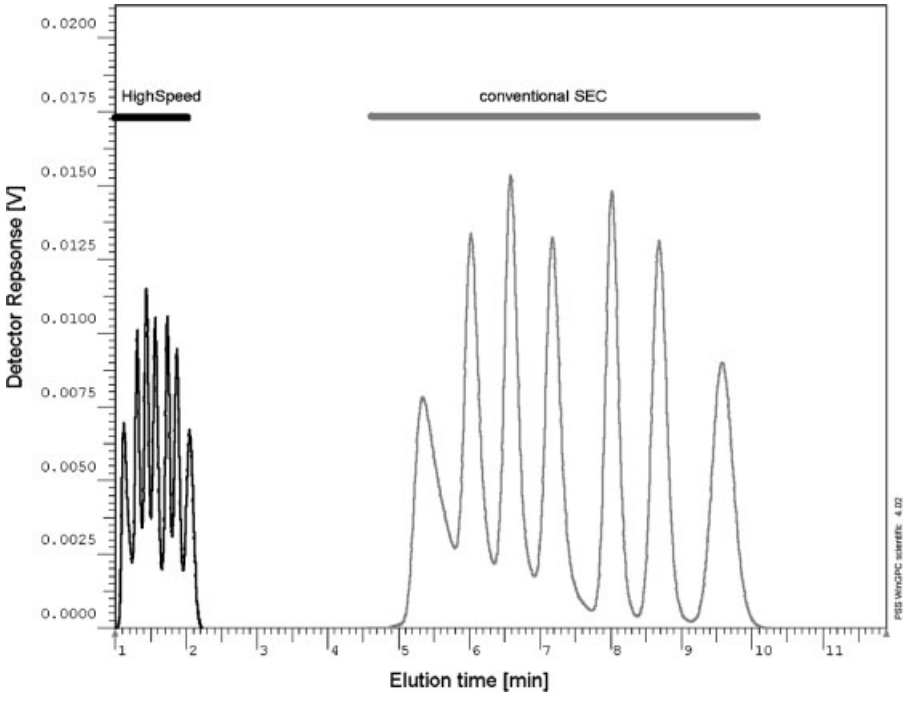


Figure 10. Comparison of a high-speed GPC calibration curve with conventional GPC calibration. (Taken from the literature.[90])

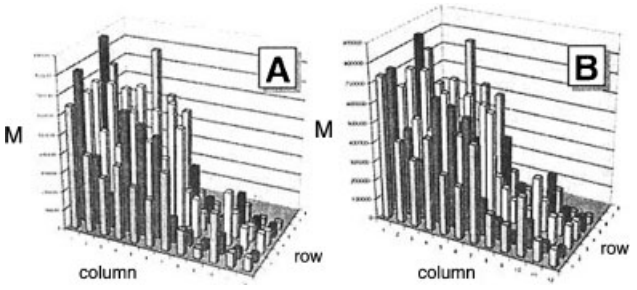


Figure 11. $\bar{M}_w$ values for a model polystyrene library obtained with: (A) GPC system with a series of three conventional 30 cm × 7.5 cm columns, (B) rapid chromatographic system (analysis time: 2 min). (Taken from the literature.[97])

MALDI-TOF-MS is a powerful analytical method, which allows the investigation of synthetic polymers with respect to molecular weight, molecular weight distribution, and end-group analysis.[98,99] Mass spectrometry in principle is a highly selective and high-throughput analytical technique, that is perfectly suited for the identification of a large number of compounds[100] even in the form of mixtures.[101] Figure 12 shows a typical workflow for the screening of a biological library by means of mass spectrometry.[100] In recent years, several studies have been performed to develop analytical MALDI methods for the fast analysis of a high number of samples.[102,103] MALDI-TOF-MS has also been used for the automated identification of proteins[103] or as a screening tool for peptide libraries.[104] Very recently, Schubert et al. showed that rapid screening of synthetic polymers using MALDI-TOF-MS is possible via automated sample preparation and spotting in a synthetic robot (Figure 13).[63,64] It should therefore be rather easy to implement MALDI screening techniques together with automated sample preparation as a standard technique for combinatorial polymer research.

Light scattering and viscosity techniques also represent powerful methods determine the molecular weight of macromolecules. A combination of both approaches was shown to be a very useful tool for the automated batch characterization of polymer solutions.[105] Light-scattering, refractometric and viscosimetric detectors were coupled with a GPC system in order to obtain reproducible $\bar{M}_w$ data for poly(vinyl pyrrolidine) (PVP) and poly(ethylene oxide) (PEO) polymers in an automated approach. Automated viscosity as a stand-alone utility can be another fast approach for the determination of molecular weights. By measuring concentration series of polymers, the intrinsic viscosity can be obtained and used to calculate the corresponding $\bar{M}_n$ values. Commercial systems that allow parallel viscosity measurements in combination with

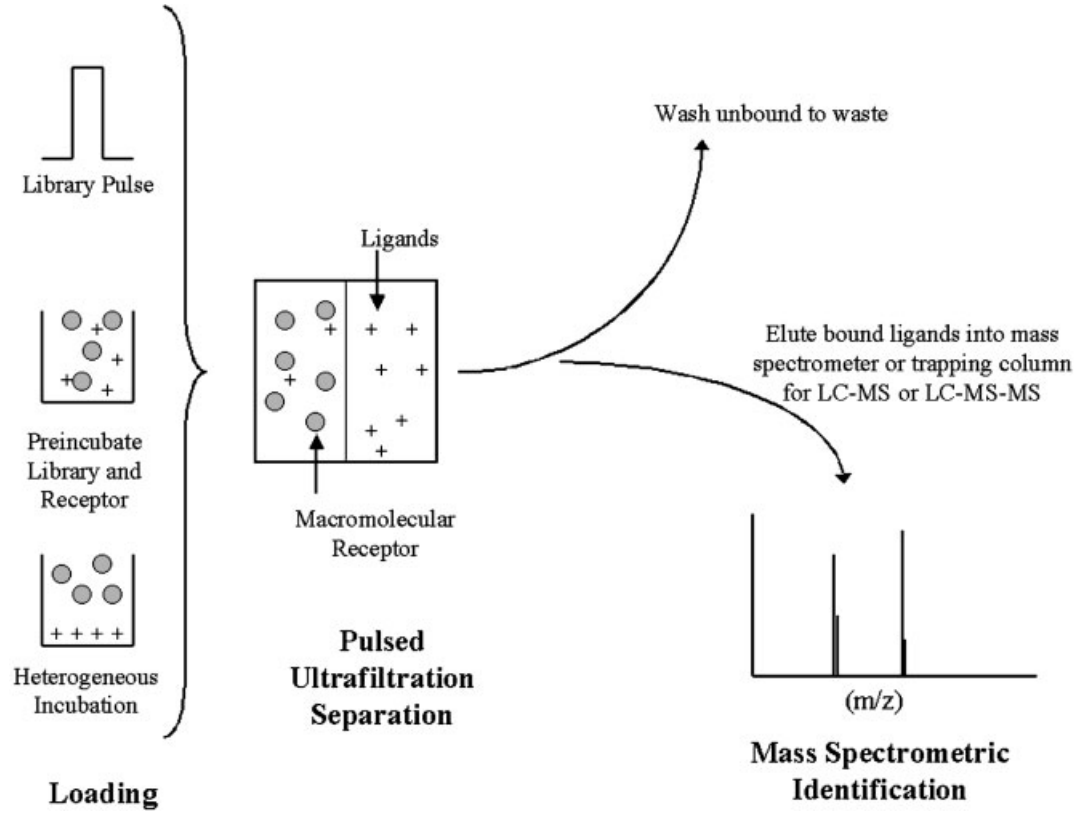


Figure 12. Typical workflow for the screening of a library by means of mass spectrometry.

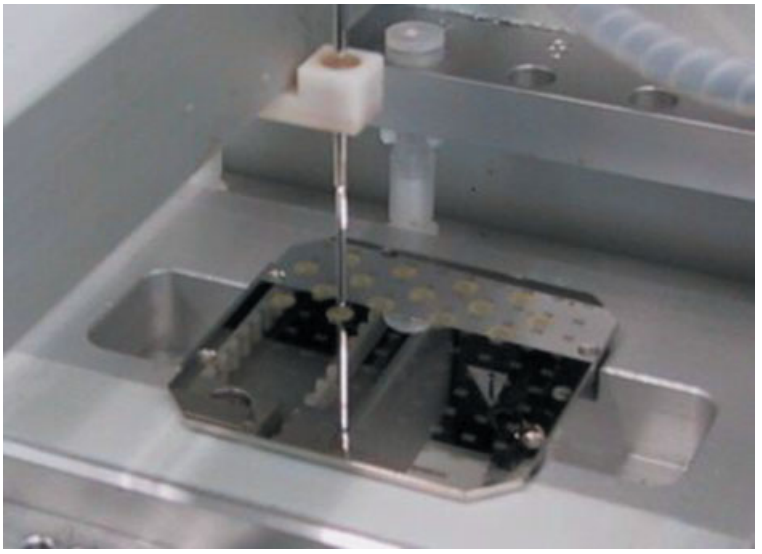


Figure 13. Automated spotting of polymer/matrix solutions on a MALDI-TOF-MS target by an automated synthesizer.

automated dilution, and a robotic system for sample handling have recently become available,[106] but no explicit application has been reported so far.

4.2 Optical Screening Methods

Optical screening methods are a well-developed tool for the high-throughput evaluation of different parameters. Laser-induced fluorescence imaging for instance has been shown to be efficient for high-throughput identification of active catalysts.[107] Parallel absorbance measurements in a 96-well microtiter plate format were used for the screening of active catalysts.[108] In analytical chemistry, optical sensing techniques are widely used for the fast evaluation of different parameters. Fluorescent imaging was shown to be applicable to pH sensing in a 96-well microtiter plate format,[109] and an optical biosensor was developed for the fast analysis of streptomycin residues in pasteurized milk.[110] Fiber optic mid-IR[111] or FT-IR[112] probes were developed for the online monitoring of polymerization reactions. Fourier-transform infrared spectroscopy (FT-IR) was used to monitor living isobutylene and ethylene oxide polymerizations,[113] as well as the copolymerization of ethene and 1-hexene.[114] The acquisition time for an FT-IR spectrum can be as low as 22 s.[113] Monitoring the monomer consumption by means of FT-IR spectroscopy was accomplished by following the intensities of monomer signals in the spectra. These bands decrease during monomer consumption and conversion can be calculated from their intensities. Attenuated total reflection FT-IR (ATR-FT-IR) is also a useful tool for the online monitoring of polymerization reactions as it was shown for the terpolymerization of butyl acetate, methyl methacrylate and vinyl acetate in emulsion,[115] and the carbocationic polymerization of isobutylene.[116] Mülhaupt et al. reported that ATR-FT-IR is applicable for the high-throughput evaluation of olefin copolymer compositions.[117] ATR-FT-IR allows a direct characterization of powders and polymers without preprocessing and is therefore highly suited for the fast evaluation of large sample amounts. The composition of ethene/propene, ethene/1-hexene and ethene/1-octane copolymers could be calculated from ATR-FT-IR spectra with an error of less than 5% by using multivariant calibration. Due to timesaving during sample preparation, the sampling rate for these ATR-FT-IR measurements can be increased to 40 samples per hour. This already high sampling rate could be further increased by using new commercially available equipment,[118] which allows the usage of standard microtiter plates in 96-, 384- and even 1596-well format for the acquisition of FT-IR spectra.

Photoluminescence and UV-vis spectroscopy were used successfully to characterize a library of 14 kinds of π-conjugated polymeric monolayers and 49 kinds of polymeric bilayer films by Muramatsu et al.[119] The library was screened for the bilayer with the strongest emission of blue light. Another successful example for the application of optical screening techniques is the evaluation of adsorption properties of fluorescence-labeled graft copolymers with an imaging set-up.[53] Thousands of new materials could be evaluated with this set-up on a weekly bases. Potyrailo et al. showed that it is possible to evaluate the weathering of polymeric materials[120] and the abrasion resistance of coating libraries[121] by spectroscopic methods. The abrasion resistance of UV-cured coatings was tested by automated light-scattering measurements with a fiber-optic arrangement.[121] This approach was shown to

be at least 10 times faster than the conventional development process for coatings. The UV-induced degradation of polycarbonate (PC), poly(butylene terephthalate) (PBT) and their 45:55 wt.-% blend with two types of pigments (rutile and carbon black) was evaluated by means of fluorescence imaging and spectroscopy.[120] Using this method the screening throughput can be 800 times higher as compared to conventional methods, such as color change or gloss loss. Altogether, optical screening techniques represent very promising methods for the online monitoring and high-throughput screening of polymer properties since they are fast, non-destructive and comparably cheap. However, the implementation of some of these techniques is complicated, as advanced chemometric tools are required. For example, sensors for surfactants could provide helpful information such as the critical micelle concentration in emulsion polymerizations.[122] Another possibility to introduce automation into this field of polymer research could be the utilization of UV-vis plate readers, which offer manifold possibilities for parallel measurements and fast data acquisition. Using beam splitting and advanced detection methods, dozens of samples can be investigated in parallel allowing the measurement of 384 UV spectra within seconds (e.g., Figure 14).[123]

4.3 Screening of Morphology and Physical Properties

Atomic force microscopy (AFM) was shown to be a useful tool for the high-throughput screening of pattern formation in symmetric polystyrene-*block*-poly(methyl methacrylate) (PS-*b*-PMMA) diblock copolymer films.[78] Thickness-gradient films of copolymers (cf. Section 3) with different molecular weights were prepared and their morphology was screened using AFM and optical microscopy with the result, that new morphology patterns of these materials were observed. Figure 15 shows a color optical micrograph of a PS-*b*-PMMA thin-film thickness library. AFM was also utilized for the evaluation of two-dimensional thickness/surface energies of PS-*b*-PMMA. The degree of the formation of islands and holes in the film was found to be dependent on differences in surface energy.[124] A novel high-throughput instrument, comparable to AFM just on the micron scale, was also used successfully to obtain film-toughness and strength data for poly(D,L-lactide)/poly(ε-caprolactone) composition-gradient libraries.[75] This instrument uses steel darts with a diameter of 500 μm to map impact strengths at multiple points of coating libraries.

Structure/property relationships of segmented polyurethaneurea libraries with gradients in curing temperature were

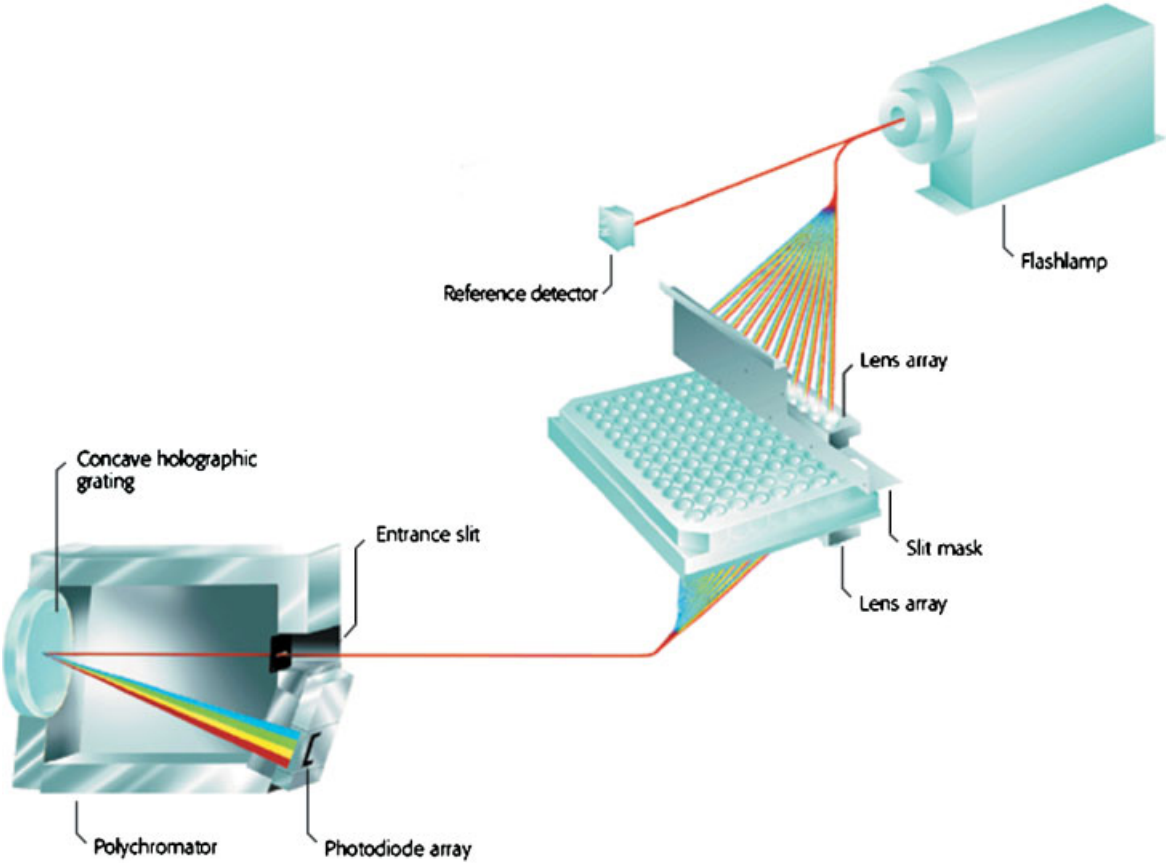


Figure 14. Schematic diagram for high-throughput screening of optical properties (microtiter plate format). (Taken from the literature.[123])

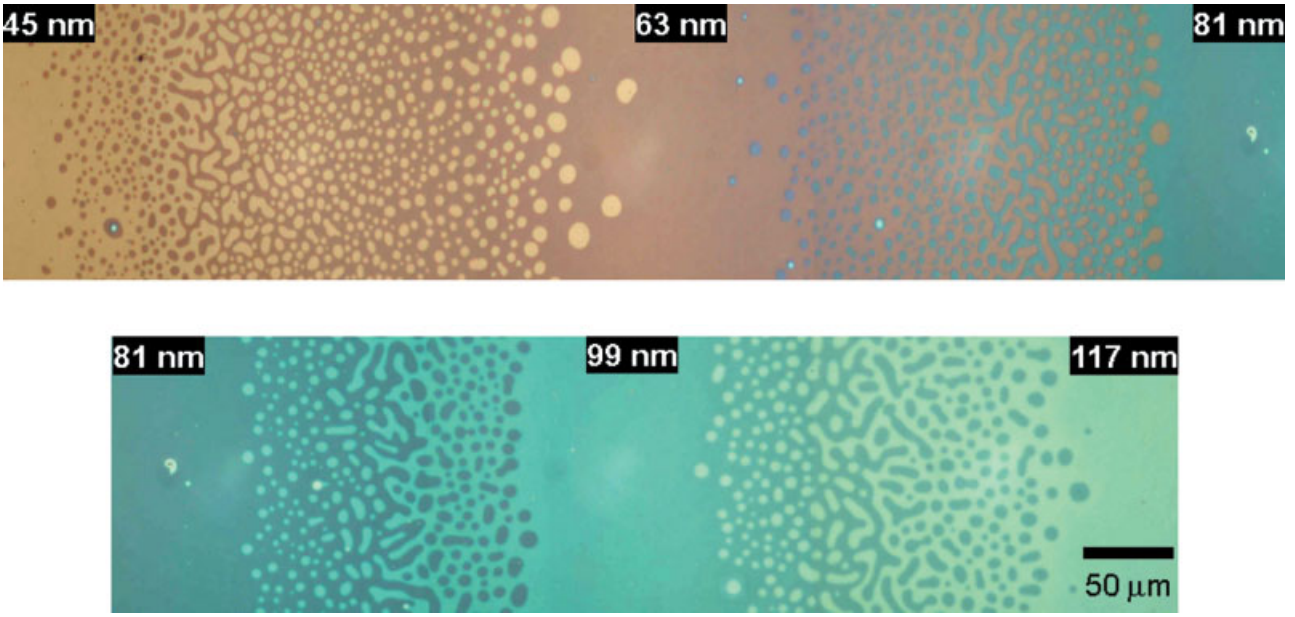


Figure 15. True-color optical micrograph of a continuous 26k polystyrene-*block*-poly(methyl methacrylate) gradient film. The lower section is a continuation of the upper section. (Taken from the literature.[78])

established by correlating AFM and FT-IR measurements with stress/strain profiles.[125] With these combinatorial methods, the optimum curing temperature for a maximum strength in mechanical properties was ascertained.

Optical microscopy was applied to two-dimensional composition/temperature libraries to detect phase separations and microstructures of polystyrene/poly(vinyl methyl ether) blends.[76] An automated set-up combined with an optical microscope was also used to evaluate the dewetting behavior of PS thin films on silicon.[77] The set-up of this high-throughput microscopy system is shown in Figure 8. By investigating thin films of PS with orthogonal, continuous variations in thickness and temperature it was possible to obtain the temperature/thickness/time dependence of dewetting structures and kinetics.

Adhesion properties of a poly(dimethylsiloxane) (PDMS) microlense library were obtained by ''pressing'' the PDMS samples against a PS-coated Si wafer and subsequently removing the wafer at constant speed.[126] An optical microscope was used to obtain digital images of this process, leading to a quantitative map of relative adhesion differences of interfaces created across the microlens array. In addition to the described techniques, Symyx introduced a sensor array modular measurement system (SAMMS^{TM}), which is utilized for applications such as the identification of phase transitions via AC calorimetry or thermal conductivity measurements of thin films.[127]

As shown in this paragraph AFM and optical microscopy are valuable methods to gain access to morphologies and physical parameters of polymers. In the future it should be possible to implement some of these techniques into combinatorial workflows for a full investigation of polymer properties and the build-up of structure/property relationships. For this purpose an automated AFM (LS7, developed by NT-MDT, Moscow, Russia) could be useful, which is equipped with a motorized sample stage that accepts large samples (up to 30 cm) and allows fully automated AFM measurements in all common modes (contact, non-contact, Kelvin, MFM, lithography, force-distance). Therefore, wafers with drop-cast gradients of block copolymers could be easily screened with this AFM. Very recently, Loos et al. reported the utilization of such equipment for the systematic investigation of the surface properties of silicone-rubber-filled carbon black nanocomposites.[128]

5 Conclusions

Even though combinatorial approaches in polymer chemistry are still in its infancy, it can be expected that large numbers of new materials will be discovered with this methodology in the near future. Up to date, several parallel and/or automated approaches have been reported toward the preparation of polymeric materials and their high-throughput characterization. The synthetic strategies applied range from simple free-radical polymerization over controlled radical polymerization and ROP to the production of polyolefins and supramolecular polymers. The degree of automation can roughly be divided into three main groups: Parallel approaches that are carried out with standard laboratory equipment belong to the first group with the lowest degree of automation. The second group can be considered as semi-automated approaches with some degree of automation, such as parallel reactor systems

with manual filling of reagents. The third group consists of approaches with a completely automated workflow, from automated dispensing of all reagents with robotic systems via automated temperature control to product purification.

In comparable manner screening of the materials can be performed with standard equipment, as is described for most combinatorial approaches in polymer chemistry, or with highly developed equipment that is optimized for a high number of samples. High-throughput screening can be utilized for many purposes, such as molecular-weight characterization, screening of optical properties, fast evaluation of morphologies, and physical properties. Many of these techniques can be easily implemented into the combinatorial workflow towards a faster discovery of new materials and the identification of structure/property relationships.

Acknowledgement: The authors would like to thank the *NWO* and *DPI* for financial support.

Received: October 2, 2002
Revised: November 18, 2002
Accepted: December 9, 2002

[1] C. H. Reynolds, *J. Comb. Chem.* **1999**, *1*, 297–306.
[2] J. N. Cawse, *Acc. Chem. Res.* **2001**, *34*, 213–221.
[3] D. T. Williamson, T. E. Long, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42*, 634–635.
[4] M. J. Anderson, J. Whitcomb, *Rubber Plastic News*, October 21, **2002**, 16–18.
[5] G.-J. Gruter, A. Graham, B. McKay, F. Gilardoni, *Macromol. Rapid Commun.* **2003**, *24*, 73–80.
[6] S. P. Rohrer, E. T. Birzin, R. T. Mosley, S. C. Berk, S. M. Hutchins, D.-M. Shen, Y. Xiong, E. C. Hayes, R. M. Parmar, F. Foor, S. W. Mitra, S. J. Degrado, M. Shu, J. M. Klopp, S.-J. Cai, A. Blake, W. W. S. Chan, A. Pasternak, L. Yang, A. A. Patchett, R. G. Smith, K. T. Chapman, J. M. Schaeffer, *Science* **1998**, *282*, 737–740.
[7] K. C. Nicolaou, A. J. Roecker, S. Barluenga, J. A. Pfefferkorn, G.-Q. Cao, *Chem. Bio. Chem.* **2001**, *2*, 460–465.
[8] G. S. Bryan, "*Edison, the Man and His Work*", Knopf, London 1930.
[9] M. Freemantle, *Chem. Eng. News* **1998**, *76*, 37–45.
[10] J. J. Hanak, *J. Mater. Sci.* **1970**, *5*, 964–971.
[11] J. J. Hanak, *Proc. 1^er Colloq. Int. Pulverisation Cathodiques Ses Appl.* **1973**, *Suppl. 165*, 177–197.
[12] L. O. Thompson, J. A. Elman, *Chem. Rev.* **1996**, *96*, 555–600.
[13] E. M. Gordon, M. A. Gallop, D. V. Patel, *Acc. Chem. Res.* **1996**, *29*, 144–154.
[14] A. Nefzi, J. M. Ostresh, R. A. Hougthen, *Chem. Rev.* **1997**, *97*, 449–472.
[15] D. R. Liu, P. G. Schultz, *Angew. Chem.* **1999**, *111*, 36–56; *Angew. Chem. Int. Ed.* **1999**, *38*, 36–54.
[16] S. Otto, R. L. E. Furlan, J. K. M. Sanders, *Drug Discovery Today* **2002**, *7*, 117–125.
[17] *Handbook of Combinatorial Chemistry*, K. C. Nicolaou, R. Hanko, W. Hartwig, Ed., Wiley-VCH, Weinheim 2002.
[18] H. E. Tuinstra, C. H. Cummins, *Adv. Mater.* **2000**, *12*, 1819–1822.
[19] B. Jandeleit, D. J. Schaefer, T. S. Powers, H. W. Turner, W. H. Weinberg, *Angew. Chem.* **1999**, *111*, 2648–2689; *Angew.Chem. Int. Ed.* **1999**, *38*, 2494–2532.
[20] X.-D. Wang, X. Sun, G. Briceño, Y. Lou, K.-A. Wang, H. Chang, W. G. Wallace-Freedman, S.-W. Chen, P. G. Schultz, *Science* **1995**, *268*, 1738–1740.
[21] E. Danielson, J. H. Golden, E. W. McFarland, C. M. Reaves, W. H. Weinberg, X. D. Wu, *Nature* **1997**, *389*, 944–948.
[22] X.-D. Sun, C. Gao, J. Wang, X.-D. Xiang, *Appl. Phys. Lett.* **1997**, *70*, 3353–3355.
[23] J. Wang, Y. Yoo, C. Gao, I. Takeuchi, X. Sun, H. Chang, X.-D. Xiang, P. G. Schultz, *Science* **1998**, *279*, 1712–1714.
[24] P. G. Schultz, X.-D. Xiang, *Curr. Opin. Solid State Mater. Sci.* **1998**, *3*, 153–158.
[25] S. M. Senkan, *Nature* **1998**, *394*, 350–353.
[26] S. J. Taylor, J. P. Morken, *Science* **1998**, *280*, 267–270.
[27] T. R. Boussie, C. Coutard, H. Turner, V. Murphy, T. S. Powers, *Angew. Chem.* **1998**, *110*, 3472–3475; *Angew. Chem. Int. Ed.* **1998**, *37*, 3272–3275.
[28] P. P. Pescarmona, J.C. van der Waal, I. E. Maxwell, T. Maschmeyer, *Catal. Lett.* **1999**, *63*, 1–11.
[29] T. Bein, *Angew. Chem.* **1999**, *111*, 335–338; *Angew. Chem. Int. Ed.* **1999**, *38*, 323–326.
[30] W. F. Maier, *Angew. Chem.* **1999**, *111*, 1294–1296; *Angew. Chem. Int. Ed.* **1999**, *38*, 1216–1218.
[31] M. T. Reetz, *Angew. Chem.* **2001**, *113*, 292–320; *Angew. Chem. Int. Ed.* **2001**, *40*, 284–310.
[32] F. Schüth, O. Busch, C. Hoffmann, T. Johann, C. Kiener, D. Demuth, J. Klein, S. Schunk, W. Strehlau, T. Zech, *Top. Catal.* **2002**, *1*, 55–66.
[33] U.S. 5,175,209 (1991), Baylor College of Medicine, invs.: K. L. Beattie, J. D. Frost; *Chem. Abstr. 119*, 112906.
[34] R. F. Service, *Science* **1997**, *277*, 474–475.
[35] E. W. McFarland, W. H. Weinberg, *Trends Biotechnol.* **1999**, *17*, 107–111.
[36] S. Brocchini, *Adv. Drug Deliv. Rev.* **2001**, *53*, 123–130.
[37] J. D. Hewes, L. A. Bendersky, *Appl. Surf. Sci.* **2002**, *189*, 196–204.
[38] R. Iden, W. Schrof, J. Hadeler, S. Lehmann, *Macromol. Rapid Commun.* **2003**, *24*, 63–72.
[39] J. C. Meredith, A. Karim, E. J. Amis, *MRS Bull.* **2002**, *27*, 330–335.
[40] S. Brocchini, K. James, V. Tangpasuthadol, J. Kohn, *J. Am. Chem. Soc.* **1997**, *119*, 4553–4554.
[41] S. Brocchini, K. James, V. Tangpasuthadol, J. Kohn, *J. Biomed. Mater. Res.* **1998**, *42*, 66–75.
[42] D. M. Lynn, D. G. Anderson, D. Putnam, R. Langer, *J. Am. Chem. Soc.* **2001**, *123*, 8155–8156.
[43] O. Lavastre, I. Illitchev, G. Jegou, P. H. Dixneuf, *J. Am. Chem. Soc.* **2002**, *124*, 5278–5279.
[44] U.S. 6,307,004, (2001), General Electric Company, invs.: J. C. Carnahan, J. P. Lemmon, T. K. Leib, R. A. Potyrailo, G. L. Warner; *Chem. Abstr. 134*, 252754.
[45] S. M. Alesso, Z. Yu, D. Pears, P. A. Worthington, R. W. A. Luke, M. Bradley, *J. Comb. Chem.* **2001**, *3*, 631–633.
[46] M. Bradley, *Polym. Prepr. (Am. Chem. Soc.; Div. Polym. Chem.)* **2001**, *42(2)*, 629.
[47] Picture of the parallel synthesizer: www.ventacon.com.
[48] D. J. Gravert, A. Datta, P. Wentworth, Jr., K. D. Janda, *J. Am. Chem. Soc.* **1998**, *120*, 9481–9495.

[49] F. Lanza, B. Sellergren, *Anal. Chem.* **1999**, *71*, 2092–2096.
[50] F. Lanza, A. J. Hall, B. Sellengren, A. Bereczki, G. Horvai, S. Bayoudh, P. A. G. Cormack, D. C. Sherrington, *Anal. Chim. Acta* **2001**, *435*, 91–106.
[51] Argonaut Technologies, Application note, No. 28.
[52] U.S. 6,346,290 (2002), Symyx Technologies Inc., invs.: P. G. Schultz, X. Xiang.
[53] D. Charmot, P. Mansky, O. Kolosov, D. Benoit, G. Klärner, M. Jayaraman, M. Piotti, H. T. Chang, V. Nava-Salgada, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42(2)*, 627–628.
[54] R. B. Nielsen, A. L. Safir, M. Petro, T. S. Lee, P. Huefner, *Polym. Mater. Sci. Eng.* **1999**, *80*, 92.
[55] G. Klaerner, A. L. Safir, H.-T. Chang, M. Petro, R. B. Nielson, *Polym. Preprints* **1999**, *40*, 469.
[56] H. Zhang, R. Hoogenboom, M. W. M. Fijten, U. S. Schubert, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2002**, *43*, 17–18.
[57] H. Zhang, M. W. M. Fijten, R. Hoogenboom, R. Reinierkes, U. S. Schubert, *Macromol. Rapid Commun.* **2003**, *24*, 81–86.
[58] A. W. Bosman, A. Heumann, G. Klaerner, D. Benoit, J. M. J. Fréchet, C. J. Hawker, *J. Am. Chem. Soc.* **2001**, *123*, 6461–6462.
[59] C. J. Hawker, A. W. Bosman, J. M. J. Fréchet, E. Harth, A. Heumann, M. Ranger, B. van Horn, G. Klaerner, D. Benoit, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42(2)*, 639–640.
[60] P. Chapon, C. Mignaud, G. Lizarraga, M. Destarac, *Macromol. Rapid Commun.* **2003**, *24*, 87–91.
[61] F. Nederberg, E. F. Conner, M. Möller, T. Glauser, J. L. Hedrick, *Angew. Chem.* **2001**, *113*, 2784–2787; *Angew. Chem. Int. Ed.* **2001**, *40*, 2712–2715.
[62] Argonaut Technologies, Application note, No. 33.
[63] R. Hoogenboom, M. W. M. Fijten, U. S. Schubert, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2002**, *43*, 969–970.
[64] R. Hoogenboom, M. W. M. Fijten, M. A. R. Meier, U. S. Schubert, *Macromol. Rapid Commun.* **2003**, *24*, 92–97.
[65] R. Hoogenboom, M. W. M. Fijten, C. Brändli, J. Schroer, U. S. Schubert, *Macromol. Rapid Commun.* **2003**, *24*, 98–103.
[66] N. Kashiwa, J. Imuta, *Catal. Surv. Jpn.* **1997**, *1*, 125–142.
[67] T. R. Boussie, V. Murphy, K. A. Hall, C. Coutard, C. Dales, M. Petro, E. Carlson, H. W. Turner, T. S. Powers, *Tetrahedron* **1999**, *55*, 11699–11710.
[68] A. Tuchbreiter, R. Mülhaupt, *Macromol. Symp.* **2001**, *173*, 1–20.
[69] M. Stork, A. Herrmann, T. Nemnich, M. Klapper, K. Müllen, *Angew. Chem.* **2000**, *112*, 4544–4547; *Angew. Chem. Int. Ed.* **2000**, *39*, 4367–4369.
[70] J. Tian, G. W. Coates, *Angew. Chem.* **2000**, *112*, 3772–3775; *Angew. Chem. Int. Ed.* **2000**, *39*, 3626–3629.
[71] U. S. Schubert, C. Eschbaumer, *Angew. Chem.* **2002**, *114*, 3016–3050; *Angew. Chem. Int. Ed.* **2002**, *41*, 2892–2926.
[72] L. Brunsveld, B. J. B. Folmer, E. W. Meijer, R. P. Sijbesma, *Chem. Rev.* **2001**, *101*, 4071–4097.
[73] S. Schmatloch, C. Brändli, H.-H. Nguyen, U. S. Schubert, *Polym. Mater. Sci. Eng.* **2002**, *87*, 237–238.
[74] Chemspeed Ltd., Application Note, No. 010.
[75] C. Meredith, J.-L. Sormana, A. Tona, H. Elgendy, A. Karim, E. Amis, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42(1)*, 649–650.
[76] J. C. Meredith, A. Karim, E. J. Amis, *Macromolecules* **2000**, *33*, 5760–5762.
[77] J. C. Meredith, A. P. Smith, A. Karim, E. J. Amis, *Macromolecules* **2000**, *33*, 9747–9756.
[78] A. P. Smith, J. F. Douglas, J. C. Mederith, E. J. Amis, A. Karim, *J. Polym. Sci. Part B: Polym. Phys.* **2001**, *39*, 2141–2158.
[79] A. P. Smith, J. F. Douglas, J. C. Meredith, E. J. Amis, A. Karim, *Phys. Rev. Lett.* **2001**, *87*, 15503(4).
[80] C. H. Davis, A. Karim, K. L. Beers, A. J. Crosby, A. P. Smith, E. J. Amis, *Polym. Mater. Sci. Eng.* **2002**, *87*, 239–240.
[81] T. A. Dickinson, D. R. Walt, *Anal. Chem.* **1997**, *69*, 3413–3418.
[82] R. A. Potyrailo, R. J. Wroczynski, J. E. Pickett, M. Rubinsztajn, *Macromol. Rapid Commun.* **2003**, *24*, 123–130.
[83] M. T. Reetz, *Angew. Chem.* **2002**, *114*, 1391–1394; *Angew. Chem. Int. Ed.* **2002**, *41*, 1335–1338.
[84] A. Hagemeyer, B. Jandeleit, Y. Liu, D. M. Poorjary, H. W. Turner, A. F. Volpe, Jr., W. H. Weinberg, *Appl. Catal., A* **2001**, *221*, 23–43.
[85] J. M. Moore, *Curr. Opin. Biotechnol.* **1999**, *10*, 54–58.
[86] X. Cheng, J. Hochlowski, *Anal. Chem.* **2002**, *74*, 2679–2690.
[87] *Column Handbook for Size Exclusion Chromatography*, C.-S. Wu, Ed., Dekker, New York 2002.
[88] *Encyclopedia of Chromatography*, online ed., J. Cazes, Ed., Dekker, New York 2002.
[89] H. Pasch, P. Kilz, *Macromol. Rapid Commun.* **2003**, *24*, 104–108.
[90] PSS HighSpeed columns, Polymer Standards Service GmbH, 55120 Mainz, Germany; www.polymer.de.
[91] Polymer Laboratories Rapid Screening column, Polymer Laboratories, Shropshire SY6 6AX, UK; www.polymerlabs.com.
[92] M. Petro, F. Svec, I. Gitsov, J. M. J. Fréchet, *Anal. Chem.* **1996**, *68*, 315–321.
[93] M. Jančo, D. Sýkora, F. Svec, J. M. J. Fréchet, J. Schweer, R. Holm, *J. Polym. Sci. Part A: Polym. Chem.* **2000**, *38*, 2767–2778.
[94] S. Lubbad, M. R. Buchmeiser, *Macromol. Rapid Commun.* **2002**, *23*, 617–621.
[95] G. Klaerner, A. L. Safir, H.-T. Chang, M. Petro, R. B. Nielson, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **1999**, *40(1)*, 469.
[96] K. P. Peil, *DECHEMA Monographs* **2001**, *137*, 71–78.
[97] M. Petro, A. Safir, R. B. Nielson, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **1999**, *40(2)*, 702.
[98] M. W. F. Nielen, *Mass Spectrom. Rev.* **1999**, *18*, 309–344.
[99] S. Weidner, G. Kühn, J. Friedrich, *Rapid Commun. Mass Spectrom.* **1998**, *12*, 1373–1381.
[100] Y. G. Shin, R. B. van Breemen, *Biopharm. Drug. Dispos.* **2001**, *22*, 353–372.
[101] D. Chelius, P. V. Bondarenko, *J. Proteome Res.* **2002**, *1*, 317–323.
[102] O. Keil, T. LeRiche, H. Deppe, D. A. Volmer, *Rapid Commun. Mass Spectrom.* **2002**, *16*, 814–820.
[103] S. Ekström, P. Önnerfjord, J. Nilsson, M. Bengtsson, T. Laurel, G. Marko-Varga, *Anal. Chem.* **2000**, *72*, 286–293.
[104] A. Graven, P. M. St. Hilaire, S. J. Sanderson, J. C. Mottram, G. H. Coombs, M. Meldal, *J. Comb. Chem.* **2001**, *3*, 441–452.
[105] R. Srelitzki, W. F. Reed, *J. Appl. Polym. Sci.* **1999**, *73*, 2359–2368.

[106] Lauda Dr. R. Wobser GmbH & Co KG, Königshofen, Germany; www.lauda.de.
[107] H. Su, Y. Hou, R. S. Houk, G. L. Schrader, E. S. Yeung, *Anal. Chem.* **2001**, *73*, 4434–4440.
[108] N. D. Morris, T. E. Mallouk, *J. Am. Chem. Soc.* **2002**, *124*, 11114–11121.
[109] G. Liebsch, I. Klimant, C. Krause, O. S. Wolfbeis, *Anal. Chem.* **2001**, *73*, 4354–4363.
[110] G. A. Baxter, J. P. Ferguson, M. C. O'Conner, C. T. Elliott, *J. Agric. Food Chem.* **2001**, *49*, 3204–3207.
[111] G. Chatzi, O. Kammona, C. Kiparissides, *J. Appl. Polym. Sci.* **1997**, *63*, 799–809.
[112] J. E. Puskas, P. Antony, Y. Kown, C. Paulo, M. Kovar, R. R. Norton, G. Kaszas, V. Altstädt, *Macromol. Mater. Eng.* **2001**, *286*, 565–580.
[113] M. Lanzendörfer, H. Schmalz, V. Abetz, A. H. E. Müller, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42(1)*, 329–330.
[114] A. Tuchbreiter, R. Mülhaupt, A. Warmbold, P. Liebertraut, B. Koppler, J. Honerkamp, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2002**, *43*, 285–286.
[115] H. Hua, M. A. Dubé, *J. Polym. Sci., Part A: Polym. Chem.* **2001**, *39*, 1860–1876.
[116] R. F. Storey, T. L. Maggio, *Macromolecules* **2000**, *33*, 681–688.
[117] A. Tuchbreiter, J. Marquardt, J. Zimmermann, P. Walter, R. Mülhaupt, *J. Comb. Chem.* **2001**, *3*, 598–603.
[118] Bruker Optics Inc., Billerica, MA, USA; www.brukeroptics.com.
[119] Y. Muramatsu, T. Yamamoto, T. Hayakawa, H. Koinuma, *Appl. Surf. Sci.* **2002**, *189*, 319–326.
[120] R. A. Potyrailo, J. E. Picket, *Angew. Chem.* **2002**, *114*, 4404–4407; *Angew. Chem. Int. Ed.* **2002**, *41*, 4230–4233.
[121] R. A. Potyrailo, B. J. Chisholm, D. R. Olsen, M. J. Brennan, C. A. Molaison, *Anal. Chem.* **2002**, *74*, 5105–5111.
[122] P. Bühlmann, E. Pretsch, E. Bakker, *Chem. Rev.* **1998**, *98*, 1593–1687.
[123] Analytik Jena AG, Jena, Germany; www.Analytik-Jena.com.
[124] A. P. Smith, A. Sehgal, J. F. Douglas, A. Karim, E. J. Amis, *Macromol. Rapid Commun.* **2003**, *24*, 131–135.
[125] J.-L. Sormana, J. C. Meredith, *Macromol. Rapid Commun.* **2003**, *24*, 118–122.
[126] A. J. Crosby, A. Karim, E. J. Amis, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42(2)*, 645–646.
[127] P. Mansky, *Polym. Prepr. (Am. Chem. Soc., Div. Polym. Chem.)* **2001**, *42(2)*, 647–648.
[128] R. Neffati, A. Alexeev, S. Saunin, J. C. M. Brokken-Zijp, D. Wouters, S. Schmatloch, U. S. Schubert, J. Loos, *Macromol. Rapid Commun.* **2003**, *24*, 113–117.