Available online at www.sciencedirect.com
SCIENCE DIRECT®
Polymer 44 (2003) 7933–7938
polymer
www.elsevier.com/locate/polymer

N-(2-Anthracene)methacrylamide: a new fluorogenic probe molecule for monitoring in situ the radiation-induced polymerization of methyl methacrylate in bulk and in solution

M.S. Frahn*, L.H. Luthjens, J.M. Warman

Department of Radiation Chemistry, IRI, Delft University of Technology, Mekelweg 15, 2629 JB Delft, The Netherlands

Received 26 March 2003; received in revised form 1 September 2003; accepted 6 October 2003

Abstract

This paper introduces the fluorogenic molecule *N*-(2-anthracene)methacrylamide, AnMA, employed as a probe for monitoring radiation-induced polymerization in situ. The relative reactivity of AnMA during free radical polymerization of methyl methacrylate (MMA) has been determined via fluorescence techniques. The ratio between propagation rate constants for the reaction of an MMA free radical chain-end with AnMA relative to the reaction with MMA is found to be 0.96. It is demonstrated that AnMA can be used for monitoring MMA polymerization quantitatively throughout the entire course of the reaction including the gel effect region. Results are also provided which demonstrate that AnMA can be used for sensitive monitoring of MMA polymerization both in bulk and in solution.

Keywords: Fluorogenic probe; Radiation; Methyl methacrylate

1. Introduction

Since the early investigations of Loutfy [1–3] into the use of fluorescent molecules for in situ monitoring of polymerization, continued research in this field has led to the development of several different types of fluorescent probes. Common to most of these probe types is sensitivity to the mobility and/or (micro-)viscosity of the molecular environment in which the probe molecules are located. This sensitivity can be achieved via a number of physical interactions; intramolecular reorientation (e.g. molecular rotors [1,4,5] and intramolecular excimer-forming probes [6,7]), diffusion-controlled interactions (e.g. intermolecular excimer-forming probes [8]), or solvent dipole stabilization of the probe's excited state (e.g. charge-transfer (CT) probes [9,10]). It is important to note that the physical origin of the sensitivity to the molecular environment for a given probe molecule is not necessarily determined by only one of the above mentioned interactions.

A new class of probe molecules which have been studied recently are the fluorogenic molecular probes [11–15]. Such molecules are completely non-fluorescent until they are incorporated, i.e. copolymerized, into growing polymer chains. This aspect, unique to the fluorogenic probes, provides unprecedented sensitivity for monitoring polymerization at the early stage of reaction. Furthermore, as is demonstrated in this paper, fluorogenic probe molecules can also be employed for monitoring solution polymerizations.

Our research group has devoted considerable effort to studying fluorogenic molecular probes. The molecular structures of some examples are provided in Fig. 1. The fluorogenic character of these molecules is achieved by the α,β-unsaturation relative to the carbonyl groups in the free radical-reactive moieties of the probes. For MPy the reactive moiety is a maleimide group, whereas for PyMA and AnMA it is a methacrylamide group. When copolymerized into growing polymer chains, the double bonds become saturated and consequently the molecule becomes fluorescent. With increasing conversion of monomer to polymer more probe molecules are converted into the fluorescent derivative resulting in a steadily increasing intensity of fluorescence. Furthermore, since the primary probe response is based upon a chemical change and not a change in the physical properties of the polymerizing medium, fluorogenic molecules can be used for sensitive monitoring

* Corresponding author. Tel.: +31-6-4840-8388; fax: +31-15-278-7421.
E-mail address: msfrahn@netscape.net (M.S. Frahn).

0032-3861/$ - see front matter © 2003 Elsevier Ltd. All rights reserved.
doi:10.1016/j.polymer.2003.10.024

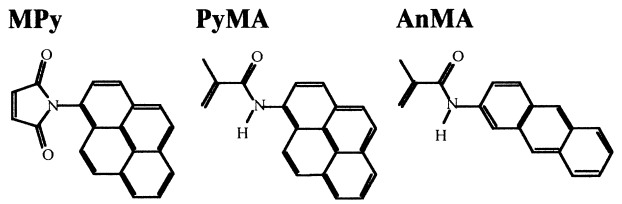

Fig. 1. Molecular structures of the fluorogenic probes *N*-(1-pyrene)maleimide (MPy), *N*-(1-pyrene)methacrylamide (PyMA) and *N*-(2-anthracene)-methacrylamide (AnMA).

of solution polymerizations which to the best of our knowledge is not possible with traditional fluorescent probes.

An important parameter required for the quantitative monitoring of polymerization via fluorogenic probes is the relative rate at which such molecules are incorporated, i.e. copolymerized, in relation to the polymerizing monomer. Because the amount of probe used is considerably less than that for typical copolymerization reactions, conventional techniques by which monomer reactivity ratios can be determined are impractical. We have previously reported a sensitive technique by which the reactivity of a probe molecule in relation to that of the monomer, i.e. the ratio between the propagation rate constants given in Fig. 2, could be determined from fluorescence experiments [14,15]. This technique has been repeated for the AnMA probe molecule and is discussed in Section 3. Using this result in combination with considerations given to density changes and changes occurring in the probe molecule's fluorescence quantum efficiency during the course of MMA polymerization, quantitative monitoring of the entire course of MMA polymerization using AnMA is possible.

We emphasize that the concentration of the probe molecules in the present experiments is only a few millimolar corresponding to less than one probe molecule per 1000 MMA molecules. The probability of propagation involving an AnMA radical end group with another AnMA molecule is therefore extremely small and can be neglected. The rate constant for the reaction of a radical AnMA end group with MMA would have to be very much smaller than

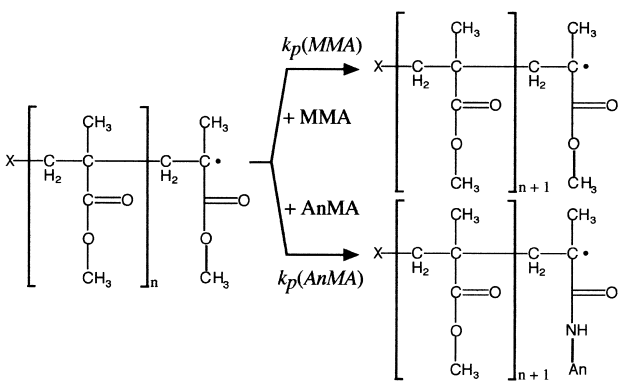

Fig. 2. Competitive propagation reactions during the free radical polymerization of MMA with a trace amount of the AnMA probe molecule present.

that for a radical MMA end group with MMA in order to have an appreciable influence on the overall polymerization process. Evidence for the lack of a significant effect of the probe on overall MMA polymerization is the fact that the dose at which the gel effect occurs is identical to that found from measurements of the monomer conversion in the absence of the probe.

One of the main advantages in using the anthracene derivative instead of the previously used pyrene derivatives is the considerably shorter lifetime of the excited state of the anthracene moiety; ca. 10 ns compared with ca. 200 ns. Because of this, quenching of fluorescence by trace impurities such as oxygen is less of a problem.

2. Experimental section

2.1. Materials

Methyl methacrylate (Merck Synthesis grade) was trap-to-trap distilled on a greaseless vacuum line at 100 °C to remove the hydroquinone stabilizer immediately prior to use. The synthesis of the fluorogenic probe AnMA was analogous to that fully described for the pyrene methacrylate derivative [15]. The freshly prepared AnMA was purified by re-crystallization from a 4:1 methylene chloride/hexane solvent mixture prior to use. Anthracene (Fluka HPLC >99%) and 9,10-diphenylanthracene (Molecular Probes HPLC >99%) were used as received. The benzene solvent, used in the MMA solution experiments, was obtained from Merck (UV spectroscopic grade) and was purified by distillation on a Fischer 'Spaltrohr HMS500' spinning-band column.

2.2. Methods

Except when mentioned otherwise, sample cells were constructed in-house from Heraeus Suprasil quartz tubing. Deaeration of the samples was carried out by three consecutive freeze-pump-thaw cycles on a vacuum line or via purging with argon gas for 15 min prior sealing with a Teflon stopper and Parafilm tape.

Optical absorption spectra were measured using a Perkin–Elmer Lambda 40 UV/VIS Spectrophotometer. Optical densities of the probe solutions were determined at 337 nm, the excitation wavelength used in fluorescence measurements.

Fluorescence spectra and quantum yields were obtained with a Photon Technology International QuantaMaster model QM-1 spectrometer. In situ fluorescence spectra were obtained using an experimental set-up described previously [12]. In situ fluorescence lifetime measurements were also obtained using an experimental flash-photolysis set-up described in a separate publication [16].

Low dose rate irradiations at ca. 0.6 kGy/h were carried out with a cobalt-60 Gammacell 200 irradiator from Atomic

Energy of Canada, Ltd. High dose rate irradiations at ca. 7 kGy/h were accomplished with an MDS Nordion cobalt-60 Gammacell 220 irradiator. Dose rates were accurately determined by Fricke dosimetry [17] and corrected for the natural decay of the source.

Direct measurement of monomer conversion was accomplished by the use of standard 20 ml scintillation vials into which a known mass (ca. 3 g) of purified and degassed MMA was administered. All manipulations with the monomer were carried out in a glove bag filled with nitrogen. Each sample was tightly closed and sealed with Parafilm tape prior to extraction from the glove bag and subsequent polymerization. After an allotted dose, the samples were removed from the source and immediately opened to atmospheric oxygen, hence rapidly terminating polymerization. The conversion to polymer was determined gravimetrically after residual monomer evaporation under reduced pressure (ca. 10 mm Hg) at room temperature.

3. Results and discussion

3.1. Anthracene methacrylamide (AnMA)

The absorption spectrum of AnMA in MMA is shown in Fig. 3 along with the fluorescence spectrum of the copolymerized derivative of the probe. The AnMA probe molecule is non-fluorescent until the double bond of the methacrylamide group becomes saturated due to copolymerization into growing polymer chains. The vibrational structure in the absorption spectrum resembles that of anthracene with an additional long wavelength absorption peak due to the electronic coupling with the methacrylamide group. The fluorescence spectrum of the copolymerized derivative is similar to that of the anthracene chromophore alone but with less sharp vibrational structure.

3.2. The relative reactivity of AnMA during MMA polymerization

In order to be able to correlate the MMA monomer conversion to the fluorescence intensity observed from a polymerizing sample containing the AnMA probe molecule it is necessary to determine the relative reactivity of AnMA to that of MMA, i.e. k_p(AnMA)/k_p(MMA) as shown in Fig. 2. The ideal situation is that in which the probe molecule and the MMA monomer both exhibit an identical reactivity to propagating free radicals. We have previously reported on a method for determining this relative reactivity parameter [14,15]. Essential to this method is the use of a reference fluorescent molecule for which the photophysical properties do not change during the low conversion region of polymerization. The reference molecule initially chosen for this purpose was unsubstituted anthracene, A.

Fig. 4 shows the integrated fluorescence intensities, I_{FL}(A) and I_{FL}(PAnMA), as a function of dose from A and AnMA sample solutions, respectively, both having an optical density at 337 nm of 0.10. As is evident in the figure, the fluorescence intensity from the A sample does not change during the initial period of polymerization whereas that for the AnMA sample steadily increases with increasing dose.

The fractional conversion of AnMA, F_C(AnMA), can be derived from the data in Fig. 4 (see Eq. (1)) if the ratio between the fluorescence efficiencies of A, ϕ_{FL}(A), and the polymerized derivative of AnMA, ϕ_{FL}(PAnMA), is known.

$$F_C(\text{AnMA}) = \frac{I_{FL}(\text{PAnMA})\phi_{FL}(\text{A})}{I_{FL}(\text{A})\phi_{FL}(\text{PAnMA})} \quad (1)$$

A value of 1.33 ± 0.05 for ϕ_{FL}(A)/ϕ_{FL}(PAnMA) has been determined from separate spectrofluorimeter measurements on solutions of A and PAnMA as described previously [14,15] using Eq. (2).

$$\frac{\phi_{FL}(\text{PAnMA})}{\phi_{FL}(\text{A})} = \frac{(1 - 10^{-\text{OD(A)}})I_{FL}(\text{PAnMA})}{(1 - 10^{-\text{OD(PAnMA)}})I_{FL}(\text{A})} \quad (2)$$

In Eq. (2), OD(A) and OD(PAnMA) are the optical densities of the solutions.

As discussed previously [15], for copolymerization with only a trace amount of probe molecules (i.e. comonomer) present, the ratio between the relevant propagation rate

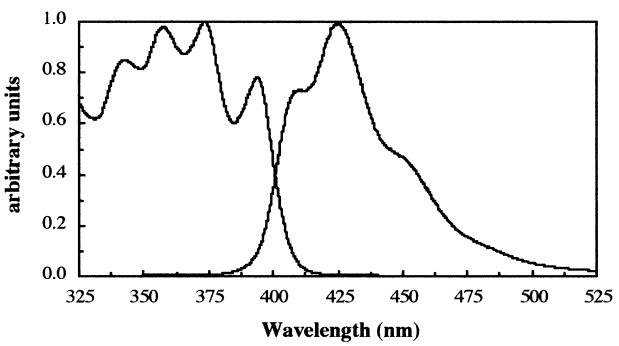

Fig. 3. The absorption spectrum of AnMA in MMA and the fluorescence spectrum of the copolymerized derivative.

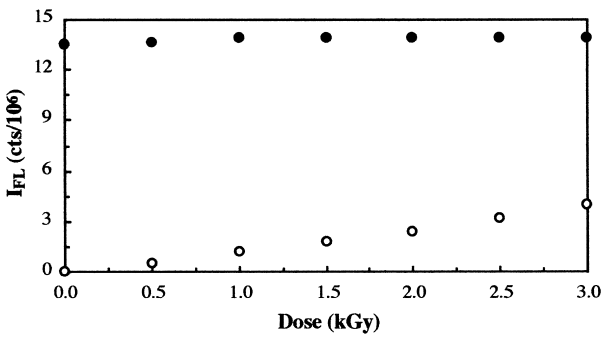

Fig. 4. Fluorescence intensities versus dose of γ-radiation on photoexcitation at 337 nm of $OD_{337} = 0.10$ solutions of anthracene (filled circles) and AnMA (open circles) in MMA.

coefficients at low conversions is given by Eq. (3):

$$\frac{k_p(\text{AnMA})}{k_p(\text{MMA})} = \frac{F_C(\text{AnMA})}{F_C(\text{MMA})} \tag{3}$$

Table 1 gives the experimental results obtained from the data in Fig. 4 and the corresponding values of MMA conversion. The average value of the results given in the final column of Table 1 is 0.93.

A repeat of the experiment conducted under identical conditions with however the choice of DPA as the reference fluorophore ($\phi_{FL}(\text{PAnMA})/\phi_{FL}(\text{DPA}) = 0.34 \pm 0.01$) gave an average value of 1.00 for the ratio between the above mentioned propagation rate constants. The average value from the two data sets is 0.96. It is therefore evident that the ideal condition in which the reactivity of the probe molecule is equal to that of the monomer is closely met.

3.3. Monitoring the gel effect in MMA using AnMA

In addition to being able to monitor the progress of polymerization in the low conversion regime, fluorogenic probes are also capable of sensitively monitoring the occurrence of the gel effect; the autoacceleration of polymerization which occurs above a certain monomer conversion. This is illustrated in Fig. 5 by the dramatic increase in fluorescence from a solution of AnMA in MMA above a total dose of ca. 3.5 kGy using the Gammacell 200 irradiator. Within the shaded area of the figure the fluorescence intensity increases by a factor of 5.5. Over the same dose range the monomer conversion is found to increase by a factor of 4.3, from 0.23 to 0.98. The fact that the increase in the fluorescence intensity is significantly larger than the increase in MMA conversion would appear to be in conflict with the close to equality of the propagation rate constants for AnMA and MMA. It is known however that the quantum yield of fluorescence can be influenced by changes in the rigidity of the medium, an effect that accompanies the gel effect.

The quantum yield of fluorescence, ϕ_{FL}, is related to the rates of radiative decay, k_r, non-radiative internal conversion, k_{ic}, and intersystem crossing, k_{isc}, by

$$\phi_{FL} = k_r/(k_r + k_{nr} + k_{isc}) \tag{4}$$

It has been argued that an increase in ϕ_{FL}, which is often observed when the environment of a fluorescent chromophore changes from a low viscosity fluid to a rigid glass, can be attributed to a decrease in the rate constant for internal conversion [18]. This argument is based on the fact that coupling between the molecule and the surrounding medium can be a determining factor in the rate of radiationless processes [19] since the medium provides an additional sink for the dissipation of the excitation energy. Since both k_r and k_{isc} are expected to be insensitive to the rigidity of the medium, a decrease in k_{ic} should result in an increase in the fluorescence lifetime, τ, according to

$$\tau = (k_r + k_{nr} + k_{isc})^{-1} \tag{5}$$

We have measured in situ the fluorescence decay of PAnMA throughout the course of MMA polymerization using flash-photolysis equipment described elsewhere [16]. The decay was monoexponential both prior to and after the autoacceleration region and the lifetimes obtained, using the Gammacell 220 irradiator, are shown in Fig. 6. As can be seen the lifetime increases from an average value of 9.6–11.2 ns on vitrification.

From Eqs. (4) and (5), the fluorescence quantum yield is

Table 1
Calculated ratios between the propagation rate constants k_p(AnMA) and k_p(MMA)

Dose (kGy)	I_{FL}(A) (cts/10^6)	I_{FL}(AnMA) (cts/10^6)	F_C(AnMA)[a]	F_c(MMA)[b]	$\frac{k_p(\text{AnMA})^c}{k_p(\text{MMA})}$
0.5	13.65	0.515	0.028	0.035	0.82
1.0	13.90	1.168	0.063	0.069	0.91
1.5	13.89	1.768	0.096	0.104	0.92
2.0	13.88	2.409	0.131	0.138	0.94
2.5	13.92	3.131	0.169	0.173	0.98
3.0	13.91	3.945	0.213	0.208	1.03

[a] Calculated using Eq. (1).
[b] Determined by monomer evaporation.
[c] Calculated using Eq. (3).

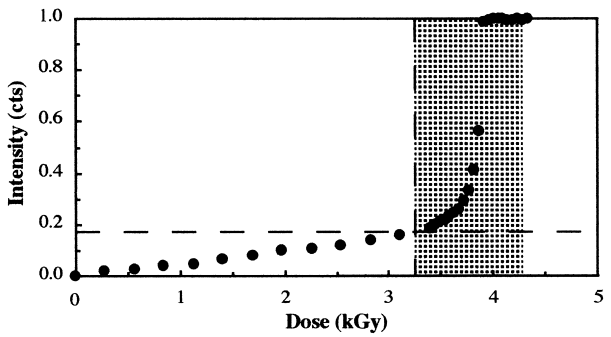

Fig. 5. Normalized fluorescent light intensity from the AnMA probe in MMA ($OD_{337\ nm} = 0.20$) versus dose in the Gammacell 200 irradiator (dose rate of 0.58 kGy/h). The shaded region of the graph corresponds to an increase in MMA conversion from 0.23 to 0.98.

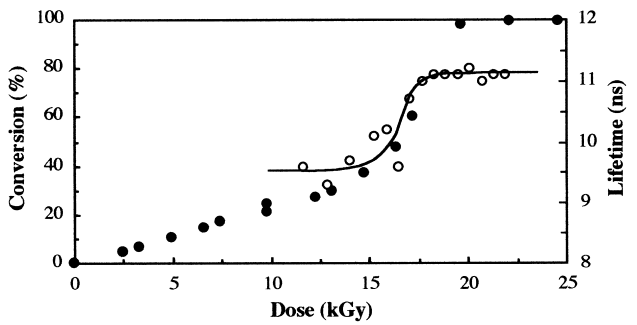

Fig. 6. MMA conversion (filled circles) and fluorescence lifetime (open circles) of the AnMA probe molecule versus γ-ray dose in the Gammacell 220 irradiator. The drawn line is a spline fit through the lifetime data points.

related to the fluorescence lifetime by Eq. (6):

$$\phi_{FL} = k_r \tau \quad (6)$$

Taking k_r to be unaffected by changes in the rigidity of the matrix, the increase in ϕ_{FL} on vitrification will be equal to the ratio of the lifetimes found, i.e. 11.2/9.6 = 1.17. When this is taken into account, the increase in fluorescence on vitrification due to causes other than incorporation of the AnMA probe is reduced from 5.5 to 4.7 which is closer to the factor of 4.3 found for the increase in MMA conversion.

A second factor which could contribute to a change in the measured fluorescence intensity is the increase in density of the matrix which accompanies vitrification. This will result in an increase in the concentration of the incorporated probe molecules and an increase in the refractive index of the medium. These effects will at least partially compensate each other as shown by Eq. (7) for the ratio of the measured intensity for a fluorophore with a given fluorescence quantum yield in a solid of refractive index n_S to that in a liquid of refractive index n_L.

$$\frac{I_S}{I_L} = \frac{(1 - 10^{-OD_S})n_L^2}{(1 - 10^{-OD_L})n_S^2} \quad (7)$$

On vitrification the density of MMA increases by a factor of 1.25 and the refractive index increases from 1.41 to 1.49. Using Eq. (7), it can be estimated that these changes should cause an increase in the measured fluorescence intensity by 6% for an initial optical density of 0.2 in the liquid phase. When this is taken into account, in addition to the increase expected on the basis of the increased quantum yield of fluorescence, the increase in fluorescence on vitrification due to the increased incorporation alone is found to be a factor of 4.4. This is in good agreement with the increase in MMA conversion observed indicating that the fluorogenic probe used is equally capable of providing a measure of the degree of polymerization even in the high conversion, gel effect regime.

3.4. Monitoring of MMA solution polymerization with AnMA

As mentioned in Section 1, the majority of previously proposed fluorescent probe molecules rely for their application on the sensitivity of one or more of their photophysical properties to changes in the viscosity and/or dielectric properties of the polymerizing medium. This limits their use to bulk or only slightly diluted polymerizing systems. More highly diluted monomer solutions, which undergo relatively small changes in physical properties on complete polymerization of the monomer, cannot be studied using these compounds. The action of fluorogenic probes on the other hand does not rely solely on changes occurring in the physical properties of the medium and therefore should be equally applicable for monitoring solution polymerization, as they have been shown to be for bulk polymerization. We demonstrate this unique capability for the first time in Fig. 7 with results on the radiation-induced polymerization of MMA in MMA/benzene mixtures with MMA volume fractions varying from 80% to as low as 20%. The results bear a strong resemblance to those found many years ago for the dependence on time of the monomer conversion for similar mixtures undergoing thermal polymerization [20].

The results serve to illustrate the strong dependence of the form of the monomer conversion versus dose on dilution. Thus, while evidence can still be seen for the occurrence of the gel effect for the 60% MMA mixture, the dose required to reach this onset is approximately a factor of 3 higher than for the bulk monomer. The results also clearly show the disappearance of the gel effect when the MMA fraction is decreased from 60 to 40%.

4. Conclusions

It was experimentally determined that the AnMA probe has a close to identical reactivity with that of the monomer during the free radical polymerization of MMA. The ratio between the propagation rate constants for the cross-over reaction in which an MMA radical chain-end reacts with AnMA relative to the homopolymerization reaction of MMA is found to be 0.96. By taking into account the change in the fluorescence quantum efficiency observed during the autoacceleration region of polymerization, in addition to the associated density change and change in the refractive index, it was possible to determine the increase in probe conversion during the gel effect of MMA. Very good agreement was found between the experimental MMA conversion and that predicted by monitoring the complete reaction with the fluorogenic probe AnMA. We have therefore demonstrated that the fluorogenic probe AnMA can be applied for sensitive, in situ monitoring of MMA polymerization throughout the entire course of the reaction.

In addition, MMA solution polymerization experiments conducted in situ involving the AnMA probe molecule demonstrate that fluorogenic probes can also be used for

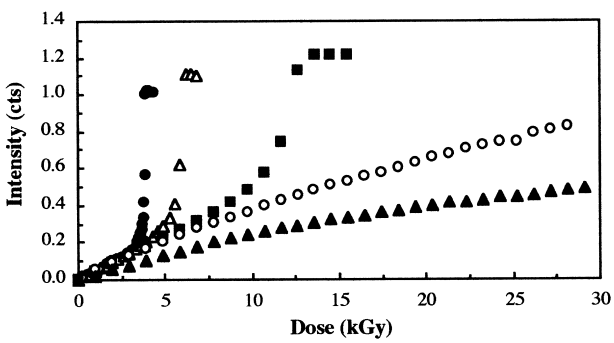

Fig. 7. The dose dependence of the fluorescence intensity from irradiated MMA/benzene mixtures containing AnMA ($OD_{337\ nm} = 0.20$) for MMA volume fractions of 1.0 (●), 0.8 (△), 0.6 (■), 0.4 (○) and 0.2 (▲).

quantitative monitoring of solution polymerizations. This ability to monitor solution polymerization by fluorescent probe techniques with high sensitivity is to the best of our knowledge unprecedented.

References

[1] Loutfy R. Macromolecules 1981;14:270.
[2] Loutfy R. Macromolecules 1983;16:678.
[3] Loutfy RO, Teegarden DM. Macromolecules 1983;16:452.
[4] Loutfy R. Pure Appl Chem 1986;58:1239.
[5] Paczkowski J, Neckers D. Macromolecules 1991;24:3013.
[6] Stroeks A, Shmorhun M, Jamieson AM, Simha R. Polymer 1988;29:467.
[7] Jing D. Polymer 1990;31:110.
[8] Valdes-Aguilera O, Pathak CP, Neckers DC. Macromolecules 1990;23:689.
[9] Rettig W. Angew Chem Int Ed 1986;25:971.
[10] van Ramesdonk H, Vos M, Verhoeven J, Möhlmann G, Tissink N, Meesen A. Polymer 1987;28:951.
[11] Warman JM, Abellon RD, Verhey HJ, Verhoeven JW, Hofstraat JW. J Phys Chem B 1997;101:4913.
[12] Warman J, Abellon R, Luthjens L, Suykerbuyk J, Verhey H, Verhoeven J. J Nucl Instr Meth Phys Res B 1999;151:361.
[13] Frahn MS, Abellon RD, Luthjens LH, Warman JM. Radiat Res Proc 1999;2:86.
[14] Frahn MS, Warman JM, Abellon RD, Luthjens LH. Radiat Phys Chem 2001;60:433.
[15] Frahn MS, Abellon RD, Jager WF, Luthjens LH, Warman JM. Nucl Instr Meth Phys Res B 2001;185:241.
[16] Frahn MS, Abellon RD, Luthjens LH, Vermeulen MJW, Warman JM. Nucl Instr Meth Phys Res B 2003;208:405.
[17] Fricke H, Hart EJ, Radiation dosimetry, vol. 2. New York: Academic Press; 1966.
[18] Kropp JL, Dawson WR, Windsor MW. J Phys Chem 1969;73:1747.
[19] Robinson GW, Frosch RP. J Chem Phys 1962;37:1962.
[20] von Schulz GV, Harborth G. Macromol Chemie 1947;1:106.