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MACROMOLECULAR CHEMISTRY AND POLYMERIC MATERIALS

Copolymers of 2-Deoxy-2-methylacrylamido-*D*-glucose with Tertiary and Quaternary Amino Groups

O. V. Nazarova, Yu. I. Zolotova, A. V. Dobrodumov, I. I. Malakhova, E. N. Vlasova, and E. F. Panarin

Institute of Macromolecular Compounds, Russian Academy of Sciences, St. Petersburg, Russia

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Abstract—Method for synthesis of graft-copolymers of a vinyl saccharide, 2-deoxy-2-methylacrylamido-*D*-glucose, and *N,N*-dimethylaminoethyl methacrylate was developed. Statistical copolymers with a controllable hydrophobic-hydrophilic balance were synthesized by alkylation of a statistical copolymer with various alkyl iodides.

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In recent decades, water-soluble diphilic polymers have been widely used for modification of low-molecular-weight biologically active substances (BAS) to control their solubility and pharmacokinetics and to prolong their action. Of particular interest among these polymers are graft- and block-copolymers, which can effectively improve BAS properties in a number of cases [1, 2]. The characteristics of polymer derivatives of BASs are strongly affected by the hydrophilic-hydrophobic properties of a polymer [1, 3–5].

The aim of our study was to develop methods for synthesis of graft-copolymers based on a vinyl saccharide, deoxy-2-methylacrylamido-*D*-glucose (MAG), and *N*,*N*-dimethylaminoethyl methacrylate (DMAEM), previously undescribed in the literature, and new statistical copolymers with variable hydrophobicity.

EXPERIMENTAL

We used azoisobutyriodinitrile (AIBN), cysteamine hydrochloride, DMAEM, and dimethyl-formamide (DMF) (Aldrich, Germany). DMAEM was distilled in a vacuum, DMF was purified by the procedure described in [6]. MAG was synthesized as described in [7]. Acrylic acid *N*-hydroxyphthalimide ester was obtained using the procedure reported in [8].

To synthesize poly-MAG with terminal amino groups (PMAG–NH_2·HCl), calculated amounts of MAG, initiator (AIBN), cystein hydrochloride, and solvent (DMF) were placed in an ampule purged with argon, and the ampule was sealed and kept in a thermostat at 60°C for 24 h. The resulting polymer was precipitated into diethyl ether, separated on a glass frit filter, and dried in a vacuum to constant weight. To remove low-molecular-weight impurities, the polymer was dissolved in water and subjected to dialysis against water. We used for this purpose Spectra/Por® dialysis bags (MWCO 1000; Spectra, USA), which can remove substances with molecular masses $M \leq 1000$.

To synthesize PMAG with terminal double bonds, calculated amounts of PMAG–NH_2·HCl and acrylic acid *N*-hydroxyphthalimide ester in a solution on DMF were mixed at room temperature in the presence of triethylamine (TEA) with a magnetic stirrer for 24 h. The resulting polymer was precipitated into diethyl ether, separated on a glass frit filter, and dried in a vacuum to constant weight.

To synthesize MAG–DMAEM graft-copolymers, calculated amounts of PMAG with terminal double bonds, initiator (AIBN), and solvent (DMF) in an ampule purged with argon and sealed were kept in a thermostat at 60°C for 24 h. The resulting polymer was precipitated with a mixture of diethyl and petroleum ethers, separated on a glass frit filter, and dried in a vacuum to a constant weight.

The statistical MAG–DMAEM copolymer was produced by radical copolymerization of the monomers in a solution in DMF at 60°C (10 wt %) in the presence of AIBN (2 wt % relative to the sum of the monomers. The initial MAG : DMAEM ratio was 50 : 50 mol %. The copolymer was isolated by precipitation into diethyl ether; the precipitate was filtered off and dried in a vacuum. The yield of the copolymer was 93%.

The MAG–DMAEM copolymer was alkylated with alkyl halides in a solution in DMF at 70 or 100°C for 4 h [9]. The resulting polymer was precipitated into diethyl ether, separated on a glass frit filter, and dried in a vacuum.

The content of terminal amino groups was determined spectrophotometrically by the known procedure [10] by measuring the absorption of a complex of the polymer with 2,4,6-trinitrobenzene sulfonic acid (λ_{max} = 420 nm), formed by primary amines or by their salts. The content of DMAEM units in the copolymers was found by potentiometric titration of DMAEM units with a 0.1 N HCl solution. The content of DMAEM-Alkl alkylated units was determined by potentiometric titration of I^- ions with a 0.01 N $AgNO_3$ solution. The characteristic viscosity of the polymers was measured at 25°C in a 0.1 M Na_2SO_4 solution. The molecular mass of PMAG was estimated using the equation $[\eta] = 8.29\times10^{-4} M^{0.49}$ [11].

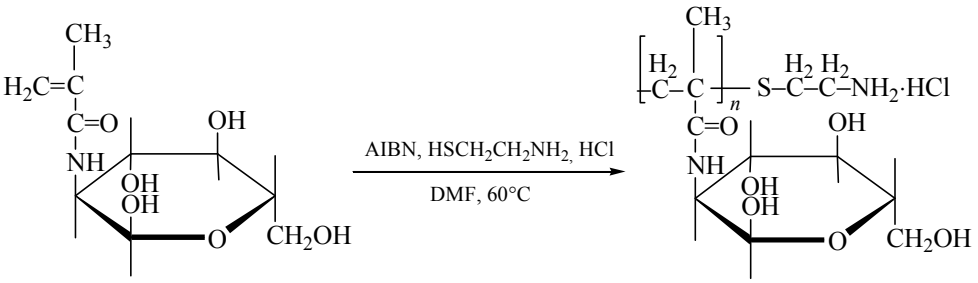
PMR spectra were recorded with a Bruker Avance 400 in D_2O, and IR spectra, with a Vertex 7 (Bruker) instrument equipped with a Pike reflection attachment.

A method to synthesize graft-copolymers is a copolymerization of one of the monomers with a macromonomer, which is a homopolymer of the other monomer containing a terminal double bond. A convenient way to introduce prescribed terminal groups into polymers is radical polymerization in the presence of chain-transfer agents and, in particular, mercaptans containing a functional group [12, 13]. We synthesized PMAG with a single terminal groups from cysteamine hydrochloride $HS–CH_2–CH_2–NH_2·HCl$ (Scheme 1).

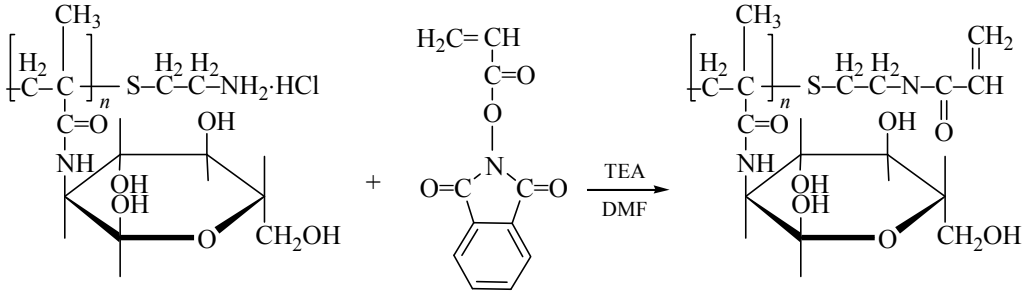
The synthesis conditions and characteristics of the polymers obtained are listed in Table 1. In run no. 1, we used large values of the [AIBN] : [MAG] and [mercaptan] : {MAG] ratios, which made it possible to obtain a polymer with low characteristic viscosity and a molecular mass (*MM*) of 1500. Raising the concentration of the monomer in the starting mixture and making smaller the [AIBN] : [MAG] and [mercaptan] : [MAG] ratios (run no. 2) resulted in that a polymer with a larger *MM* of 8500 was obtained.

The PMAG macromonomer containing a single double bond was synthesized via interaction of macromolecules containing a terminal amino group with activated acrylic acid *N*-hydroxyphthalimide ester (Scheme 2).

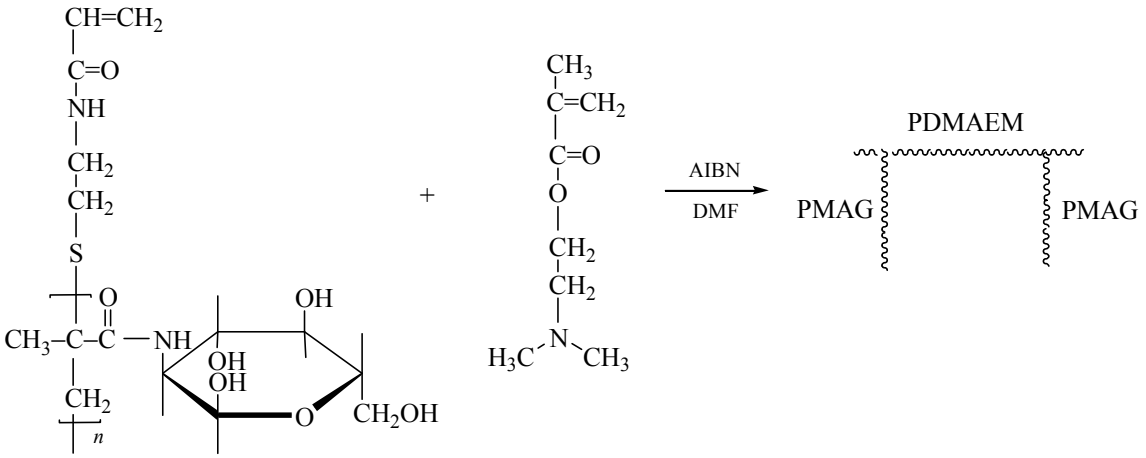
Scheme 1.



Scheme 2.



Scheme 3.



Graft-copolymers with the PDMAEM backbone and grafted PMAG chains were synthesized by radical copolymerization of DMAEM with a synthesized macromonomer in the presence of AIBN initiator (Scheme 3).

Characteristics of the graft-copolymers are listed in Table 2. It is known that the DMAEM homopolymer is soluble in alcohols, whereas the MAG homopolymer is insoluble. The difference in the solubility of the homopolymers enabled separation of the polymers obtained into fractions enriched in MAG (part insoluble in methanol) and DMAEM (part soluble in methanol).

At the initial [macromonomer (run no. 1) : [DMAEM] ratio of 50 : 50 mol % (run no. 3), the methanol-

Table 1. Synthesis conditions and properties of PMAG with terminal amino groups

Run no.	Synthesis conditions			PMAG properties			
	[MAG], M	[AIBN], mol % relative to the monomer	[SII], mol % relative to the monomer	yield, wt %	[η], dl g^{-1}	M_η	[terminal NH2 groups], wt %
1	0.39	5.0	15.3	73	0.03	1500	3.1
2	0.61	2.5	7.7	67	0.07	8500	2.1

Table 2. Synthesis of MAG–DMAEM graft-copolymers

Run no.	Starting PMAG[b]	Initial $[M_1]:[M_2]$ ratio[a]		Yield, wt %	Copolymer parameters					
					part insoluble in CH_3OH			part soluble in CH_3OH		
		wt %	mol %		fraction, wt %	$[M_1]:[M_2]$, mol %	[η], dl g^{-1}	fraction, wt %	$[M_1]:[M_2]$, mol %	[η], dl g^{-1}
3	1	50:50	39:61	77	56	75.2:24.8	0.08	44	33.0:67.0	0.06
4	1	86:14	80:20	56	95	97.5:2.5	0.04	5	[c]	[c]
5	1	28:72	20:80	68	0	–	–	100	33.8:66.2	0.09
6	2	50:50	39:61	69	45	73.8:26.2	0.12	55	40.2:59.8	0,1
7	2	60:40	49:51	69	57	72.5:27.5	0.10	43	30.5:69.5	0.08

[a] M_1, macromonomer (PMAG); M_2, DMAEM. [b] Starting PMAG obtained in run no. 1 (2) (Table 1). [c] Not determined.

Table 3. Influence exerted by the nature of the alkylating agent and by the reagent ratio on properties of MAG–DMAEM–DMAEM Alkl copolymers

Run no.	Alkylation		Parameters of alkylated copolymer			
	Alk	[DMAEM] : [Alkl], mol %	[DMAEM·Alkl], mol %	degree of alkylation, %	solubility in water	[η], dl g^{-1}
8	C_2H_5	1 : 1 .1	28 .3	64 .3	Soluble	Not determined
9	C_2H_5	1 : 1 .3	29 .8	67 .7	Soluble	Not determined
10	C_2H_5	1 : 1 .5	30 .3	68 .9	Soluble	0 .10
11	C_8H_{17}	1 : 1 .5	38 .5	87 .5	Soluble	0 .03
12	$C_{12}H_{25}$	1 : 1 .5	33 .2	75 .5	Insoluble	Not determined
13	$C_{12}H_{25}$	1 : 0 .75	22.3	50 .7	Soluble	0 .02

insoluble and -soluble fractions constitute 56 and 44% of the polymer mass, respectively. The PMR spectra of both fractions contain all the signals characteristic of PMAG and PDMAEM. The compositions of the fractions obtained were determined by potentiometric titration of DMAEM units with 0.1 N HCl solution (Table 2) and from PMR spectra. The results furnished by these two techniques are close. It was found that the methanol-insoluble part is strongly enriched in MAG units, compare with the starting mixture, and the soluble part is somewhat enriched in DMAEM units (Table 2). However, the insoluble part contains a substantial amount of DMAEM (~25 mol %), and the soluble part includes 33 mol % MAG. These results indicate that the target graft-polymer is formed. The characteristic viscosities of both fractions exceed [η] of the starting macromonomer (Table 2).

We studied the fractions obtained (Table 2, run no. 3) by the TLC method with PTSKh-S-A plates (silica gel on a glass substrate). We found the conditions in which PMAG and DMAEM can be separated. For example, with DMF used as eluent, R_f = 0 for PDMAEM and R_f = 0.99 for PMAG. Under these conditions, R_f = 0 for the metanol-soluble and -insoluble fractions, with a weak PMAG-related spot observed in the chromatogram of the insoluble part.

By contrast, with another eluent, a mixture of isopropanol, water, and diethylamine (7.0 : 1.2 : 0.5), PMAG remains at the start and R_f = 0.52 for PDMAEM. The chromatogram of the methanol-insoluble part (Table 2, run no. 3) shows only a single spot at the start. The chromatogram of the soluble fraction contains two spots: one at the start and the other, weaker spot with $R_f \approx 0.52$.

The TLC data confirm that the target graft-copolymer is formed and contains PMAG and PDMAEM homopolymers, but their content is lower than that of the graft-copolymer.

Run no. 4 (Table 2) was performed with a [macromonomer] : [DMAEM] ratio of 86 : 14 wt %. It was found that almost the whole amount of the polymer (95%) is insoluble in methanol, with the insoluble part containing an insignificant amount of DMAEM units (2.5 mol %) and the characteristic viscosity of the fraction nearly coinciding with [η] of the starting PMAG (Table 2). Presumably, the graft-copolymer is almost not formed at all under the conditions used.

At a [macromonomer] : [DMAEM] ratio of 28 : 72 wt % (Table 2, run no. 5), the whole amount of the polymer obtained is soluble in methanol, with no insoluble fraction present. The soluble polymer contains about 34 mol % MAG units, which indicates that the target copolymer is formed.

An important parameter of block- and graft-copolymers is the ratio between the block lengths of each monomer. We synthesized graft-copolymers with a PMAG monomer with MM 8500 (Table 1, run no. 2). In this case, the component ratios were 50 : 50 and 60 : 40 wt %. It was found that the methanol-insoluble fractions of both copolymers contain DMAEM units, and soluble fractions, MAG units, in considerable amounts. Thus, the target MAG–

DMAEM graft-copolymers were also synthesized with the PMAG monomer (Table 1, run no. 2).

To synthesize polymers with controlled hydrophilic-hydrophobic balance, we produced by radical copolymerization of MAG and DMAEM a statistical copolymer containing 44.0 mol % DMAEM units, with a characteristic viscosity [η] = 0.09 dl g^{-1}. By alkylating this copolymer with ethyl-, octyl-, or dodecyl iodide, we synthesized ternary copolymers containing, together with MAG and DMAEM units, units of DMAEM–Alkl quaternary ammonium bases (Scheme 4).

The alkylation conditions and parameters of the polymers are listed in Table 3. Compared with the IR spectra of the starting copolymers, the spectra of the alkylation products show an increase in the absorption intensity in the range of stretching vibrations of CH_2 groups at 2700–2900 cm^{-1}, which points to the presence of alkylated units. At the same time, no changes of this kind are observed in spectra of the alkylated MAG homopolymer, which confirmed that the reaction occurs just at the nitrogen atom. Data furnished by potentiometric titration of iodine ions demonstrated (Table 3) that varying the [DMAEM unit] : [C_2H_5I] molar ratio from 1 : 1.1 to 1 : 1.5 in alkylation of the copolymer with ethyl iodide only slightly affects the maximum achievable degree of alkylation (Table 3, run nos. 8–10). The copolymers contained about 30 mol % DMAEM C_2H_5I units, with the degree of alkylation equal to 63–69%. All the copolymers are readily soluble in water.

At a [DMAEM unit] : [C_8H_{17}] ratio of 1 : 1.5, the degree of alkylation with octyl iodide is 87%, and the content of alkylated units, 38 mol % (Table 3, run no. 11). The alkylated copolymer is also soluble in water.

On passing to dodecyl iodide at the same [DMAEM unit] : [$C_{12}H_{25}I$] molar ratio of 1 : 1.5, the degree of alkylation and the content of alkylated units was lower than that in the case of octyl iodide (Table 1, run no. 1). However, the copolymer obtained was insoluble in water, which is presumably due to the higher hydrophobicity of the $C_{12}H_{25}$ group, compared with C_8H_{17}. Lowering the [DMAEM unit] : [$C_{12}H_{25}I$] molar ratio to 1 : 0.75 (Table 3, run no. 13) made it possible to obtain a water-soluble copolymer with lower content of DMAEM $C_{12}H_{25}I$ units.

Scheme 4.

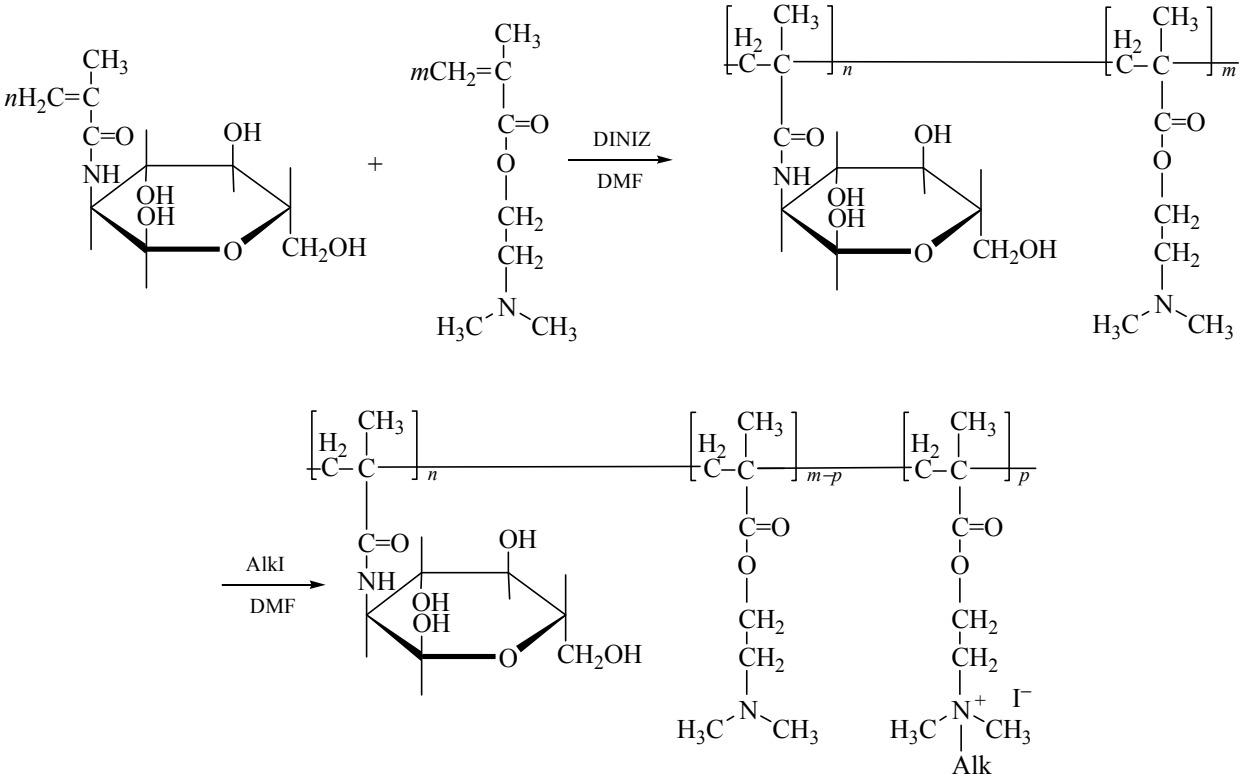


Table 3 lists values of the characteristic viscosities of the polymers. It can be seen that, for the polymer alkylated with C_2H_5I (Table 3, run no. 10), [η] = 0.10 dl g^{-1}, which almost coincides with the value [η] = 0.09 dl g^{-1} for the starting nonalkylated MAG–DMAEM copolymer. For the copolymers containing DMAEM $C_8H_{17}I$ and DMAEM–$C_{12}H_{25}I$ units, smaller values were found, [η] = 0.03 and 0.02 dl g^{-1}, which is indicative of compactification. The results obtained are consistent with published data, because it is known [4, 5, 14] that intrachain hydrophobic interactions become stronger as the length of the alkyl radical in the side chain grows.

CONCLUSIONS

New graft- and ternary statistical copolymers with varied hydrophobicity, based on a vinyl saccharide, *N*-methacryloylamino glucose, and *N*,*N*-dimethylamino-ethyl methacrylate were synthesized and characterized.

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