Neutron Scattering Studies of the Structure of a Polyelectrolyte Globule in a Water−Acetone Mixture

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ABSTRACT: The polycationic polyelectrolyte poly(methacryloylethyltrimethylammonium methyl sulfate) (PMETMMS) of high molecular weight (ca. 26×10^6 g mol^{-1}) in dilute solutions with acetone−water mixtures displays a coil−globule transition above a certain threshold of acetone. SANS observations of this system indicate that the collapsed state of this molecule consists of a sequence of dense spheres with smooth surfaces connected by regions of loose polymer. The radius of the spheres, 28 nm, is smaller than either the hydrodynamic radius (R_H = 39 nm) or the radius of gyration (R_G = 42 nm). The number of such collapsed regions is small (3−4), the volume fraction of polymer inside them being about 8%.

Introduction

Under suitable circumstances, when the solvent quality is modified, polyelectrolyte molecules undergo a volume transition from an extended coil state in water to a globular state.[1] The conformation of a polyelectrolyte in its collapsed state may differ significantly from the structure of the globules of uncharged polymers. Thus, rigid macromolecules, such as double-stranded DNA chains in aqueous solutions of poly(ethylene oxide), form a toroidal doughnut-like globule.[2] Kantor and Kardar suggested that a weakly charged polymer may adopt a necklace-like conformation in which compact beads are joined by flexible chain segments.[3] This idea has been developed theoretically by Dobrynin et al.[4] However, the necklace-like model is still conceptual and, therefore, controversial.[5]

The theoretically predicted model of the necklace-like polyelectrolyte globule[3,4] matches well the "molten globular" state known for uncharged polymers.[6,7] For polymers with high molecular weight the coil−globule transition may proceed through a number of such molten globular states. The concept of the molten globule may also be relevant to the kinetics of the collapse of flexible polyelectrolytes.

Dynamic and static light scattering measurements have shown that poly(methacryloylethyltrimethylammonium methyl sulfate) (PMETMMS) with molecular weight in the range $M_w = (10-28) \times 10^6$ g/mol displays a coil-to-globule transition in water−acetone mixtures when the acetone mass fraction in the solvent composition (γ) increases beyond 0.80.[8−11] The conductivity of the solutions decreases drastically in the transition region. However, the remaining charges of the polycation, as well as the hydrophobic parts of the polymer soluble in acetone, prevent aggregation of PMETMMS globules in a dilute solution. For this linear polyelectrolyte, previous investigations showed that the hydrodynamic radius R_H in the collapsed state is significantly smaller than the radius of gyration R_G. This result, which is characteristic of an open structure, is surprising for this state where the second virial coefficient is negative. Studies on dynamic birefringence in flow have shown that in the collapsed state the asymmetry parameter equals unity.[12] This is direct experimental evidence that the globule is a symmetrical particle displaying no macroform effect; the PMETMMS globule is thus both compact and completely symmetrical. However, the structure of the PMETMMS globule is uncertain. A hollow sphere (polymer micelle), a toroidal doughnut-like structure similar to DNA,[2] or a spherical structure consisting of a sequence of separate collapsed regions, like a string of pearls,[4] may also describe the compact conformation of PMETMMS globule.

To obtain further information on this system beyond that afforded by visible light scattering, higher spatial resolution is required. To this end, we report small-angle neutron scattering (SANS) measurements, in addition to dynamic and static light scattering observations.

Experimental Section

Materials and Methods. The sample of poly(methacryloylethyltrimethylammonium methyl sulfate) (PMETMMS) was prepared by free-radical polymerization using a twice precipitated monomer.[13] The monomer was prepared by quaternizing a commercial monomer, (dimethylamino)ethyl methacrylate (Yarcintez, Yaroslavl, Russia) with dimethyl sulfate. The polydispersity index of the resulting sample is estimated to be $M_w/M_n = 1.6-1.8$.[8] The structure of the polymer repeat unit is shown in Figure 1.

Samples of PMETMMS were examined by light scattering both in 1 M aqueous sodium nitrate solutions and in the collapsed state in water−acetone mixtures.[8−11] The molar masses were found to be identical in both solvents. Sedimentation−diffusion analysis gives similar results, indicating a narrow molar mass distribution of the sample.[10] Molar mass measurements were repeated several times during the period

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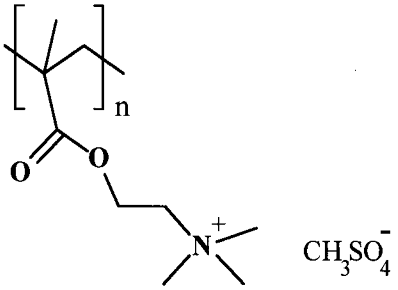

Figure 1. Chemical structure of the repeat unit of PMETMMS.

of study to check the stability of the polymer. The concentration dependence of PMETMMS size distribution in water–acetone mixtures has been studied in detail; see experimental results of refs 9 and 11.

A weighed amount of polymer was first dissolved in deuterium oxide (SDS, France, >99.9% deuteration), and when this solution was homogeneous, aliquots of deuterated acetone (SDS, >99.8% deuteration) were successively added to obtain the final acetone mass fraction $\gamma = 0.82$. In this solvent system, to a good approximation (ca. 1%), a given value of γ corresponds to the same acetone volume fraction, independent of whether acetone and water are either both deuterated or both protonated.

The SANS measurements were made on the D22 instrument at the Institut Laue Langevin, Grenoble. Two sample detector distances were used, 6 and 15 m, with the wavelength selector working at 8 and 14 Å, respectively, and a wavelength spread of 10%. Counting times were between 1 and 2 h. The solutions were contained in quartz scattering cells with a 2 mm path length. Standard corrections were made for electronic noise and background scattering from the cells. The incoherent background was removed by subtracting the signal from the pure deuterated water–acetone mixture.[14] Signals were normalized using a standard water sample, the cross section of which is known.[15]

Quasi-elastic and static light scattering measurements were also made on the system, using a Spectra Physics SP1161 laser working at 488 nm in conjunction with a Malvern 7035 correlator. The slow mode in the relaxation time distribution, observed for the concentrated solution in this study, is due to the presence of a small fraction of intermolecular aggregates. These aggregates disappear at lower concentrations.

Results

PMETMMS is known to adopt a globular conformation in the water–acetone mixtures with the acetone mass fraction (γ) above 0.80.[8–11] The value $\gamma = 0.82$ was therefore chosen for this study of the internal structure of PMETMMS in its collapsed state.

Two sets of SANS measurements were made. The first observations were performed on a deuterated solution containing 0.000 50 g cm^{-3} polymer 2 months after preparation of the sample. A second sample containing 0.0010 g cm^{-3} polymer was measured 24 h after preparation. The SANS response from the latter, shown in Figure 2, displays a characteristic Guinier plateau at small wave vectors from which the radius of gyration $R_G = 42$ nm can be calculated (Figure 3). Static light scattering measurements on the same sample yielded $R_G = 43$ nm, in agreement with the SANS result. At angles below 60°, however, the light measurements display excess scattering, betraying the presence of larger molecular associations. Such associations were not observed in the more dilute sample, which had, however, been allowed to come to equilibrium for a longer time.

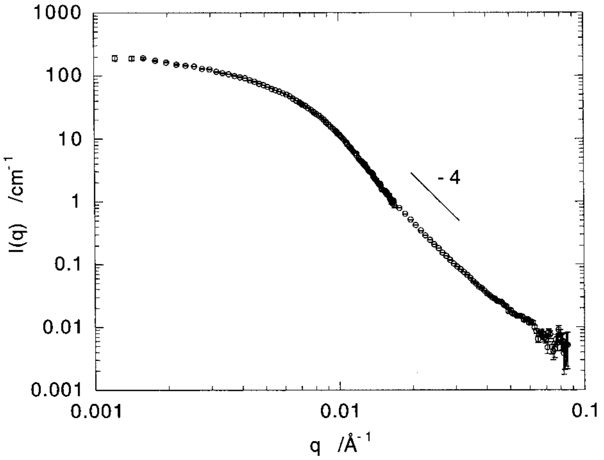

Figure 2. SANS curve from 0.0010 g cm^{-3} solution of PMETMMS in deuterated acetone–water mixture at $\gamma = 0.82$. Comparison with the straight line of slope −4 shows the region where surface scattering prevails.

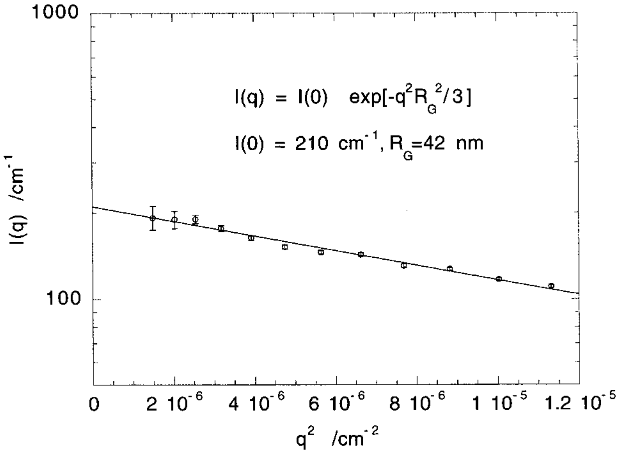

Figure 3. Guinier representation of the low q region in Figure 1. The slope of the least-squares straight line through the data points yields $R_G = 42$ nm.

In the dynamic light scattering measurements of the more concentrated solution, the hydrodynamic radius, R_H, displays a distinct bimodal distribution when analyzed using CONTIN.[16] This behavior is consistent with the molecular associations mentioned above that cause the extra scattering observed at small angles. The smaller of the two radii, i.e., that of the single macromolecules, was found to be $R_H = 39 \pm 6$ nm. The somewhat large uncertainty in this result is a consequence of the bimodal decomposition. Previous measurements of R_H in this system[9,10] yielded 35 ± 2 nm, a value that is consistent with the present observation. The present and the earlier investigations thus converge toward the conclusion that $R_G/R_H > 1$. To place this result in perspective, for a uniform solid sphere, R_G/R_H is equal to $(3/5)^{1/2}$, while for an infinitely thin impenetrable spherical shell, this ratio is equal to 1. Thus, although the virial coefficient A_2 is negative and the coil is collapsed, the present results reveal that the hydrodynamic flow of solvent molecules penetrates inside the volume occupied by the radius of gyration. The collapsed coil state is therefore an open structure.

From the calculated neutron contrast factor for this PMETMMS-deuterated acetone–water system, $\Delta\rho^2 = 2.60 \times 10^{21}$ cm^{-4}, and the density $d = 1.277$ g cm^{-3} of the dry polymer, the Guinier plots of the SANS measurements yield the apparent molecular weights

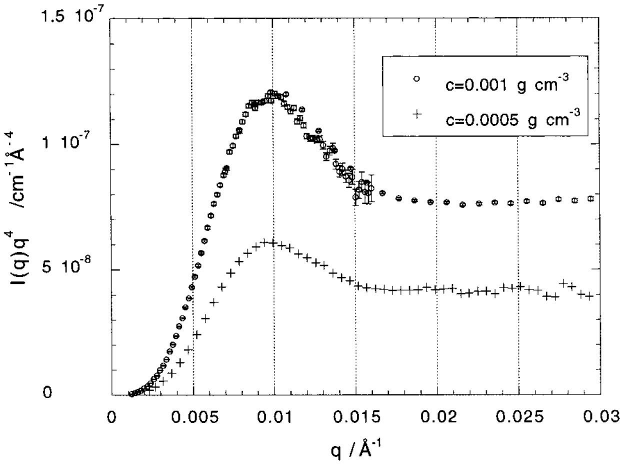

Figure 4. Porod plot of SANS data in the surface scattering region, from PMETMMS solutions at $c = 0.000\,50$ g cm^{-3} and 0.0010 g cm^{-3}.

30×10^6 and 38×10^6 g/mol for the two concentrations $c = 0.000\,50$ and $c = 0.0010$ g cm^{-3}, respectively. These values are consistent with the extrapolated estimate $M_w = 26 \times 10^6$ g/mol and the negative value of the second virial coefficient A_2 reported previously for this system.[11]

Figure 4 shows the SANS data plotted in a Porod representation, $I(q)q^4$ vs q. Both samples exhibit a distinct peak at $q_{max} \approx 0.0097$ Å^{-1}, followed by a plateau at higher values of the wave vector q. Such patterns are characteristic of scattering from mildly polydisperse compact structures (e.g., a solution of spheres) displaying a surface that is smooth at the resolution scale of the observation.[17] For uniform spheres of radius R, the position of the maximum in this representation would be located at $q_{max}R = 2.744$, thus yielding for the present case $R = 28.3$ nm. Since, however, other regular structures can give rise to an analogous maximum, this feature does not constitute a proof that spheres are present here.

Furthermore, from the plateau region in this figure where the quantity $I(q)q^4$ is constant, the surface area Σ of the condensed internal structure of the molecule may be evaluated using the relation[17]

$$\frac{\Sigma}{V} = \frac{I(q)q^4}{2\pi\Delta\rho^2\varphi} \tag{1}$$

where V is the volume occupied by the polymer. Equation 1 is usually expressed without the factor φ ($=c/d$), representing the polymer volume fraction. The volume V would then be that of the overall sample, rather than that of the polymer alone, as here. The heights of the two plateaux in Figure 4 yield

$$\Sigma/V = 6.5 \times 10^{-3}\ \text{Å}^{-1} \quad (c = 0.000\,50\ \text{g cm}^{-3}) \tag{2}$$

and

$$\Sigma/V = 6.0 \times 10^{-3}\ \text{Å}^{-1} \quad (c = 0.0010\ \text{g cm}^{-3}) \tag{3}$$

Clearly, since these internal surfaces form a smooth barrier, solvent molecules do not pass freely through them.

Discussion

The simplest physically reasonable forms that can be envisaged to describe the internal structure of the collapsed molecule are the following: 1, solid spheres; 2, ellipsoids of revolution; 3, rods or disks; 4, toroids; and 5, strings of pearls. Each of these possibilities is discussed in turn below.

For a uniform solid sphere, the external radius R found from the surface-to-volume ratio is

$$R = 3V/\Sigma \tag{4}$$

which, from eq 3, would yield $R \approx 50$ nm. Such a large structure cannot explain the maximum observed in Figure 4, corresponding to $R = 28.3$ nm. Furthermore, the resulting sphere does not possess the requisite open structure. A model consisting of a simple sphere is untenable and must be excluded.

Conformations 2–4 all display cylindrical symmetry. If any of these anisotropic configurations were present, it would give rise to depolarized light scattering. None is detected. In addition, the samples display no measurable flow birefringence.[12] Cylindrical configurations must therefore also be excluded from consideration.

The fifth configuration, that of the string of pearls, does have the required symmetry. In this case, the collapsed PMETMMS molecule would consist of a sequence of freely jointed dense spheres with smooth surfaces. However, the radii of the component spheres, calculated from eqs 2–4, are 46 and 50 nm; i.e., their apparent surface-to-volume ratio is only about two-thirds of that suggested by the value of the intensity maximum in Figure 4. This discrepancy can, however, be understood if the polymer connecting the different beads is not in a collapsed state:[4] the calculation of the specific surface area in eq 1 assumes that *all* of the polymer is condensed. For consistency with the value of R obtained from the maximum in Iq,^{4} we are thus led to conclude that only two-thirds of the polymer material is in the form of condensed beads, the other third being in a loosely expanded state between beads. The effective value of φ in eq 1 is thus lower than that measured. It is also probable that the beads may occasionally interpenetrate to a certain extent; this feature would explain the absence of correlation peak in the SANS spectrum of Figure 2.

Although the following model is oversimplified owing to the presence of uncondensed polymer, it serves to illustrate the situation. We assume that the molecule is a freely jointed chain composed of n structural elements, each of which is a dense sphere of diameter $2R$. Then, for a random walk,

$$n(2R)^2 = 6R_G{}^2 \tag{5}$$

where the factor 6 relates the radius of gyration to the end-to-end length.[18] Equation 6 yields a value for n between 3 and 4. The hydrodynamic radius of such a small assembly, although difficult to calculate analytically, is nonetheless expected to be intermediate between R and R_G, as is observed experimentally. Moreover, on the basis of the results of the specific surface area, it can be assumed that two-thirds of the total mass of the molecule M_w is concentrated into three roughly equal spheres of radius $R = 28.3$ nm. It follows that the polymer volume fraction inside each sphere, $^2/_3M_w/4\pi R^3d$ (where, as before, $d = 1.277$ g cm^{-3} is the density of the pure polymer), is close to 8%. In this concentration range three-body repulsive interactions tend to become significant, and it is therefore reasonable to suppose that these forces oppose further collapse.

Our numerous observations show that the stability of the solutions of the collapsed PMETMMS depends on the molecular weight of the polymer. The formation of supermolecular structures (aggregates) was traced using light scattering as well as flow birefringence. In solutions of samples with $M_w = (20-30) \times 10^6$ having polymer concentration of $c < 10^{-5}$ g cm^{-3}, no intermolecular aggregation or precipitation was observed over many weeks. (The observed aggregation in the present sample is probably related both to the higher concentration and to the short interval between sample preparation and measurement compared with the time required to disentangle such large molecules.) For samples with $M_w = (10-15) \times 10^6$ in the same range of polymer concentration, however, aggregation starts within 2−5 days, while in PMETMMS solutions with $M_w = (2-5) \times 10^6$ all molecules were found to be aggregated after the transition. If the PMETMMS globule investigated here consists of three spheres, then the mass of each sphere will be roughly one-third of the total mass of the molecule, i.e., $(7-10) \times 10^6$. This result may suggest that spheres of this molecular weight have the surface-to-volume and/or mass ratio that makes a globule or an ensemble of such globules stable in solution. Samples with M_w less than $(7-10) \times 10^6$ aggregate because of lack of the polymer material to form a stable globule.

Finally, in Figure 2 it can be observed that at the highest values of q the slope of the curve drops less steeply than indicated by the Porod relation. This change in behavior is expected, as it reflects the scattering signal both from the internal structure of the beads and from the loose polymer outside.

Conclusions

SANS observations of the cationic polyelectrolyte PMETMMS of high molecular weight (ca. 26×10^6 g mol^{-1}) in dilute solutions with acetone−water mixtures indicate that the collapsed state of this molecule consists of a short sequence of dense spheres with smooth surfaces, connected by regions of loose polymer. The radius of the spheres, about 28 nm, is smaller than either the hydrodynamic radius ($R_H = 39$ nm) or the radius of gyration ($R_G = 42$ nm). The number of such collapsed regions in the molecule is small (ca. 3), the volume fraction of polymer inside them being close to 8%.

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