

NOTES

Conformation of Amylose and Excluded-Volume Effects on Its Chain Dimensions

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Considerable attention has been paid to dilute-solution behavior of amylose, but our understanding of it still leaves much to be desired. Aqueous solutions of this polysaccharide are notoriously unstable over a broad range in molecular weight M from 8×10^3 to 1.6×10^5 .^{1,2} Because of this unfavorable property, much experimental work³⁻⁵ so far done could only show that the global conformation of amylose in aqueous solution at room temperature is a random coil having a characteristic ratio C_∞ of about 5 at infinite M . On the other hand, dimethyl sulfoxide (DMSO) dissolves the polymer of any molecular weight. However, the chain conformation in this organic solvent has long been controversial as to whether it is a random coil or a semiflexible helix.⁶ Conformational energy calculations^{2,7-9} predict that amylose should be flexible but locally helical.

Very recently, Nakanishi *et al.*¹⁰ demonstrated from light scattering, sedimentation equilibrium, and viscosity measurements on narrow-distribution samples of synthetic amylose at 25°C that the chain conformation in DMSO is a random coil expanded by excluded-volume effect if M is higher than 10^5 . They, analyzing intrinsic viscosity ($[\eta]$) data for

M between 342 (the dimer) and 10^4 in terms of the unperturbed helical wormlike (HW) chain,^{11,12} derived the following conclusions from the estimated HW model parameters: (1) The value of C_∞ in DMSO (at 25°C) is about 5, so that without excluded-volume effect, the global conformations in DMSO and aqueous solvents are similar, and (2) though flexible, the amylose chain in DMSO has some helical nature locally. This molecular picture, *i.e.*, an irregular helical conformation with high flexibility, appears to resemble that predicted from conformational calculations.^{2,8,9} It is thus intriguing and probably significant to make a quantitative comparison between them in terms of some conformation-dependent property. The most relevant for such a comparison may be the mean-square radius of gyration $\langle S^2 \rangle$, for which experimental data¹⁰ in DMSO are available though limited to high M where excluded-volume effects are significant.

In the present work, we computed $\langle S^2 \rangle$ of amylosic chains with or without excluded volume by a Monte Carlo method, and compared the results with the experimental data of Nakanishi *et al.*¹⁰ and the $\langle S^2 \rangle$ values derived from the HW parameters. The last set

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of $\langle S^2 \rangle$ without excluded volume may be taken to reflect the actual viscosity behavior of amylose for M below 10^4 in DMSO. It should also be noted that though the mean-square end-to-end distance $\langle R^2 \rangle$ for amylose chains with or without excluded volume has already been studied by a few groups⁷⁻⁹ on the basis of the rotational isomeric state approximation or Monte Carlo methods, the simulation data for $\langle S^2 \rangle$ presented below are new.

MONTE CARLO CALCULATIONS

Amylose model chains were generated according to the method of Jordan *et al.*⁸ with the conformational energy map for maltose constructed elsewhere.⁹ The bond angle for the glucosidic oxygen atom was taken as 118° and the virtual bond length l as 0.44 nm. In computing $\langle S^2 \rangle$, each glucose unit was replaced by a hard-core sphere of radius a whose center is located at the center of mass of the glucose unit. This radius represents the magnitude of the excluded-volume repulsion between a pair of glucose residues; no attraction was considered for excluded-volume interactions. Thus, the simulated chains with $a=0$ are unperturbed.

Radii of gyration were obtained for $a=0, 0.1, 0.2, 0.3,$ and 0.4 nm over a range of x (the number of glucose residues or virtual bonds) from 2 to 800. The Monte Carlo sample for each x contained 5000 chains, but for self-avoiding chains with $x > 20$, the computation was effected by use of the Wall-Erpenbeck *s-p* method¹³ of chain enrichment, as in the work by Kitamura *et al.*⁹ on $\langle R^2 \rangle$ (see ref 9 for the details of the procedures). The results all refer to 25°C . The value of C_∞ obtained was 5.0.

The above-mentioned replacement of each glucose unit by a sphere underestimates $\langle S^2 \rangle$ by Δ :

$$\Delta = \frac{\sum_i m_i \langle s_i^2 \rangle}{\sum_i m_i}$$

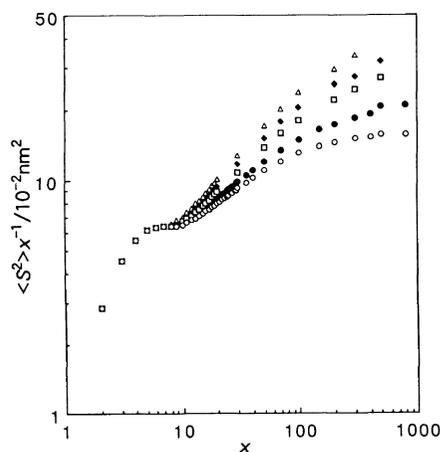


Figure 1. Simulation data of $\langle S^2 \rangle/x$ plotted against x for amylose chains with $a=0$ (\circ), 0.1 nm (\bullet), 0.2 nm (\square), 0.3 nm (\blacklozenge), and 0.4 nm (\triangle).

where m_i denotes the mass of atom i (numbered within the unit) and s_i , the distance between the atom i and the mass center of the unit; the sum of m_i over i equals the molar mass of each glucose residue. The value of Δ should be about or slightly larger than $5 \times 10^{-16} \text{ cm}^2$ (depending on the treatment of the hydrogen atoms of the hydroxyl groups), so that its neglect introduces considerable errors in $\langle S^2 \rangle$ for x smaller than 22, *i.e.*, for $M < 3.6 \times 10^3$. However, no correction for Δ was made in this work, since $\langle S^2 \rangle$ from the HW model parameters, to be compared with Monte Carlo $\langle S^2 \rangle$ for such small x , is concerned only with the chain contour.

The Monte Carlo data of $\langle S^2 \rangle$ obtained are shown in Figure 1. The ratio $\langle S^2 \rangle/x$ for $a=0$ has a hump around $x=7$, reflecting the well-known tendency toward helical loop formation^{2,8,9} in the amylose Monte Carlo chain. As x increases, this ratio rises and almost levels off for $x > 500$. Thus, the unperturbed chain is non-Gaussian at least up to $x=500$. For any nonzero a , $\langle S^2 \rangle/x$ coincides with that for $a=0$ in the region of x below 8 and begins to deviate from it when x exceeds 8–11. Such a critical x for the onset of volume effect is perhaps too small. This may have something

to do with the replacement of each glucose unit with a hard-core sphere. We note, however, that for $a=0.1$ nm, which is relevant to amylose in DMSO (see the analysis below), the volume effect stays rather small up to $x \approx 40$.

DISCUSSION

The mean-square radius of gyration of an HW bead chain with excluded volume¹⁴⁻¹⁶ is determined by five parameters: L (the contour length of the chain), λ^{-1} (the stiffness parameter), κ_0 (the differential-geometrical curvature at the minimum energy of the chain), τ_0 (the differential-geometrical torsion), and B (the dimensionless parameter representing the strength of the excluded-volume interaction between a pair of beads). The first four parameters determine $\langle S^2 \rangle_0$ (the value of $\langle S^2 \rangle$ in the unperturbed state) or more generally any dimensional properties of an unperturbed HW chain.¹¹ The last parameter (measured in units of λ^{-1}) is proportional to the binary cluster integral and related to the excluded-volume parameter z by¹⁴⁻¹⁶

$$z = (3/2\pi)^{3/2} B(\lambda L)^{1/2} \quad (1)$$

If λ^{-1} is equal to the bead spacing, z reduces to the familiar excluded-volume parameter in the conventional two-parameter theory.¹⁷ The contour length is related to M by $L = M/M_L$, with M_L being the molar mass per unit contour length.

Nakanishi *et al.*¹⁰ estimated M_L , λ^{-1} , κ_0/λ , and τ_0/λ for amylose in DMSO at 25°C to be 500 nm⁻¹, 4 nm, 3.5, and 4, respectively, from $[\eta]$ data for $M < 10^4$. This parameter set with $l = 0.425$ nm (instead of 0.44 nm) gave $C_\infty = 4.5$, but these authors corrected it to a value of 5 by referring to the experimental fact¹⁸ that the current polymer hydrodynamic theory^{12,17,19} somewhat overestimates the Flory viscosity factor $[\equiv [\eta]M/(6\langle S^2 \rangle)^{3/2}]$ for long flexible chains in the unperturbed state. To diminish such a theoretical error, we here take λ^{-1} to be 4.5 nm with M_L , κ_0/λ , and τ_0/λ kept at the

above values; note that this λ^{-1} value is hardly beyond the uncertainty of λ^{-1} (4 ± 0.5 nm)¹⁰ originally estimated.

The experimental $\langle S^2 \rangle$ of amylose in DMSO at 25°C¹⁰ varies in proportion to $M^{1.2}$. This exponent of 1.2 is the asymptotic value expected for long flexible chains in good solvents.¹⁹ Thus the remaining parameter B was estimated from the $\langle S^2 \rangle$ data and the HW parameter values ($M_L = 500$ nm⁻¹, $\lambda^{-1} = 4.5$ nm, $\kappa_0/\lambda = 3.5$, and $\tau_0/\lambda = 4$) using the asymptotic relation^{20,21} between the radius expansion factor α_s and z :

$$\alpha_s^2 \equiv \langle S^2 \rangle / \langle S^2 \rangle_0 = 1.53z^{2/5} \quad (z \rightarrow \infty) \quad (2)$$

Here, $\langle S^2 \rangle_0$ of a long HW chain is given by¹¹

$$\langle S^2 \rangle_0 = (4\lambda^2 + \tau_0^2)M / [6\lambda M_L(4\lambda^2 + \kappa_0^2 + \tau_0^2)] \quad (\lambda M/M_L \rightarrow \infty) \quad (3)$$

The B value obtained was 0.28.

With the values for the HW parameters and B together with the known expression for $\langle S^2 \rangle_0$ of the HW chain with finite $\lambda M/M_L$ (see eq 23 of ref 11), $\langle S^2 \rangle$ was calculated as a function of M from the combination¹⁵ of the Yamakawa–Stockmayer–Shimada perturbation theory^{14,16} for the HW bead chain (including the wormlike bead chain) and the Domb–Barrett equation²² for flexible chains, *i.e.*, from

$$\alpha_s^2 = \left[1 + 10\tilde{z} + \left(\frac{70\pi}{9} + \frac{10}{3} \right) \tilde{z}^2 + 8\pi^{3/2} \tilde{z}^3 \right]^{2/15} \times [0.933 + 0.067 \exp(-0.85\tilde{z} - 1.39\tilde{z}^2)] \quad (4)$$

Here, \tilde{z} is the scaled excluded-volume parameter defined by¹⁴⁻¹⁶

$$\tilde{z} = (3/4)Kz \quad (5)$$

with K being a known function of λL .¹⁶ In the coil limit, \tilde{z} becomes identical to z and eq 4 reduces to the original Domb–Barrett equation.²² We note that typical experimental data of α_s^2 for flexible polymers²³⁻²⁵ are accurately described down to low M by eq 4 with eq 5.

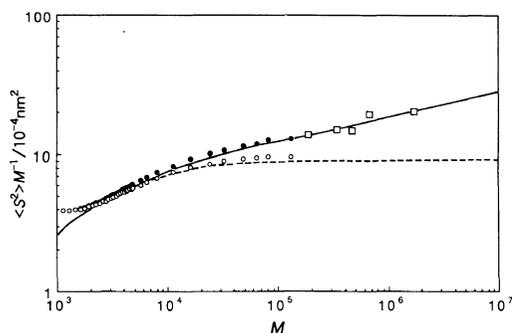


Figure 2. Simulation data of $\langle S^2 \rangle / M^2$ for amylose chains with $a=0.1 \text{ nm}$ (●) and $a=0$ (○) compared with light scattering data¹⁰ (□) for synthetic amylose samples in DMSO and calculated values for HW chains with $B=0.28$ (—) and $B=0$ (---). The HW model parameters determining $\langle S^2 \rangle_0$ are $\lambda^{-1}=4.5 \text{ nm}$, $M_L=500 \text{ nm}^{-1}$, $\kappa_0/\lambda=3.5$, and $\tau_0/\lambda=4.0$.

Figure 2 compares the Monte Carlo $\langle S^2 \rangle / M$ values for $a=0.1 \text{ nm}$ (the filled circles) with the experimental data¹⁰ in DMSO (the squares) and also with the calculated values for the perturbed HW chain with the above parameter set (the solid curve). The unfilled circles and the dashed line in the figure refer to the Monte Carlo $\langle S^2 \rangle_0$ (for $a=0$) and the calculated $\langle S^2 \rangle_0$ for the unperturbed HW chain ($B=0$), respectively. It can be seen that the simulation data for $a=0.1 \text{ nm}$ are connected smoothly with the measured values in DMSO and that both are fitted closely by the solid curve for $M > 2 \times 10^3$, *i.e.*, for $x > 12$. In this M range, the simulation data without excluded volume also come close to the dashed line. The latter is based entirely on the $[\eta]$ data of Nakanishi *et al.* for $M < 10^4$, for which excluded-volume effects on $\langle S^2 \rangle$ are seen to be practically insignificant. Thus, it may be concluded that the present simulation data (for $M > 2 \times 10^3$) are consistent with the measured $[\eta]$ and $\langle S^2 \rangle$ in DMSO. In other words, some irregular helical conformation, which has long been suspected for amylose in solution on the basis of conformation analysis, must be similar to that deduced from dilute-solution data with the aid of the HW model. For M below 2×10^3 , the

simulation data (with or without excluded volume) deviate appreciably from the solid line. Although this deviation ought to be seen by further study, it is attributable, to a large extent, to the length scale adopted by Nakanishi *et al.* in the analysis of the viscosity data, as explained below.

Yamakawa and coworkers²⁶ found that the Monte Carlo data of Jordan *et al.*⁸ for the persistence vector and $\langle R^2 \rangle$ (without excluded volume) are accurately reproduced by HW theoretical curves (for an appropriately chosen set of parameters) over the entire possible range of x (≥ 2) when the HW chain contour is taken along the helical sequence. The chain contour so chosen corresponds to a small length scale of one glucose residue. On the other hand, if the contour is taken along the helix axis, *i.e.*, if a large length scale of a few glucose units (relevant to analysis of data for steady-state transport coefficients)²⁶ is adopted, the HW theoretical curve for $\langle R^2 \rangle$ deviates downward from the Monte Carlo data at x smaller than 10 (see Figure 9 of ref 26). The analysis of $[\eta]$ made by Nakanishi *et al.* indeed refers to such a large length scale of two to four glucose residues.

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