Single molecule electrophoresis of star polymers through nanopores: Simulations

H. H. Katkar^{a)} and M. Muthukumar^{b)}

Department of Polymer Science and Engineering, University of Massachusetts, Amherst, Massachusetts 01003, USA

(Received 17 March 2018; accepted 21 May 2018; published online 10 July 2018)

We study the translocation of charged star polymers through a solid-state nanopore using coarse-grained Langevin dynamics simulations, in the context of using nanopores as high-throughput devices to characterize polymers based on their architecture. The translocation is driven by an externally applied electric field. Our key observation is that translocation kinetics is highly sensitive to the functionality (number of arms) of the star polymer. The mean translocation time is found to vary non-monotonically with polymer functionality, exhibiting a critical value for which translocation is the fastest. The origin of this effect lies in the competition between the higher driving force inside the nanopore and inter-arm electrostatic repulsion in entering the pore, as the functionality is increased. Our simulations also show that the value of the critical functionality can be tuned by varying nanopore dimensions. Moreover, for narrow nanopores, star polymers above a threshold functionality do not translocate at all. These observations suggest the use of nanopores as a high-throughput low-cost analytical tool to characterize and separate star polymers. *Published by AIP Publishing.* <https://doi.org/10.1063/1.5029980>

I. INTRODUCTION

Polymers with specific architectures find many useful applications. Recent developments in polymer chemistry have enabled synthesis of new polymer architectures such as ring polymers, branched polymers, etc. Star polymers are a subset of branched polymers with only one branch point. These polymers are used in the industry as viscosity modifiers, in manufacturing of adhesives and in optically clear resins.¹ Biological star polymers made of DNA also exist, typically as three arm² and as four arm³ DNA. Recently, synthetic star polymers with specific functionalities have also been synthesized.⁴ Being architecturally different from linear polymers, star polymers exhibit rich dynamics unique to their architecture under various experimental conditions. For example, the mechanism of transport of star polymers is different from the reptation mechanism by which linear polymers move in concentrated polymer solutions and in gels.^{5–7}

On the theoretical front, two different mechanisms have been suggested to understand the transport of star polymers in the presence of obstacles such as a concentrated polymer solution. The reptation mechanism by which linear polymers diffuse in such a system has been extended to star polymers.^{8–10} This extended model assumes that for the center-of-mass of the star polymer to move by a unit distance, all but two of its arms have to retract toward its branch point. Such

a move would involve an entropic penalty that reduces the diffusion coefficient dramatically, with a multiplicative factor of $\exp(-\alpha N_b)/N_b$ relative to the linear polymer reptation model, where N_b is the number of monomers in each arm and α is a constant related to the entropic potential.⁸ A similar exponential dependence is predicted for a three arm star polymer based on the entropic penalty of one arm retracting toward the branch point.⁹ With a similar approach, a different choice of the potential related to the arm retraction results in a similar scaling with N_b .¹⁰ An alternative mechanism by which a star polymer can diffuse is by dragging all but two of its arms along the direction of motion of the branch point. In other words, the arm ends are not required to retract to the branch point, but are dragged along with it. This mechanism, in combination with reptation, predicts a scaling of $\exp(-\alpha_2 N_b) \times N_b$ relative to the linear polymer reptation model.^{11,12}

Understanding the behavior of star polymers under confinement has been the focus of various recent studies.^{13–24} This is an interesting problem because the distribution of arms of the star polymer under confinement can lead to rich dynamics, and developing such an understanding is required in devising many applications. For asymmetric star polymers, arm length has been found to have a profound effect on the transport properties of the polymer in gel electrophoresis.¹³ For a three arm star polymer confined in a channel, simulation results reveal that an arm from the doubly occupied side of a channel flips to the other side using an equilibrium barrier crossing mechanism.¹⁴ The corresponding free energy landscape has been calculated for varying radii of the channel, showing features that are specific to star polymers.¹⁵ Langevin dynamics simulations show that a four arm star polymer inside a channel with all arms on

^{a)}Present address: Department of Chemistry, The University of Chicago, Chicago, IL 60637, USA

^{b)}Electronic mail: muthu@polysci.umass.edu

the same side of the branch point transitions to the equilibrium configuration of two arms on each side by flipping one arm at a time by forming a loop near the branch point instead of retracting the arm end.¹⁶ In the presence of a flowing fluid, conformational dynamics of a star polymer become even more important.¹⁷ The critical capture rate of a star polymer into a nanopore is predicted to have a strong dependence on its functionality (number of arms) for certain arm lengths.^{18,19} The star polymer is found to migrate away from the walls of a wide cylindrical pipe due to the flow along the pipe axis, with the extent of migration increasing for stars with higher functionalities.²⁰ Consequently, star polymers with higher functionality flow with a higher average velocity through the cylindrical pipe. These studies are focused on dynamics and equilibrium conformations of star polymers in infinitely long channels. Even in the absence of any confinement, flow of the fluid in the neighboring regions of the star polymer is affected to different extents depending on the functionality of the polymer.²¹ Individual arms of the star polymer show a coil-stretch transition similar to linear chains in the presence of a fluid undergoing either extensional flow or shear flow.^{22,23} The critical strain rate or critical shear rate at which such a transition occurs is related to the arm-length instead of the molecular weight of the star polymer. Furthermore, star polymers exhibit a non-monotonic dependence of their mobility on the strength of the flow field.²⁴ This is found to be related to the conformational changes in the star polymer induced by the flow, whereby the size of the star polymer first decreases and then increases as a function of the field strength. Thus, conformational dynamics of a star polymer are strongly dependent on its functionality.

Translocation of linear polymers through finite-length nanopores itself is a rich problem due to the large number of parameters involved. For example, in the simplest case of charged linear polymers, the charge patterns along the polymer and the pore-polymer interactions play an important role in the translocation kinetics, as seen from simulations,^{25–27} experiments,²⁸ and from the theoretical description based on the Fokker-Planck formalism.^{29–31} In the experimental work involving DNA translocation, the ionic size of the salt used³² and the mismatch in salt concentration across the nanopore³³ change the kinetics of translocation. The problem gets even more complex when the polymer architecture is added to the parameter space. For short finite-length pores, hydrodynamics simulations show that the time taken by a star polymer to translocate across the nanopore depends on the initial distribution of arms inside and outside the nanopore.³⁴ If the translocation process begins with one or more leading arms inside the nanopore, the translocation time is found to increase with the increasing number of trailing arms in the initial configuration. On the other hand, the increasing number of leading arms beyond small numbers does not affect the translocation time significantly. However, increasing the number of arms in either case leads to an increase in the molecular weight of the star polymer. To arrive at specific conclusions about the role of polymer functionality, a systematic comparison between translocation kinetics of star polymers with varying functionalities but with the same molecular weight is necessary. In other words, simulating star polymers with equal molecular

weight but different architectures allow us to isolate the effect of topology on translocation kinetics.

In this paper, we study the translocation kinetics of charged star polymers of varying functionalities driven through narrow finite length nanopores. We find that the mean translocation time varies non-monotonically with the functionality of the star polymers for a nanopore of given dimensions. We also find that the nanopore dimensions affect the observed translocation kinetics significantly. Details of our simulation strategy are described in Sec. II, followed by the observed trends in the kinetics of translocation obtained from these simulations and a discussion of the observed non-monotonic dependence in Sec. III. Key conclusions are summarized in Sec. IV.

II. SIMULATION DETAILS

Coarse-grained three-dimensional Langevin dynamics simulations are used to study the translocation of charged star polymers of varying functionalities through an uncharged solid-state nanopore under the influence of an electric field. Figure 1 shows a schematic of the simulation setup.

A. Coarse-grained models for the polymer and the nanopore

A bead-spring model is used to represent the star polymer. A spherical bead is used to represent a monomer, which is connected to other such beads using harmonic springs. The mass and length scales are chosen such that the mass of a monomer bead is $m = 1$ (corresponding to a mass-scale of 130 g/mol) and size of a monomer bead is $\sigma = 1$ (corresponding to a lengthscale of 3 Å). Independently, we choose the energy scale to be $k_B T$, where k_B is the Boltzmann constant and $T = 300$ K is the temperature in Kelvin. All other quantities are converted to reduced units using these scales and are reported in this work in reduced units henceforth, unless noted otherwise.

The total number of monomer beads is kept constant at $N = 121$ for star polymers of different functionalities. f linear arms, each made of L_f beads connected linearly, are connected to a central bead (branch point) to form a star polymer with a functionality f . Consequently, each arm consists of

FIG. 1. Schematic of the simulation setup for a star polymer of functionality $f = 3$. The nanopore is made of static beads shown as black circles. The arms of the star polymer are made of charged beads shown as red circles, connected to a central uncharged bead (branch point) shown as a black circle.

$L_f = (N - 1)/f$ beads. The branch point is uncharged, while the rest of the beads carry a unit negative charge of $q = -1e$. Here, e is the elementary charge.

The functionality f of the star polymer is varied between 2, 3, 4, 5, 6, 8, and 10. For comparison, we also study the translocation of a star polymer with a very high functionality ($f = 120$) that mimics a soft colloid. A slightly modified model is used for the $f = 120$ star polymer. The uncharged branch point bead is connected to 10 arms, each consisting of 2 beads, to form the first “shell” of monomers. The free ends of each of these arms are then connected to 10 beads each, to form a second “shell” that envelops the first “shell.” This modified model is only used for $f = 120$ and helps to avoid large inter-monomer electrostatic and excluded volume repulsions that would arise if the original model was naïvely used for $f = 120$. The soft colloid represents a very large functionality star polymer of the same molecular weight as the rest of the star polymers used in this study and serves as a limiting case of a “structureless” star polymer with diminishing arm entropy. However, it should be noted that the soft colloid with $f = 120$ has a different mechanism of translocation than the rest of the star polymers studied.³¹

The three-dimensional simulation box is divided into two compartments, cis and trans, by a M unit thick rigid membrane modeled using two vertical walls. A small hole of radius r_p is present in both the walls and is connected using a cylindrical nanopore of length M . The membrane and nanopore walls are made of immobile and uncharged spherical beads of sizes 1 and 0.25, respectively.

B. Driving electric field

In a typical translocation experiment, an electric potential difference is applied across the nanopore using electrodes located in the two compartments. The electric potential drops significantly only inside the nanopore and varies only weakly outside the nanopore. To simulate this in an approximate sense, we set a constant electric field $E = 0.061$ inside the nanopore, acting along the nanopore axis (z -direction in Fig. 1). The electric field is set to zero in the cis and trans compartments.

C. Pair-wise interactions

Excluded volume between a pair of beads (monomer-monomer or monomer-wall) is modeled using the truncated Lennard-Jones interaction potential (U_{LJ}),

$$U_{LJ} = \begin{cases} 4\epsilon_{LJ} \left[\left(\frac{\sigma}{r} \right)^{12} - \left(\frac{\sigma}{r} \right)^6 \right] + \epsilon_{LJ} & \text{for } r \leq 1.12\sigma, \\ 0 & \text{for } r > 1.12\sigma, \end{cases}$$

where ϵ_{LJ} is the depth of the potential well, σ is the mean of the size of the two beads, and r is the distance between them. Two additional monomer-monomer interactions exist. The springs between bonded monomers are modeled using a pair-wise harmonic bond potential (U_b),

$$U_b = K(r - r_0)^2,$$

where $K = 15\,480$ is the harmonic spring constant, r is the distance between the two connected beads, and $r_0 = 1$ is the

equilibrium bond length. Electrostatic interactions between monomer beads are modeled using the Debye-Hückel potential (U_e),

$$U_e = \frac{1}{\epsilon_r} \frac{q_1 q_2}{r} e^{-\kappa r},$$

where $\epsilon_r = 80$ is the dielectric constant of the medium, q_1 and q_2 are the charges on the two monomer beads, and $\kappa^{-1} = 3.25$ is the Debye length corresponding to a 0.1M monovalent salt.

D. Equations of motion

In Langevin dynamics, the above potentials are used to solve the equations of motion for each monomer bead,

$$m \frac{d^2 r}{dt^2} = -\zeta \frac{dr}{dt} - \nabla(U_{LJ} + U_b + U_e) + F_r + F_{ext},$$

where r is the position of the monomer bead, t is the time, ∇ is the gradient operator, and $F_{ext} = qE$ is the force due to the external electric field. The drag force with drag coefficient $\zeta = 1$ and random force F_r acting on the monomer bead due to an implicit solvent is related by the fluctuation-dissipation theorem. The three-dimensional equations are integrated using the velocity-Verlet algorithm implemented in the molecular dynamics package LAMMPS³⁵ using a time step $dt = 0.005$.

Before setting up the simulation runs, 40 different equilibrium conformations are generated for each functionality f . These equilibrated initial conformations consist of the star polymer with one terminal bead just inside the nanopore and the rest of the beads on the cis side of the nanopore. Our motivation behind choosing such an initial conformation for the star polymer versus choosing an interior bead of an arm just inside the nanopore (corresponding to a hairpin initial conformation) is that the star polymer would be stretched as it is captured from the bulk of the donor compartment toward the nanopore entrance due to the electro-osmotic flow and the extended electric field near the nanopore.²⁹ The electrostatic repulsion between beads of the same arm can further make the hairpin initial configuration less likely. Hence, we exclude the hairpin initial conformations from our study. For each functionality f , 2000 statistically independent simulation runs are performed. Each simulation run starts with an initial conformation selected from the 40 different equilibrium conformations. Random initial velocities are assigned to all the monomer beads in each run. The equations of motion are then integrated in time until all the beads are on the either side of the nanopore. A translocation run is considered as successful if the entire star polymer translocates across the nanopore onto the trans side. Translocation time is defined only for successful runs and is calculated as the time taken by the terminal bead of the last arm to exit the nanopore. The time taken by all the successful runs is averaged to calculate the mean translocation time $\langle \tau \rangle$. Separately, radius of gyration (R_g) of isolated star polymers is recorded as a function of f , based on 9 simulations and in the absence of the nanopore. We observe that the radius of gyration systematically decreases with increasing f (for $f = 2, 3, 4, 5, 6, 8$, and 10 and $R_g = 11.0, 9.9, 8.7, 7.9, 7.1, 6.1$, and 5.3, respectively).

III. RESULTS AND DISCUSSION

A typical simulation run proceeds as follows (see Figs. 1 and 7). As mentioned earlier, the translocation is nucleated at the beginning of the simulation run by placing one monomer bead belonging to the end of an arm (leading arm) just inside the nanopore entrance. With translocation progressing, more and more beads of the leading arm enter the nanopore. This continues until the branch point is at the nanopore entrance. Depending on the functionality of the star polymer, there can be a delay before the branch point enters the nanopore. Once the branch point is absorbed into the nanopore, the rest of the arms (trailing arms) are dragged along with the branch point into the nanopore. The translocation continues until all the trailing arms exit the nanopore. This closely follows the drag mechanism proposed in Ref. 11, albeit in the presence of a drift. In the presence of strong driving forces such as the force due to the electric field present in our simulations and in typical translocation experiments, the trailing arms of the polymer do not get enough time to retract to the branch point to undergo relaxation. Instead, they are dragged along with the branch point and are forced into the nanopore. The free end of each trailing arm enters the nanopore only after the rest of the arm is already inside the nanopore, in contrast to the reptation mechanism for star polymers.^{8–10}

A. Effect of polymer functionality

The nanopore dimensions are kept fixed at $M = 16$ and $r_p = 3.2$ to study the translocation of star polymers with different functionalities f . Figure 2 shows the mean translocation time $\langle\tau\rangle$ as a function of f . Error bars denote the width of the translocation time distribution. For lower functionalities, the mean translocation time decreases as the functionality of the star polymer increases. This continues until a critical value of the functionality ($f^* = 6$). Above this functionality, the mean translocation time increases with the functionality. For the soft colloid mimicked with $f = 120$, however, the mean translocation time is the lowest. The error bars in Fig. 2 correspond to standard deviation in the data and indicate that the width of the translocation time distribution increases

FIG. 2. Mean translocation time $\langle\tau\rangle$ of star polymers of equal mass as a function of their functionality f for a nanopore of radius $r_p = 3.2$ and length $M = 16$. Note: The polymer with $f = 120$ is used to simulate very large functionality and is architecturally different than other star polymers. See text for details.

beyond the critical functionality. Figure 3 shows the corresponding translocation time histograms, emphasizing that for star polymers with $f > f^*$, the translocation time distribution is wider and is characterized by a longer tail. The distribution is narrow and more symmetric for lower functionality star polymers.

These results can be understood in terms of two competing forces. Before the branch point enters the nanopore, the polymer charge density inside the nanopore is the same across different functionalities since only the leading arm of the polymer is inside the nanopore. In the later stages of translocation, however, the number of trailing arms of the polymer and hence the charge density inside the nanopore is higher for polymers with higher functionality. Consequently, in the later stages, the average driving force due to the electric field acting on the polymer increases with increasing functionality. At the same time, an increase in charge density also leads to higher inter-arm repulsion between the trailing arms of the polymer and hence an increase in the resistance of the trailing arms to enter the nanopore. A balance between the driving electric force and the opposing confinement effect results in a minimum in the mean translocation time, as observed in Fig. 2. In the extreme case of the soft colloid ($f = 120$), the driving force is maximum while the confinement effect is negligible since the nanopore is wider than the effective size of the colloid. Hence, in Fig. 2, the shortest translocation time is observed for $f = 120$. It is essential to distinguish between the translocation of polymers through nanopores and the transport of a soft colloid through a nanopore. Although it is tempting to think of the translocation of polymers through nanopores in terms of an equivalent structure-less particle translocating through a nanopore of a longer effective length, it is important to realize that the two mechanisms of transport are fundamentally different.³¹

B. Effect of nanopore radius

Nanopore radius r_p affects the confinement without affecting the driving electric force and hence changes Fig. 2 significantly. The variation in mean translocation time of star polymers with different functionalities translocating across nanopores of different radii is shown in Fig. 4(a), and the corresponding probability of successful translocations, $p(\text{success})$,

FIG. 3. Translocation time distribution for polymers with different functionalities f for a nanopore of radius $r_p = 3.2$ and length $M = 16$. Note that for better visualization, each of these curves is obtained by fitting a log-normal distribution to the translocation time histogram obtained from the 2000 statistically independent simulation runs.

FIG. 4. (a) Mean translocation time $\langle \tau \rangle$ and (b) probability of successful translocation $p(\text{success})$ of star polymers as a function of their functionality f for different nanopore radii r_p . The nanopore length is $M = 16$.

is shown in Fig. 4(b). The probability of successful translocations is calculated as the fraction of the total simulation runs that lead to a successful translocation across the nanopore. All the features observed for $r_p = 3.2$ (Fig. 2) are also observed for smaller nanopore radii. The mean translocation time shows a non-monotonic dependence on the functionality of the star polymer. For the lowest functionality star polymer ($f = 2$), making the nanopore narrow only slightly affects the mean translocation time. For polymers with all other functionalities, the mean translocation time increases with narrowing of the nanopore. The critical functionality f^* , corresponding to the shortest translocation time, also decreases with decreasing nanopore radius. Additionally, a cutoff functionality f_c exists, beyond which the translocation is “never” successful. For example, for $r_p = 1.4, 1.7, 1.8$, and 2.1 , the cutoff is $f_c = 4, 5, 5$, and 6 , respectively. To ensure that these values of cutoff functionalities are not sensitive to the total simulation time for each statistical run, we performed test simulations with a small number of longer simulation runs to find that the probability of successful translocation approaches zero beyond the cutoff functionality even when the simulation time is up to two orders of magnitude longer than the mean translocation time (results not shown).

Clearly, for star polymers with higher functionalities, the resistance of trailing arms to enter the nanopore increases as the nanopore becomes narrower. This effect is irrelevant for linear polymers ($f = 2$) since there is only one trailing arm. Hence, nanopore radius does not seem to affect the translocation time significantly for $f = 2$, within the range of

nanopore radii studied. For higher functionality polymers, the resistance is stronger for smaller nanopore radii, resulting in an increase in the mean translocation time. The critical functionality corresponding to the slowest translocation time is a consequence of the balance between the driving electric force and the opposing confinement, both governed by the polymer charged density inside the nanopore. The linear charge density along the nanopore axis does not change dramatically with decreasing nanopore radii. Thus, the driving electric force is comparable across nanopores with varying radii. However, the radial charge density inside the nanopore is higher for narrower nanopores, which increases the resistance of trailing arms to enter the nanopore due to inter-arm repulsion. Hence, the critical functionality is found to be affected by the nanopore radius. The existence of a cutoff functionality for a given nanopore radius can similarly be attributed to the increased number of trailing arms for polymers with higher functionalities that makes their entrance into the nanopore prohibitive.

C. Effect of nanopore length

In the following simulations, the nanopore radius is kept fixed at $r_p = 3.2$, while its length M is varied between 2 and 32 units. For systematic comparison, we choose to keep the electric field inside the nanopore to be constant, instead of the alternative choice of keeping the electric potential drop across the nanopore to be constant. The latter choice would significantly reduce the driving force during the nanopore filling stage for longer pores and hence would decrease the number of simulation runs that result in a successful translocation. Hence, we keep the electric field constant, noting that the driving electric force increases with increasing nanopore length.

Figure 5 shows the variation in mean translocation time with the nanopore length M . For a given functionality, the translocation time decreases with an increase in the nanopore length for shorter pores. For longer pores, however, the translocation time is independent of the nanopore length. Additionally, we also observe that the sensitivity of translocation kinetics toward the functionality of the polymer reduces with increasing nanopore length. Histograms for the translocation time distribution corresponding to three nanopore lengths for

FIG. 5. Mean translocation time $\langle \tau \rangle$ of star polymers as a function of the nanopore length M . The nanopore radius is $r_p = 3.2$. Different symbols indicate different functionalities. Note that the electric field inside the nanopore is kept constant. Hence, longer pores have a stronger driving force.

FIG. 6. Translocation time histograms for star polymers of different functionalities f for different nanopore lengths M . (a) $M = 2$, (b) $M = 4$, (c) $M = 6$, (d) $M = 8$, (e) $M = 16$, and (f) $M = 32$. Within statistical variations, the histograms in (e) are similar to the corresponding histograms in Fig. 3. Note that for better visualization, each of these curves is obtained by fitting a log-normal distribution to the translocation time histogram obtained from the 2000 statistically independent simulation runs.

three different polymer functionalities are shown in Fig. 6. Histograms for different functionalities have distinct peaks for all nanopore lengths. Additionally, increasing the nanopore length also results in a narrower distribution.

In summary, a clear non-monotonic trend exists between the functionality of star polymers and the kinetics of their translocation through uncharged nanopores. Remarkably, the translocation time histograms for architecturally different polymers can be made distinct by tuning the nanopore dimensions.

IV. MECHANISM OF TRANSLOCATION

A. One leading arm vs two leading arms

We initialize all our simulations by placing an end of one of the arms of the star polymer just inside the nanopore. The following paragraph justifies this choice of the initial polymer conformation, in contrast to having ends of two or more arms inside the nanopore.

As mentioned in Sec. II, in typical experiments, a significant drop in the applied electric potential takes place across the nanopore, while the potential only weakly changes far from the nanopore. The star polymer has to diffuse to a certain distance near the nanopore, termed as the critical capture radius, before it is drifted toward the nanopore due to the electric field while also undergoing diffusion.²⁹ It is energetically unfavorable to have ends of two arms of the star polymer to simultaneously enter the nanopore. Nevertheless, we also simulate this unlikely situation to show that even if such a two-arm capture did take place, most of such translocation attempts would fail,

resulting in one or both arms of the star polymer retracting out of the nanopore. For a nanopore with dimensions $r_p = 3.2$ and $M = 16$, we initialize translocation simulations by placing the ends of two arms of the star polymer just inside the nanopore. These simulations show that less than 10% of the statistical runs with two leading arms result in successful translocation (the predicted values for the probability of successful translocations are 0.074, 0.049, 0.071, and 0.055 for $f = 2, 3, 4$, and 5, respectively). The low probability of success observed in these simulations additionally supports our choice of initial polymer conformations.

As discussed in Sec. I, two distinct mechanisms have been proposed in the literature to model the transport of star polymers. The reptation model proposes that for a star polymer with f arms, $f - 2$ of its arms have to retract to the branch point for the center of mass of the polymer to move by a unit distance⁸ along the direction of the leading arm. An alternative mechanism is where the branch point of the star polymer moves along with the leading arm, while the rest $f - 1$ arms are dragged along.¹¹ Based on the results of our simulations with two leading arms, we conclude that the latter mechanism is dominant in driven translocation of star polymers. In contrast to our observation, fluid flow driven star polymer translocation shows a non-monotonic trend in the probability of successful translocation with respect to the number of leading arms.¹⁷ The flow field drags the star polymer into the nanopore and makes conformations with more than one leading arm more probable, giving rise to a maximum probability of successful translocation for more than one leading arm.

B. Stages of translocation

In order to understand the results shown in Fig. 4, we divide the translocation of the star polymer into the following four stages (see Fig. 7):

- Branch point absorption: The branch point approaches and enters the nanopore from the cis side, starting from the initial configuration of having the end of the leading arm just inside the nanopore.
- Trailing arms filling: The branch point is translocated across the nanopore as the trailing $f - 1$ arms fill the nanopore completely.
- Trailing arms threading: The trailing $f - 1$ arms thread through the nanopore until all trailing arm ends enter the nanopore.
- Nanopore depletion: Ends of the $f - 1$ arms translocate across the nanopore and exit the nanopore.

We compute the average time taken for the above stages based on successful simulation runs corresponding to Fig. 4

FIG. 7. Simulation snapshots showing a three arm star polymer ($f = 3$) in its initial conformation with one leading arm just inside the nanopore, branch point absorption, branch point translocation, trailing arms threading, and nanopore depletion. The nanopore dimensions are $r_p = 3.2$ and $M = 16$.

and denote these by $\langle\tau_a\rangle$, $\langle\tau_b\rangle$, $\langle\tau_c\rangle$, and $\langle\tau_d\rangle$, respectively. Figure 8 shows the average time taken by each of these stages.

The length of each arm of the star polymer L_f decreases as its functionality increases. For example, a linear polymer has the longest contour length from end to end ($=2L_f = 120$) compared to the contour length from the end of the leading arm to the end of a trailing arm for a three arm star polymer ($=80$). Thus, the total path that the star polymer needs to travel while undergoing translocation decreases with increasing functionality. Beyond branch point absorption, the driving force due to the electric field increases with increasing functionality of the star polymer. This is due to the increased linear charge density inside the nanopore due to the increased number of trailing arms. At the same time, an increase in the number of trailing arms causes an increase in their resistance to enter the nanopore due to inter-arm electrostatic repulsion. We now describe our results in terms of the above three effects: path length, driving force, and trailing arms resistance.

Higher functionality star polymers encounter a shorter path in the first stage of translocation, causing the average time for the first stage $\langle\tau_a\rangle$ to decrease with f . However, as the number of trailing arms increases, $\langle\tau_a\rangle$ increases. The increase is expected to arise from the trailing arms interacting with the membrane walls, which causes a delay as the branch point gets closer to the nanopore. Additionally, the branch point also brings nearby monomers of the trailing arms closer as it enters the nanopore, causing an additional increase in $\langle\tau_a\rangle$ with increasing f .

The average time taken for the branch point threading stage $\langle\tau_b\rangle$ increases with the number of trailing arms and hence with f . For a star polymer of a certain functionality, $\langle\tau_b\rangle$ decreases with increasing nanopore radius. The time taken for this stage is governed by the balance between the resistance of the monomers of the trailing arm to enter the nanopore due to electrostatic repulsion and the increase in driving force due to increased charge density inside the nanopore. For narrow nanopores, the former causes the average time of this stage to

FIG. 8. Detailed time taken for each stage of the translocation for different nanopore radii corresponding to Fig. 4(a). Average time for (a) branch point absorption τ_a , (b) trailing arms filling τ_b , (c) trailing arms threading τ_c , and (d) nanopore depletion τ_d is plotted for star polymers of functionality f and nanopore radii r_p ($M = 16$).

FIG. 9. Average time for the trailing arms threading stage $\langle\tau_c\rangle$ for star polymers of different functionalities plotted versus the star polymer arm length $L_f = 120/f$ ($M = 16$).

increase with f . As the nanopore radius increases, the resistance of trailing arms to enter the nanopore becomes weaker as the nanopore is able to accommodate trailing arms with larger inter-arm distance, making the inter-arm electrostatic repulsion weaker.

The translocation velocity during the trailing arm threading stage is nearly constant for different functionality star polymers, as seen from Fig. 9. This is because for every monomer belonging to a trailing arm that enters the nanopore, another monomer comes out from the cis side. There is no net change in the average charge density inside the nanopore and hence in the resistance of trailing arms to enter the nanopore. The path that the star polymer has to traverse during this stage decreases with increasing functionality. Hence, $\langle\tau_c\rangle$ decreases with f . The average velocity of the star polymer during this stage slightly increases as the nanopore radius increases since the arms are slightly less stretched in a wider nanopore, and hence the nanopore accommodates slightly more charges. This reduction in stretching is evident from the observation that the time taken for trailing arms threading through a 3.2 radius nanopore $\langle\tau_c\rangle \sim 0$ for the star polymer with functionality $f = 6$ (the arm length $L_f = 20$), even when the nanopore length $M = 16$. This indicates that the arms of the polymer are not fully stretched inside the nanopore.

The time taken for the nanopore depletion stage $\langle\tau_d\rangle$ is not sensitive to the functionality of the star polymer or the nanopore radius. During this stage, the driving electric force and the confinement act synergistically to drive the polymer out of the nanopore quickly.

V. CONCLUSION

We have studied the translocation of charged star polymers of varying functionalities through an uncharged nanopore driven by an electric field. The key observation made in this work is the strong dependence of the mean translocation time on the functionality of the star polymer. More significantly, the dependence is non-monotonic, with the presence of a minimum for a critical functionality. We also demonstrate that this minimum can be shifted by tuning the dimensions of the nanopore such that the histograms of translocation time are able to distinguish between star polymers of different functionalities.

Gel electrophoresis is used as an analytical method for detecting and isolating star polymers based on their architecture, in addition to more sophisticated techniques such as nuclear magnetic resonance (NMR) spectroscopy. However, techniques such as gel electrophoresis are indirect in the sense that one can only distinguish between polymers based on their sizes. Our simulation results show that the translocation time histograms (which are directly measured in translocation experiments) of architecturally different polymers translocating through nanopores are clearly distinguishable with distinct peaks for certain nanopore dimensions. This suggests that nanopores can be used as a high-throughput low-cost alternative analytical tool for detection and separation of charged polymers based on their architecture. Tuning the pore-polymer interactions in combination with dialing the pore diameter can further amplify the discriminatory power of distinguishing various star-like architectures.

ACKNOWLEDGMENTS

This is made to the National Institutes of Health (Grant No. R01HG002776-11), Air Force Office of Scientific Research (Grant No. FA9550-17-1-0160) and National Science Foundation (Award No. 1504265).

- ¹G. S. Grest, L. J. Fetters, J. S. Huang, and D. Richter, *Adv. Chem. Phys.* **94**, 67 (1996).
- ²W. D. Volkmuth, T. Duke, R. H. Austin, and E. C. Cox, *Proc. Natl. Acad. Sci. U. S. A.* **92**, 6887 (1995).
- ³L. Bell and B. Byers, *Anal. Biochem.* **130**, 527 (1983).
- ⁴J. Xing, L. Tan, F. Cao, and Y. Wang, *J. Appl. Polym. Sci.* **129**, 1179 (2013).
- ⁵D. M. Heuer, S. Saha, and L. A. Archer, *Electrophoresis* **24**, 3314 (2003).
- ⁶S. Saha, D. M. Heuer, and L. A. Archer, *Electrophoresis* **27**, 3181 (2006).
- ⁷I. Hoffmann, P. M. de Molina, B. Farago, P. Falus, C. Herfurth, A. Laschewsky, and M. Gradzielski, *J. Chem. Phys.* **140**, 034902 (2014).
- ⁸P. G. de Gennes, *J. Phys.* **36**, 1199 (1975).

- ⁹W. W. Graessley, *Adv. Polym. Sci.* **47**, 67–117 (1982).
- ¹⁰M. Doi and N. Y. Kuzuu, *J. Polym. Sci., Polym. Lett. Ed.* **18**, 775 (1980).
- ¹¹J. Klein, D. Fletcher, and L. J. Fetters, *Faraday Symp. Chem. Soc.* **18**, 159 (1983).
- ¹²D. S. Pearson and E. Helfand, *Macromolecules* **17**, 888 (1984).
- ¹³D. M. Heuer, S. Saha, A. T. Kusumo, and L. A. Archer, *Electrophoresis* **25**, 1772 (2004).
- ¹⁴A. Milchev, M. Müller, and L. Klushin, *Macromolecules* **47**, 2156 (2014).
- ¹⁵J. M. Polson, A. F. Tremblett, and Z. R. N. McLure, *Macromolecules* **50**, 9515 (2017).
- ¹⁶D. Račko and P. Cifra, *J. Mol. Model.* **21**, 186 (2015).
- ¹⁷Q. Chen, L. Zhang, M. Ding, X. Duan, Y. Huang, and T. Shi, *Eur. Phys. J. E* **39**, 109 (2016).
- ¹⁸F. Brochard-Wyart and P. G. de Gennes, *C. R. Acad. Sci., Ser. IIB: Mec., Phys., Astron.* **323**, 473 (1996).
- ¹⁹P. G. de Gennes, *Adv. Polym. Sci.* **138**, 91 (1999).
- ²⁰Z. Li, Y. Li, Y. Wang, Z. Sun, and L. An, *Macromolecules* **43**, 5896 (2010).
- ²¹M. Ripoll, R. G. Winkler, and G. Gompper, *Eur. Phys. J. E* **23**, 349 (2007).
- ²²J. Hernández Cifre, M. López Martínez, and J. García de la Torre, *J. Non-Cryst. Solids* **307**, 818 (2002).
- ²³J. Hernández Cifre, R. Pamies, M. López Martínez, and J. García de la Torre, *Polymer* **46**, 6756 (2005).
- ²⁴S. P. Singh, G. Gompper, and R. G. Winkler, *J. Chem. Phys.* **148**, 084901 (2018).
- ²⁵K. Luo, T. Ala-Nissila, S.-C. Ying, and A. Bhattacharya, *Phys. Rev. Lett.* **99**, 148102 (2007).
- ²⁶M. G. Gauthier and G. W. Slater, *J. Chem. Phys.* **128**, 175103 (2008).
- ²⁷S. Mirigian, Y. Wang, and M. Muthukumar, *J. Chem. Phys.* **137**, 064904 (2012).
- ²⁸G. Maglia, M. R. Restrepo, E. Mikhailova, and H. Bayley, *Proc. Natl. Acad. Sci. U. S. A.* **105**, 19720 (2008).
- ²⁹M. Muthukumar, *Polymer Translocation* (Taylor & Francis, Boca Raton, 2011).
- ³⁰H. H. Katkar and M. Muthukumar, *J. Chem. Phys.* **140**, 135102 (2014).
- ³¹M. Muthukumar and H. H. Katkar, *Biophys. J.* **108**, 17–19 (2015).
- ³²S. W. Kowalczyk, D. B. Wells, A. Aksimentiev, and C. Dekker, *Nano Lett.* **12**, 1038 (2012).
- ³³B. J. Jeon and M. Muthukumar, *J. Chem. Phys.* **140**, 015101 (2014).
- ³⁴Z. Liu, J. Liu, M. Xiao, R. Wang, and Y.-L. Chen, *Biomicrofluidics* **8**, 054107 (2014).
- ³⁵S. Plimpton, *J. Comput. Phys.* **117**, 1 (1995).