Comparison of the single molecular dynamics of linear and circular DNAs in microfluidic planar extensional flows

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ABSTRACT

Understanding the elongational flow behavior of macromolecules is of great interest due to its wide application in turbulent drag reduction [1], fiber spinning [2], injection molding [3], and DNA/polymer purification techniques [4-6]. It was predicted by P. G. de Gennes in 1974 [7] that above a critical extension rate, a coiled linear polymer experiences a steady state coil-stretch transition (CST) to adopt a relatively open configuration, and conformation hysteresis occurs when the molecular weight becomes sufficiently high. This remarkable prediction has been proven to be true for linear polymers by many techniques, such as flow birefringence [8], microfluidic trap [9], and Brownian dynamics simulation [10]. However, due to the experimental difficulty in synthesizing high molecular weight circular polymers [11], it remains unknown whether ring-like polymers exhibit a similar CST, and, if so, how their steady state and transient elongational flow behavior compares with that of linear counterparts.

DNA, due to its biological significance [4], easiness to be fluorescently labeled [9], and uniformity in molecular structure, is used in the present study as a model polymer. We, for the first time, examine the dynamics of large circular and linear DNA molecules with the goal of providing fundamental data in order to compare the CST [7, 9], conformation hysteresis [12], and molecular individualism (MI) [9] for the two types of macromolecules. To do this, we combined the biological synthesis of DNA molecules following the procedures of Douglas E. Smith and coworkers [13] with direct imaging of fluorescently labeled DNA at the stagnation point in a novel microfluidic device that has a cross-slot design to develop a stagnation point extensional flow that permits direct imaging of the fluorescently labeled DNA [14].

We found that the relaxation time of linear DNA is about 3.2 times of that of circular counterpart of the same molecular weight, and this is true for both 45 and 115 kbp (kilo base pair). Also, as shown in Figure 1, both linear and circular DNAs exhibit a sharp transition in molecular configurations at a Weissenberg number Wi of about 0.4 – 0.5 (Wi = flow rate multiplied by

relaxation time). However, for both 45 and 115 kbp circular DNA, the transition is shallower compared with the linear DNA. Unlike linear DNA which tends to adopt folded configurations when MI occurs during a transient stretch [9], the 115 kbp circular DNA prefers forming a kink near the middle of the chain; however, 45 kbp circular DNA always shows a dumbbell-like configuration without any molecular individualism. Therefore, molecular individualism not only is a function of molecular weight, but also closely relates to molecular topology. Within the range of molecular weight investigated in this work (45 and 115 kbp), conformation hysteresis is not observed for circular DNAs.

To summarize, circular DNAs, due to their unique molecular structure, possess shorter relaxation times, shallower slopes in steady state CST curves, and different transient MI behaviors, comparing with linear DNAs. This implies that quite different physics [15], such as entropic elasticity, viscous drag, hydrodynamic interactions, excluded volume effects, internal viscosity, and self-entanglement, could be involved in the flows of circular and linear DNAs. Therefore, this comparison of circular and linear macromolecules serves as an ideal platform for scientists to explore the structure-property relationship of macromolecules, both experimentally and in a simulation sense.

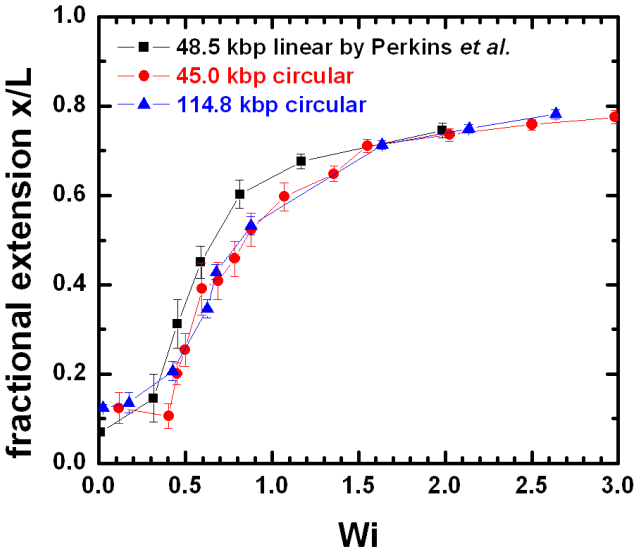


Figure 1. Steady state fractional extension x/L as a function of Weissenberg number Wi for three DNA samples: 48.5 kbp linear, 45 and 115 kbp circular.

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