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Probing the Alignment-Dependent Mechanical Behaviors and Time-Evolutional Aligning Process of Collagen Networks

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Note S1. Coarse-Grained Model

This coarse-grained (CG) model is derived and validated from a higher-fidelity all-atomistic (AA) model with explicit water solvents, in which the hydrodynamic interaction between collagen (COL) and water significantly impacts mechanical behaviors from bending to shearing of COL. Therefore, by fitting the CG forcefield parameters to match a series of mechanical loadings, e.g., tension, compression, shearing, and bending, to the higher-fidelity solvent-explicit AA model, the effects of solvent and hydrodynamic interactions from the fluid shear are inherently taken into account within this implicit-solvent CG potential forcefield. More importantly, the extremely flexible and solvent-sensitive property, persistence length of tropocollagen, usually measured by experiments, can also be accurately captured with this CG model, which also validates the predictability of the CG model for COL solution. We further validate the accuracy and our implementation of the CG simulation framework with uniaxial tensile and three-point bending tests of a single tropocollagen molecule (~308 nm) by comparing against existing literature[1] in **Note S4**. Our simulated single tropocollagen molecule is in a calculated concentration of 0.32×10^{-4} mg/mL, which is about four orders of magnitude more dilute than our most dilute experimental COL solution (0.5 mg/mL)[2]; therefore, we are only simulating the very initial tethering stage of the COL aligning for gaining some qualitative and fundamental insight in the smaller scale of nano- to micro-meters.

Note S2. Collagen Tethering

We apply the COL tethering by applying a point force at one end of the tropocollagen to mimic the experimental beginning condition of the single tropocollagen adhering/tethering to the gelatin

during spinning. In this manner, other portions of the COL will be "dragged" with the facilitating flow generated from the rotating spindle. The point force is applied to the centroid of the specific end bead (an infinitesimal entity). There is no difference between the application of tension or shear to the entity. Therefore, it is assumed that the point force should mimic the shear applied in experiments. The forcefield parameters (**Table S1**) of this CG model are set strictly following previous works[3] which were well-validated. The tethering threshold force values are determined by systematic simulations with trial-and-error and statistically averaged approaches. The number of COL units in an oligomer is rationally selected to qualitatively elucidate the chain length effects.

Note S3. P_2 Parameter

We use an alignment parameter, P_2, to characterize the conformation of the tropocollagen and bulk COL. It is extensively used to characterize the alignment of a single-chain/bulk polymer in previous works[4]. This parameter was used to effectively describe the main chain orientation, hydrogen bonds, and benzene segments of the uniaxially stretched poly(vinylphenol)[4a]; it was also used to analyze the strain-dependent conformational change of polymer chain and correlated to the enhanced thermal conductivity[4b]; it was effectively applied to track the alignment of bulk polyethylene under thermal drawing[4c]. In conclusion, P_2 is a validated qualitative measure which fluctuates between perfectly aligned (value of 1) and perfectly disordered (value of 0). The P_2 value is increasing over time until it reaches a plateau, which indicates the chain becomes more aligned over time.

Note S4. Validating the CG Forcefield for Mechanical Behaviors of a Single Tropocollagen Molecule

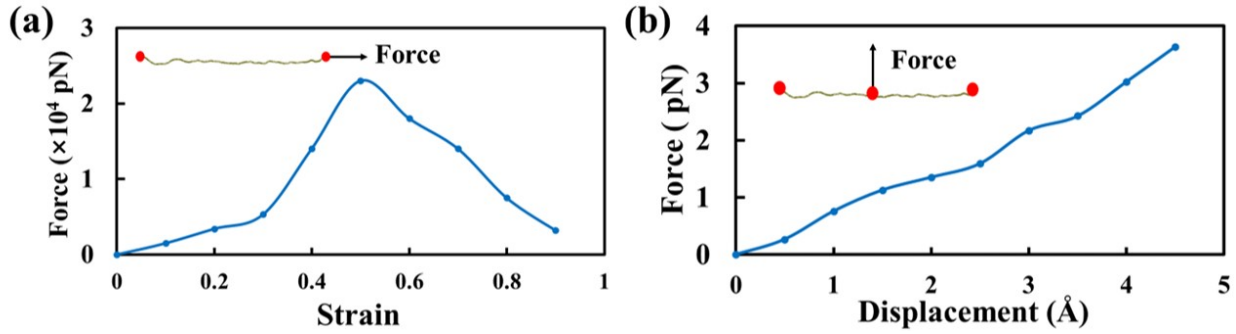

Figure S1. **(a)** CG-simulated uniaxial tensile behavior of a single tropocollagen molecule (308 nm) with a deformation rate of 0.1 m/s. **(b)** CG-simulated three-point bending behavior of a fraction of tropocollagen molecule (8.4 nm) with a deformation rate of 0.075 m/s.

To validate the accuracy of the simulation framework in our CG modeling, we first perform uniaxial tensile and three-point bending tests of a single tropocollagen molecule (~308 nm) to compare against existing literature[1] using the forcefield parameters. The tensile force gradually increases when the strain is below ~0.3 while increasing more steeply thereafter (**Figure S1(a)**), indicating the hyper-elastic behavior at large strains. The molecule finally breaks at a strain of ~0.5. The mechanical behavior corresponds well to a previous study[1] that investigated the same properties in tropocollagen molecule but with a length of 301.7 nm. Similarly, the bending force gradually increases as a function of the displacement of the middle bead for a small tropocollagen molecule with a length of 8.4 nm (**Figure S1(b)**). This yields a bending stiffness of 9 $.26 \times 10^{-29}\, N \cdot m^2$, corresponding well with the stiffness of $9.71 \times 10^{-29}\, N \cdot m^2$ in previous work[3b].

Table S1. CG model parameters.

Parameter	Collagen	Cross-link
Dispersive parameter (σ_{LJ}, Å)	14.72	
Dispersion energy parameter (ε_{LJ}, kcal/mol)	11.06	
Tensile stiffness parameter (k_1, kcal/mol/Å^2)	17.13	0.2
Tensile stiffness parameter (k_2, kcal/mol/Å^2)	97.66	41.84
Equilibrium angle (β_0, degree)	170 – 180	
Bending stiffness parameter ($k_{angular}$, kcal/mol/rad^2)	14.98	
Equilibrium bead distance (r_0, Å)	14	10
Critical hyperelastic distance(r_1, Å)	18.2	12
Bond breaking distance (r_{break}, Å)	21	14.68

Table S2. Computational fracture strains where the collagen breaks.

Cross-link density	Strains
0	0.69
10%	0.65
20%	0.63
40%	0.62
60%	0.58
80%	0.55

Table S3. Computational paired, bonded, and angular interactional energies (kcal/mol) per bead of collagen with different chain lengths. The 5-unit collagen oligomer shows higher repulsive paired energy but similar bonded and angular energy per bead compared with the 1-unit collagen monomer.

Chain length	1-unit	5-unit
Paired	– 0.487	– 0.348
Bonded	0.073	0.083
Angular	0.142	0.138

References

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