Supporting Information

Effect of scaffold morphology and cell co-culture on tenogenic differentiation of HADMSC on centrifugal melt electrospun poly (L-lactic acid) fibrous meshes

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Supplemental Table S1 qPCR Primers

	Genbank ID	Primer sequences (5'→3')	Product size (bp)
18S	NR_003286	F: GAGAAACGGCTACCACATCC	170
		R: CACCAGACTTGCCCTCCA	
SCX	NM_001080514.2	F: AGCGATTCGCAGTTAGGAGG	185
		R: GTCTGTACGTCCGTCTGTCC	
COL1	NM_000088.3	F: CTACGATGGCTGCACGAGTC	151
		R: GACAGGGCCAATGTCGATGC	
COL3	NM_000090.3	F: CGCCCTCCTAATGGTCAAGG	161
		R: TTCTGAGGACCAGTAGGGCA	
TNMD	NM_022144.2	F: AATGAACAGTGGGTGGTCCC	164
		R: TTGCCTCGACGGCAGTAAAT	
TNC	NM_002160.3	F: AAAGCGGGGAATGTTGGGAT	139
		R: CCTGTAAGCTTTTCCCAAGTG	

1. Effects of different parameters on fiber orientation

Abbreviations: T: spinning temperature; R: rotational speed, V: voltage; C: distance of collection; M: mass of raw PLLA materials; P: production rate of fibers

1.1 Effects of spinning voltage on fiber orientation

The fiber orientation was generally improved with increasing voltage in the range of 5-45 kV (Fig. S1). However, the fiber orientation slightly declined, when the voltage was too high.

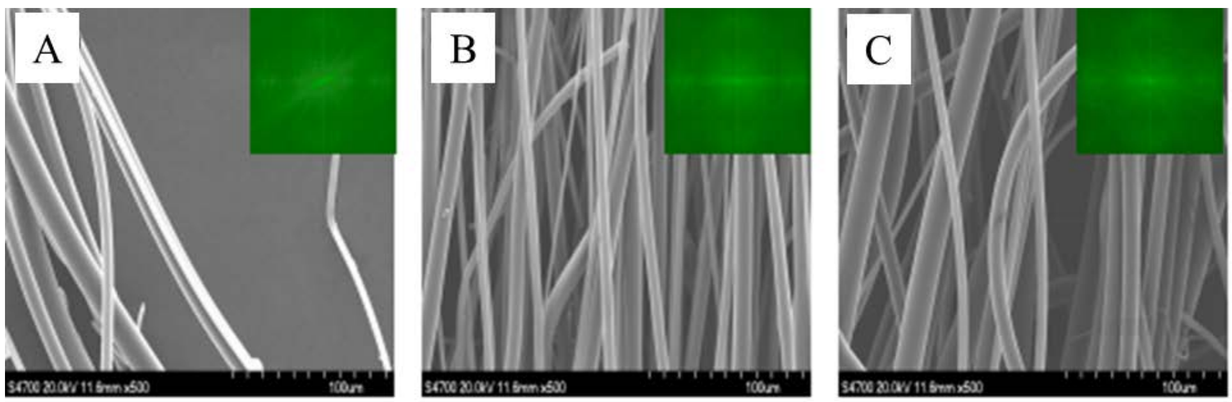

Figure S1 SEM images and fast fourier transform (FFT) spectrograms of fibers prepared at different voltages: (A) 5 kV, (B) 35 kV, (C) 45 kV. Other spinning parameters remained stable: T=230 °C, C=10 cm, R=900 rpm.

1.2 Effects of spinning temperature on fiber orientation

The fiber orientation presented a significantly increasing trend with the increase of temperature from 215 to 265 °C (Fig. S2).

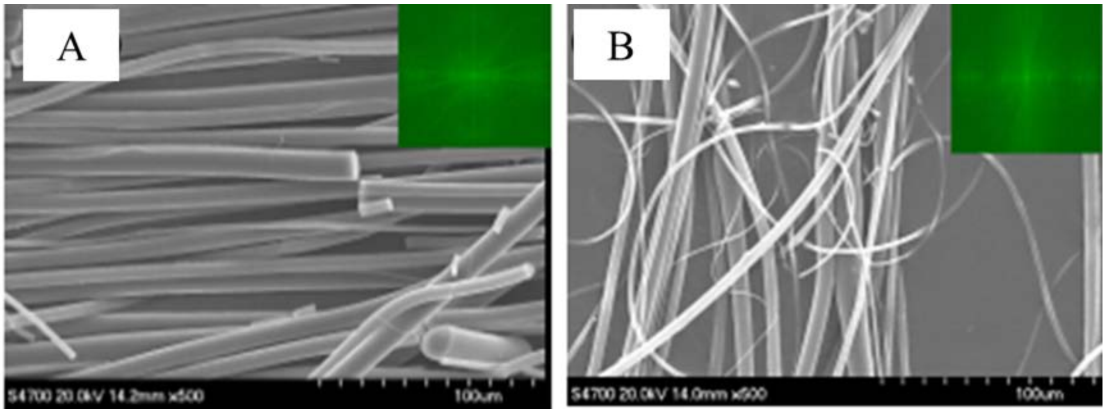

Figure S2 SEM images and FFT spectrograms of fibers prepared at different temperatures: (A) 265 °C, (B) 215 °C. Other spinning parameters remained stable: V=35 kV, C=10 cm, R=1140 rpm.

1.3 Effects of rotational speed on fiber orientation

The fiber orientation ascended initially and then descended with rotational speed increased from 780 to 1500 rpm (Fig. S3).

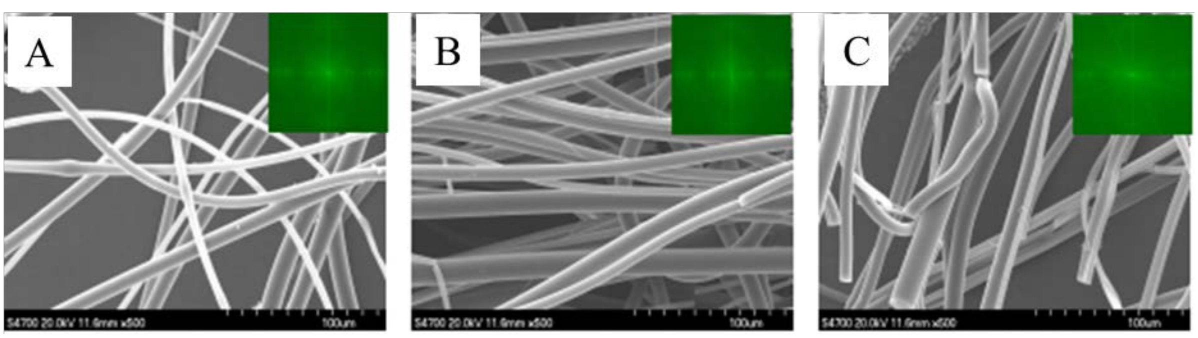

Figure S3 SEM images and FFT spectrograms of fibers at different rotational speeds: (A) 780 rpm, (B) 1140 rpm, (C) 1500 rpm. Other spinning parameters remained stable: V=30 kV, C=10 cm, T=230 °C.

2. Effects of different parameters on fiber diameter

2.1 Effects of spinning voltage on fiber diameter

Fiber diameter was measured by using Image J software. Five points of interest of 30 fibers were taken for measurement as shown in Fig. S4. As seen in Fig. S4, S5, during the CME process, increasing spinning voltage lead to polymeric fibers being discretely stretched towards the collection ring. As shown in Fig. S4, S5, the electric field excerted a tensile effect. With increasing voltage, the fiber was fully stretched during the collection process, with a concomitant decrease in fiber diameter. At a voltage above fiber stability, the stretch of the jet was influenced, leading to an increase of mean nanofiber diameter (Fig. S5).

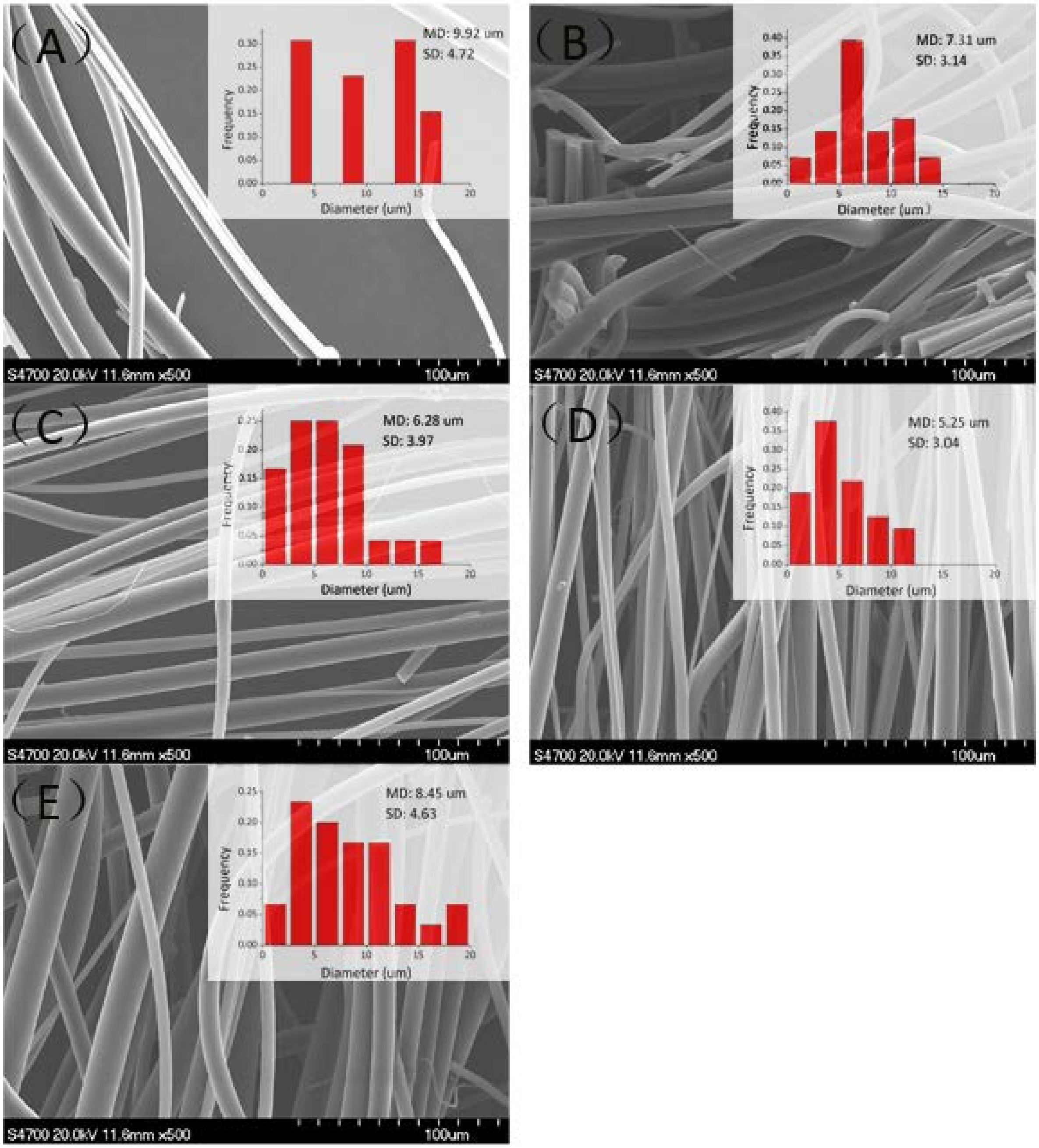

Figure S4 SEM images and fiber diameter distributions (A-E) at different spinning voltages: (A) 5 kV, (B) 15 kV, (C) 25 kV, (D) 35 kV, (E) 45 kV. Other spinning parameters remained stable: T=230°C, C=10cm, R=900 rpm.

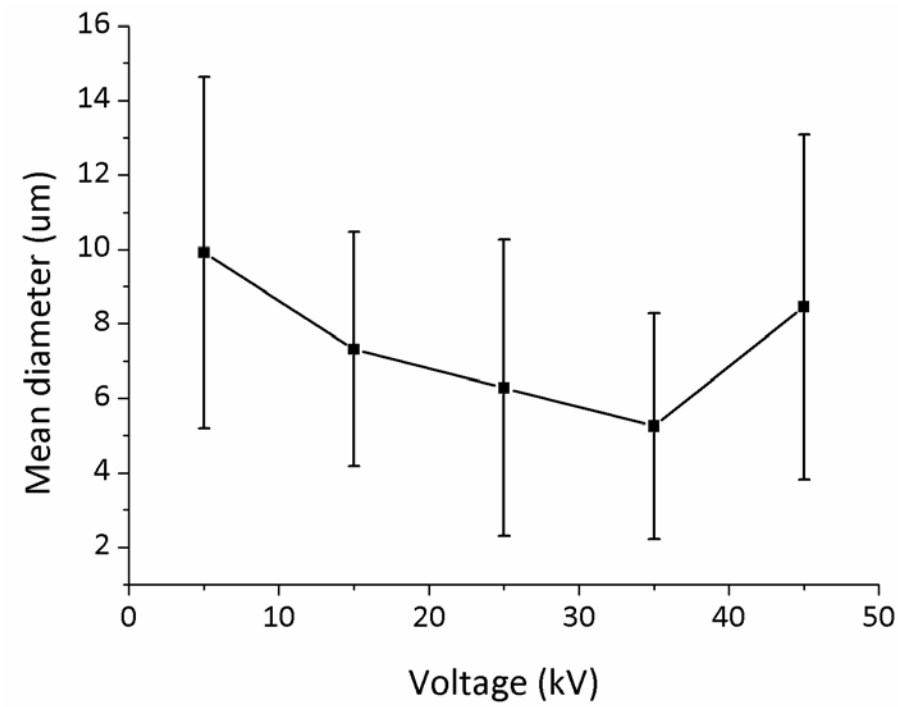

Figure S5 Relationship between spinning voltage and mean diameters of fibers.

2.2 Effects of temperature on fiber diameter

At temperatures below the minimum limit value of the melting point, a spinning jet could not be formed. With increasing temperature, raw material was fully molten and the jet was stretched out faster. Above the melting point, viscosity of spinning materials decreased with increasing temperature, allowing the jet to be easily stretched into ultra-fine fibers as shown in Fig. S6, S7. The initial jet and its stretch were undermined by the strong effect of quenching present due to the temperature difference between the inside and outside of the fiber.

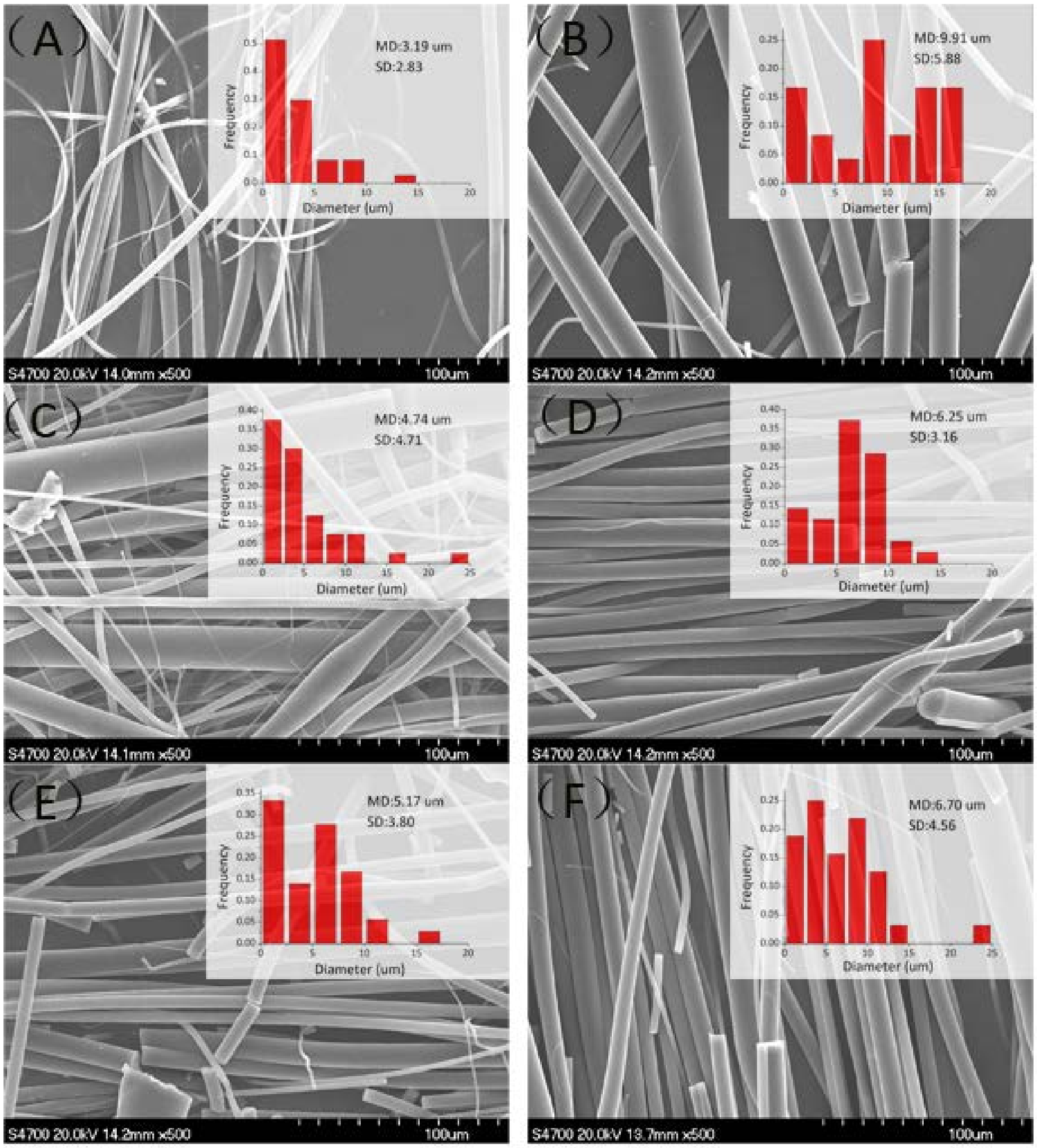

Figure S6 SEM images and fiber diameter distributions at different spinning temperature: (A) 215 °C, (B) 225 °C, (C) 235 °C, (D) 245 °C, (E) 255 °C, (F) 265 °C. Other spinning parameters remained stable: R=1020 rpm, C=10 cm, V=35 kV.

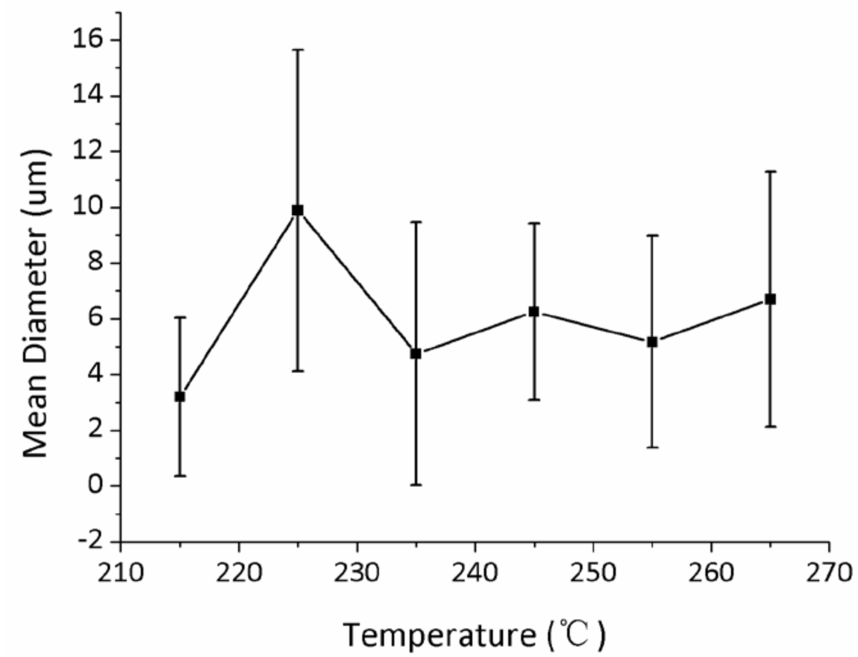

Figure S7 Relationship between spinning temperature and mean diameter of fibers.

2.3 Effects of rotational speed on the diameter of fibers

In the process of spinning, increasing rotational speed lead to an increase in jet speed and jet throw distance, with more serious floating of fibers. Experiment results showed an initial decrease in fiber diameter followed by progressive increase as shown in Fig. S8, S9. When the rotational airflow increased significantly, which acted as a countervailing power to centrifugal force, the jet dried faster and the jet walk path was more unstable.

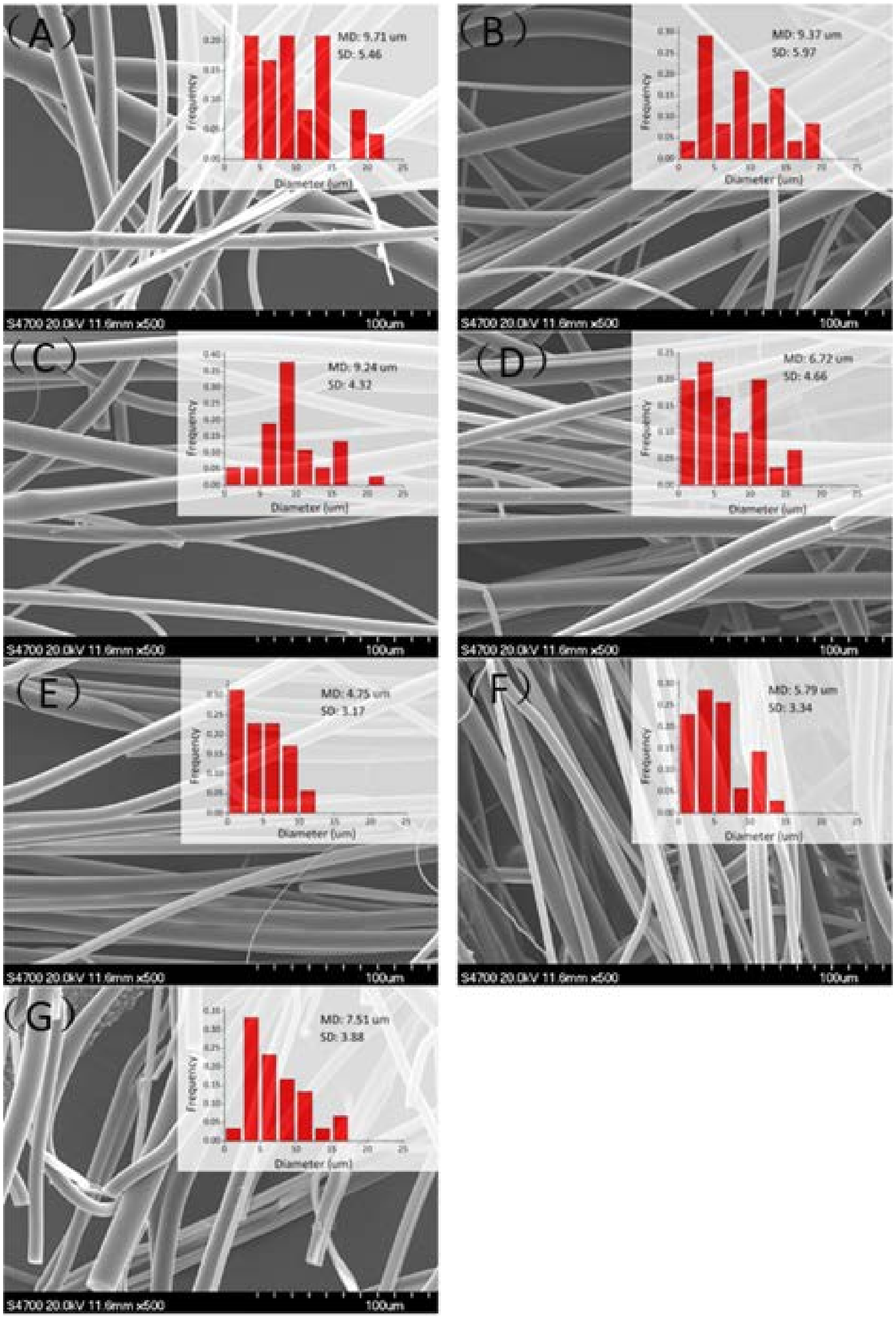

Figure S8 SEM images and fiber diameter distributions at different rotational speed: (A)780 rpm, (B)900rpm, (C)1020rpm, (D) 1140rpm, (E) 1260rpm, (F) 1380rpm, (G) 1500rpm. Other spinning parameters remained stable: V=30 kV, C=10 cm, T=230 °C.

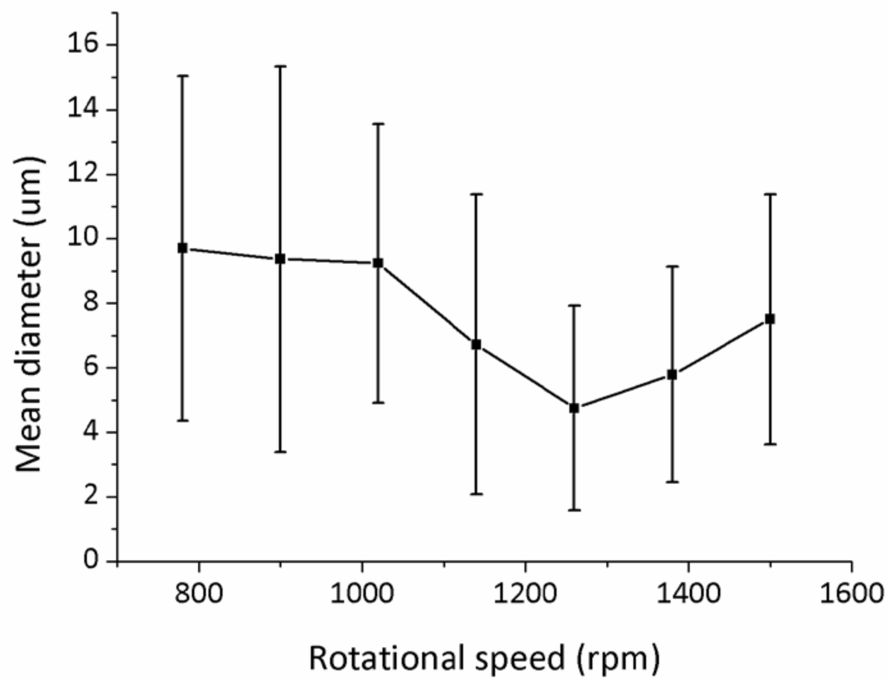

Figure S9 Relationship between rotational speed and mean diameter of fibers.

Additionally, we chose measured the diameter of 20 ultra-fine nanofibers prepared at different voltages. The mean diameter of the twenty ultra-fine nanofibers and the SEM images of ultra-fine nanofibers fabricated by CME method were shown in Fig. S10. The mean diameter decreased with the increase of voltage.

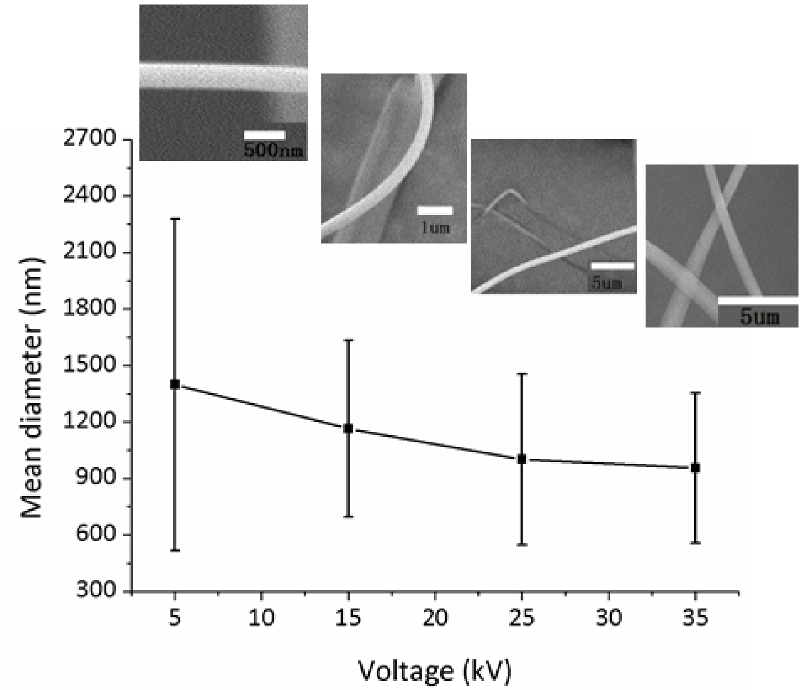

Figure S10 Relationship between spinning voltage and mean diameter of the obtained most ultra-fine nanofibers.

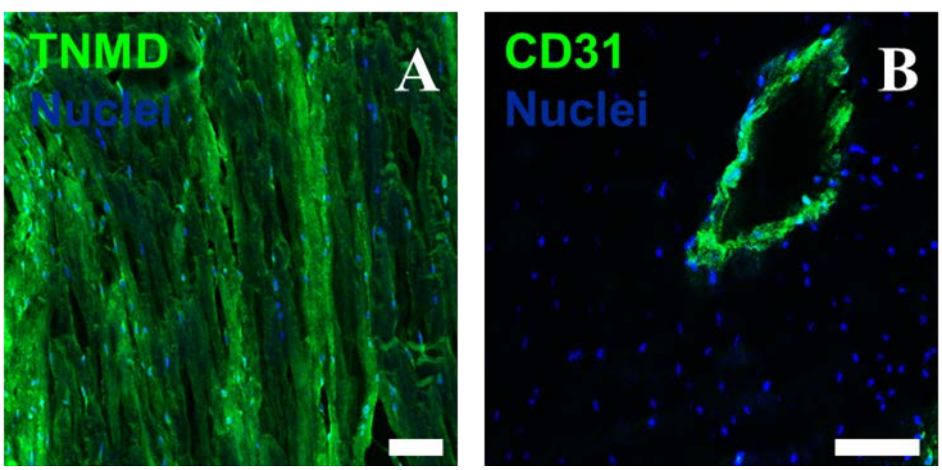

Figure S11 Immunofluorescent staining of (A) TNMD (green), nuclei (blue), and (B) CD31 (green), nuclei (blue) for cross-sectional native human tendon. Scale bars: 100 μm for (A) and (B).