Supporting Information

Influence of substituted aromatics on the formation and stability of β-sheet-based peptide hydrogels

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1. Peptide synthesis

Table S1. Code, sequence, chemical formula, molecular weight, HRMS results and yield of all synthesized peptides.

Code	Sequence	Chemical formula	Molecular weight TFA salt (g/mol)	HR-MS (ESI+), m/z		Yield (%)
				Calculated [M+H]+	Found [M+H]+	
SBL-HG-063	H-FQFQFK-NH_2	$C_{43}H_{58}N_{10}O_8$	1071.05	843.4512	843.4537	56
SBL-HG-092	H-FQFQF(4-NO_2)K-NH_2	$C_{43}H_{57}N_{11}O_{10}$	1116.04	888.4363	888.4321	38
SBL-HG-273	H-F(4-NO_2)QF(4-NO_2)QF(4-NO_2)K-NH_2	$C_{43}H_{55}N_{13}O_{14}$	1206.04	978.4064	978.4088	31
SBL-HG-095	H-FQFQF(4-CN)K-NH_2	$C_{44}H_{57}N_{11}O_8$	1096.06	868.4464	868.4423	22
SBL-HG-089	H-F(4-CN)QF(4-CN)QF(4-CN)K-NH_2	$C_{46}H_{55}N_{13}O_8$	1146.08	918.4369	918.4367	35
SBL-HG-224	H-FQFQF(4-CF_3)K-NH_2	$C_{44}H_{57}F_3N_{10}O_8$	1139.04	911.4386	911.4103	48
SBL-HG-220	H-F(4-CF_3)QF(4-CF_3)QF(4-CF_3)K-NH_2	$C_{46}H_{55}F_9N_{10}O_8$	1275.04	1047.4133	1047.3789	45
SBL-HG-211	H-FQFQF(4-OMe)K-NH_2	$C_{44}H_{60}N_{10}O_9$	1101.07	873.4618	873.4565	48
SBL-HG-090	H-F(4-OMe)QF(4-OMe)QF(4-OMe)K-NH_2	$C_{46}H_{64}N_{10}O_{11}$	1161.12	933.4829	933.4850	51
SBL-HG-088	H-FQFQYK-NH_2	$C_{43}H_{58}N_{10}O_9$	1087.04	859.4461	859.4445	63
SBL-HG-306	H-YQYQYK-NH_2	$C_{43}H_{58}N_{10}O_{11}$	1119.04	891.4359	891.4398	45

2. UV-absorbance

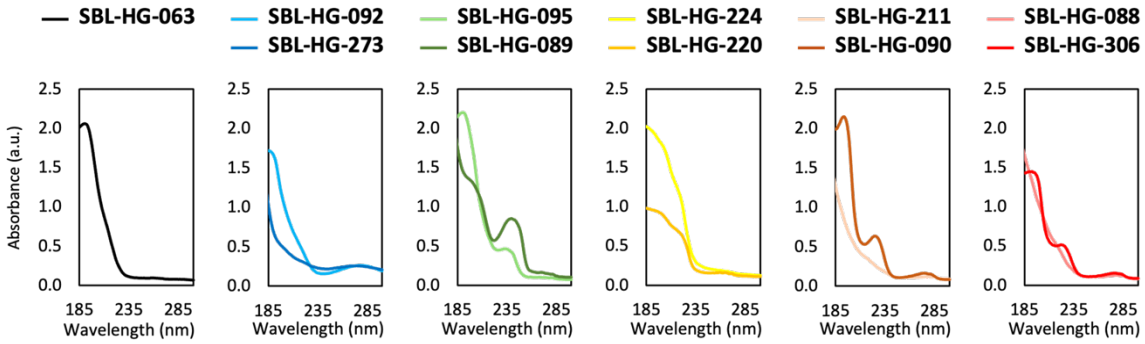


Figure S1. UV-absorbance spectra for the 2% w/v hydrogels in physiological saline in the spectral range of 185-300 nm.

3. Rheology

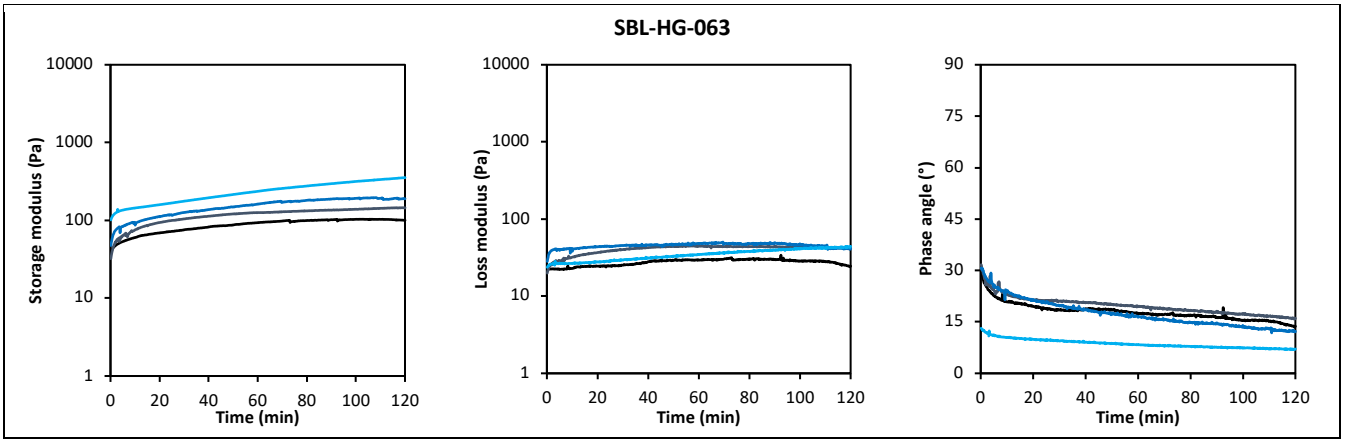


Figure S2. Dynamic rheology of **SBL-HG-063** with storage modulus (left), loss modulus (middle) and phase angle (right) plotted as a function of time. The measurement was performed at 37°C, with a strain of 0.5% and oscillation frequency of 0.15 Hz. Colour shades represent replicate measurements.

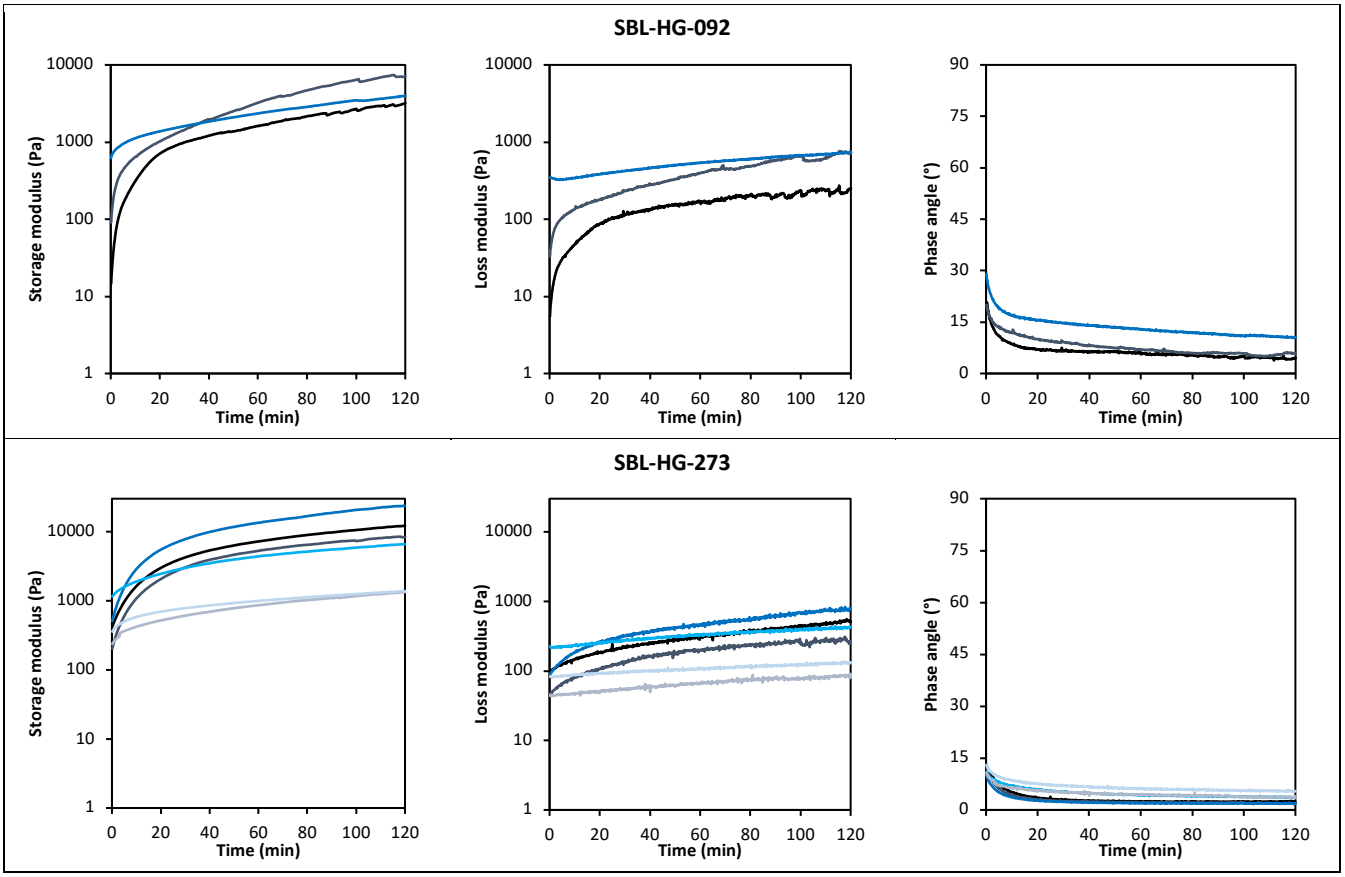


Figure S3. Dynamic rheology of **SBL-HG-092** and **SBL-HG-273** with storage modulus (left), loss modulus (middle) and phase angle (right) plotted as a function of time. The measurement was performed at 37°C, with a strain of 0.5% and oscillation frequency of 0.15 Hz. Colour shades represent replicate measurements.

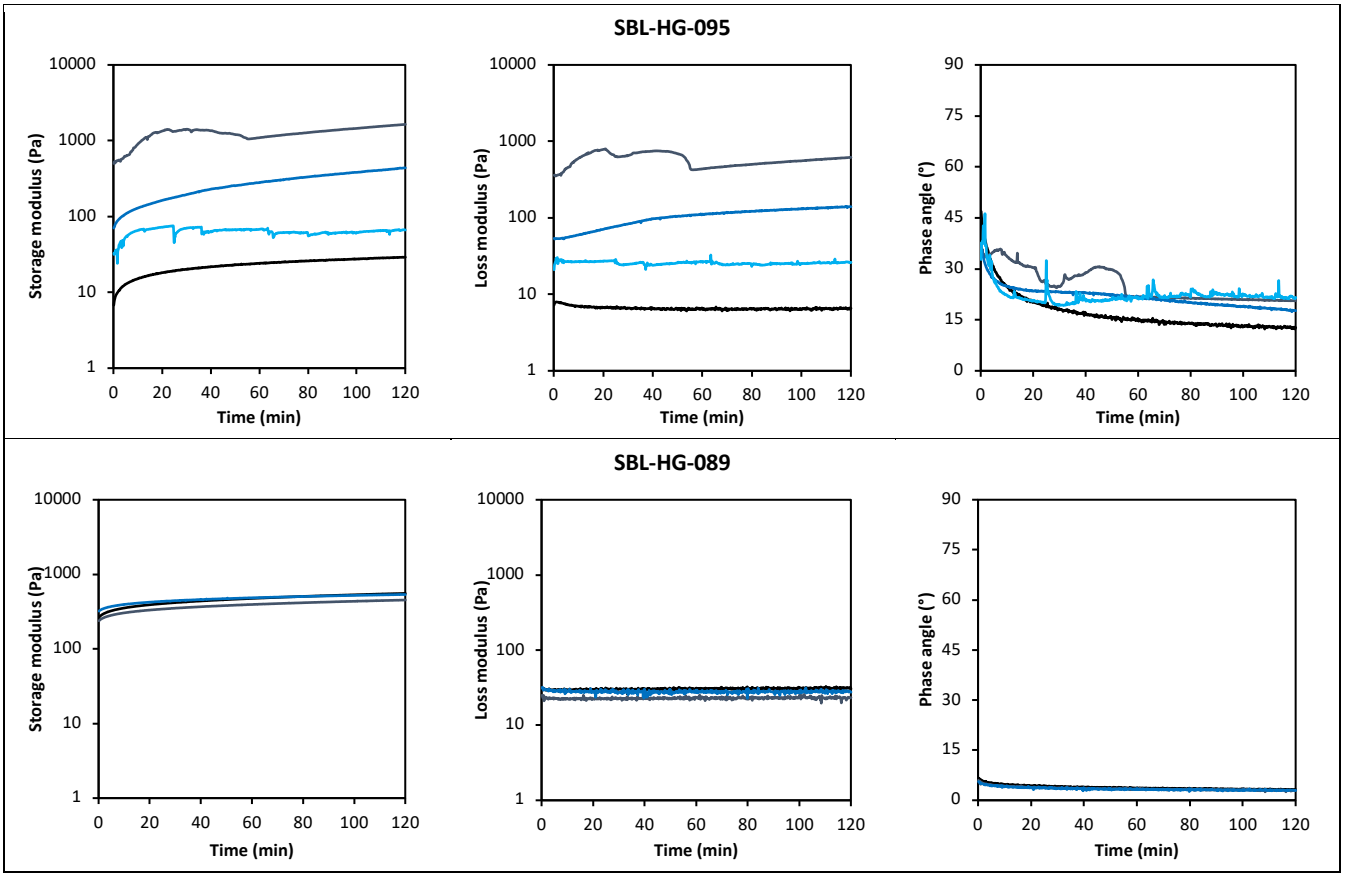


Figure S4. Dynamic rheology of **SBL-HG-095** and **SBL-HG-089** with storage modulus (left), loss modulus (middle) and phase angle (right) plotted as a function of time. The measurement was performed at 37°C, with a strain of 0.5% and oscillation frequency of 0.15 Hz. Colour shades represent replicate measurements.

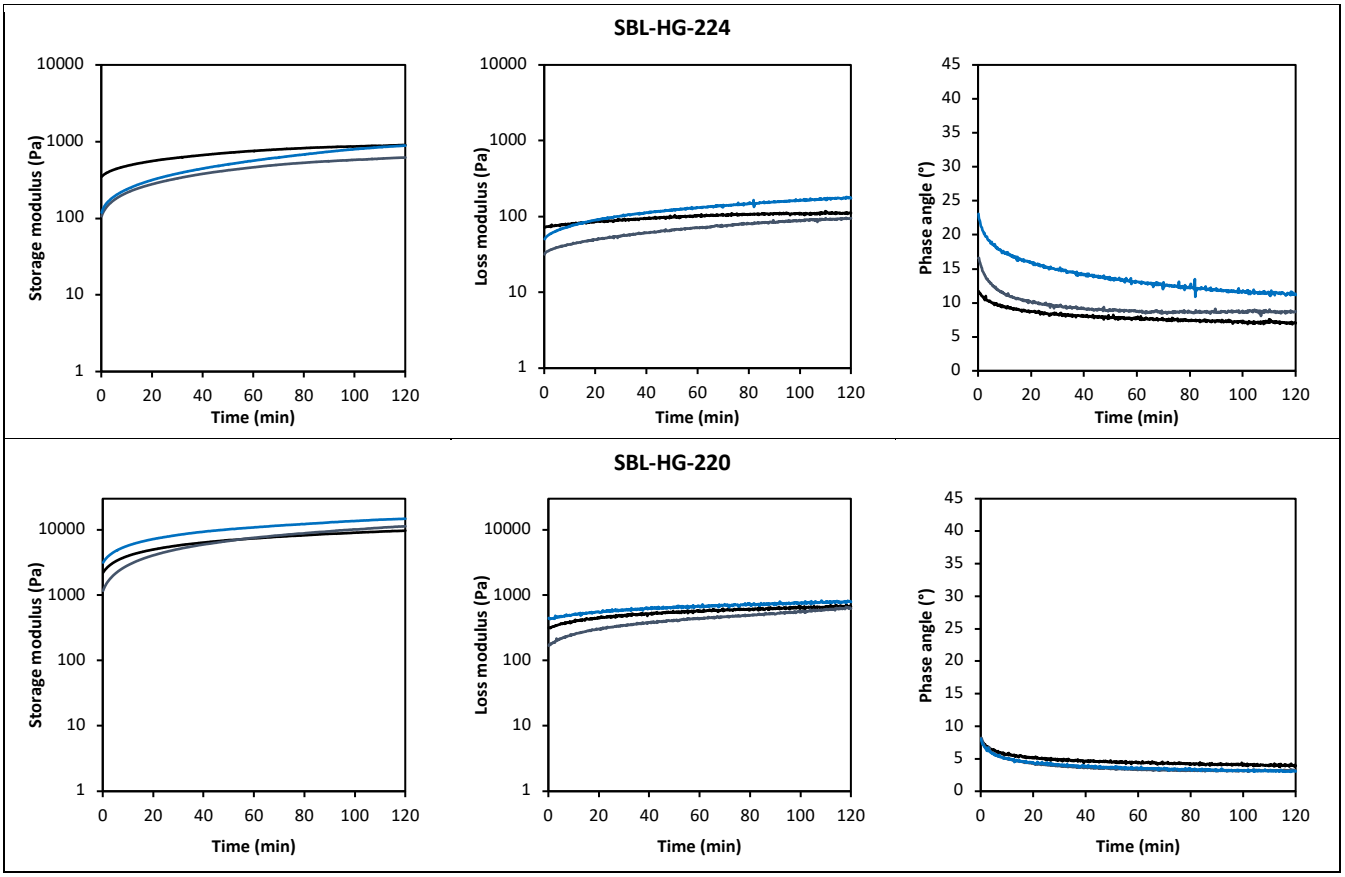


Figure S5. Dynamic rheology of **SBL-HG-224** and **SBL-HG-220** with storage modulus (left), loss modulus (middle) and phase angle (right) plotted as a function of time. The measurement was performed at 37°C, with a strain of 0.5% and oscillation frequency of 0.15 Hz. Colour shades represent replicate measurements.

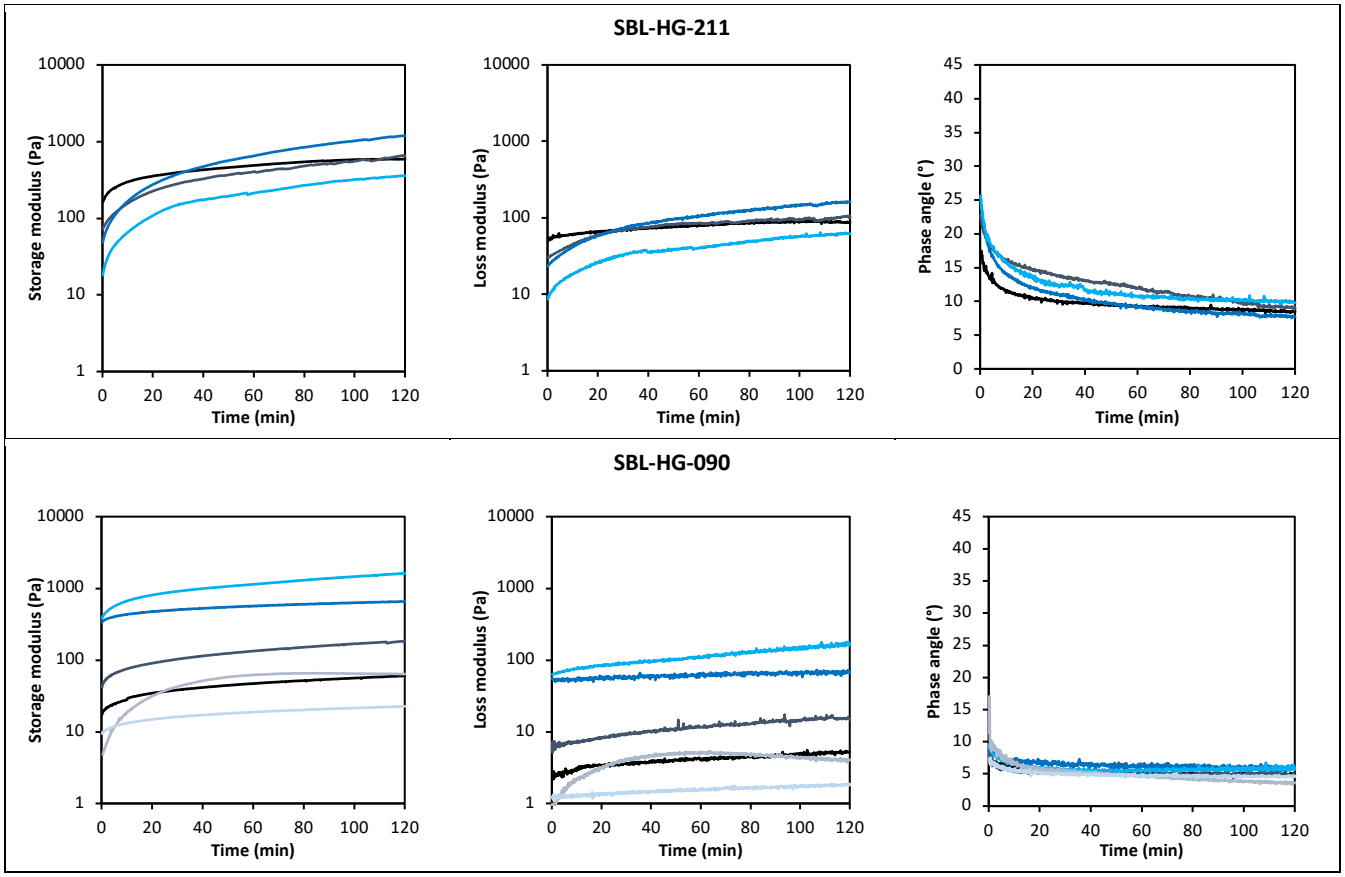


Figure S6. Dynamic rheology of **SBL-HG-211** and **SBL-HG-090** with storage modulus (left), loss modulus (middle) and phase angle (right) plotted as a function of time. The measurement was performed at 37°C, with a strain of 0.5% and oscillation frequency of 0.15 Hz. Colour shades represent replicate measurements.

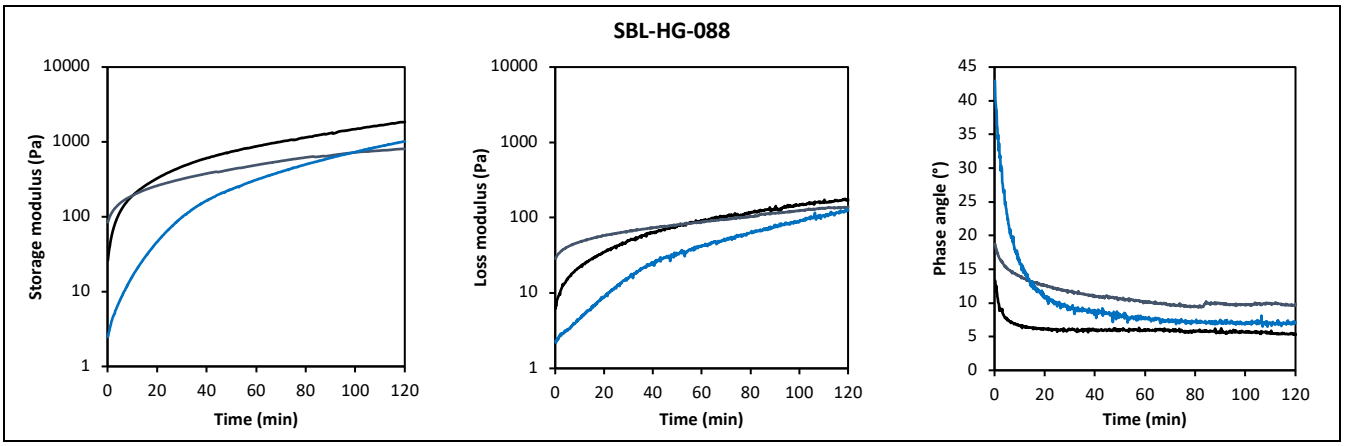
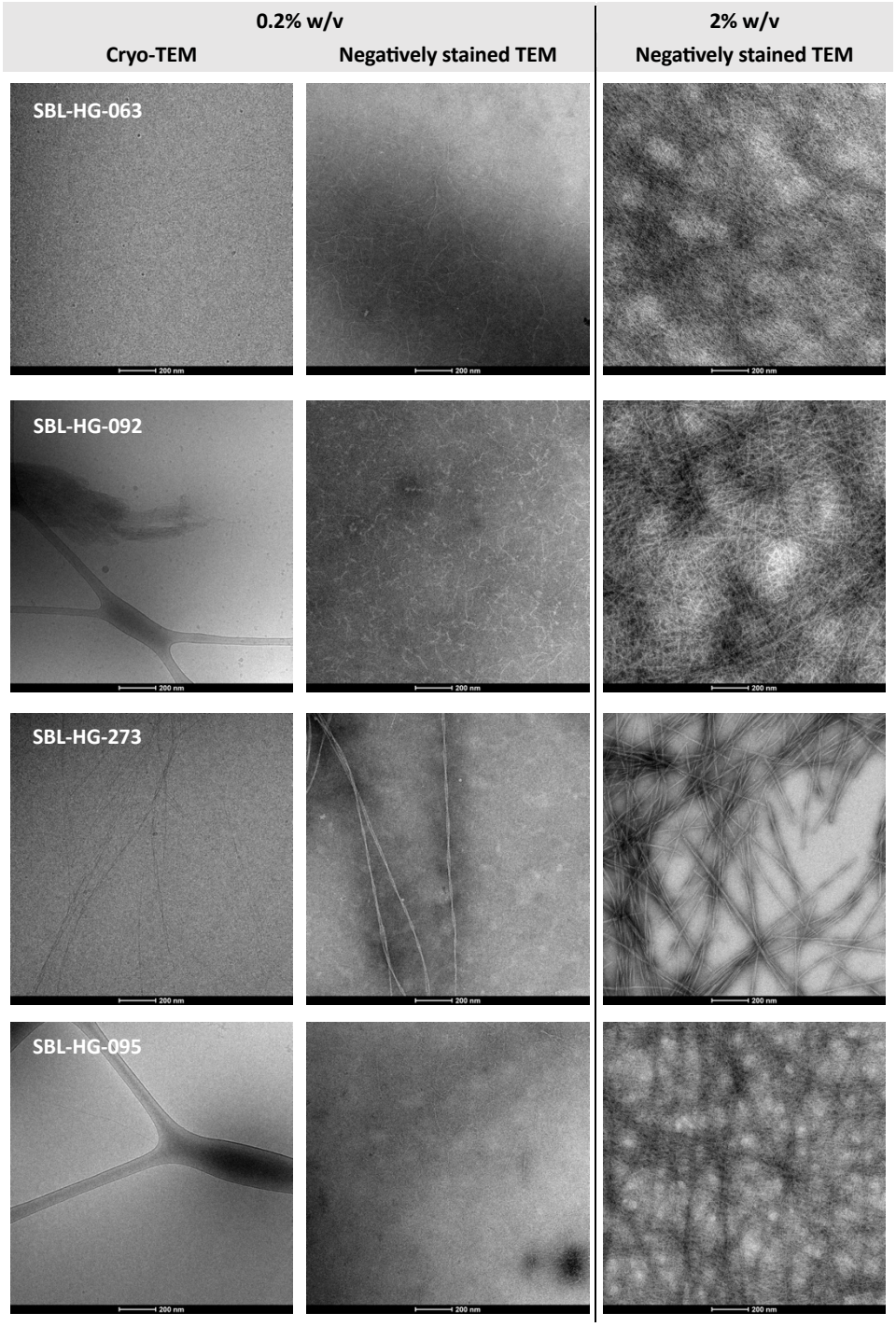
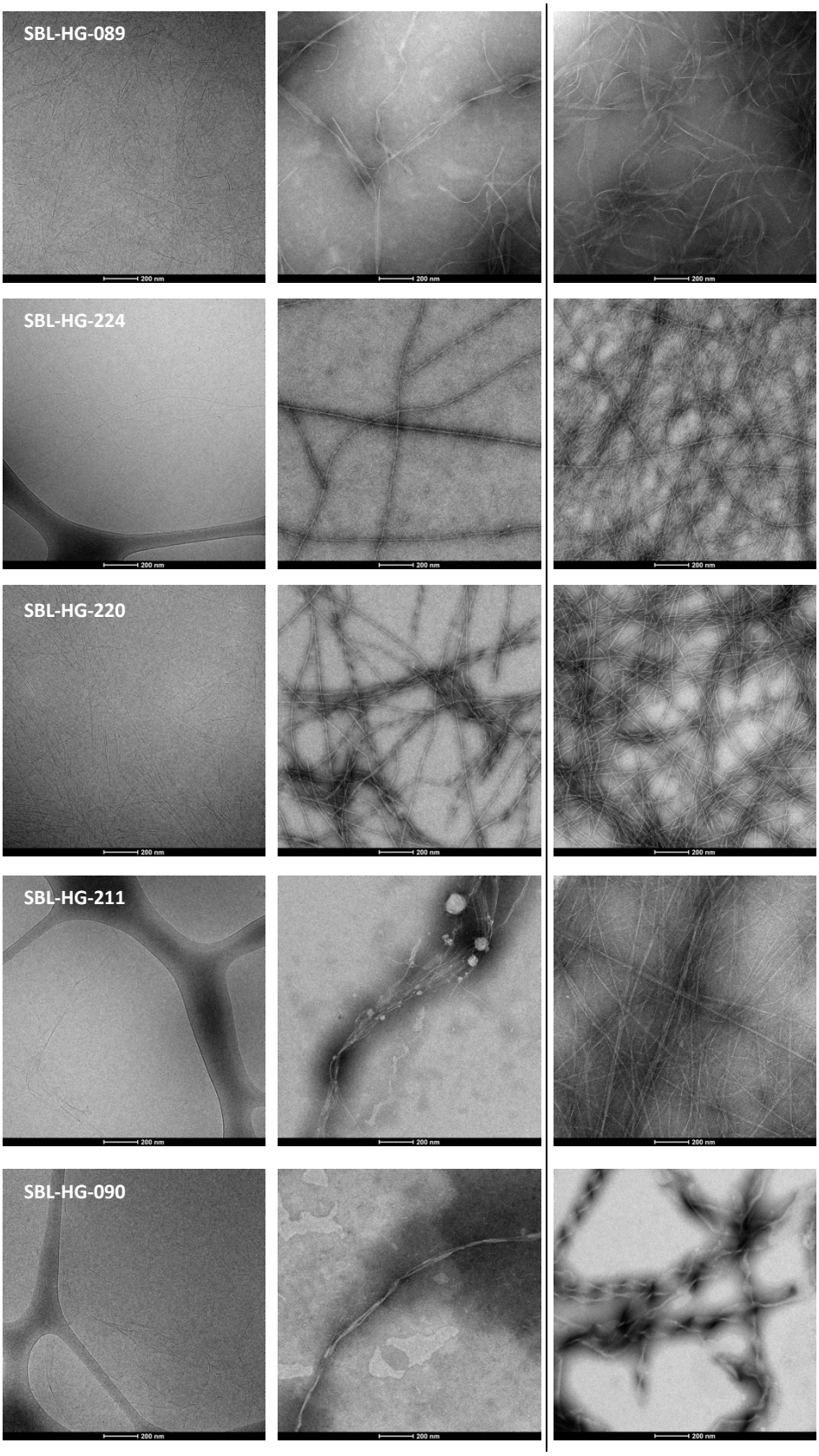


Figure S7. Dynamic rheology of **SBL-HG-088** with storage modulus (left), loss modulus (middle) and phase angle (right) plotted as a function of time. The measurement was performed at 37°C, with a strain of 0.5% and oscillation frequency of 0.15 Hz. Colour shades represent replicate measurements.

4. Microscopy

4.1. Electron microscopy




SBL-HG-089
SBL-HG-224
SBL-HG-220
SBL-HG-211
SBL-HG-090

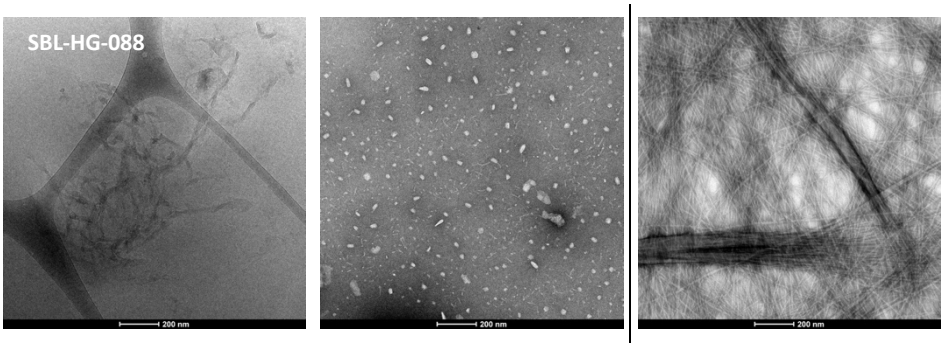


Figure S8. Additional cryo-TEM and negatively stained TEM images. Scale bars represent 200 nm.

4.1. Atomic force microscopy

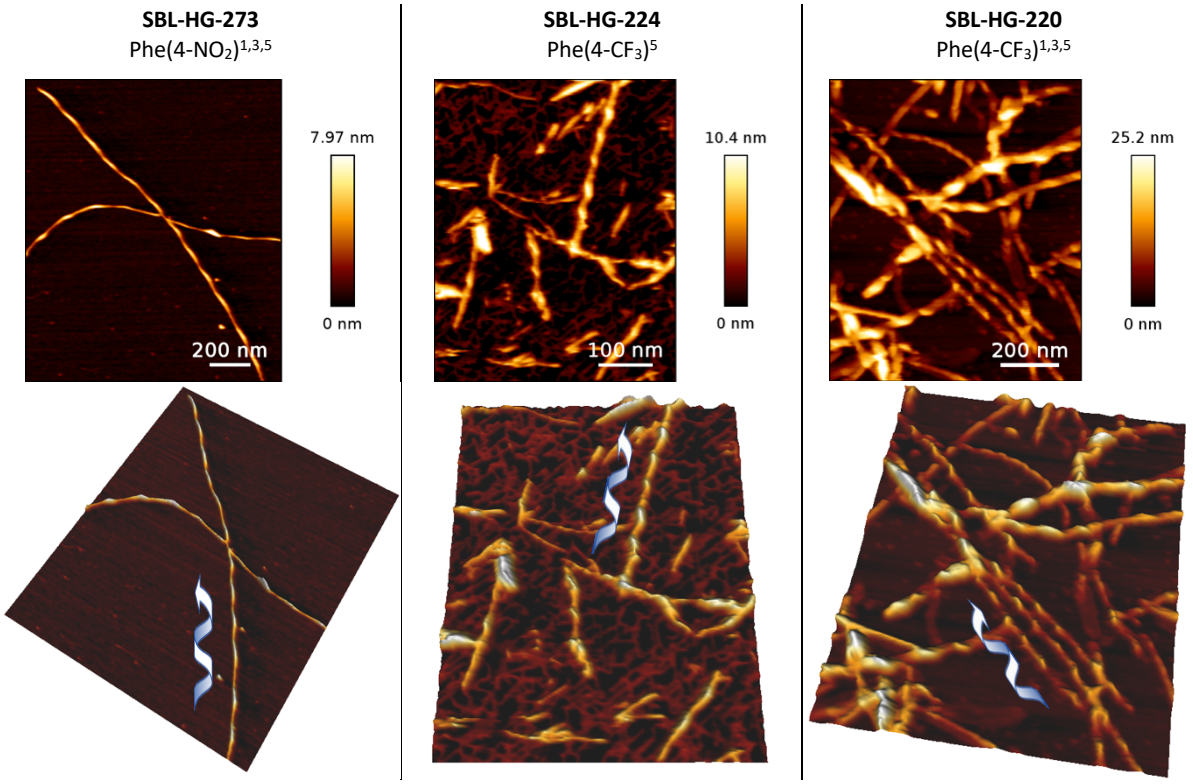


Figure S9. AFM height images (2D and 3D) of type 2 hydrogels at 0.2% w/v in physiological saline. The twisting handiness is indicated on the 3D images.

4.2. Fibril dimensions

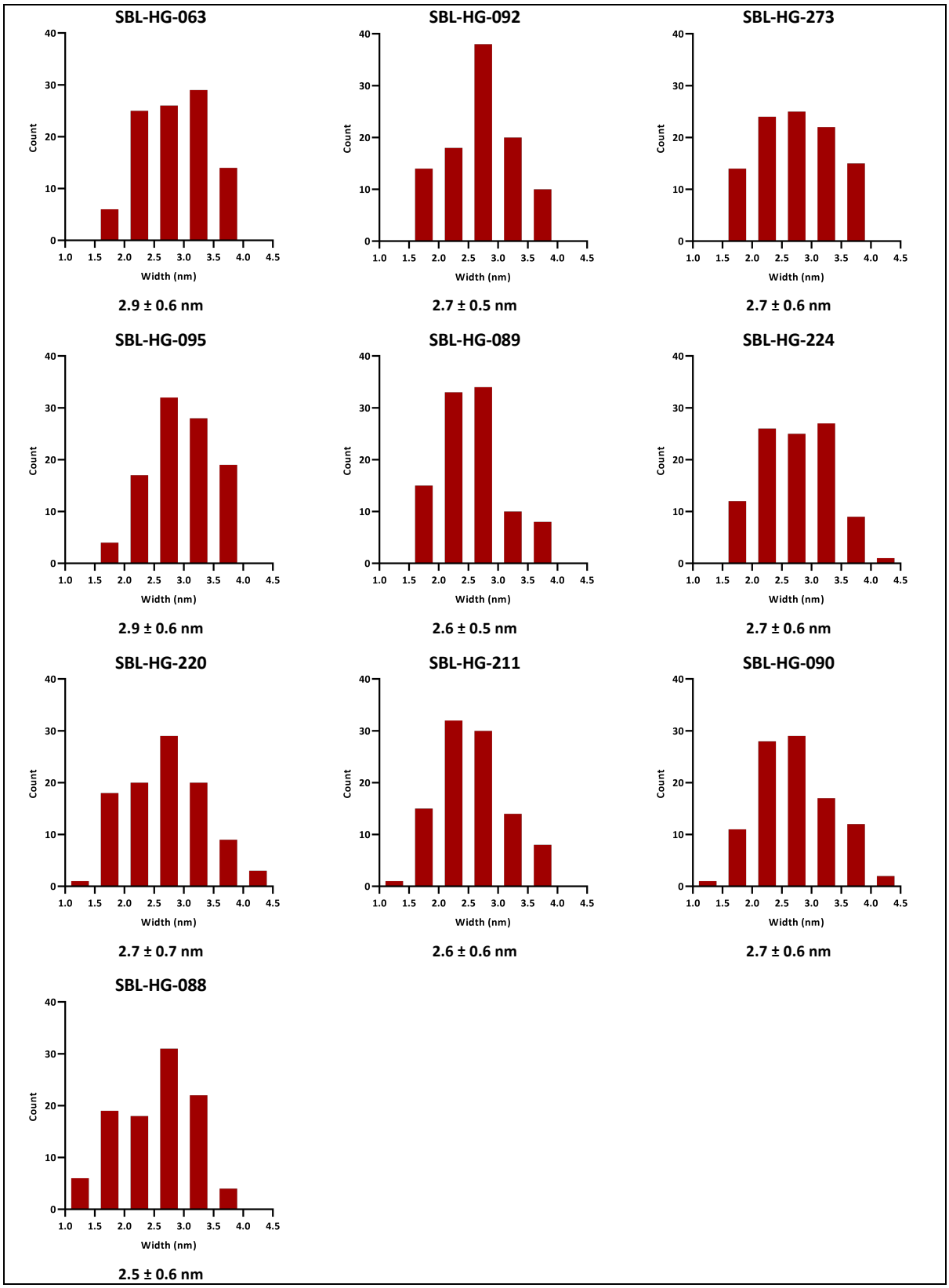


Figure S10. Protofibril widths measured from the 0.2% w/v TEM images of all 10 peptides, mean ± SD shown beneath each histogram, n=100.

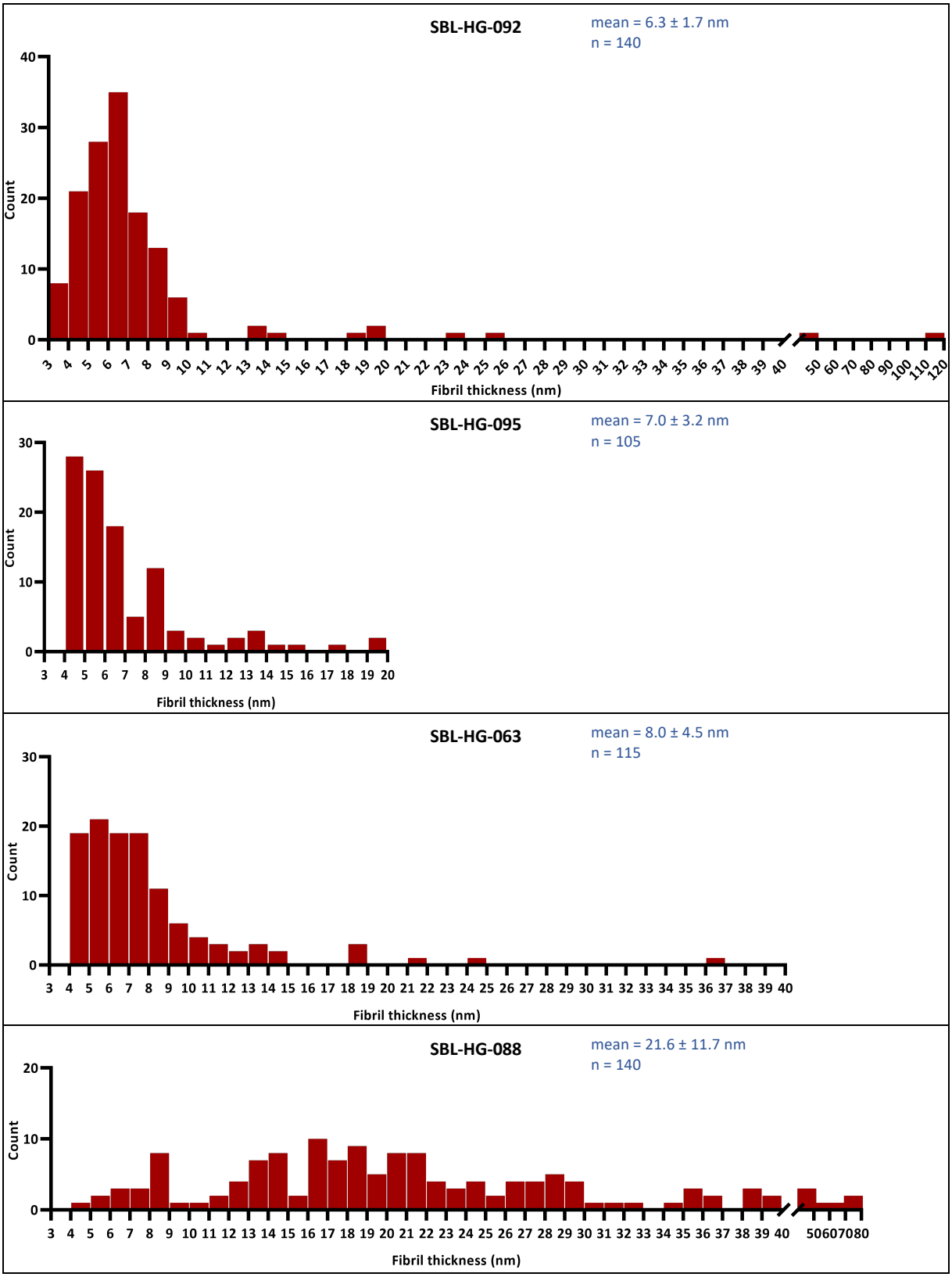


Figure S11. Fibril thickness of **SBL-HG-092**, **SBL-HG-095**, **SBL-HG-063** and **SBL-HG-088** with predominantly type 1 fibril networks. Measurements deducted from TEM images at 0.2% w/v, mean ± SD shown for each histogram.

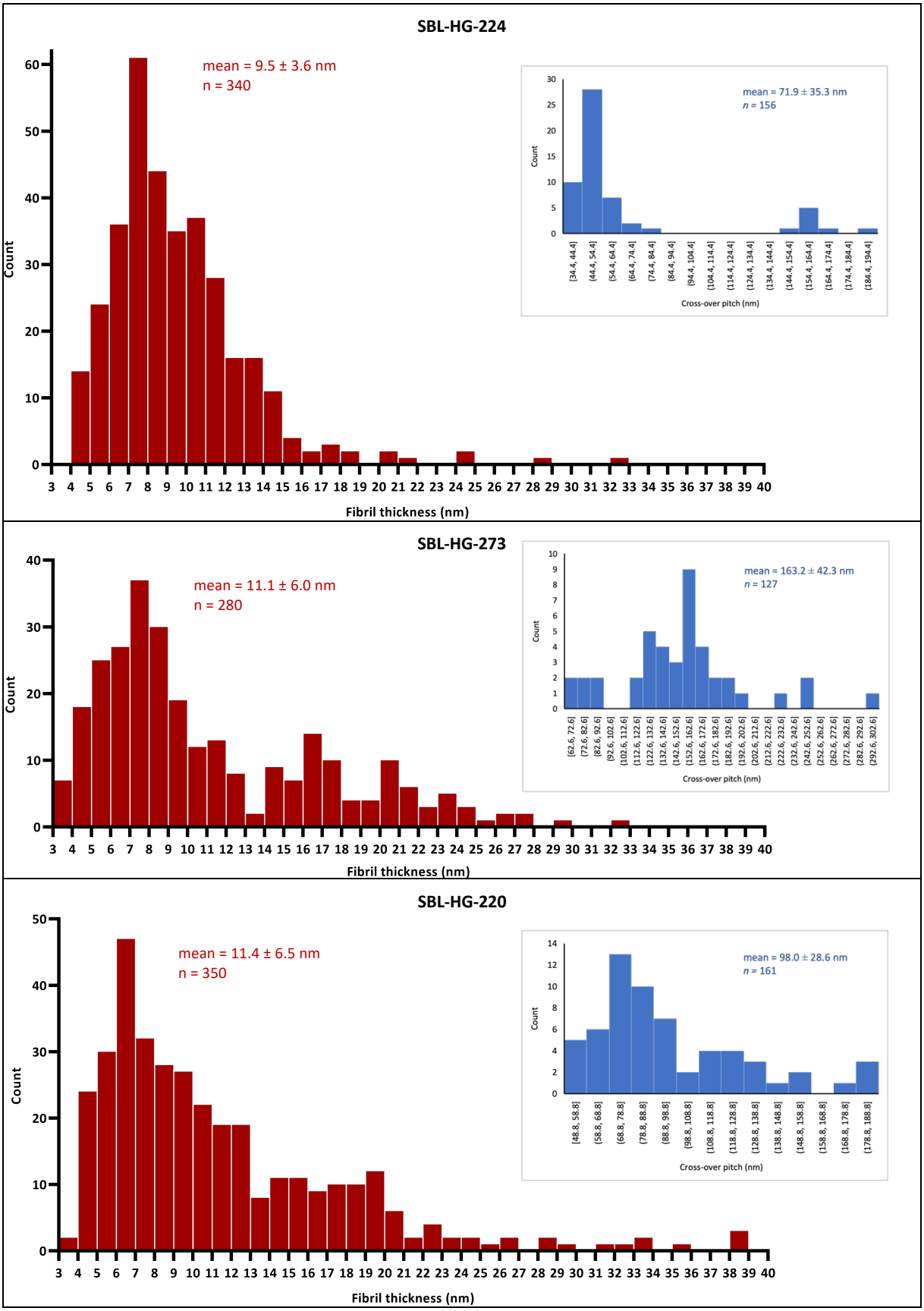


Figure S12. Fibril thickness (red) and cross-over pitch (blue) of **SBL-HG-224**, **SBL-HG-273** and **SBL-HG-220** with predominantly type 2 fibril networks. Measurements deducted from AFM and TEM images at 0.2% w/v, mean ± SD shown for each histogram. For the dimensions on the AFM images, maximal heights of fibril cross-sections were employed.

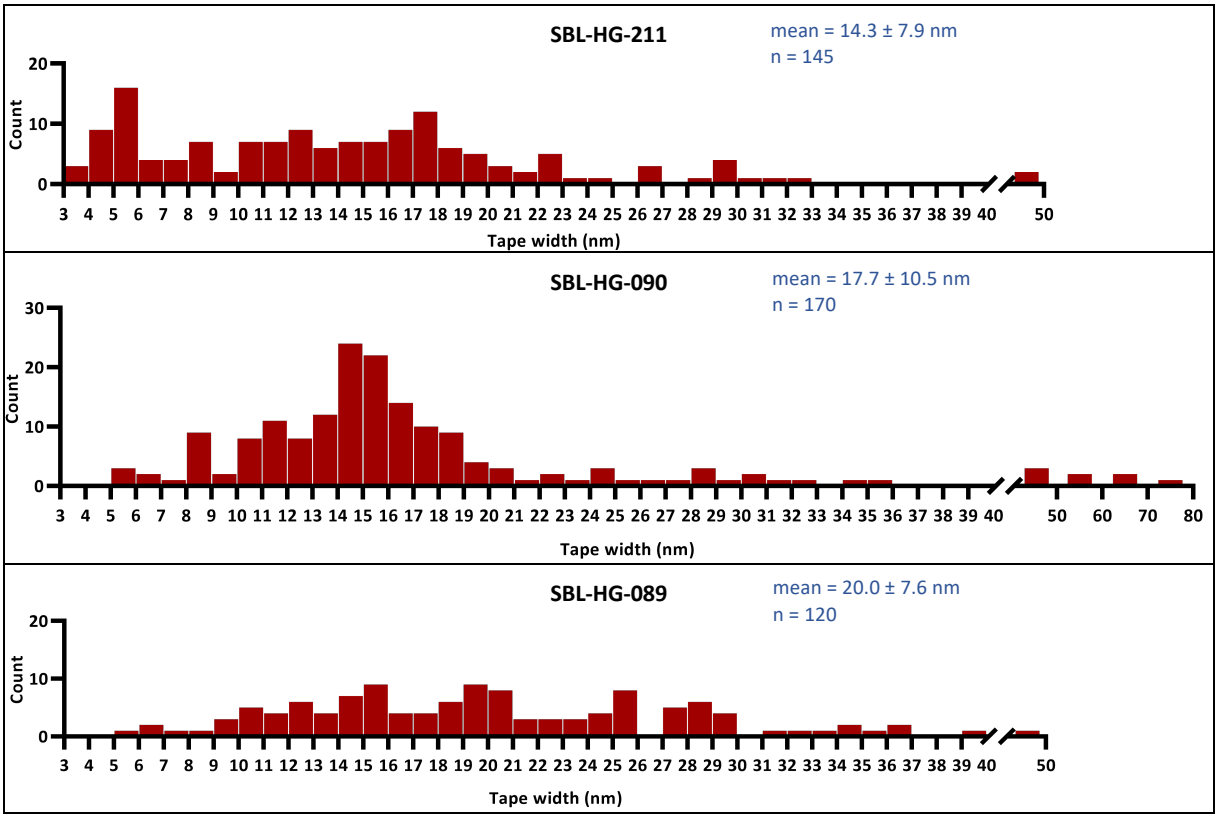


Figure S13. Tape width of **SBL-HG-211**, **SBL-HG-090** and **SBL-HG-089** with predominantly type 3 fibril networks. Measurements deducted from TEM images at 0.2% w/v, mean ± SD shown for each histogram.