Electronic Supplementary Material (ESI) for Materials Advances.

Supporting Information

Towards the Understanding of Halogenation in Peptide Hydrogels: a Quantum Chemical Approach

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This PDF file includes:

I. Benchmarking of quantum mechanical methods for non-covalent interactions in halogenated stacked dimers

Table S1. Comparison of the interaction energies computed with different quantum mechanical methods for a set of stacked Phe···Phe dimers.[a]

System	CCSD(T)-F12b	M06	CCSD-F12b	DF-MP2	DF-SCS-MP2-F12
H	-5.88	-4.43	-4.05	-9.78	-5.23
***o*F**	-6.12	-4.53	-4.09	-10.56	-5.56
***m*F**	-5.76	-4.35	-3.83	-9.91	-5.18
***p*F**	-5.89	-4.50	-3.90	-10.26	-5.40
***o*Cl**	-6.48	-4.86	-4.32	-11.32	-5.95
***m*Cl**	-6.47	-4.88	-4.24	-11.43	-5.93
***p*Cl**	-6.40	-5.04	-4.28	-11.20	-5.91
MAE	-	1.49	2.04	4.50	0.55
RMSE	-	2.22	4.17	20.36	0.30

[a] The interaction energies for explicitly-correlated methods were computed with the aug-cc-pVDZ for all atoms, whereas the M06 energies were evaluated with the aug-cc-pVTZ basis set. All these energies were obtained from single-point calculations on the optimized geometries at the M06/cc-pVTZ level of theory. No counterpoise corrections were considered in the benchmark.

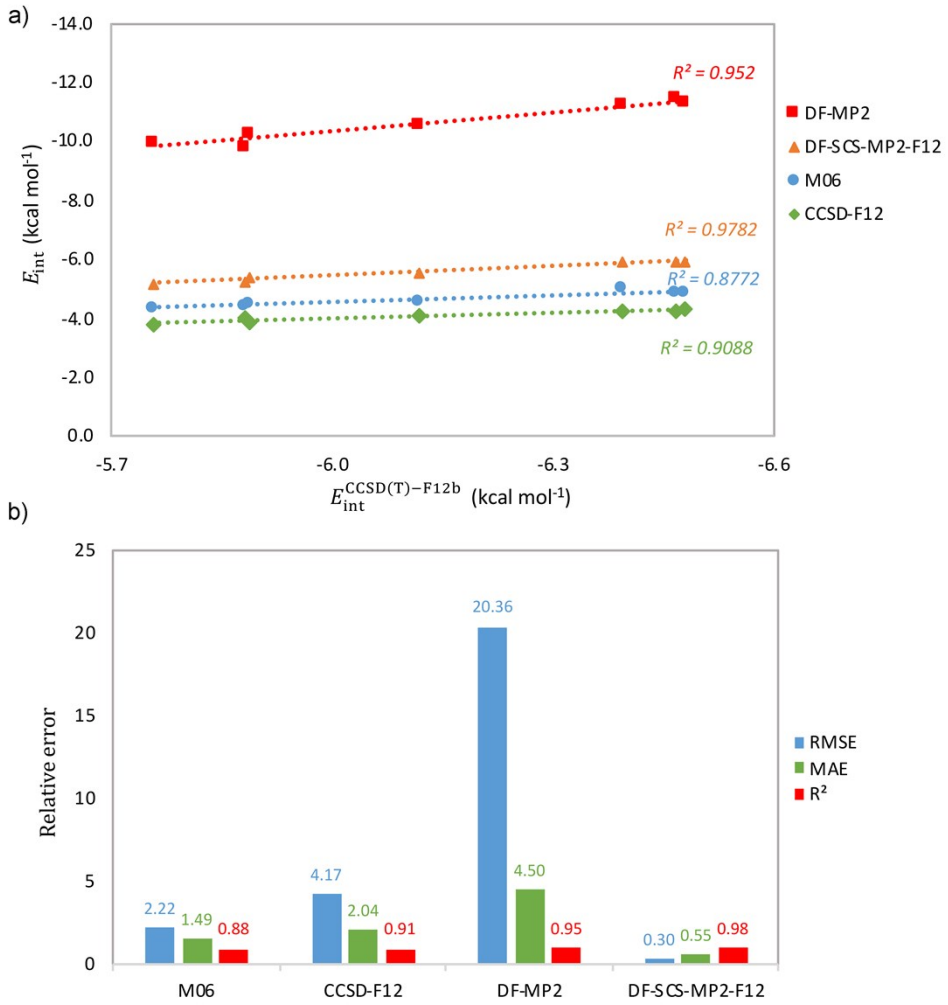


Figure S1. Mean absolute error (MAE) and root-mean-square error RMSE (in kcal mol^{-1}) relative to CCSD(T)-F12 for describing the interaction energies of selected stacked Phe···Phe dimers. The correlation coefficient (R^2) for the different methods is also shown.

II. Systematic search over stacked dimer configurations

Table S2. Relative electronic energies (in kcal mol^{-1}) for different stacked dimer configurations.

Conf.	E_{rel} [a]	E_{rel}^{CP} [b]	E_{rel} [a]	E_{rel}^{CP} [b]	E_{rel} [a]	E_{rel}^{CP} [b]	E_{rel} [a]	E_{rel}^{CP} [b]
	H		***o*F**		***mF***		***p*F**	
1	0.56	0.88	0.21	0.12	1.34	0.65	0.51	0.21
2	0.55		0.57	**0.00**	0.32	0.00	0.00	**0.00**
3	0.57		0.30	0.19	0.00	0.29	0.92	1.06
4	0.00	0.57	0.35	0.30	0.88	0.36	0.34	0.24
5	0.05	**0.00**	0.00	0.19	0.41	0.30		
6	0.52	0.31	0.34	0.20	1.49	0.75		
	o*F*o		***o*F*m***		***o*F*p***		***m*F*m***	
1	0.96		0.78		0.00	0.18	1.02	
2	0.54		0.16	**0.00**	0.12	**0.00**	0.88	
3	2.01		0.21		0.95		0.53	0.98
4	0.53		0.00	0.12	0.68		0.96	
5	1.33		0.54		0.26	0.52	0.00	**0.00**
6	0.19	**0.00**	1.18		0.84		1.15	
7	0.80		1.14				0.65	0.41
8	0.74		0.89				1.61	
9	0.51	0.46	0.34				0.56	
10	0.58		0.68				0.68	
11	0.00	0.05	0.14	0.28			0.78	
12	0.92		0.58				1.43	
	m*F*p		***p*F*p***					
1	0.48		1.11					
2	0.69		0.81					
3	0.31		0.84					
4	0.19	0.66	0.00	**0.00**				
5	0.00	**0.00**	0.49	0.44				
6	0.28	0.56	0.75	0.08				

[a] Relative energies computed from the ZPVE-corrected electronic energies at the M06/cc-pVTZ level of theory. [b] For the most stable dimer configurations, the relative energies were recomputed at the M06/aug-cc-pVTZ level of theory including the Counterpoise correction.

III. Mutual relationships between interaction energies, integrated volumes of the NCI regions and intermonomer distances

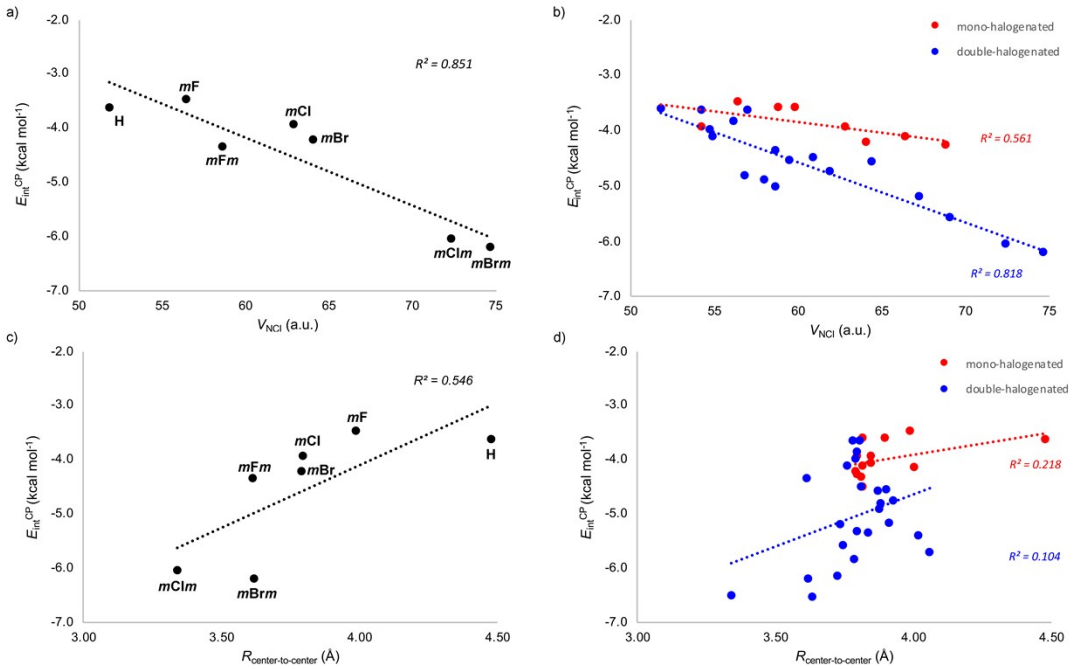


Figure S2. Correlation between the counterpoise-corrected interaction energies and the integrated volume of the NCI region and the centroid-centroid distance, respectively. a) and c) show the correlations for *meta-substituted* Phe···Phe dimers, whereas b) and d) correspond to the full set of dimers as a function of the number of heteroatoms.

IV. Energy decomposition analysis for the full set of mono- and double-halogenated dimers

Table S3. Energy decomposition analysis on the mono- and double-halogenated dimers. The energy components (in kcal mol^{-1}) correspond to electrostatic interactions (ΔE_{elst}), orbital interaction (ΔE_{oi}) and Pauli repulsion (ΔE_{Pauli}) between deformed monomers.[a]

System	ΔE_{elst}	ΔE_{Pauli}	ΔE_{oi}	System	ΔE_{elst}	ΔE_{Pauli}	ΔE_{oi}
H	-15.3	40.4	-27.9	***o*F*o***	-17.12	48.92	-34.37
***o*F**	-16.1	44.3	-31.0	***o*F*m***	-18.41	50.62	-35.55
mF	-16.4	49.6	-36.4	***o*F*p***	-16.74	46.56	-32.86
***p*F**	-16.2	46.1	-32.8	***m*F*m***	-20.93	50.62	-33.63
***o*Cl**	-17.0	45.9	-32.1	***m*F*p***	-18.46	47.18	-32.34
***m*Cl**	-19.0	53.7	-38.4	***p*F*p***	-14.06	38.34	-26.83
***p*Cl**	-16.9	47.8	-34.2	***o*Cl*o***	-18.27	51.82	-37.61
***o*Br**	-17.2	46.5	-32.6	***o*Cl*m***	-17.63	46.91	-33.38
***m*Br**	-18.8	52.7	-37.7	***oClp***	-17.56	49.25	-35.91
***p*Br**	-16.7	43.5	-30.7	***m*Cl*m***	-24.28	60.53	-41.79
***o*I**	-15.7	42.9	-30.6	***m*Cl*p***	-20.04	51.72	-36.05
***m*I**	-19.1	54.5	-39.3	***p*Cl*p***	-17.18	48.84	-35.02
***p*I**	-17.5	48.5	-35.1	***o*Br*o***	-17.41	47.38	-34.25
				o*Br*m	-17.40	46.09	-33.04
				o*Br*p	-19.60	52.99	-38.10
				m*Br*m	-24.89	61.48	-42.44
				m*Br*p	-20.58	52.12	-36.77
				p*Br*p	-15.74	44.87	-33.42
				o*I*o	-19.69	54.55	-39.71
				o*I*m	-23.76	59.65	-41.81
				o*I*p	-17.62	46.25	-33.61
				m*I*m	-25.49	63.79	-44.57
				m*I*p	-21.40	54.97	-39.18
				p*I*p	-14.87	31.74	-21.64

V. Material and reagents

All the amino acids (Fmoc protected), the rink amide resin (loading 0.47 mmol/g) and the coupling reagent (HBTU) were purchased from IRIS Biotech GMBH. The reagents 4-methylpiperidine, diisopropylethylamine, triisopropylsilane and dimethyl sulfoxide (DMSO) were obtained from Sigma-Aldrich. Trifluoroacetic acid (TFA) was purchased from Fluorochem. The solvents *N*,*N*-dimethylformamide 99.5% (DMF) and methanol 99.8% (MeOH) were purchased from Acros Organics, while the solvents dichloromethane analytical grade (DCM) and the acetonitrile HPLC gradient grade were purchased from Fisher Chemical. The Milli-Q water was obtained after purification through a Millipore Simplicity UV system.

VI. Experimental methods

Peptide synthesis and purification

Peptides were synthesized manually using standard Fmoc-strategy solid-phase peptide synthesis (SPPS) on Rink amide resin. All couplings were performed using 3 equivalents (equiv.) of Fmoc-protected amino acids, 3 equiv. of coupling reagent (HBTU) and 5 equiv. of DIPEA in DMF for 45 min. Fmoc deprotection was carried out by treatment of the resin with 20% 4-methylpiperidine in DMF for 5 and 15 min. After completion of the sequence, the peptide was cleaved from the resin and the protecting groups were concomitantly removed by using a mixture of TFA/TIS/H_2O (95:2.5:2.5) for 4 h at room temperature.

The crude peptides were purified by preparative reverse phase high-performance liquid chromatography (Gilson Middleton system with Gilson 322 pumps, controlled by the software package Unipoint) using water (containing 0.1% TFA) and acetonitrile (containing 0.1% TFA) as eluents. The pure peptides (>95% purity) were obtained after lyophilization and were characterized by electrospray ionization mass spectrometry on a Micromass Q-Tof Micro spectrometer.

Peptide gelation

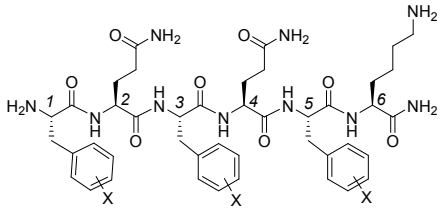
The peptide gelation was performed by dissolving the TFA salt of the hydrogelator (6 mg) in 300μL of buffer solution (PBS). After successive vortexing and sonication, the formed hydrogel was left to rest overnight at room temperature.

Dynamic rheometry

By using dynamic rheometry, the G’ and G” are determined. The G' refers to the elastic modulus and reflects elastic behavior of a material when deformed. The G" refers to the viscous modulus, which reflects the flow of a material while it is deformed. Mechanical properties of the hydrogels, prepared as described above, were evaluated at 34°C (AR-G2 Rheometer from TA Instruments (Delaware, USA) 24h after gelation, samples were transferred with a spatula onto a profiled cross hatched 15 mm parallel plate geometry, after which the gap height was adjusted to 500 nm. Water evaporation was minimized by covering the sample edges with low viscous mineral oil (Sigma Aldrich, BioReagent, for molecular biology). After loading, the stabilization of the gels was determined with a first 2h time sweep (frequency of 1.6Hz). Frequency sweep tests with the frequency ranging from 0.01 to 10Hz were conducted at a strain within the linear viscoelastic region (1% strain). Followed by a strain sweep performed starting from 0.1% to 100%. Then the recovery of samples was evaluated by the implementation of a dynamic shear step, which consisted of the application of 500% oscillatory strain during 5 s (frequency of 1.6Hz), after which the restructuration of the gels was evaluated by returning to analytical conditions during a 2h time sweep (frequency of 1.6Hz). The final G’ were obtained by taking the average of the G’ value during the frequency sweep at 1.6Hz for 3 measurements. The experimental data of the frequency test for all investigated peptide hydrogelators (measured in triplicate) is included at the end of this supporting information.

VII. Peptide characterization

Table S4. Compound number, sequences and characterization of the halogenated peptide library synthesized from the parent sequence H-FQFQFK-NH_2.



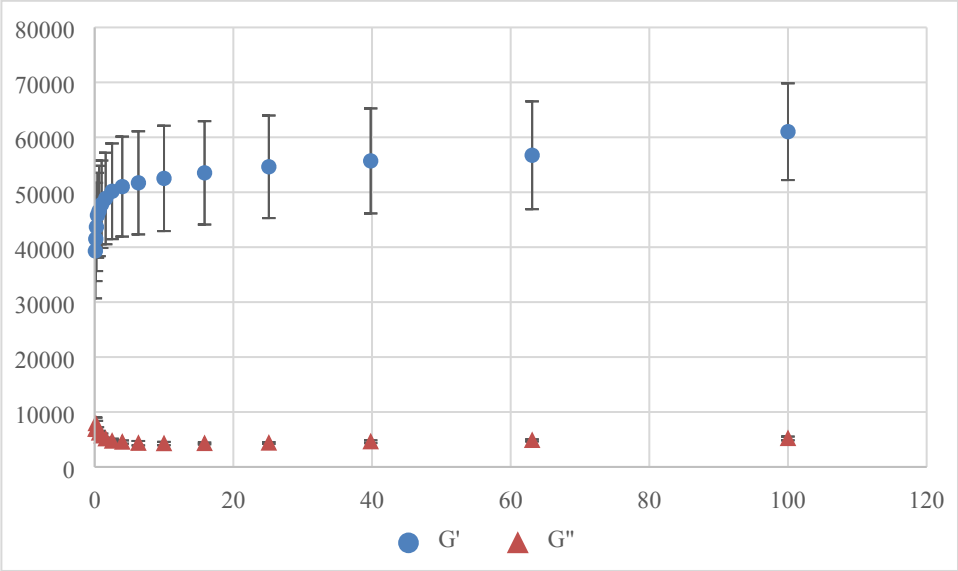
1

N°	Sequence	Yield (%)	R_t[a] (min)	HRMS-[H^+] Calc	HRMS-[H^+] Found
1	H-F[1]QF[3]QF[5]K-NH_2	59	2.73	843.4439	843.4421
5	H-F(p-I)QFQFK-NH_2	42	2.91	969.3406	969.3484
6	H-FQF(p-I)QFK-NH_2	10	2.91	969.3406	969.3484
7	H-FQFQF(p-I)K-NH_2	39	2.90	969.3406	969.3391
8	H-F(m-I)QFQFK-NH_2	45	2.91	969.3406	969.3456
9	H-FQF(m-I)QFK-NH_2	38	2.92	969.3406	969.3406
10	H-FQFQF(m-I)K-NH_2	34	2.93	969.3406	969.3402
11	H-F(o-I)QFQFK-NH_2	46	2.86	969.3406	9693.484
12	H-FQF(o-I)QFK-NH_2	53	2.89	969.3406	969.3484
13	H-FQFQF(o-I)K-NH_2	57	2.88	969.3406	969.3484
14	H-F(p-I)QFQF(p-I)K-NH_2	64	3.05	1095.2372	1095.2358
15	H-F(m-I)QFQF(m-I)K-NH_2	43	3.04	1095.2372	1095.2394
16	H-F(o-I)QFQF(o-I)K-NH_2	43	3.06	1095.2372	1095.2394
17	H-F(p-I)QF(p-I)QF(p-I)K-NH_2	23	3.25	1221.1338	1221.1417
18	H-F(m-I)QF(m-I)QF(m-I)K-NH_2	42	3.27	1221.1338	1221.1417
19	H-FQFQF(p-Br)K-NH_2	47	2.85	921.3544	921.3596
20	H-F(m-Br)QFQFK-NH_2	43	2.86	921.3544	921.3568
21	H-FQF(m-Br)QFK-NH_2	40	2.84	921.3544	921.3544
22	H-FQFQF(m-Br)K-NH_2	43	2.85	921.3544	921.3568
23	H-F(p-Br)QFQF(p-Br)K-NH_2	52	3.05	1001.7910	1001.7979
24	H-F(m-Br)QFQF(m-Br)K-NH_2	54	2.93	1001.7910	1001.7920
25	H-F(p-Br)QF(p-Br)QF(p-Br)K-NH_2	41	3.20	1080.6930	1080.6955
26	H-F(m-Br)QF(m-Br)QF(m-Br)K-NH_2	55	3.01	1080.6930	1080.6938
27	H-FQFQF(p-Cl)K-NH_2	44	2.86	877.4049	877.4057
28	H-F(m-Cl)QFQFK-NH_2	35	2.85	877.4049	877.4128
29	H-FQF(m-Cl)QFK-NH_2	40	2.87	877.4049	877.4128
30	H-FQFQF(m-Cl)K-NH_2	57	2.89	877.4049	877.4015
31	H-F(m-Cl)QFQF(m-Cl)K-NH_2	39	2.96	911.3660	911.3666
32	H-F(p-Cl)QF(p-Cl)QF(p-Cl)K-NH_2	54	3.12	911.3660	911.3612
33	H-F(m-F)QFQFK-NH_2	38	2.75	861.4345	861.4423
34	H-FQF(m-F)QFK-NH_2	50	2.77	861.4345	861.4423
35	H-FQFQF(m-F)K-NH_2	52	2.90	861.4345	861.4324
36	H-F(m-F)QFQF(m-F)K-NH_2	45	3.05	879.4251	879.4246

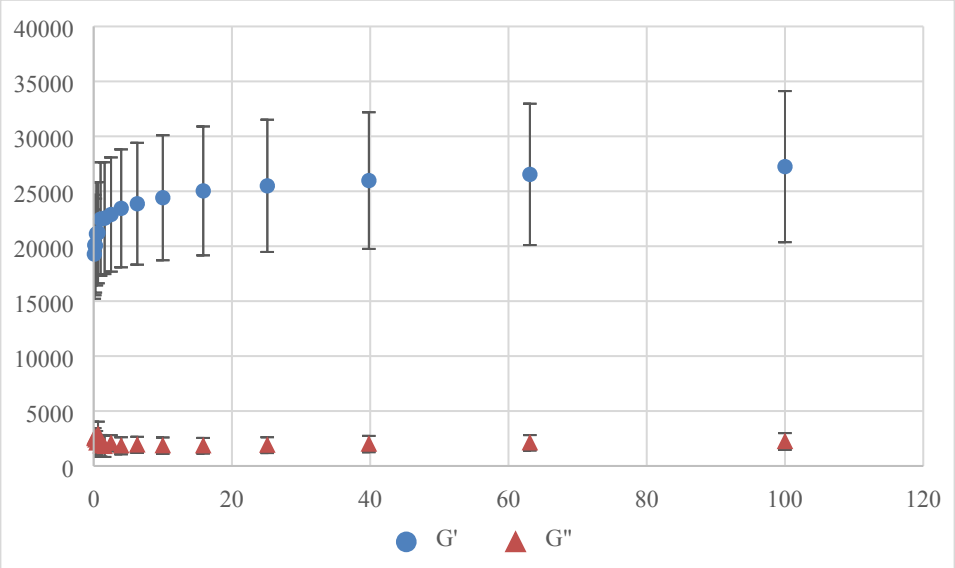
[a] Retention time obtained by the use of a Hitachi Chromaster system (HPLC 5260 autosampler, Chromaster HPLC 5160 Pump, Chromaster HPLC 5310 column and Chromaster HPLC 5430 diode array detector). Peptides were analyzed using a Chromolith column (HR RP-18 50-4.6 mm) with a gradient from 1% to 100% of acetonitrile over 5 min at a flow rate of 3 mL/min. The mobile phase consists of 0.1% TFA in acetonitrile and 0.1% TFA in Milli-Q water.

VIII. Rheological data (frequency sweep)

1. H-FQFQFK-NH_2



5. H-F(p-I)QFQFK-NH_2



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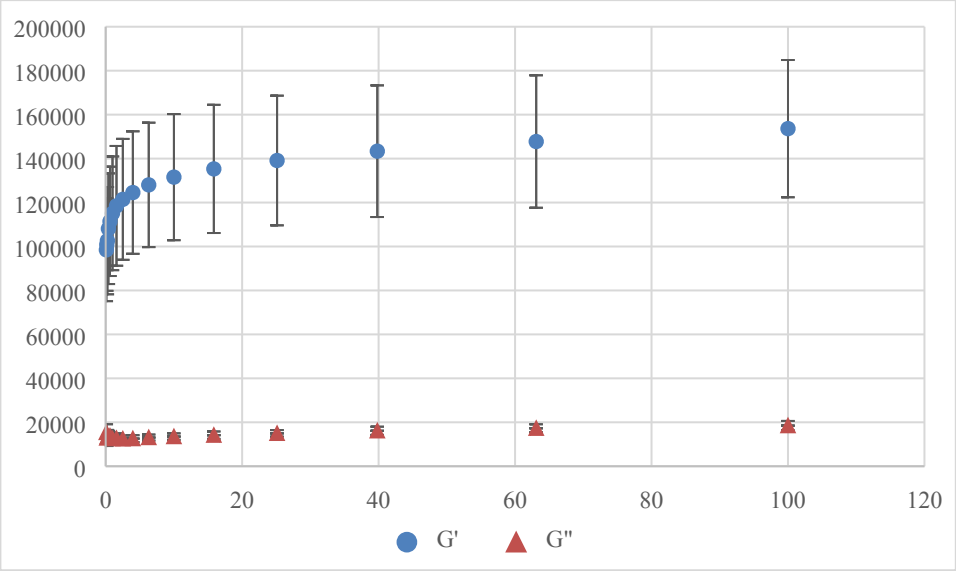
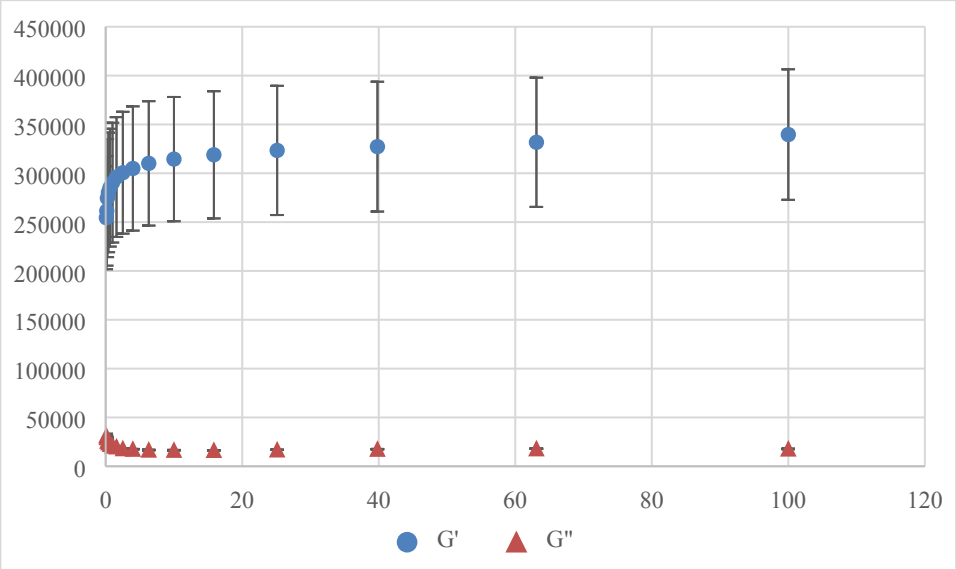
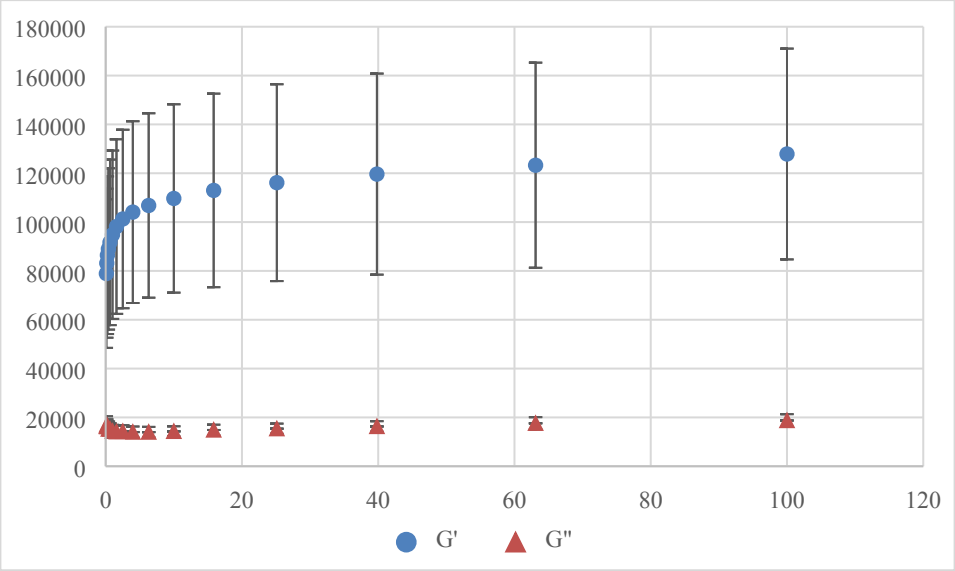


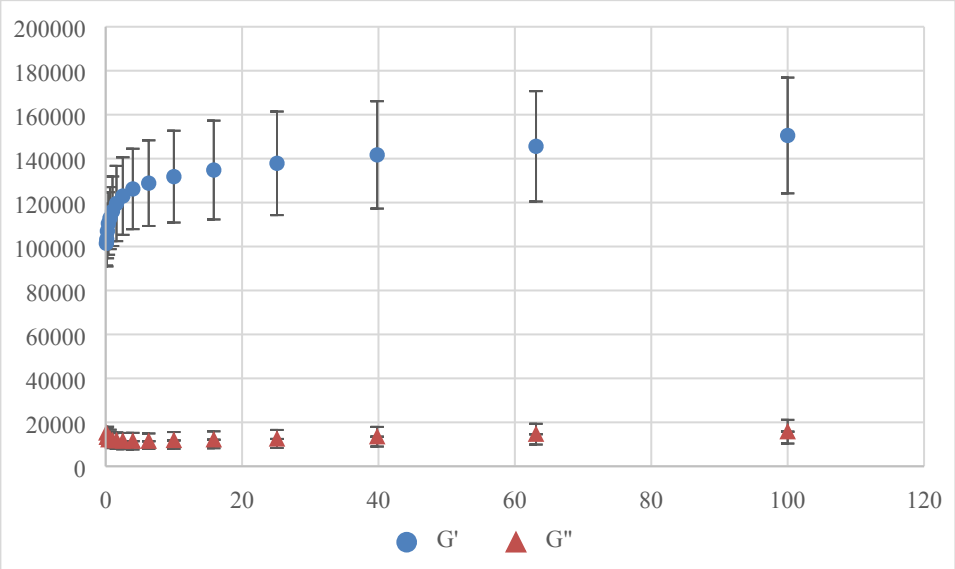
7. H-FQFQF(p-I)K-NH_2



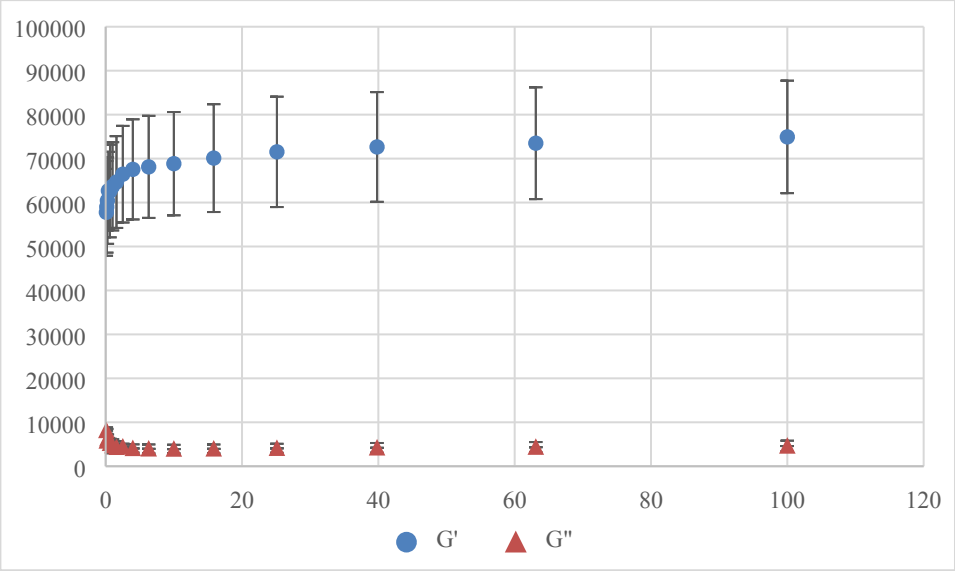
8. H-F(m-I)QFQFK-NH_2



9. H-FQF(m-I)QFK-NH_2



11. H-F(o-I)QFQFK-NH_2



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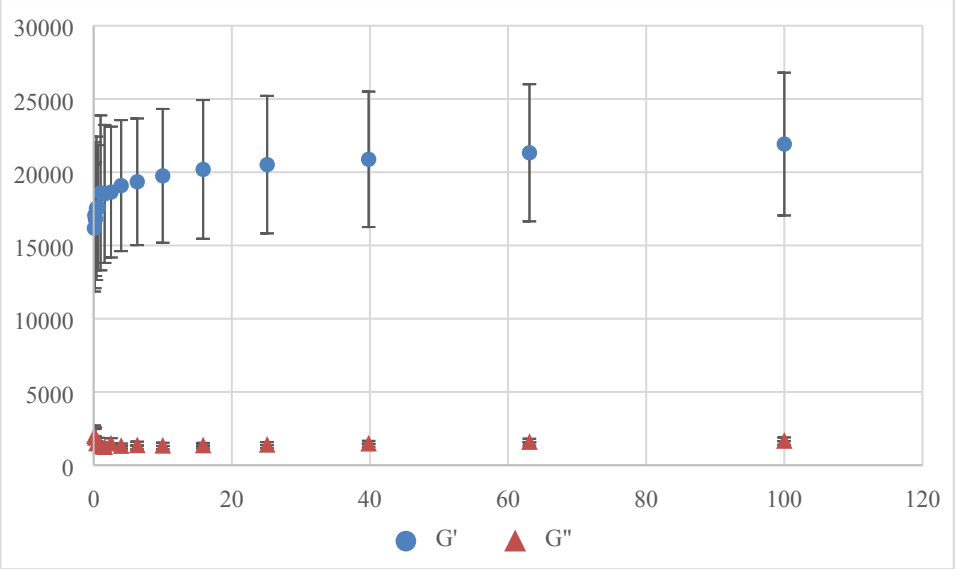

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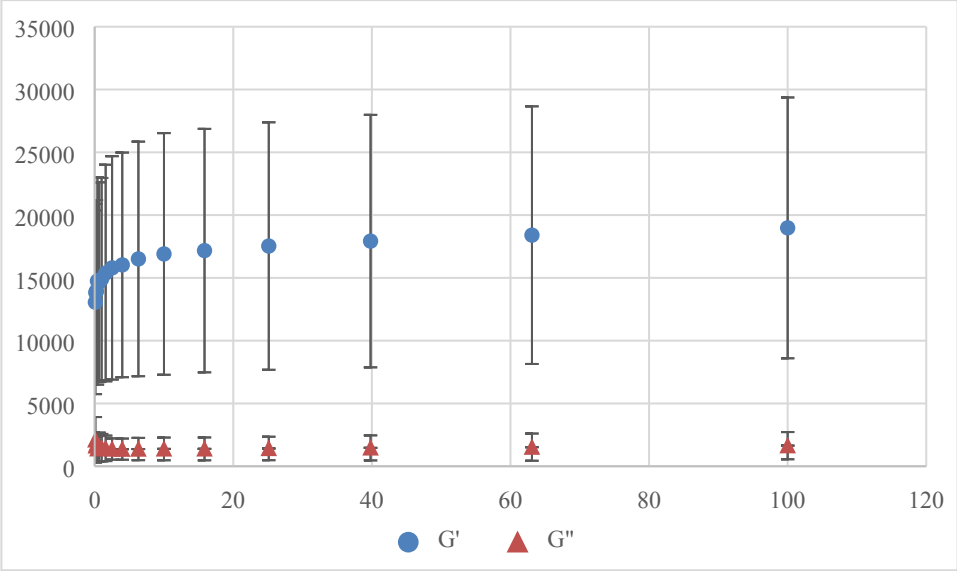

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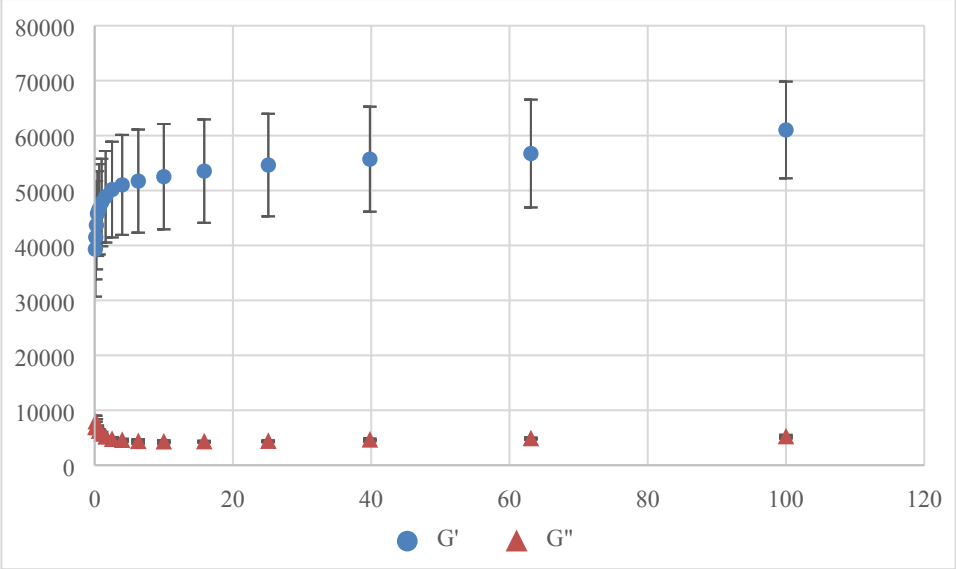
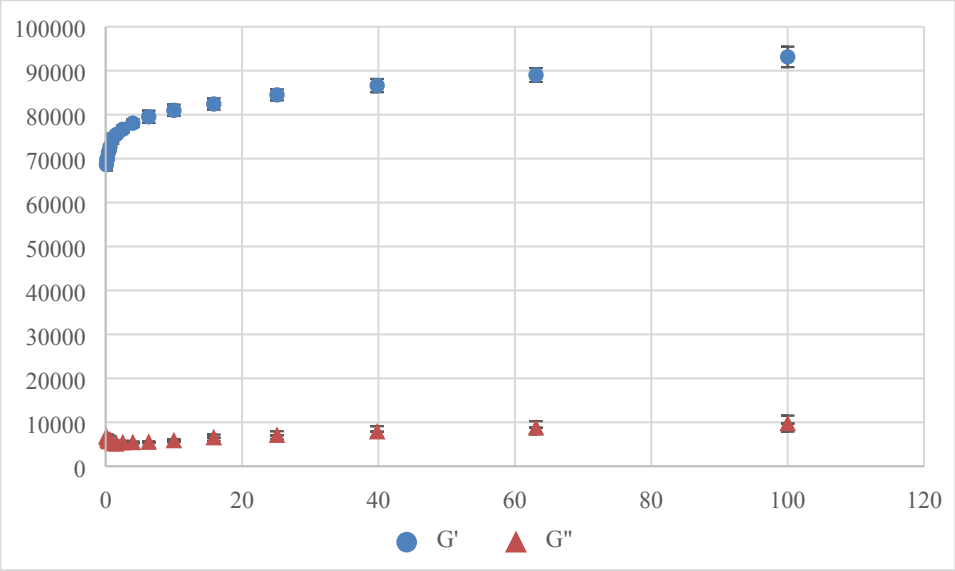


18. H-F(m-I)QF(m-I)QF(m-I)K-NH_2



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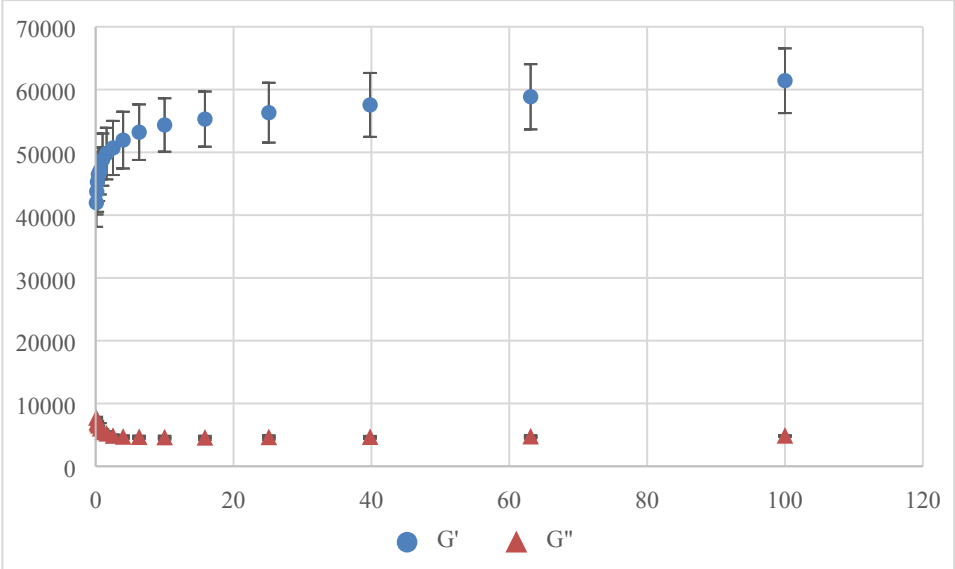

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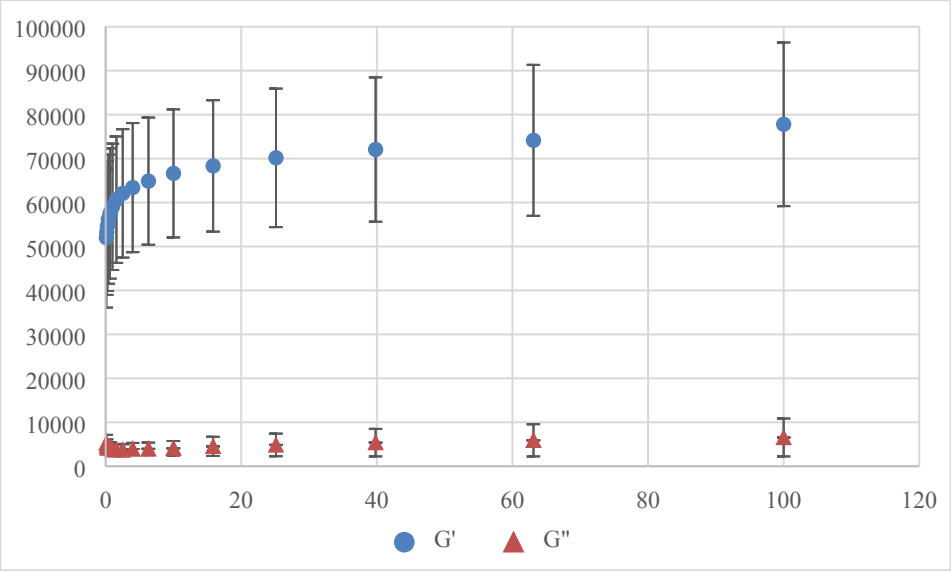
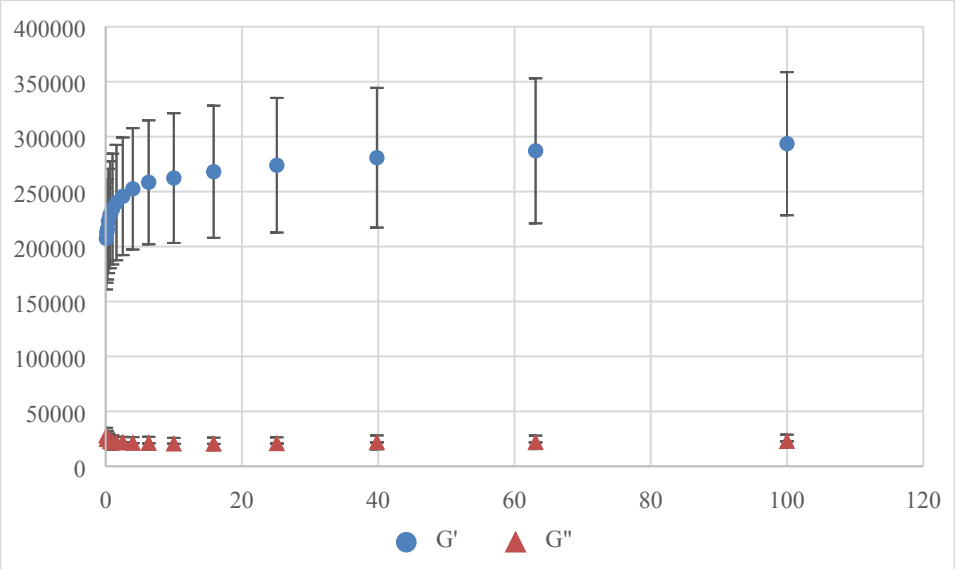
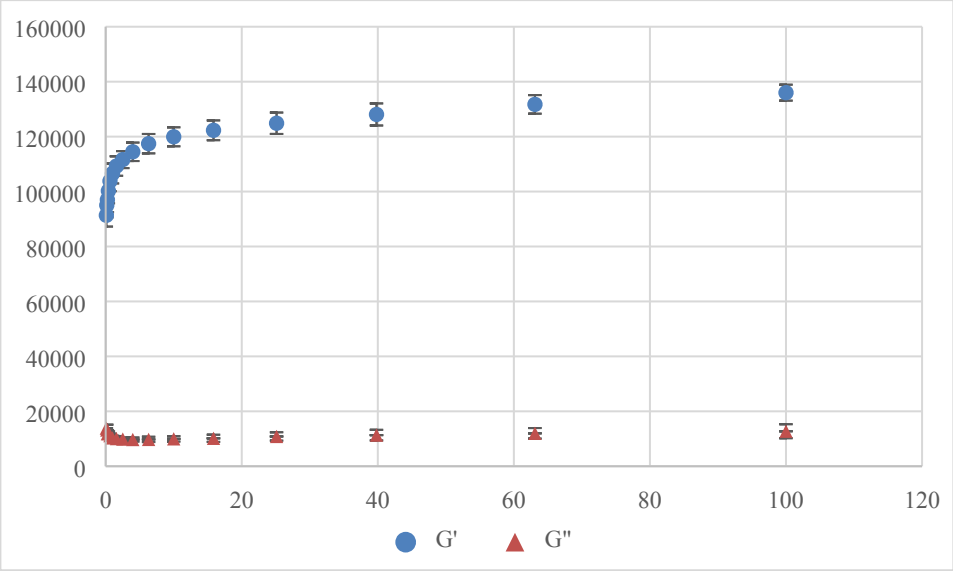


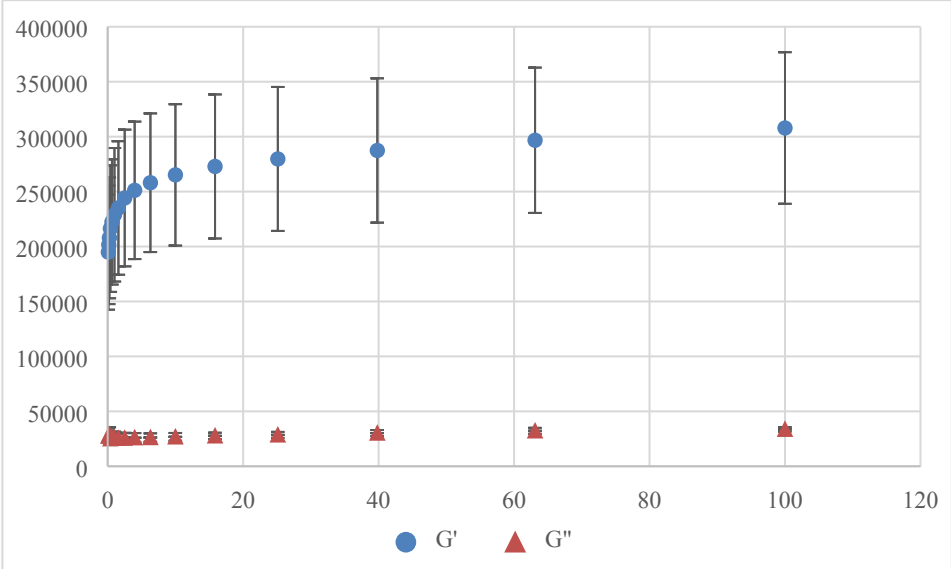
21. H-FQF(m-Br)QFK-NH_2



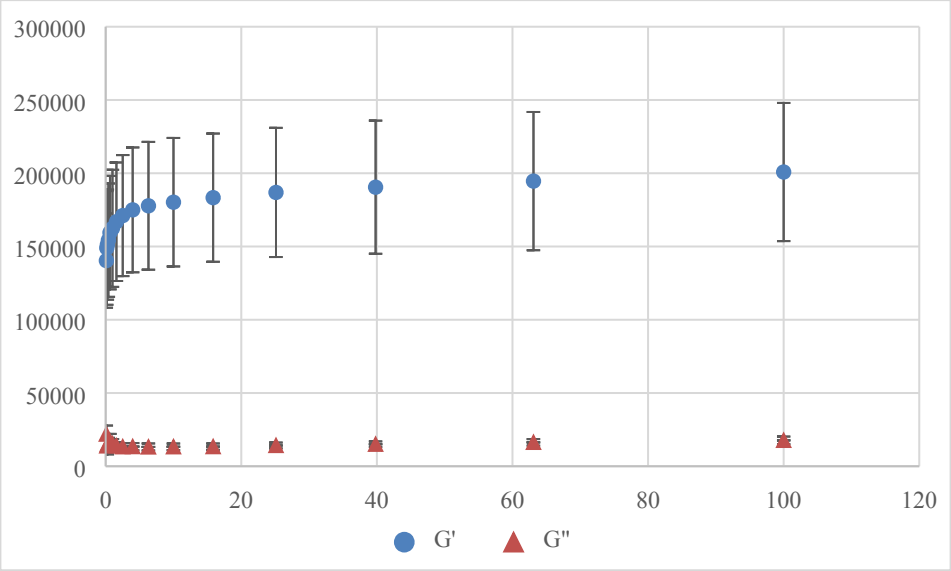
22. H-FQFQF(m-Br)K-NH_2



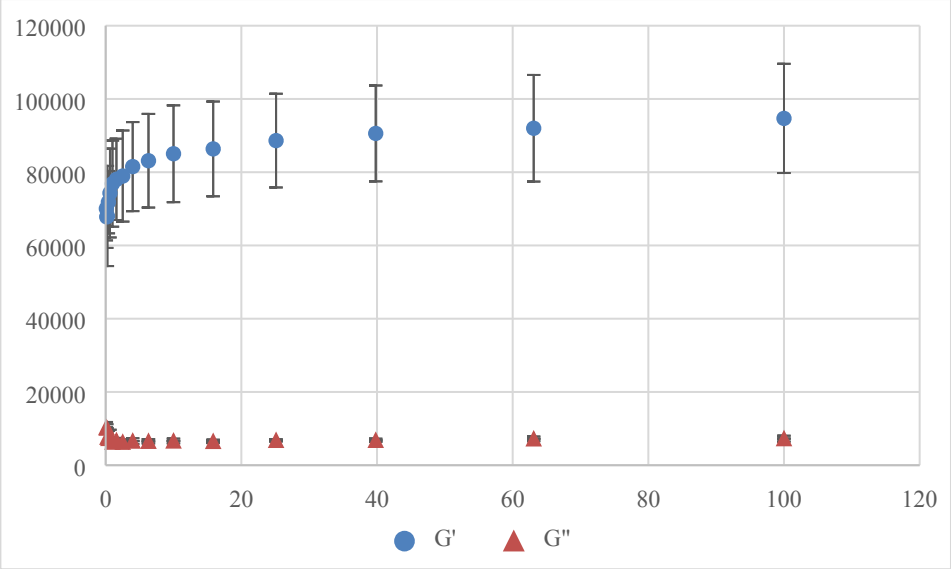
24. H-F(m-Br)QFQF(m-Br)K-NH_2



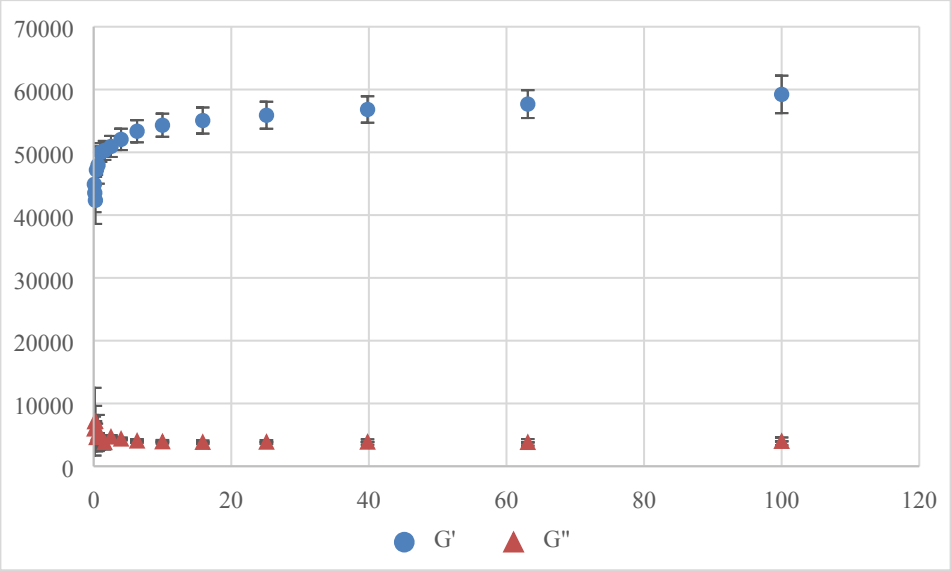
26. H-F(m-Br)QF(m-Br)QF(m-Br)K-NH_2



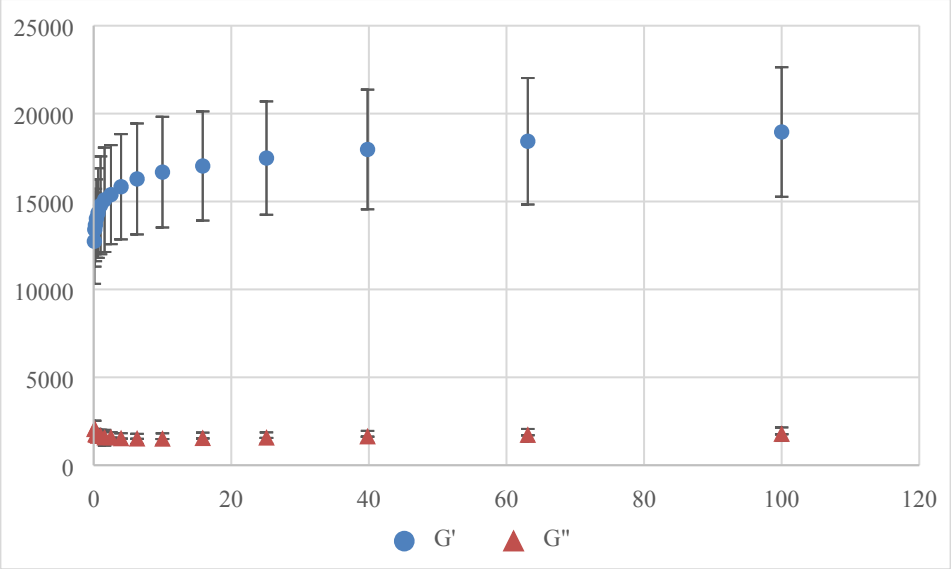
27. H-FQFQF(p-Cl)K-NH_2



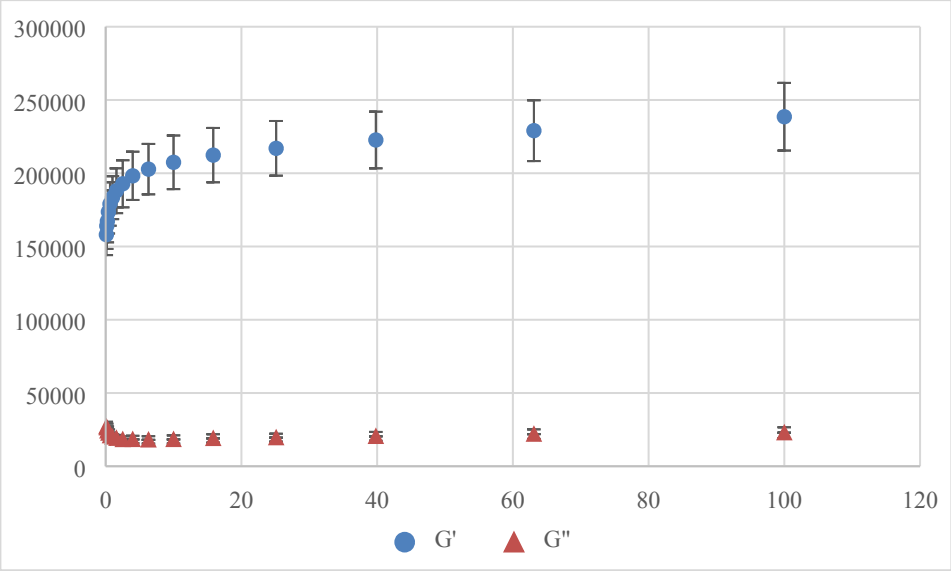
28. H-F(m-Cl)QFQFK-NH_2



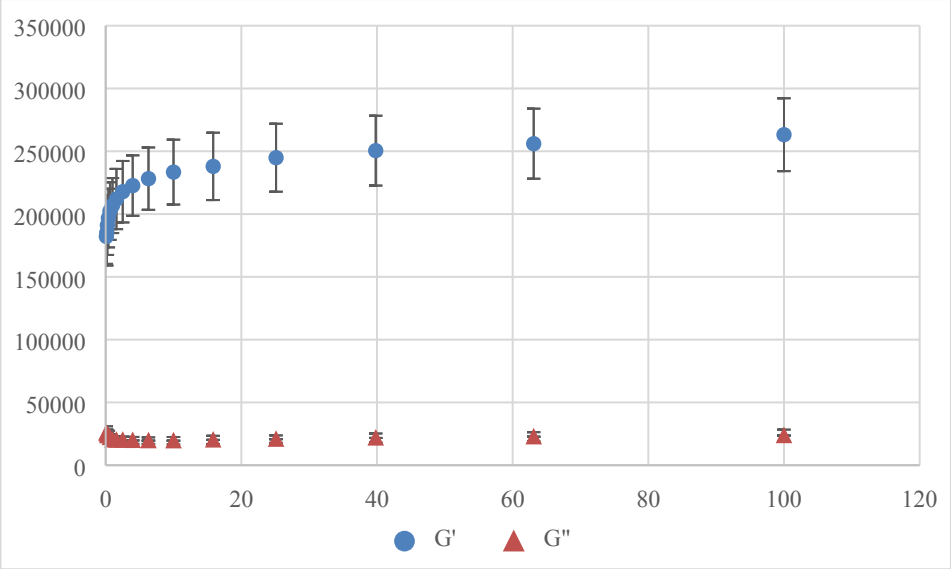
29. H-FQF(m-Cl)QFK-NH_2



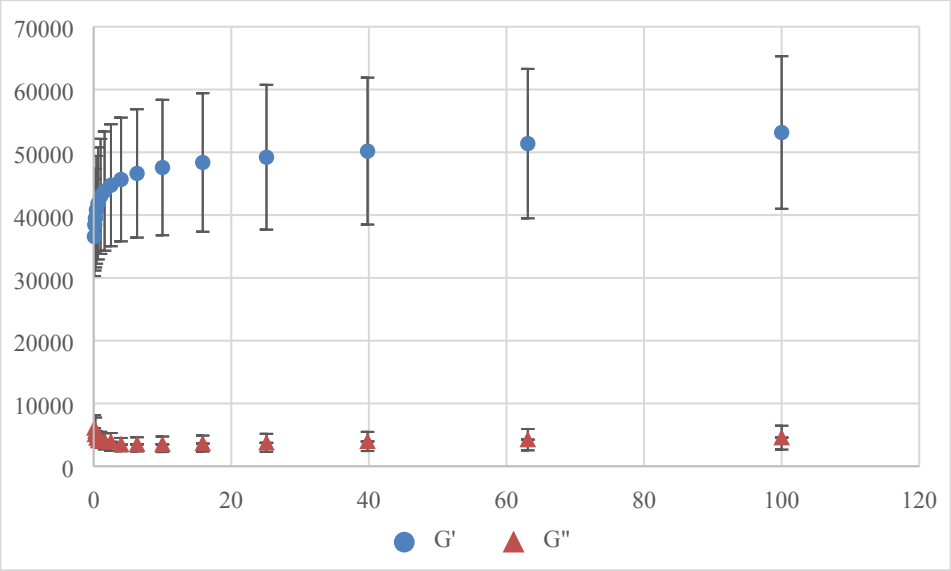
30. H-FQFQF(mCl)K-NH_2



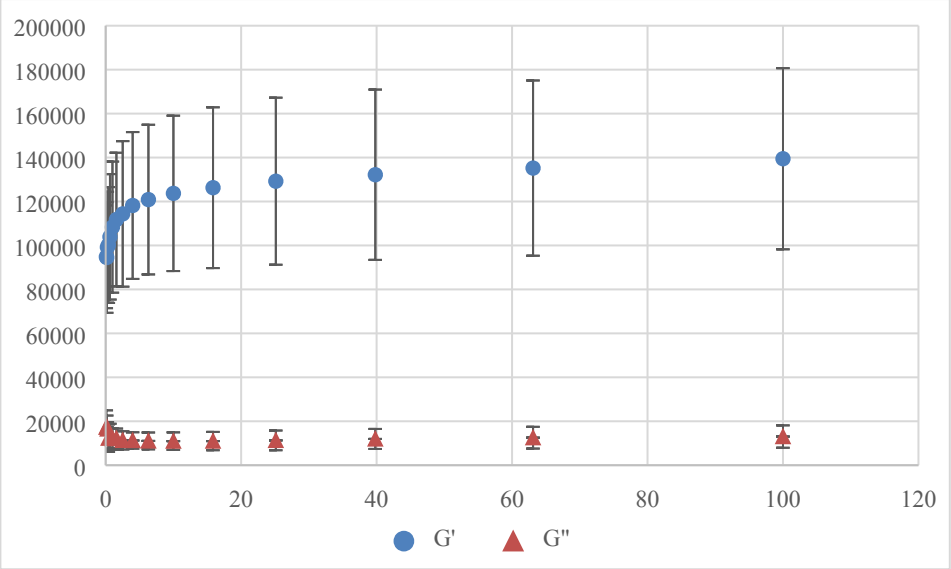
31. H-F(m-Cl)QFQF(m-Cl)K-NH_2



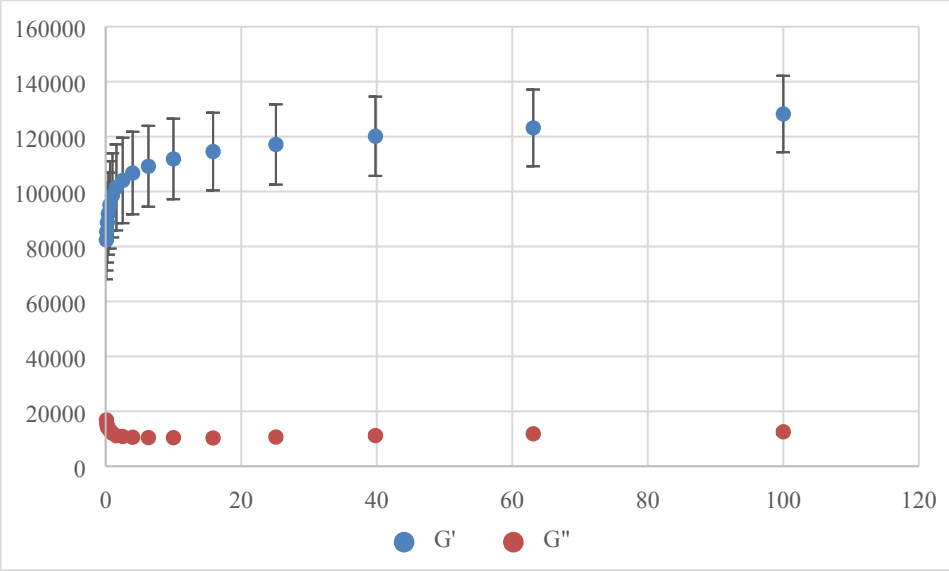
33. H-F(m-F)QFQFK-NH_2



34. H-FQF(m-F)QFK-NH_2



35. H-FQFQF(m-F)K-NH_2



36. H-F(m-F)QFQF(m-F)K-NH_2