Electronic Supplementary Material (ESI) for Soft Matter.

The Interplay of Aggregation, Polymerization and Gelation of An Unexpected Low Molecular Weight Gelator: Glycylalanylglycine in Ethanol/Water

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Supporting Information

Calculation of effects of turbulence on dichroism

In this paragraph we estimate the influence of turbidity on the measured dichroism values of our UVCD spectra. Turbidity can increase the overall absorptivity and thus upshift the baseline of the absorption spectra. The ellipticity θ of the transmitted light is proportional to the difference ΔA between the absorptivity of right- and left-handed circular polarized light:

$$\theta = \frac{180° \ln(10)}{4\pi} \Delta A \qquad \text{(S1)}$$

Ellipticity is converted into molar dichroism Δε by using the Beer-Lambert relationship and the well established relationship between molar ellipticity and molar dichroism:[1]

$$\Delta\varepsilon = \frac{180^0 \cdot \ln 10}{4\pi \cdot b \cdot c \cdot 32980} \cdot \Delta A \qquad \text{(S2)}$$

where b is the pathlength of the cuvette and c the molar concentration of the peptide. The variation δΔA caused by the turbidity of the sample can be calculated as (α-1)·ΔA, where α is the factor by which the absorptivity is increased from values observed for a transparent sample. Propagation of error therefore yields:

$$\delta\Delta\varepsilon = \frac{180^0 \cdot \ln 10}{4\pi \cdot b \cdot c \cdot 329800} \cdot \delta\Delta A = \frac{180^0 \cdot \ln 10}{4\pi \cdot b \cdot c \cdot 329800} \cdot (\alpha - 1)\Delta A \text{ [1]} \qquad \text{(S3)}$$

The absorbance of a 55mol% solution of ethanol and water without peptide at 221nm was used as the reference for calculation of α. The plotted results are shown in SI Figure 1.

SI Table 1: Spectral parameters obtained from decomposing the FTIR spectrum shown in Figure 6 using the program MULTIFIT.

A. Integrated Intensities in arbitrary units

	Time [min]	**3**	**5**	**10**	**15**	**20**	**25**	**30**	**35**	**40**	**45**	**50**	**55**	**60**
1329cm^{-1}	10°C	23047	6637	7667	6417	3076	2154	1805	1467	1392	1442	1332	1296	1301
	16°C	2250	2193	2308	1889	1750	1738	1714	1671	1640	1656	1651	1595	1606
	23°C	1553	2284	3053	2884	2699	2574	2463	2294	2246	1909	1766	1631	1806
1341cm^{-1}	10°C	692	357	332	311	369	249	255	266	277	286	287	293	294
	16°C	659	567	559	572	542	567	572	553	548	558	557	555	550
	23°C	3103	2471	2478	1922	1728	1650	1528	1286	1234	1215	1108	923	794
1364cm^{-1}	10°C	6057	3985	3820	3575	2335	1483	1208	915	843	850	738	625	624
	16°C	3725	3521	3349	3193	3013	2989	2974	2909	2876	2883	2889	2841	2831
	23°C	2226	2074	2045	2093	2107	2081	2036	1951	1925	1565	1420	1263	1252
1389cm^{-1}	10°C	34095	30211	30454	31182	25244	24018	21180	17741	16463	15567	14634	13427	13408
	16°C	35834	36404	36181	36581	37480	37230	37371	37862	37968	37482	37595	37622	37925
	23°C	15862	16119	15651	16291	16903	17062	17291	17729	17823	13281	11040	9072	8909
1418cm^{-1}	10°C	0	42528	43665	41318	24175	14537	11253	7987	7351	6974	6312	5564	5628
	16°C	26090	25101	25750	25365	24698	24875	24874	24708	24591	24868	24935	24581	24577
	23°C	27455	26844	26380	25265	24622	24172	23573	22155	21872	18972	17172	15148	15126

1449cm^{-1}	10°C	24614	15824	15150	14892	11436	9159	7729	6234	5803	5665	5178	4571	4574
	16°C	26655	26625	26086	25915	25889	26098	25962	25811	25824	25301	25520	25689	25694
	23°C	11438	11538	13541	14517	15252	15471	15626	15636	15609	10578	7946	5536	4926
1476cm^{-1}	10°C	24473	20778	20665	22156	19554	15256	12607	9736	8601	8219	7380	6550	6468
	16°C	21883	21151	20376	19711	19358	19534	19417	19357	19256	19813	19317	19129	19113
	23°C	19305	19362	18541	18083	17928	17808	17541	16946	16826	14150	12593	11109	12097
1511cm^{-1}	10°C	2949	198	11	18	133	444	396	341	338	299	300	281	290
	16°C	2261	1630	1322	1256	1127	1252	1221	1208	1170	1082	1186	1026	1111
	23°C	2086	2053	1358	1223	1172	1046	911	807	756	668	649	590	432
1551cm^{-1}	10°C	944	2337	1205	1080	838	745	724	666	639	606	562	543	533
	16°C	335	611	1143	1017	857	997	976	955	905	889	943	728	845
	23°C	537	446	1366	1259	1182	1042	929	834	790	691	675	633	594
1564cm^{-1}	10°C	13318	809	4313	3333	385	336	362	500	591	601	753	895	949
	16°C	1184	664	215	237	256	251	251	259	258	260	251	246	254
	23°C	1274	1605	231	302	317	327	329	317	311	308	309	333	326
1627cm^{-1}	10°C	6227	2796	3218	3224	1483	1655	1923	2319	2539	2637	2994	3377	3462
	16°C	530	665	746	1325	1639	1746	1882	1981	2049	2092	2156	2170	2242
	23°C	1224	1510	1311	1675	1893	2023	2119	2184	2231	2403	2536	2688	2726
1646cm^{-1}	10°C	5390	9450	11934	12438	13230	13248	12773	12600	12482	12466	12147	11677	11655
	16°C	2571	3420	5609	6364	6720	6880	7023	7120	7194	7214	7272	7328	7372

	23°C	2199	2298	5067	5803	6130	6305	6405	6458	6503	6328	6240	6066	6088
1670cm^{-1}	10°C	52087	34807	29328	26868	20961	18010	16115	13545	12691	12198	11490	10504	10489
	16°C	32501	33195	35741	32900	31511	30241	29658	29234	28877	28619	28393	27989	27944
	23°C	25673	25671	28143	25739	23995	23016	22494	21778	21399	18728	16973	15072	14948
1729cm^{-1}	10°C	23944	14505	13495	13246	10384	9289	8399	7193	6668	6395	5893	5241	5214
	16°C	17782	16697	12964	12243	11969	11434	11271	11167	11058	11039	10970	10831	10821
	23°C	16621	17160	10993	9957	9588	9371	9259	9101	9022	8234	7663	6982	6995
1817cm^{-1}	10°C	10227	8178	9745	6851	527	517	414	373	357	402	430	768	795
	16°C	159	153	133	211	229	269	277	286	297	295	298	323	312
	23°C	106	173	337	387	414	434	436	446	462	526	510	428	464
1879cm^{-1}	10°C	529	1988	1363	1292	2471	2269	2042	1790	1670	1619	1455	1309	1275
	16°C	3803	3475	4985	3411	3427	3801	3828	3842	3859	3801	3789	3926	3881
	23°C	1357	2513	4069	3688	3692	3626	3511	3463	3454	2178	1794	1673	1650
1917cm^{-1}	10°C	8679	10095	10166	10879	11979	10808	9467	7439	6816	6640	6107	5369	5367
	16°C	12565	12911	13006	12504	12353	12107	11986	11962	11877	11898	11815	11617	11710
	23°C	13020	12222	12595	12207	11763	11543	11430	11276	11207	10115	9270	8111	8277

B. Lorentzian or Gaussian Primary Halfwidths, denoted (L)/(G)

	Time [min]	3	5	10	15	20	25	30	35	40	45	50	55	60
1329cm^{-1} (G)	10°C	76.7	45.4	45.8	42.3	33.7	28.6	26.8	25.3	25.0	25.8	25.3	25.9	25.9
	16°C	41.3	39.2	36.2	31.6	29.9	29.6	29.0	28.4	27.9	28.1	27.9	27.4	27.3
	23°C	81.3	61.5	62.5	51.7	46.3	44.1	42.0	38.9	38.0	37.8	37.5	36.6	37.7
1341cm^{-1} (G)	10°C	17.5	16.0	11.3	9.9	10.7	7.9	8.0	8.2	8.4	8.6	8.6	8.8	8.8
	16°C	21.7	19.5	15.9	14.7	13.9	13.9	13.7	13.3	13.1	13.1	13.0	13.1	12.8
	23°C	42.2	37.9	35.5	30.0	27.5	26.6	25.6	23.3	22.7	23.4	22.8	21.4	20.5
1364cm^{-1} (G)	10°C	14.8	12.6	12.2	11.8	11.2	10.6	10.6	10.8	10.9	11.2	11.1	11.1	11.1
	16°C	13.8	13.5	13.2	13.0	12.7	12.6	12.6	12.5	12.5	12.5	12.5	12.4	12.4
	23°C	12.1	11.6	11.4	11.4	11.4	11.3	11.2	11.3	11.2	11.4	11.7	12.1	12.1
1389cm^{-1} (L)	10°C	3.7	4.4	4.2	5.2	10.5	19.2	21.4	23.6	23.6	22.4	23.3	23.7	23.6
	16°C	11.2	11.8	11.7	11.9	12.6	12.7	12.7	13.0	13.1	12.9	12.8	12.9	13.1
	23°C	3.3	3.9	0.7	1.6	2.8	3.1	3.7	6.5	7.0	6.4	6.3	7.0	6.6
1418cm^{-1} (G)	10°C	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6
	16°C	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6	69.6
	23°C	67.9	67.9	67.9	67.9	67.9	67.9	67.9	67.9	67.9	67.9	67.9	67.9	67.9
1449cm^{-1} (L)	10°C	22.7	19.2	18.5	18.3	20.4	21.4	21.7	22.2	22.2	22.5	22.4	22.1	22.0
	16°C	14.8	15.5	15.7	15.8	15.9	16.2	16.2	16.0	16.1	15.5	15.7	16.0	16.1
	23°C	0.8	5.2	12.2	15.1	17.1	17.9	18.5	19.9	20.0	18.1	15.7	11.2	8.6
1476cm^{-1}	10°C	3.7	5.0	8.7	12.6	14.9	13.5	12.8	11.8	10.1	10.0	8.6	8.2	7.7

(L)	16°C	4.9	4.2	1.4	0.7	0.7	0.7	0.7	0.7	0.7	2.0	0.7	0.7	0.7
	23°C	4.8	4.4	4.8	4.9	5.1	5.6	5.7	5.8	5.8	6.7	7.1	8.4	13.6
1511cm^{-1} (G)	10°C	63.2	14.5	7.0	6.2	10.0	12.4	11.9	11.4	11.4	10.8	10.9	10.9	11.0
	16°C	44.2	35.4	22.7	21.4	20.5	20.9	20.6	20.4	20.2	19.6	20.0	19.2	19.6
	23°C	39.8	38.7	24.5	22.6	22.0	21.0	19.7	18.8	18.1	18.4	18.7	18.8	17.0
1551cm^{-1} (G)	10°C	30.3	35.9	22.3	19.8	15.1	13.6	13.6	13.8	13.7	13.2	12.8	12.8	12.6
	16°C	23.3	25.4	28.9	25.3	21.9	23.6	22.9	22.3	21.2	20.7	21.6	16.8	19.5
	23°C	28.3	22.9	28.8	25.6	23.6	20.9	18.4	16.2	15.2	13.7	13.9	13.8	13.1
1564cm^{-1} (G)	10°C	76.0	42.3	62.8	60.1	17.5	14.8	15.8	21.3	24.4	24.5	28.8	32.4	33.5
	16°C	37.6	31.7	23.3	22.6	20.7	22.8	22.2	21.9	20.8	20.1	20.9	15.3	18.7
	23°C	43.3	45.4	24.2	24.6	23.6	21.2	18.1	15.5	14.5	13.9	14.6	16.1	15.5
1627cm^{-1} (G)	10°C	77.9	126.8	45.7	41.6	31.0	31.7	33.3	37.3	39.3	40.3	42.8	44.9	45.4
	16°C	33.7	23.0	20.3	23.3	24.4	24.7	25.1	25.4	25.6	25.6	25.8	25.9	26.0
	23°C	36.4	30.2	23.3	23.9	24.3	24.7	24.9	25.1	25.3	26.3	27.1	28.2	28.3
1646cm^{-1} (L)	10°C	3.6	11.6	11.6	10.9	11.7	12.1	12.1	12.8	12.9	13.0	12.7	12.3	12.2
	16°C	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7
	23°C	24.5	18.2	17.1	16.8	16.7	16.7	16.7	16.7	16.7	16.7	16.7	16.7	16.7
1670cm^{-1} (L)	10°C	43.5	34.9	30.6	28.6	24.9	23.0	21.7	19.7	18.8	18.3	17.6	16.6	16.5
	16°C	12.8	18.3	36.1	33.9	32.9	31.9	31.5	31.2	30.9	30.7	30.5	30.2	30.1
	23°C	3.5	6.4	34.0	33.5	31.8	30.8	30.3	29.7	29.3	27.8	26.6	25.3	25.1
1729cm^{-1} (L)	10°C	48.3	35.2	30.7	29.8	26.1	25.3	24.4	23.3	22.6	22.2	21.5	20.7	20.6
	16°C	41.9	40.4	34.6	32.8	32.1	31.1	30.8	30.6	30.4	30.3	30.2	29.8	29.9

	23°C	40.9	42.5	32.2	29.7	28.7	28.2	27.8	27.6	27.4	26.9	26.6	26.0	26.0
1817cm^{-1} (G)	10°C	170.9	324.8	233.8	166.9	44.0	45.3	43.4	44.6	45.2	47.9	51.2	66.7	68.1
	16°C	22.7	23.2	22.2	27.4	28.4	30.0	30.5	30.9	31.3	31.4	31.6	31.8	32.0
	23°C	18.3	23.3	30.3	33.0	34.1	34.8	35.0	35.5	35.9	36.7	36.4	34.7	36.2
1879cm^{-1} (L)	10°C	0.7	41.4	32.3	29.3	43.2	42.2	40.3	38.5	37.9	37.7	36.8	35.4	34.9
	16°C	70.0	69.1	95.8	67.2	65.8	66.9	66.1	65.8	65.2	65.0	64.2	63.8	64.1
	23°C	0.7	65.1	73.5	65.0	61.2	58.6	56.3	54.9	54.1	14.3	0.7	0.7	0.7
1917cm^{-1} (L)	10°C	41.5	45.5	45.1	46.5	50.6	50.9	50.0	48.3	48.3	49.0	50.1	51.8	52.0
	16°C	56.0	57.4	58.8	55.8	55.2	54.0	53.4	53.3	53.0	53.1	52.7	51.9	52.3
	23°C	60.2	56.6	57.5	55.1	52.9	51.7	51.1	50.8	50.5	49.7	49.4	48.0	48.9
1955cm^{-1} (G)	10°C	348.2	300.4	271.5	211.6	12.8	12.8	13.0	12.8	12.8	12.7	12.5	12.6	12.5
	16°C	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0
	23°C	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0

C. Gaussian halfwidths of Voigtian profiles

	Time [min]	3	5	10	15	20	25	30	35	40	45	50	55	60
1389cm^{-1}	10°C	18.6	17.1	17.6	17.2	20.1	16.8	15.7	12.9	12.2	13.0	11.5	9.5	9.4
	16°C	12.3	11.8	12.3	11.9	11.1	11.4	11.4	10.8	10.7	11.2	11.1	11.1	10.8
	23°C	25.6	26.1	28.1	28.0	27.6	27.6	27.4	26.2	26.0	26.7	26.2	25.3	25.4
1449cm^{-1}	10°C	-	-	-	-	-	-	-	-	-	-	-	-	-
	16°C	13.3	12.3	11.5	11.3	11.1	10.8	10.8	11.0	11.0	11.4	11.2	10.9	10.8
	23°C	26.0	23.1	18.9	17.0	15.5	14.8	14.3	13.5	13.4	14.7	15.7	17.9	19.0
1476cm^{-1}	10°C	28.2	25.8	21.3	17.8	14.7	13.4	12.8	11.8	12.3	12.0	12.4	11.9	12.3
	16°C	28.9	29.0	30.2	30.2	30.0	30.0	30.0	30.0	29.9	29.3	29.9	29.8	29.8
	23°C	31.2	32.3	31.0	30.7	30.5	30.2	30.1	29.9	29.9	29.5	29.1	28.0	25.0
1646cm^{-1}	10°C	22.2	11.9	8.6	8.9	8.4	7.9	7.8	6.7	6.5	6.3	6.4	6.7	6.7
	16°C	25.0	19.6	17.2	16.8	16.7	16.8	16.8	16.8	16.8	16.8	16.8	16.9	16.9
	23°C	-	-	-	-	-	-	-	-	-	-	-	-	-
1670cm^{-1}	10°C	-	-	-	-	-	-	-	-	-	-	-	-	-
	16°C	36.2	31.2	-	-	-	-	-	-	-	-	-	-	-
	23°C	44.5	41.4	9.9	1.8	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7
1879cm^{-1}	10°C	29.0	0.7	0.7	6.3	0.7	0.7	0.7	0.7	0.7	0.7	0.7	3.8	3.4
	16°C	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7
	23°C	59.8	3.9	0.7	0.7	1.2	3.4	5.6	7.7	8.5	45.2	52.4	52.0	51.7

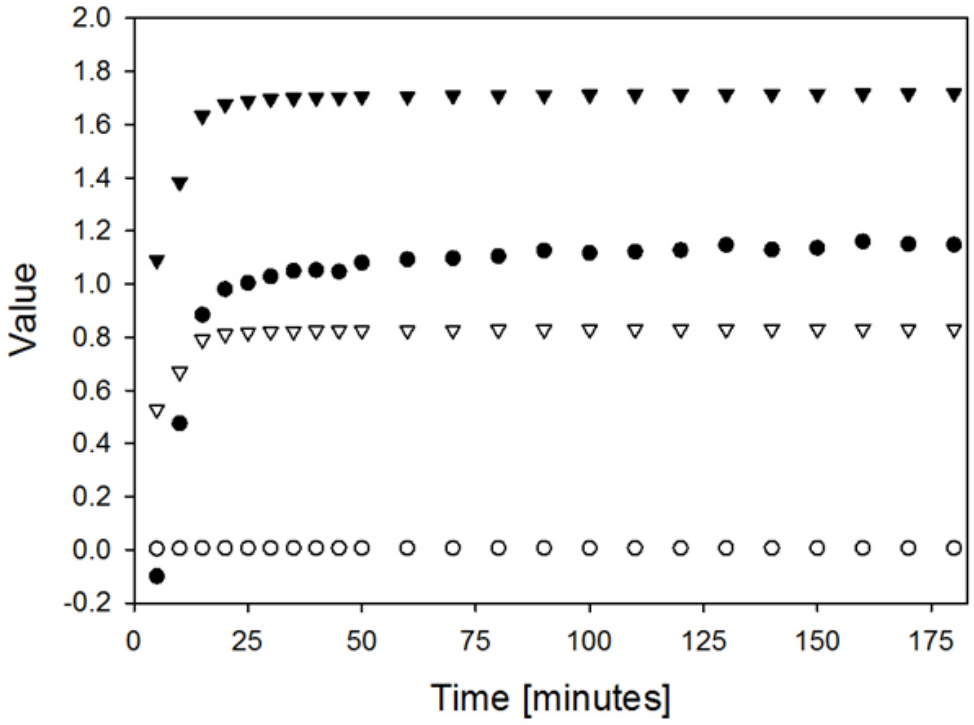

Figure S1: Effects of turbidity on molar dichroism calculated using eq. (S3). The figure depicts the dichroism $\Delta\varepsilon_{221}$[$M^{-1}cm^{-1}$] (black circles) of GAG in 55%mol ethanol/45mol% water at 10^0 C as a function of time, the respective turbidity induced changes $\Delta\Delta\varepsilon_{221}$ [$M^{-1}cm^{-1}$] (white circles), the time dependence of the absorbance A_{221}(black triangles) and the corresponding changes in absorbance ΔA_{221}(white triangles).

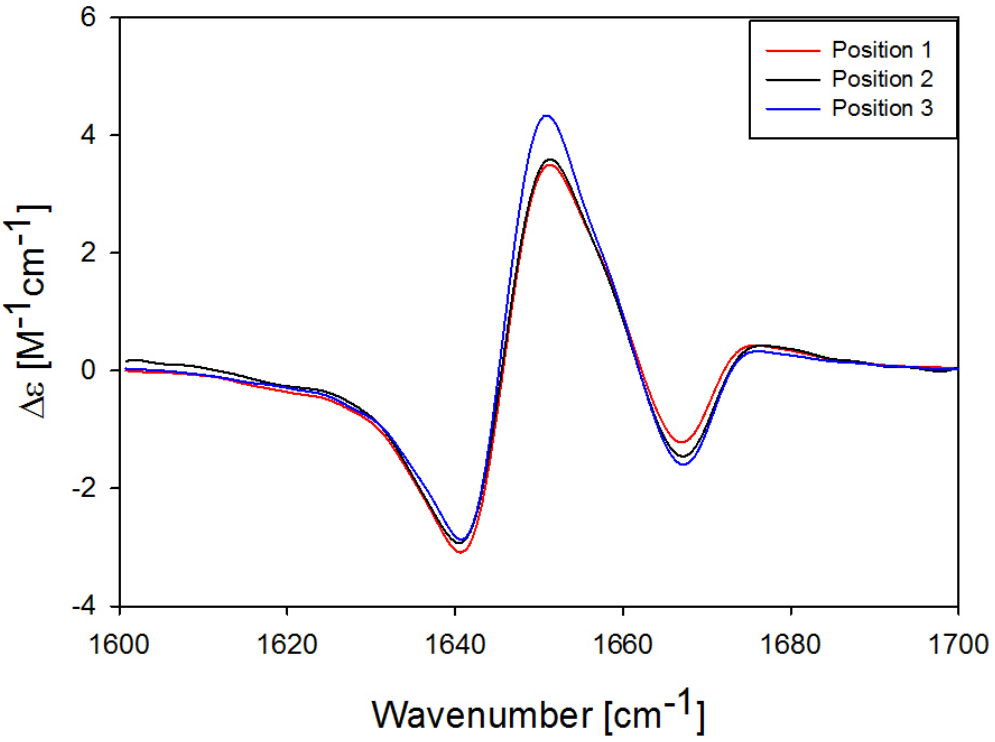

Figure S2: VCD scans of the amide I' region of 220mM GAG in 55mol% d-ethanol/45mol% D_2O for different orientations of the sample, which was rotated 120^o clockwise for each position. The spectra were taken at 23^o C 60 minutes after incubation.

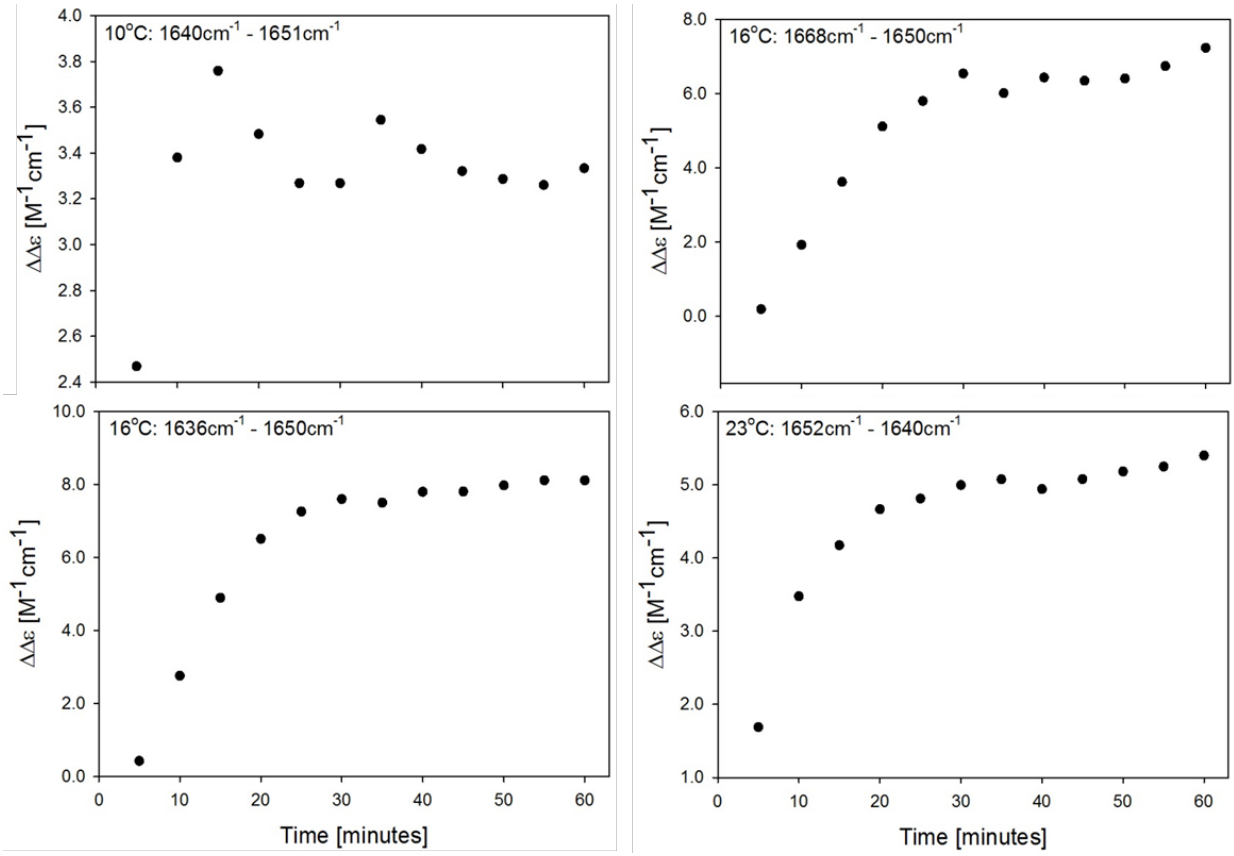

Figure S3: $\Delta\Delta\varepsilon$ measured as the differences between the dichroism values of the positive and negative maxima of VCD profile of AI_1' recorded at 10°, 16°, and 23° C. For 16° C, the respective differences between the negative maximum and both positive maxima are shown.

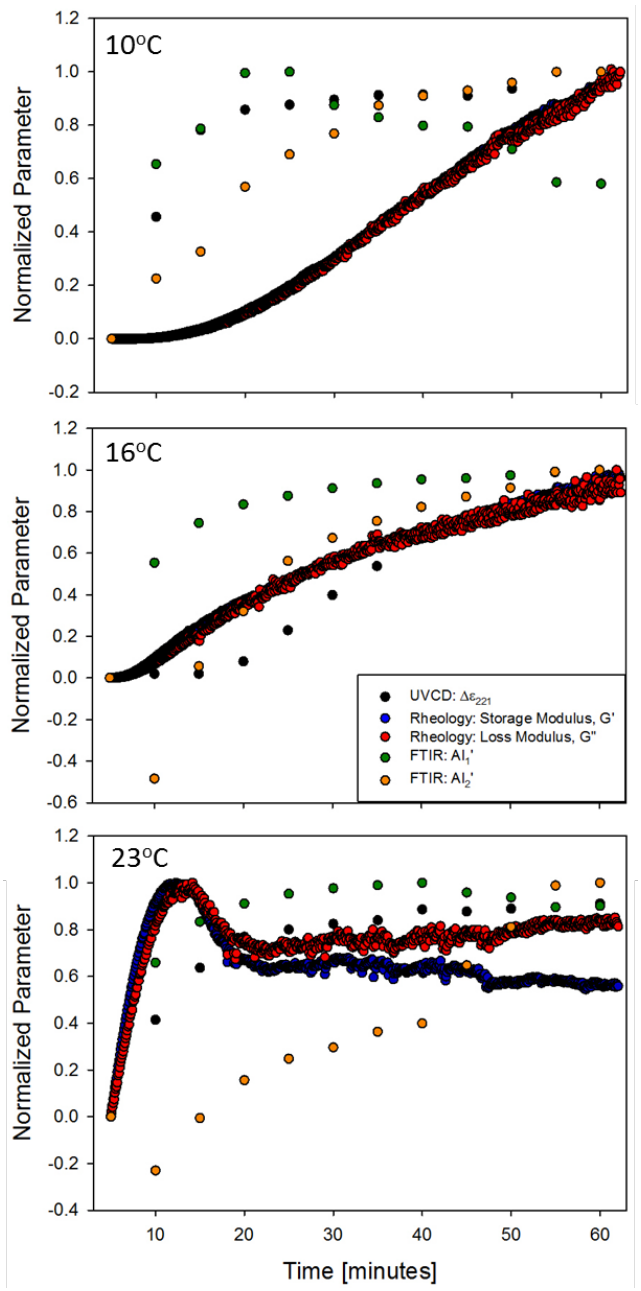

Figure S4: Normalized plots of the AI_1' and AI_2' integrated intensities, rheology storage and loss moduli, and UVCD molar dichroism development over time at 10^o, 16^o, and 23^o C. The AI_2' curve was normalized to its lowest value as 1 due to the exponential decreasing nature of the curve to allow for comparison to other methods which increase over time.

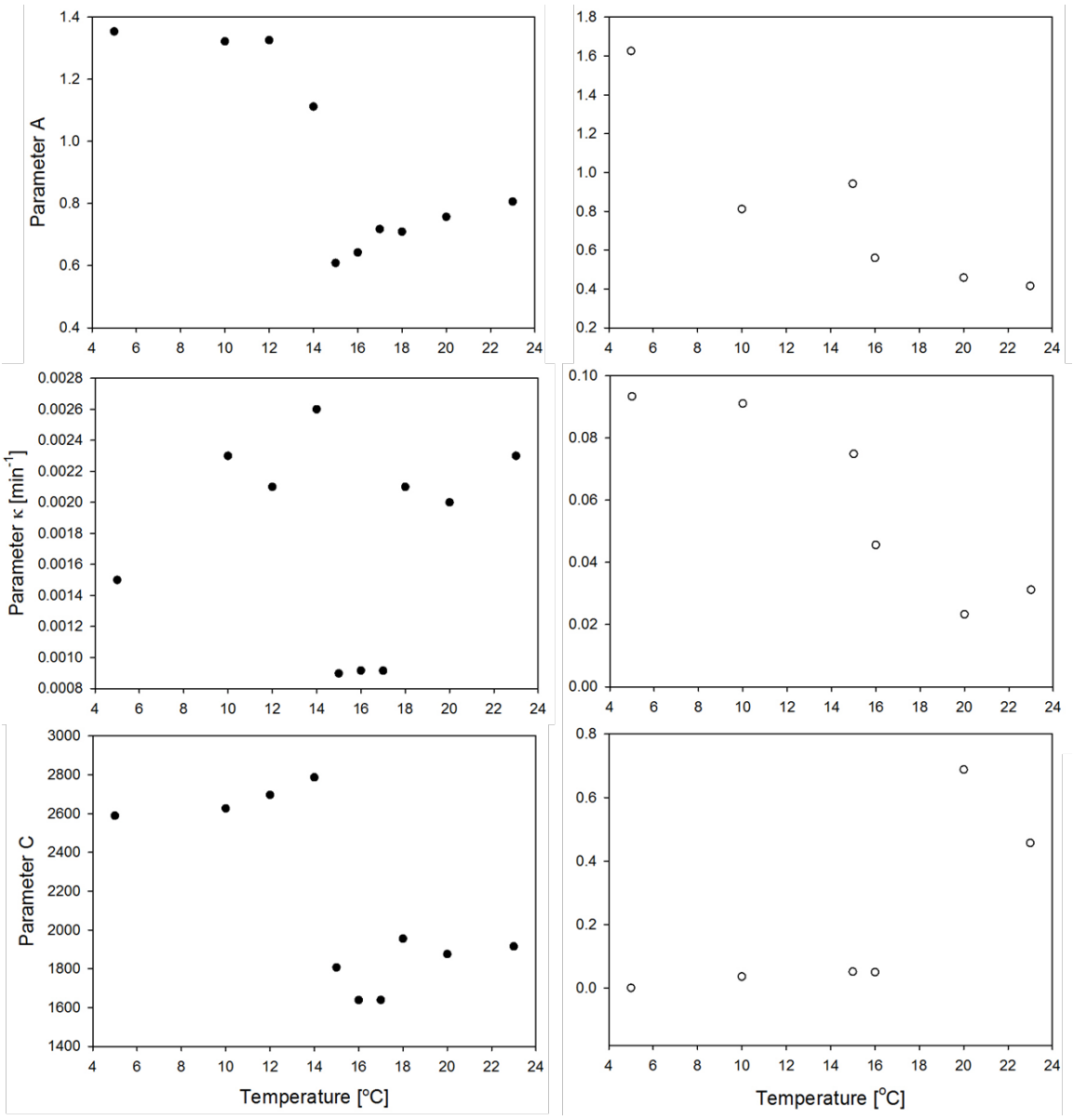

Figure S5: Parameters A, κ, and C obtained from the fits of eq. (6) to the data in Figures 3 and 4 plotted as a function of temperature. Measurements taken by going directly to temperature are shown as black circles; those obtained after quenching the sample from 50° C to indicated temperatures are shown as white circles.

References

1. Schweitzer-Stenner, R.; Soffer, J. B., Optical Spectroscopy. In *Biophysical Techniques for Structural Characterization of Macromolecules*, Dyson, H. J., Ed. Oxford Academic Press: Oxford, 2012; Vol. 1, pp 533-591.