-Supporting Information-

Opposing temperature dependence of the stretching response of single PEG and PNiPAM polymers

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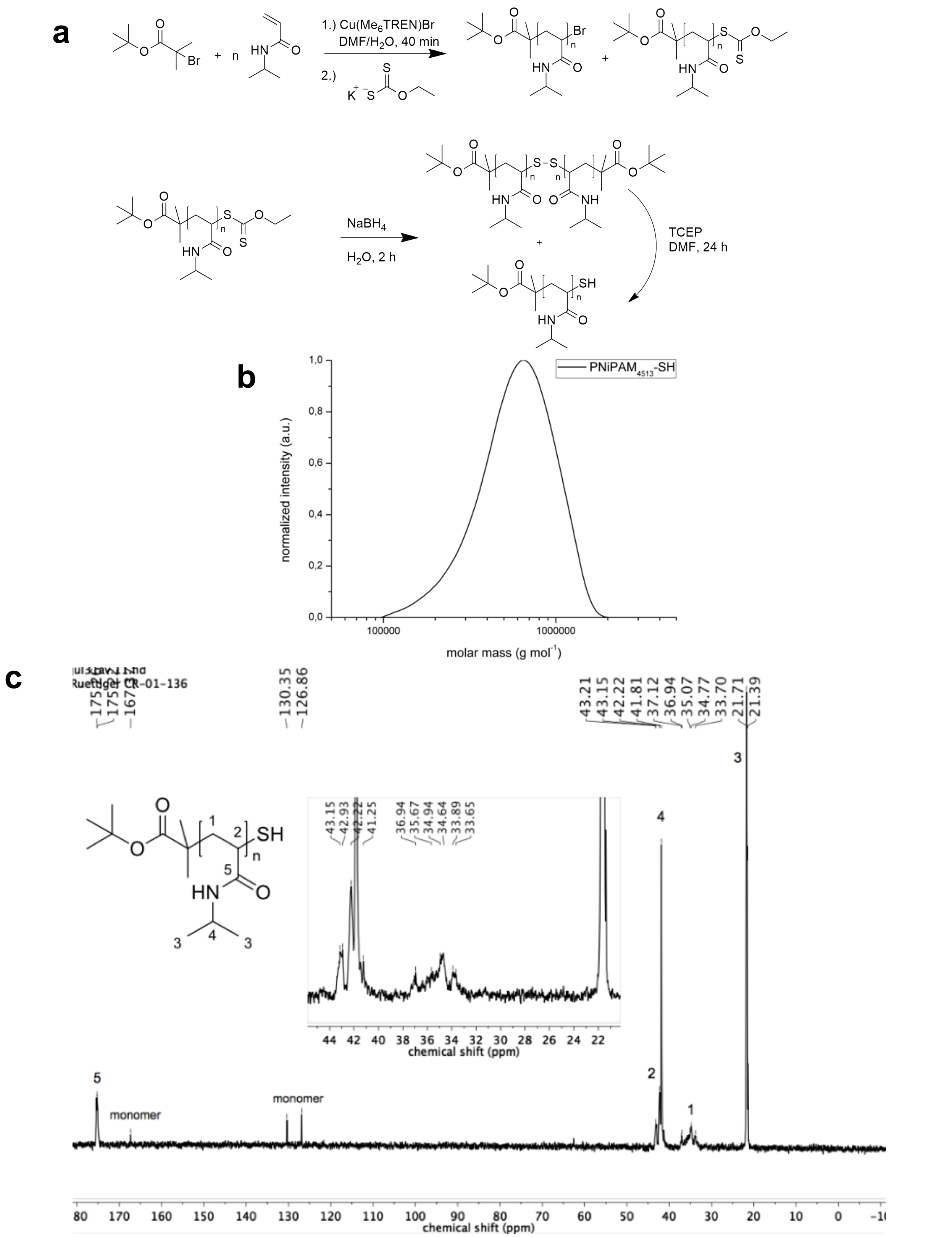

Figure S1. Synthesis of SH-PNiPAM. **a** The reaction path in modification of the following references.[1,2] **b** Resulting GPC data leading to M_n: 510 kDa, $Đ$ = 1.28. **c** ^{13}C-NMR data (125,8 MHz, D_2O as solvent) of the product with a zoom-in in the region between 22 ppm and 45 ppm, indicating that the SH-PNiPAM is atactic according to Idota *et al.*[3]

Force-extension curves for PEG

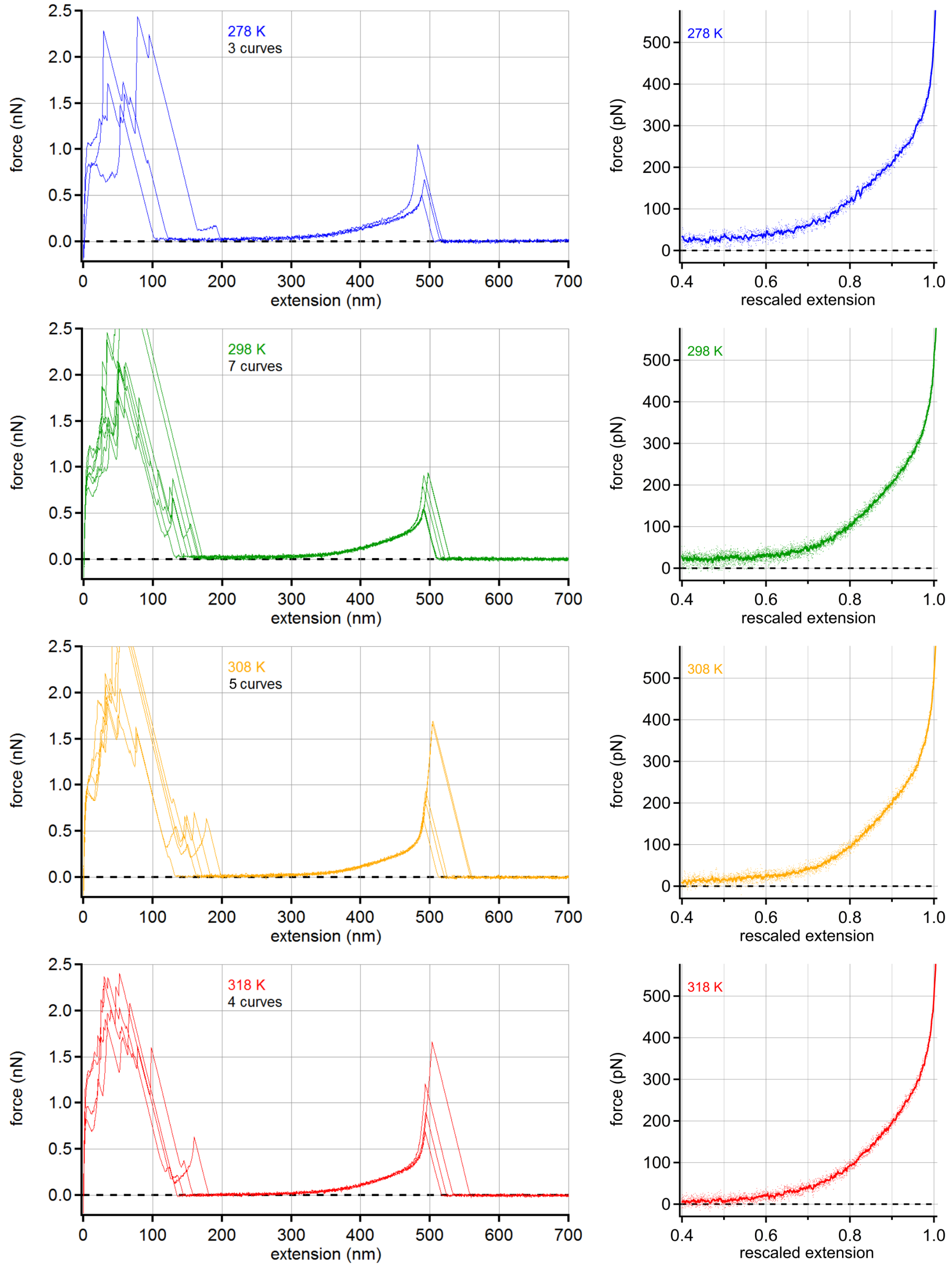

Figure S2. Experimental force-extension curves and master curves of PEG at different temperatures. For a master curve all traces with rupture events higher than 500 pN were selected (left). The extension of each trace was then rescaled to its extension at 500 pN followed by binomial smoothing with a smoothing parameter of 20 (right). The rescaled curves used to obtain the master curves are given as dots while the master curves are shown as solid lines. The number of force-extension curves used for master curve determination is: 278 K – 3 curves, 298 K – 7 curves, 308 K – 5 curves, 318 K – 4 curves.

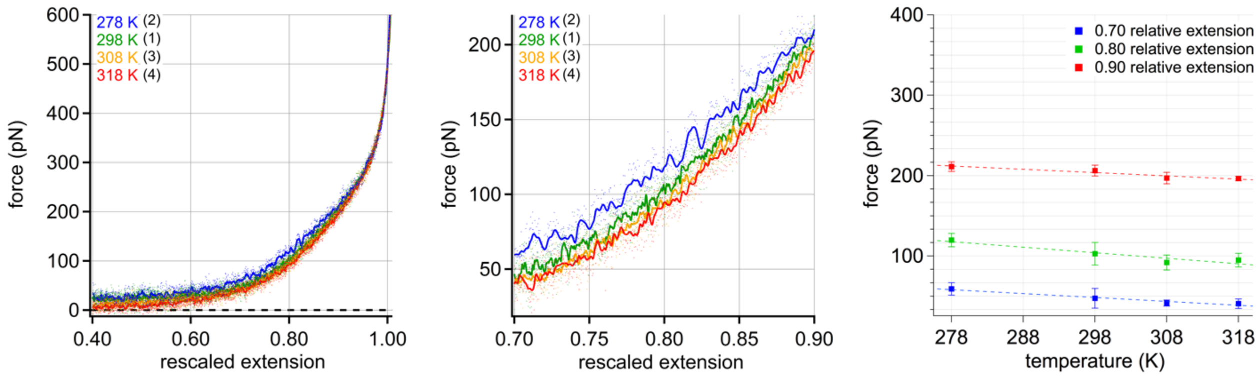

Figure S3. Master curves for different temperatures obtained experimentally for PEG (left) and zoom-in (middle) corresponding to Figure S2. The rescaled curves used to obtain the master curves are given as dots while the master curves are shown as solid lines. The order of data acquisition is given in brackets, i.e. force-extension curves were first taken at 298 K, then at 278 K, followed by 308K and 318 K. Furthermore, force-temperature curves for relative extensions of 0.70, 0.80 and 0.90 are shown, respectively (right). The broken lines serve as a guide to the eye and the error bars represent the standard deviation.

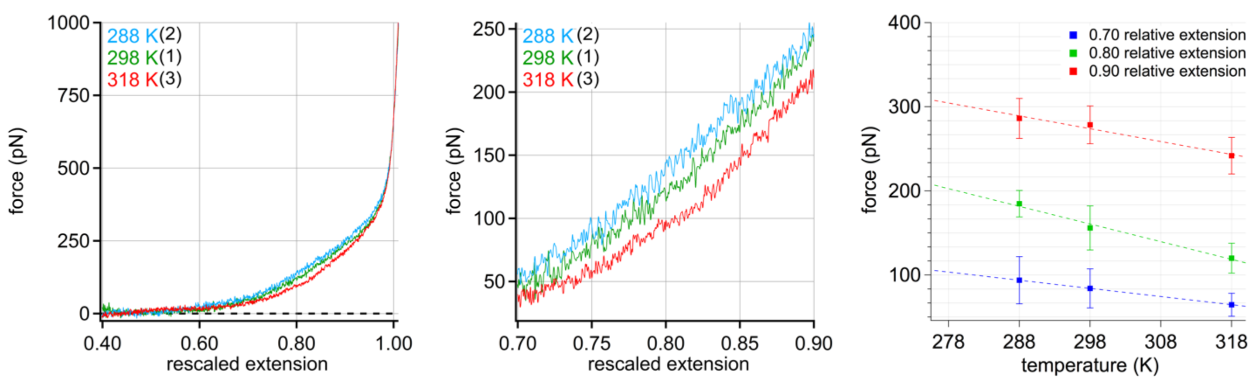

Figure S4. Master curves for different temperatures for a second experimental PEG data set (left) and zoom-in (middle). The number of force-extension curves used for the master curve determination is: 288 K – 7 curves, 298 K – 7 curves, 318 K – 9 curves. The order of data acquisition is given in brackets. Furthermore, force-temperature curves for relative extensions of 0.70, 0.80 and 0.90 are shown, respectively (right). The broken lines serve as a guide to the eye and the error bars represent the standard deviation.

Force-extension curves for PNiPAM

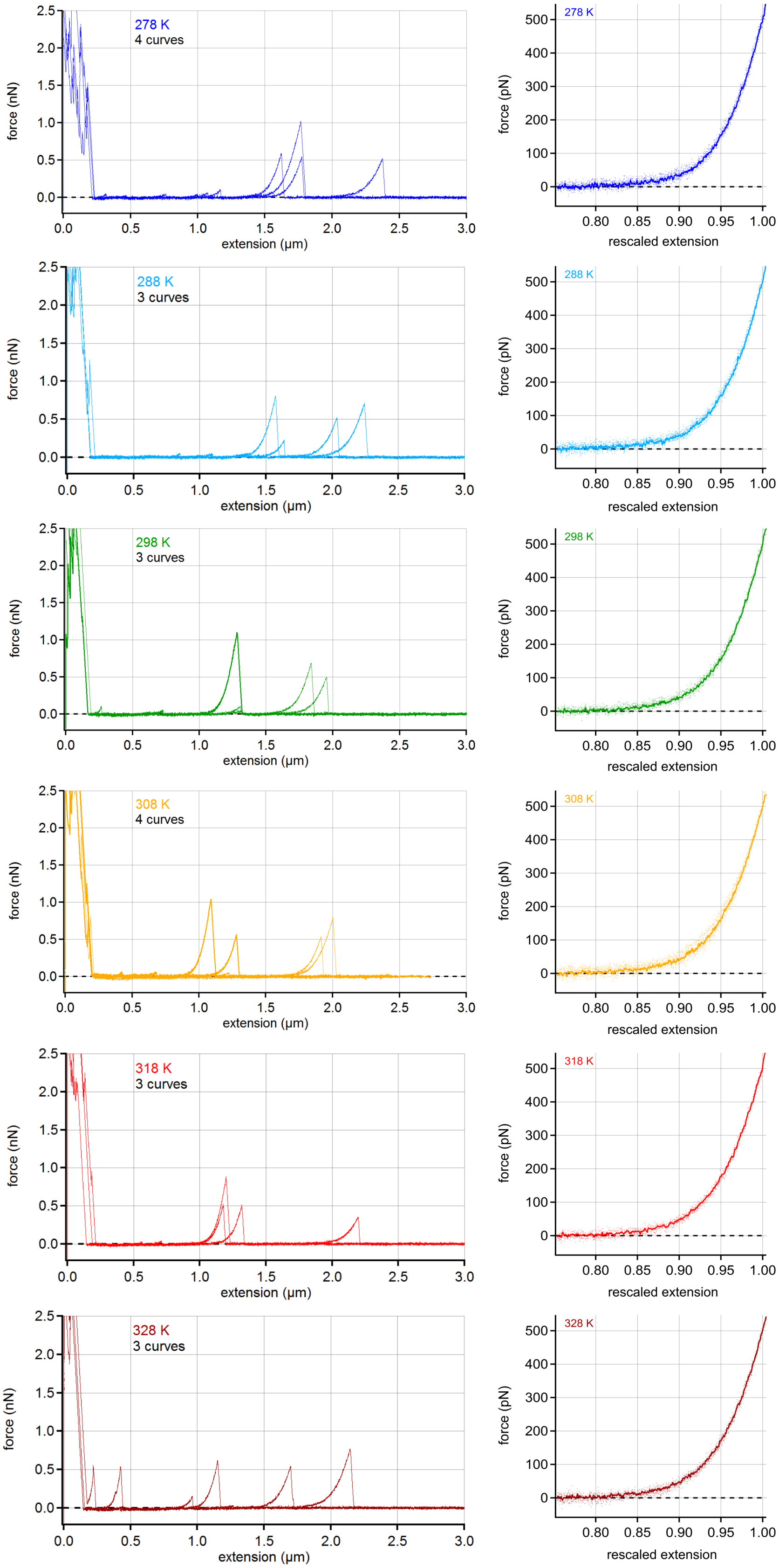

Figure S5. Experimental force-extension curves and master curves of PNiPAM at different temperatures. For a master curve all traces with rupture events higher than 500 pN were selected (left). The extension of each trace was then rescaled to its extension at 500 pN followed by binomial smoothing with a smoothing parameter of 20 (right). The rescaled curves used to obtain the master curves are given as dots while the master curves are shown as solid lines. The number of force-extension curves used for master curve determination is: 278 K – 4 curves, 288 K – 3 curves, 298 K – 3 curves, 308 K – 4 curves, 318 K – 3 curves, 328 K – 3 curves.

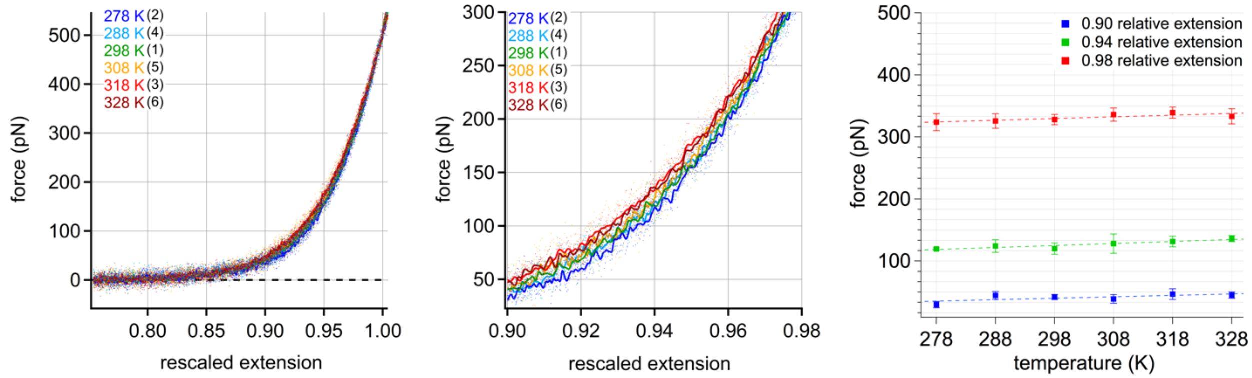

Figure S6. Master curves for different temperatures obtained experimentally for PNiPAM (left) and zoom-in (middle) corresponding to Figure S5. The rescaled curves used to obtain master curves are given as dots while the master curves are shown as solid lines. The order of data acquisition is given in brackets. Furthermore, force-temperature curves for relative extensions of 0.90, 0.94 and 0.98 are shown, respectively (right). The broken lines serve as a guide to the eye and the error bars represent the standard deviation.

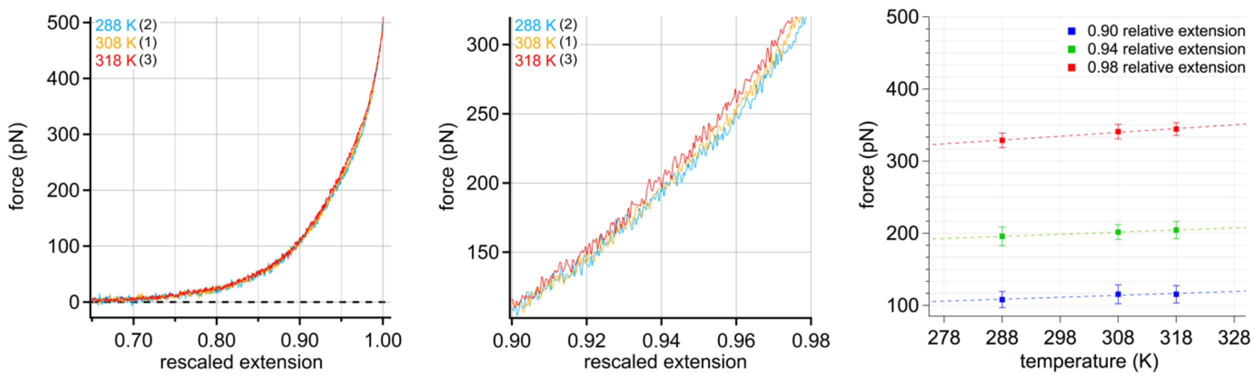

Figure S7. Master curves for different temperatures for a second experimental PNiPAM data set (left) and zoom-in (middle). The smaller effect compared to Figures 2 and S6 can be explained by the smaller temperature range of 40 K and the different lengths of the measured polymers. The first data set shows contour lengths of about 1.0 to 2.5 µm which is approx. two to four times longer than the contour lengths of the second data set. The number of force-extension curves used for rescaling is: 288 K – 18 curves, 308 K – 18 curves, 318 K – 16 curves. The order of data acquisition is given in brackets. Furthermore, force-temperature curves for relative extensions of 0.90, 0.94 and 0.98 are shown, respectively (right). The broken lines serve as a guide to the eye and the error bars represent the standard deviation.

Polymer elasticity models

To further investigate the elasticity of the polymers, the MD simulation data of PEG was fitted with the linear force-extension relation (linear response theory) in the limit of $fb \ll k_B T$ [4–6]:

$$\frac{z_{ete}}{L_c} \approx \frac{fb}{3k_B T} \tag{S1}$$

This allows for the determination of the Kuhn length b by fitting the force-extension data in the range of 1 to 4 pN. The fitting of the slope has been done using the conventional method of weighting each point by its squared error, which was obtained by block averaging.[7] The course of the Kuhn length suggests no significant change over a wide range of temperatures (Table S1). Therefore, we use the mean of the simulated Kuhn lengths, namely $b = 0.75$ nm to fit the experimental data.

Temperature (K)	**Kuhn length b (nm)**
250	0.74 ± 0.02
300	0.75 ± 0.05
350	0.73 ± 0.03
400	0.76 ± 0.02

Table S1: Kuhn length fit parameter obtained by fitting the low force – extension (1 to 4 pN) range of MD simulation data for PEG using eq. (S1). No significant change of the Kuhn length over a temperature range of 150 K could be found.

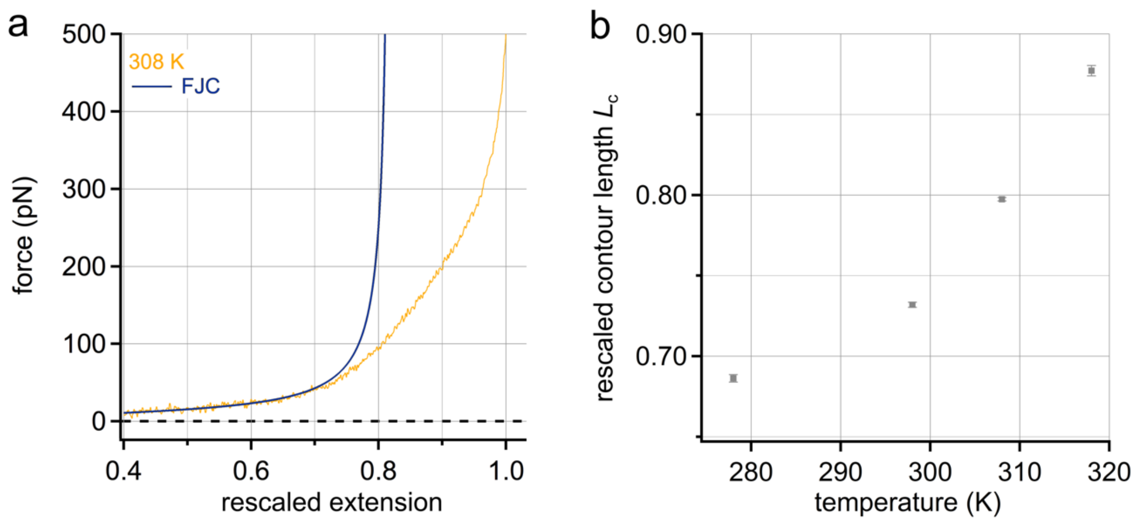

Figure S8. **a** Master curve of PEG at 308 K with an exemplary FJC fit (blue line, eq. 1). The parameter for the Kuhn length is b = 0.75 nm (see Table S1) and the fit range was chosen between 0 and 80 pN. **b** The resulting rescaled contour length Lc (with respect to the extension at a force of 500 pN) for each master curve is plotted against the respective temperature. The FJC model fits the experimental traces very well at low and intermediate forces. For higher forces the bond elasticity has to be taken into account.[6,8,9] Furthermore, fitting the force-extension curves of PEG for different temperatures using the FJC model leads to clear increase of the contour length with increasing temperature. This is consistent with the change of the fraction of monomers that are in the trans conformation, as discussed in the main text.

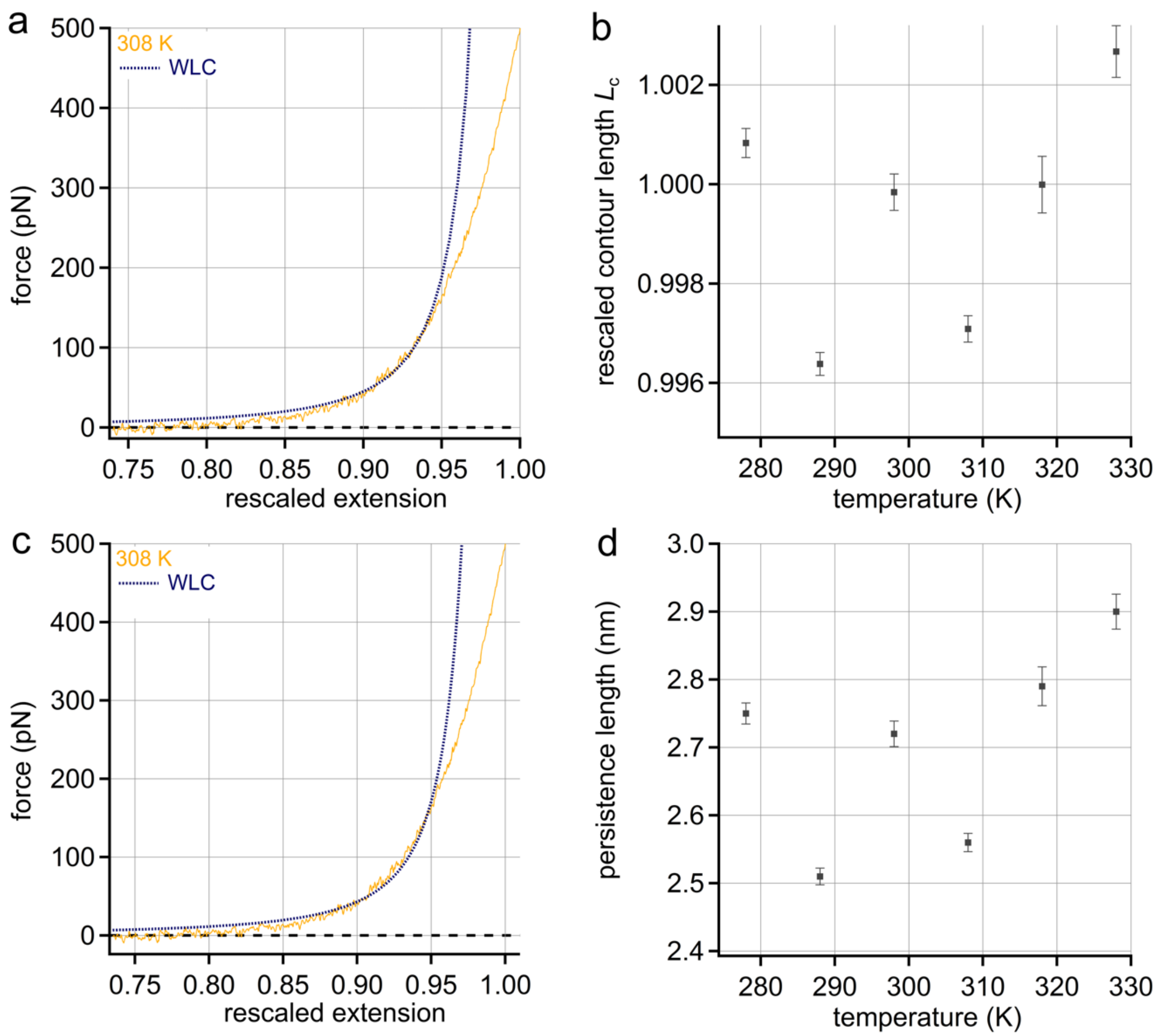

Figure S9. **a** Master curve of PNiPAM at 308 K with an exemplary WLC fit (dashed blue line, eq. 2). The parameter for the persistence length was kept constant at 2.5 nm [10] and the fit range was chosen between 0 and 80 pN. **b** The resulting rescaled contour length L_c (with respect to the extension at a force of 500 pN) for each master curve is plotted against the respective temperature. Keeping the persistence length constant there is very little variation for the rescaled contour length for different temperatures. **c** Master curve of PNiPAM at 308 K with an exemplary WLC fit (dashed blue line). The parameter for the contour length was kept constant at 0.9995 nm (average value of data points shown in b). **d** The resulting persistence length for each master curve is plotted against the respective temperature. Here we keep the rescaled contour length constant (0.9995 nm) for each of the WLC fits and obtain the respective persistence lengths for the different temperatures. In summary, the temperature-dependent stretching response of PNiPAM can neither be clearly assigned to the contour length nor to the persistence length if the WLC model is applied.

Coefficients of variation

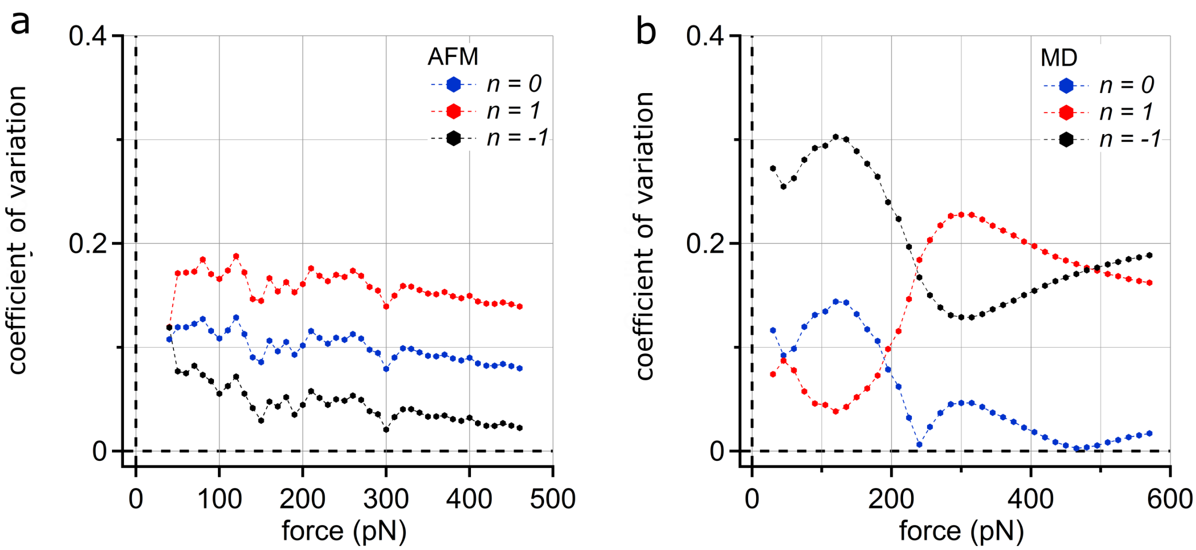

Figure S10. Coefficient of variation vs force for the PEG data given in Figure 4 for **a** AFM experiments and **b** MD simulations. The coefficient of variation indicates the standard deviation over the mean value and is presented for: the stretching free energy *F* per *Lo (*the extension at a force of 500 pN) which represents the case of n=0, the stretching free energy *F* per *Lo* divided by k_BT (n=1) and the stretching free energy *F* per *Lo* divided by k_BT^{-1} (n=-1). For experimental data the coefficient of variation was determined in steps of 10 pN while for the MD simulations each data point was used. The lines between the points are a guide to the eye.

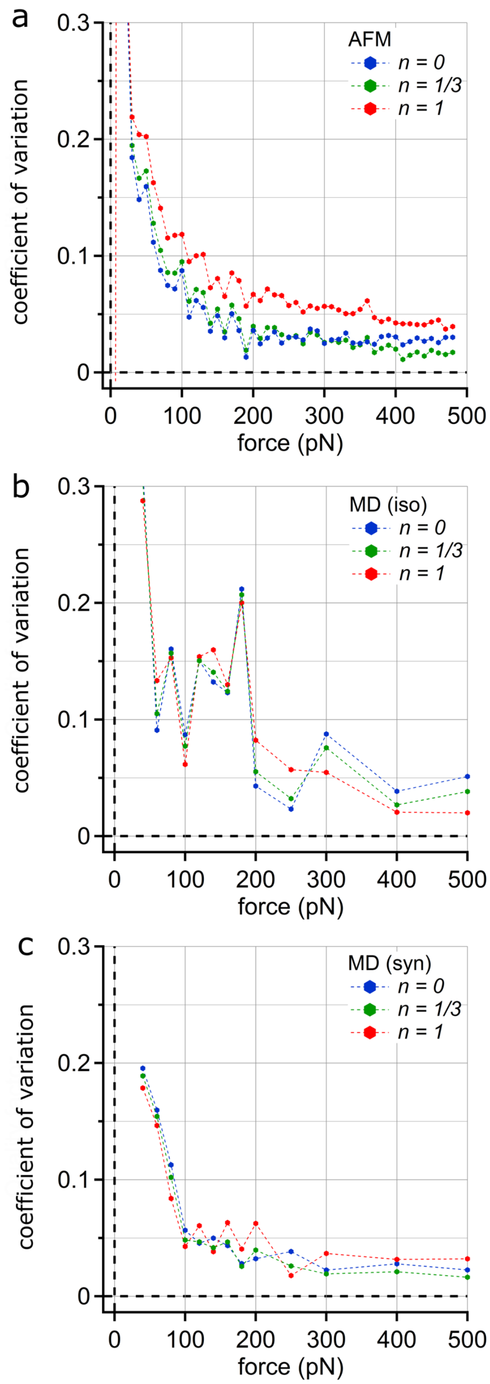

Figure S11. Coefficient of variation vs force for the PNiPAM data given in Figure 6 for **a** AFM experiments and **b** MD simulations. The coefficient of variation indicates the standard deviation over the mean value and is presented for: the stretching free energy *F* per *Lo* (the extension at a force of 500 pN) which represents the case of *n*=0, the stretching free energy *F* per *Lo* divided by k_BT (*n*=1) and the stretching free energy *F* per *Lo* divided by $k_B\,T^{1/3}$ (n=*1/3*). For experimental data the coefficient of variation was determined in steps of 10 pN while for the MD simulations each data point was used. The lines between the points are a guide to the eye.

References

(1) Djerassi, C.; Gorman, M.; Markley, F. X.; Oldenburg, E. B. Studies in Organic Sulfur Compounds. VII. 1 Lithium Aluminum Hydride Reduction of Xanthates to Mercaptans. Synthesis of Substituted β-Mercaptoethanols. *J. Am. Chem. Soc.* **1955**, *77*, 568–571.

(2) Scales, C. W.; Convertine, A. J.; McCormick, C. L. Fluorescent labeling of RAFT-generated poly(N-isopropylacrylamide) via a facile maleimide-thiol coupling reaction. *Biomacromolecules* **2006**, *7*, 1389–1392.

(3) Idota, N.; Nagase, K.; Tanaka, K.; Okano, T.; Annaka, M. Stereoregulation of Thermoresponsive Polymer Brushes by Surface-Initiated Living Radical Polymerization and the Effect of Tacticity on Surface Wettability. *Langmuir* **2010**, *26*, 17781–17784.

(4) Fixman, M.; Kovac, J. Polymer conformational statistics. III. Modified Gaussian models of stiff chains. *J. Chem. Phys.* **1973**, *58*, 1564–1568.

(5) Dittmore, A.; McIntosh, D. B.; Halliday, S.; Saleh, O. A. Single-molecule elasticity measurements of the onset of excluded volume in poly(ethylene glycol). *Phys. Rev. Lett.* **2011**, *107*, 148301.

(6) Marko, J. F.; Siggia, E. D. Stretching DNA. *Macromolecules* **1995**, *28*, 8759–8770.

(7) Flyvbjerg, H.; Petersen, H. G. Error estimates on averages of correlated data. *J. Chem. Phys.* **1998**, *91*, 461–466.

(8) Liese, S.; Gensler, M.; Krysiak, S.; Schwarzl, R.; Achazi, A.; Paulus, B.; Hugel, T.; Rabe, J. P.; Netz, R. R. Hydration Effects Turn a Highly Stretched Polymer from an Entropic into an Energetic Spring. *ACS Nano* **2017**, *11*, 702–712.

(9) Hugel, T.; Holland, N. B.; Cattani, A.; Moroder, L.; Seitz, M.; Gaub, H. E. Single-Molecule Optomechanical Cycle. *Science* **2002**, *296*, 1103–1106.

(10) Abbott, L. J.; Tucker, A. K.; Stevens, M. J. Single Chain Structure of a Poly(N-isopropylacrylamide) Surfactant in Water. *J. Phys. Chem. B* **2015**, *119*, 3837–3845.