Electronic Supporting Information for;

Ultra-Low Dispersity Poly(vinyl alcohol) Reveals Significant Dispersity Effects on Ice Recrystallization Inhibition Activity

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Experimental

Materials

Phosphate-buffered saline (PBS) solutions were prepared using preformulated tablets (Sigma-Aldrich) in 200 mL of Milli-Q water (>18.2 Ω mean resistivity) to give [NaCl] = 0.138 M, [KCl] = 0.0027 M, and pH 7.4. Vinyl acetate (>99%) was purchased from Sigma-Aldrich and was filtered through a plug of basic alumina to remove inhibitors prior to use. 4,4′-Azobis(4-cynaovaleric acid) (>98%) was recrystallized from methanol and stored at -18 °C in the dark. Carbon disulphide, methyl bromoacetate, sodium ethoxide and amberlite IR120 hydrogen form were purchased from Sigma-Aldrich. Potassium hydroxide was purchased from Fisher Scientific. All solvents were purchased from VWR or Sigma-Aldrich and used without further purification.

Physical and Analytical Methods

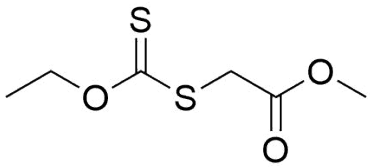
^{1}H and ^{13}C NMR spectra were recorded on Bruker DPX-300 and DPX-400 spectrometers using deuterated solvents purchased from Sigma-Aldrich. Chemical shifts are reported relative to residual non-deuterated solvent. Infrared data was recorded on a Bruker Vector 22 GI003097. ESI mass spectrometry was carried out on an Agilent 6130B single quad mass spectrometer coupled with an isocratic Agilent 1100 HPLC (without column) as an automatic sample delivery system. SEC was carried out in THF and run on an Agilent 390-LC MDS instrument equipped with differential refractive index (DRI) and dual wavelength UV detectors. The system was equipped with 2 x PLgel Mixed D columns (300 x 7.5 mm) and a PLgel 5 µm guard column. The eluent is THF with 2 % TEA (triethylamine) and 0.01 % BHT (butylated hydroxytoluene) additives. Samples were run at 1 mL.min^{-1} at 30 °C. Poly(methyl methacrylate) and polystyrene standards (Agilent EasyVials) were used for calibration. Analyte samples were filtered through a PVDF membrane with 0.22 μm pore size before injection. Respectively, experimental molar mass ($Mn_{,SEC}$) and dispersity ($Đ$) values of synthesized polymers were determined by conventional calibration using Agilent GPC/SEC software. The calibration range is 500 - 2,000,000 Da. Ice wafers were annealed on a Linkam Biological Cryostage BCS196 with T95-Linkpad system controller equipped with a LNP95-Liquid nitrogen cooling pump, using liquid nitrogen as the coolant (Linkam Scientific Instruments UK, Surrey, U.K.). An Olympus CX41 microscope equipped with a UIS-2

20x/0.45/∞/0−2/FN22 lens (Olympus Ltd., Southend on sea, U.K.) and a Canon EOS 500D SLR digital camera was used to obtain all images. Image processing was conducted using ImageJ, which is freely available from http://imagej.nih.gov/ij/.

Ice recrystallization inhibition 'splat' assay

A droplet of polymer solubilised in PBS solution was dropped from 2 m onto a glass slide cooled to -78 °C using dry ice. On impact with the glass, the droplet spreads out and freezes instantly, forming a single layer of ice crystals. The wafer is then placed in a liquid nitrogen cooled cryostage held at -8 °C. Micrographs of the ice crystals are taken at 20 x, 10 x and 4 x zoom under cross polarisers, the wafer is then left to anneal for another 30 minutes. Three further micrographs at 20 x, 10 x and 4 x zoom are taken at this point. The photos are then analysed using Image J to determine the average crystal size. The average crystal size is obtained by counting the crystals visible in the photo and dividing by the area of the photo (determined using a standard of 100 µm wide gold tracks printed on a glass slide). This value is averaged across three repeats and then compared to a standard PBS solution giving the average crystal size compared to PBS as a %, termed mean grain size (MGS).

Synthesis of methyl (ethoxy carbonothioyl) sulfanyl acetate (CTA 1)



Ethanol (140 mL) was added to a round bottomed flask equipped with a stirrer bar. Potassium hydroxide (22.9 g, 0.41 mol) was added and left to dissolve for 1 h. Carbon disulphide (24.2 mL, 0.40 mol) was then added dropwise, forming a yellow solution that was left to stir for 5 h. Methyl bromoacetate (13 mL, 0.012 mol) was added dropwise and the solution left to stir overnight. The solution was washed with cold ethanol then filtered and concentrated *in vacuo*. The crude product was partitioned in a DCM and saturated brine solution; the organic fraction then concentrated *in vacuo*. This product was then dissolved in ethyl acetate and filtered to remove impurities. Finally the product was concentrated *in vacuo* and thoroughly dried under vacuum. Yield 9.10 g 39 %. ^{1}H NMR (400 MHz, $CDCl_3$): **δ** = 1.44 (2H, t, $CH_3C\mathbf{H}_2$), 3.78 (3H, s, $C\mathbf{H}_3O$), 3.93 (2H, d, $SC\mathbf{H}_2$), 4.67 (3H, q, $C\mathbf{H}_3CH_2$). ^{13}C NMR ($CDCl_3$): δ = 13.7 (CH_2-$\mathbf{C}H_3$),

37.9 (S-**C**H_2), 61.9 (CO_2-**C**H_3), 70.6 (**C**H_2-CH_3), 167.8 (**C**=O), 212.6 (**C**=S). Other peaks due to solvent (ethanol).

Polymerisation of vinyl acetate using CTA 1

As a representative example: vinyl acetate, was filtered three times through a silica plug. The vinyl acetate (2 g, 23 mmol), CTA 1 (0.45 mg, 2.32 mmol) and ACVA ((4,4' azobis(4-cyanovaleric acid); 13 mg, 0.464 mmol) were then added to a stoppered vial containing a stirrer bar. The solution was degassed under N_2 for 20 minutes before being left to polymerise for typically 18 h at 68 °C. The reaction was quenched by plunging into liquid nitrogen and the resulting yellow solid was dissolved in the minimum amount of THF (tetrahydrofuran) and stirred into hexane and the polymer precipitated via centrifugation three times, decanting off the hexane each time. The final polymer was recovered as a sticky yellow solid after drying under vacuum. Representative characterisation data for $PVAc_{11}$: ^{1}H NMR (400 MHz, $CDCl_3$) **δ** = 2.06 (3H, br, COC**H**$_3$), 1.87 (2H, br, C**H**$_2$) 4.88 (1H, br, C**H**OCH_2); M_n^{SEC}(THF) = 1200 Da, Đ = 1.14.

Separation of poly(vinyl acetate) oligomers *via* column chromatography

As a representative example: 10 g of vinyl acetate (DP (NMR) ≈ 11) was loaded onto a silica column (6.5 cm diameter, 20 cm silica gel). Vinyl acetate oligomers were separated using a 75:25 ethyl acetate: hexane solvent. 80 fractions of approximately 20 mL were obtained. Fractions were taken until no separation could be seen via TLC. TLC spots were visualised under long wave UV light or by brief immersion in potassium permanganate solution followed by heating. The fractions were dried *in vacuo*. The mass spectrum of each fraction was obtained and used to determine Đ and purity %.

Hydrolysis of poly(vinyl acetate) to poly(vinyl alcohol)

As a representative example: vinyl acetate (0.152 g), ethanol (15 mL) and THF (5 mL) were added to a round bottomed flask equipped with a stirrer bar. The solution was cooled over ice. Sodium ethoxide (1 mL, 0.018 mol) was added dropwise forming a deep orange solution, the reaction was allowed to proceed for one hour. The solution was then neutralised by passing through a column packed with Amberlite ion exchange resin until a neutral pH was reached. The product was then dried *in vacuo*, before dissolving in deionised water and freeze drying. Impurities were removed by dissolving the freeze dried product in deionised water and filtering off any undissolved solids before freeze drying again. The final product was obtained as a

brown powder. Representative characterisation data for PVA (DP (NMR) ≈ 12, Đ 1.008): ^{1}H NMR (400 MHz, D_2O) δ = 1.62 (2H, br, C**H**$_2$), 3.95 (1H, br, C**H**OH).

Figures

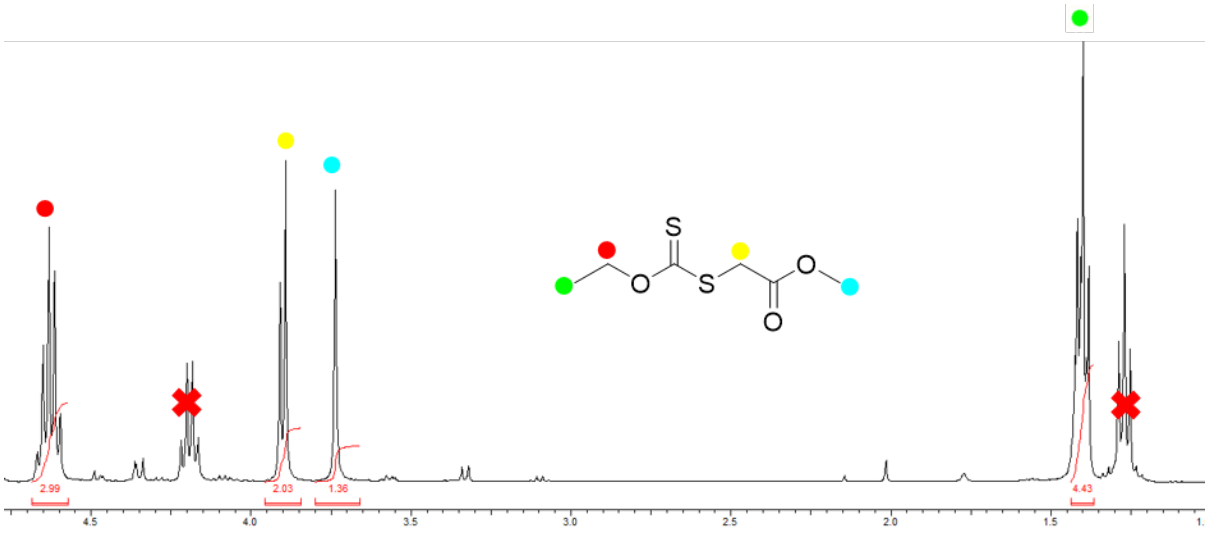


Figure S1 – Assigned ^{1}H NMR of methyl (ethoxy carbonothioyl) sulfanyl acetate (CTA)

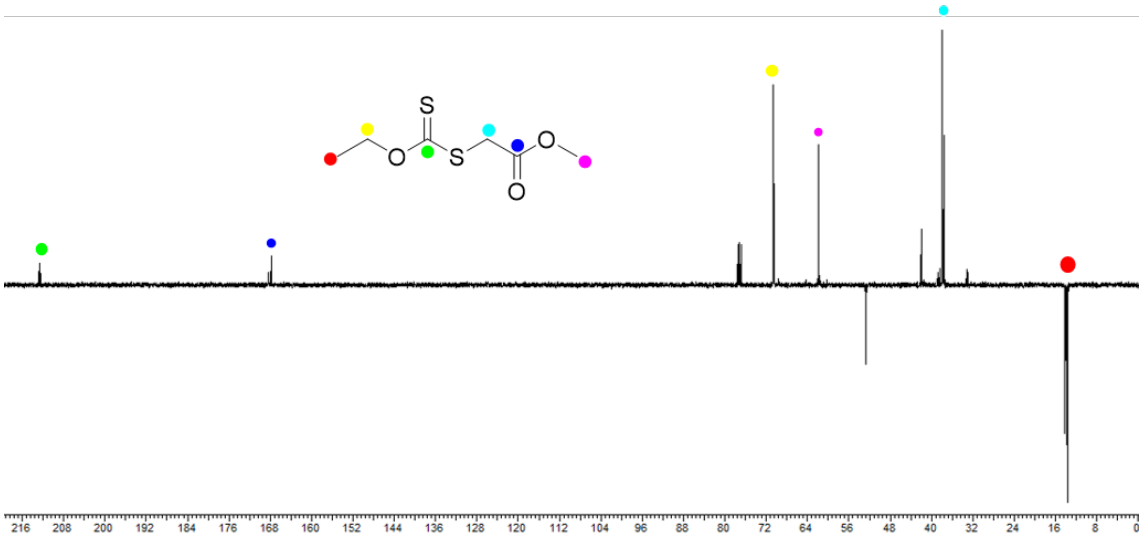


Figure S2 – Assigned ^{13}C NMR of methyl (ethoxy carbonothioyl) sulfanyl acetate (CTA)

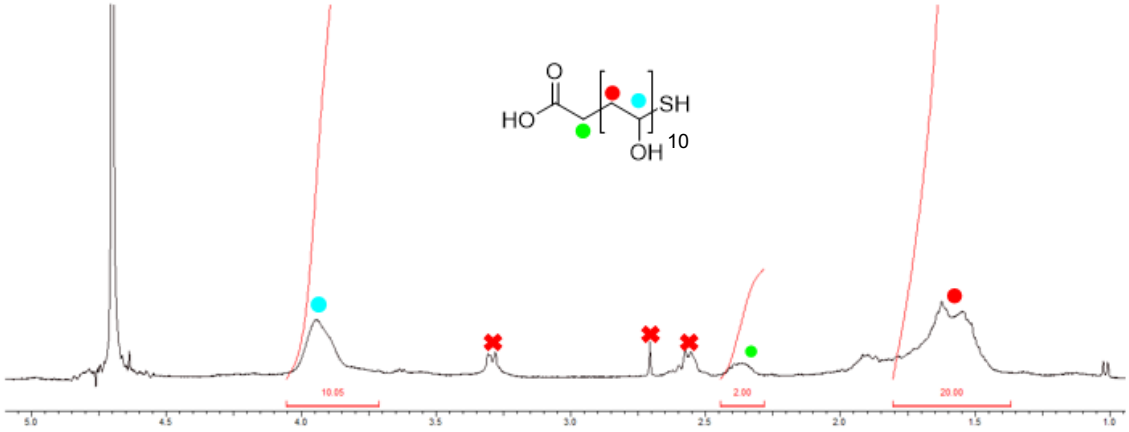


Figure S3 – ^{1}H NMR of PVA_{10}.

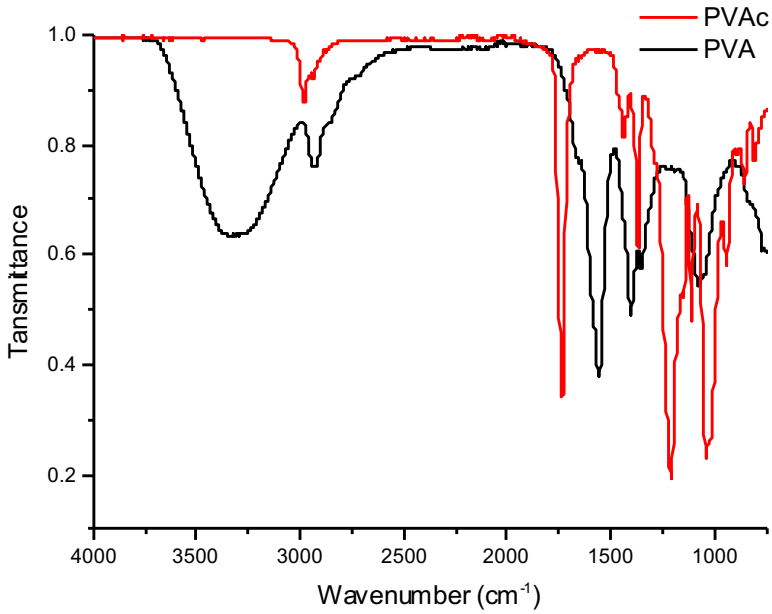


Figure S4 – IR showing the deprotection of PVAc to PVA, the removal of the acetate peak (~ 1800 cm^{-1}) and the emergence of OH peak (3300 cm^{-1}) are clearly visible.

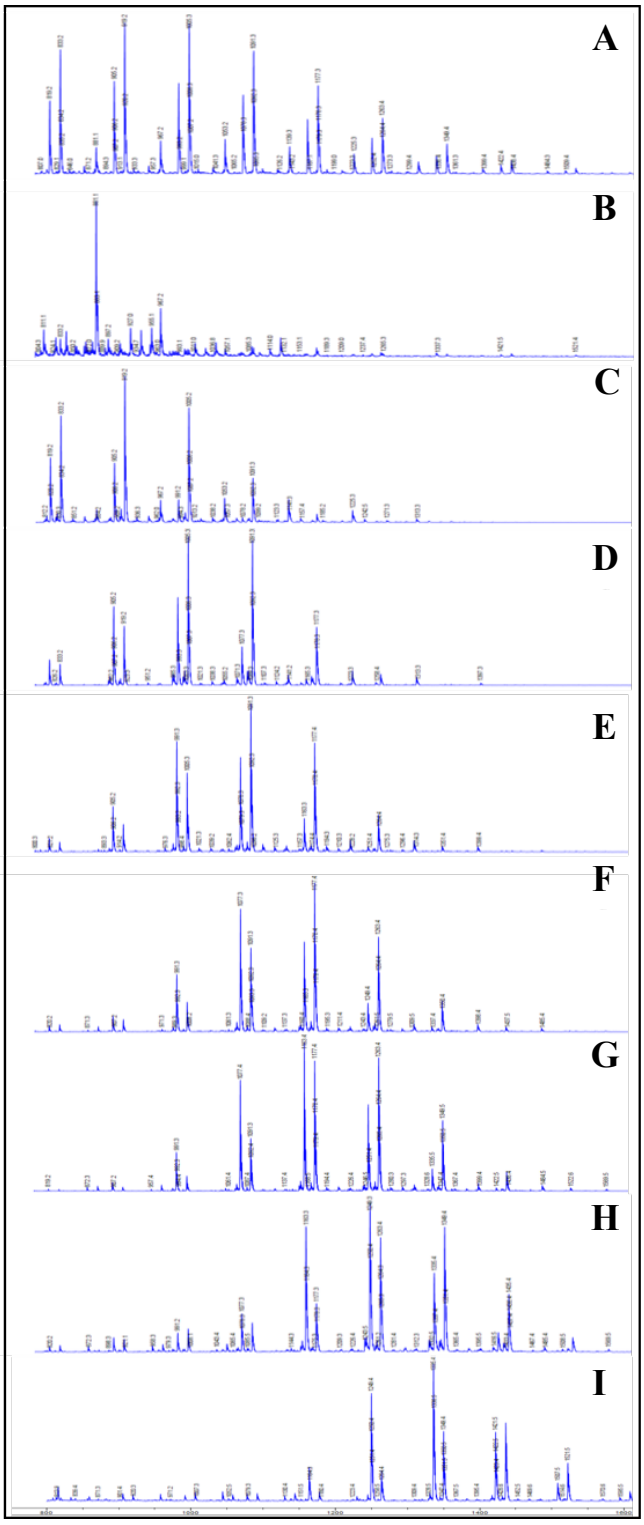


Figure S5 – Example ESI-Mass spectra of $PVAc_{16}$ (**A**) and of PVAc obtained by separation of $PVAc_{16}$ by column chromatography (**B - I**) to highlight how multiple, narrower, distributions can be isolated from a single precursor. Note – only fractions significantly enriched in oligomer (>50 %) were used for IRI testing.

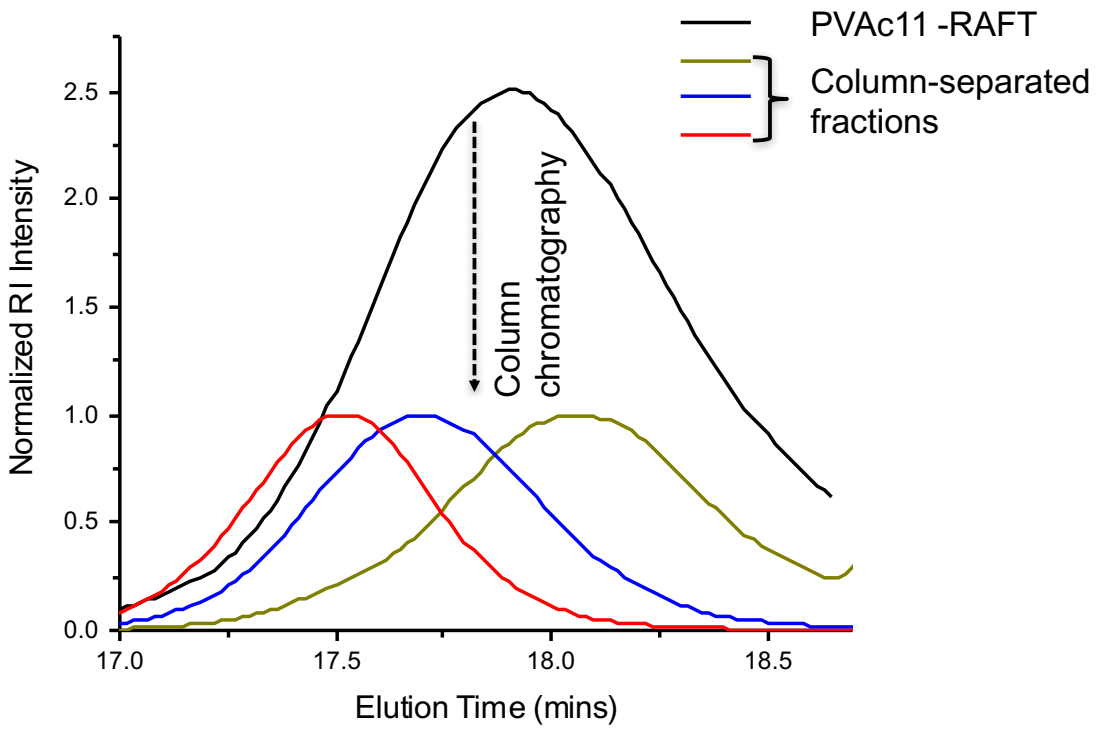


Figure S6 – Example SEC traces for PVAc (black) and PVAc fractions which have been separated by column chromatography.

Table S1: PVAc characterisation data from SEC, fractions were collected from the separation of RAFT $PVAc_{11}$, narrower dispersity was reported in each column fraction. Not SEC does not have the resolution for the ultra-low dispersities hence MS was used.

Polymer Sample	**M_n (g.mol^{-1})**	**Đ**
RAFT PVA_{11}	1200	1.14
Fraction 29	920	1.05
Fraction 34	1150	1.06
Fraction 39	1500	1.05