Structure and Properties of Poly(vinyl alcohol) Hydrogels Obtained by Freeze/Thaw Techniques

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Summary: The relationships between the structure and the viscoelastic properties of freeze/thaw PVA hydrogels obtained by repeatedly freezing and thawing dilute solutions of PVA in D_2O(11% w/w PVA) in as-prepared and rehydrated states are investigated. Our results indicate that the PVA chains and solvent molecules are organized at different hierarchical length scales, which include the presence of micro- and macro-pores, into a network scaffolding. The porous network is ensured by the presence of crystallites, which act as knots interconnected by portions of PVA chains swollen by the solvent. X-ray diffraction and SANS techniques are used to obtain structural information at short (angstroms) and medium (nanometers) ranges of length scales, concerning the crystallinity, the size of small crystalline aggregates and the average distance between crystallites in PVA hydrogels. Indirect information concerning the structural organization on the large length scales (microns) are provided by viscoelastic measurements. The dynamic shear elastic moduli at low frequency and low strain amplitude, G', are determined and related to the degree of crystallinity. These data indicate that a minimum crystallinity of 1% is required for these PVA samples to exhibit gel behaviour and have allowed obtaining the order of magnitude of the average mesh size in these gels. Finally, it is shown that the negative effect of aging, inducing worse physical and mechanical properties in these systems, may be prevented using a drying/re-hydration protocol able to keep the physical properties of the as-prepared PVA hydrogels.

Keywords: freeze/thaw technique; hydrogels; poly(vinyl alcohol); SANS; X-ray

Introduction

Poly (vinyl alcohol) (PVA) is a semicrystalline synthetic polymer able to form physically crosslinked hydrogels by different methods.[1,2] (PVA) hydrogels prepared by repeatedly freezing and thawing diluted PVA aqueous solutions have attracted much attention in the last years for their many attractive properties as for instance high water content (80-90%wt), dimensional stability at room temperature, high mechanical strength, rubber-like elasticity, lack of toxicity and biocompatibility.[1,3-6]

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A limit of PVA hydrogels for practical applications is due to the fact that these gels cannot be stored for a long time, before usage, because their physical and mechanical properties are strongly affected by aging.[7] However, the outstanding physical and mechanical properties of as-formed PVA gels can be preserved even for a long time, drying the sample immediately after the preparation (to avoid aging) and then restored when needed, upon rehydration of the dried samples.[8]

Distinct phenomena may occur during the gelation of PVA water solution through freeze/thaw cycles: phase separation, crystallization and hydrogen bonding. Liquid-liquid phase separation results in a PVA-rich and PVA-poor regions. Then, in the polymer-rich regions, due to the specific properties of PVA, crystallization and hydrogen bonding may take place. As a consequence, the gelation of aqueous PVA solutions through freeze/thaw cycles results in the formation of a porous network in which polymer crystallites act as junction points.[1-3,8-20]

The porous structure of freeze/thaw PVA hydrogels, along with their chemical and mechanical stability, makes freeze/thaw PVA hydrogels attractive matrices in a large variety of biotechnological and biomedical applications.[1,3,21]

The outstanding physical properties of PVA hydrogels derive from their complex structure, where PVA chains and the solvent molecules are organized at different hierarchical scales.

Quantitative information concerning the structural organization of PVA gels formed in mixtures of dimethyl sulfoxide (DMSO) and water on various length scales have been obtained by Kanaya *et al.* using wide[15,17]and small[15-17,19] angle neutron scattering and light scattering[16-18] techniques. These studies confirmed that cross-linking in these gels is ensured by small crystallites.

However, in spite of the numerous studies concerning the use and properties of PVA hydrogels prepared using freeze/thaw techniques, the structure of these systems is still unclear.

In this paper the relationships between the structure and the physical properties of freeze/thaw PVA hydrogels, at different numbers of freeze/thaw cycles, in as-prepared and rehydrated state is analyzed. X-ray diffraction and SANS techniques are used to obtain structural information at short and medium range of length scales, concerning the crystallinity, the size of small crystalline aggregates and the average distance between crystallites in PVA hydrogels and then the results of the structural analysis are related to the viscoelastic properties of freeze/thaw PVA hydrogels.

Experimental Section

Materials. All experiments utilized commercial grade PVA (Aldrich, ref. 36,315-4) with an average molecular weight, $\overline{M}_w$, of about 115000, and a degree of hydrolysis of 98-99%. The ^{13}C NMR spectrum analysis of PVA in deuterated water solution showed that the percentages of *mm*, *mr* and *rr* configurational triads are 22.1, 50.1 and 27.8%, respectively.

PVA hydrogels preparation. Aqueous solutions of PVA of 11%w/w concentration were prepared by dissolving the PVA polymer in deuterated water at 96° C, under reflux and stirring, for about 3 hours. The polymer was entirely dissolved and the obtained homogeneous solutions were slowly cooled to room temperature. We checked that the solutions do not jelly and remain transparent when left at room temperature in a sealed text tube for more then one month.

The freshly prepared PVA solutions were kept for one night, in order to eliminate air bubbles and then poured between glass slides with 1 mm spacers, at room temperature.

Strong physical PVA hydrogel films were obtained by subjecting the polymer aqueous solutions to several repeated freeze/thaw cycles, consisting of a 20h freezing step at –22°C followed by a 4h thawing step at 25°C. In the following sections, the as-formed PVA hydrogels obtained by 1 to 9 freeze/thaw cycles are denoted as GEL-1 to GEL-9 samples.

Dried PVA hydrogel specimens were obtained by keeping in air, at room temperature, the as-formed PVA GEL-*n* immediately after the last *n*-th freeze/thaw cycle. The drying procedure was performed until achieving a constant weight for the PVA hydrogel samples.

Rehydrated PVA hydrogel films were obtained by dipping the so obtained “dried gels” in deuterated water for 1 day (24 hours) or two weeks (14 days).

Gravimetric measurements. Polymer weight concentrations of as-formed and rehydrated PVA hydrogels were determined by weighing each sample in the swollen and in the corresponding dried state.

X-ray measurements. Wide-angle X-ray powder diffraction profiles were collected at room temperature, with a Philips diffractometer using Ni filtered CuKα radiation (λ=1.5418Å) and scans at 0.005 deg(2θ)/s in the 2θ range from 10 to 60°. In order to prevent the sample from drying during the experiment, the diffraction profiles were recorded using a home made brass sample holder placed in a special brass chamber covered with an out-of-focus Mylar film, in an atmosphere saturated with vapors of the mother solution. During the time needed for recording the diffraction patterns (≈ 3h), the weight loss of the sample was less than 2 %wt.

Apparent crystalline dimensions along the $[10\bar{1}]$ lattice direction were calculated by measuring the half-width of the corresponding Bragg reflection and applying the Scherrer formula:[22]

$$t = \frac{k \cdot \lambda}{\beta \cdot \cos\theta} \qquad (1)$$

where t is the apparent crystalline dimension along a given lattice direction, k is a constant (k= 0.89 rad), λ is the wave length of the X-rays, β is the half-width (expressed in radians) and θ is the Bragg angle. Due to the low intensity of the Bragg peak at 2θ=19.4° in the crystalline PVA hydrogels, the standard deviation associated to the so-determined apparent crystalline dimensions is of the order of 3Å.

SANS measurements. Small angle neutron scattering measurements were performed at the KWS2 facility located at the Forschungszentrum of Jülich, Germany. Samples were contained in 1 mm path length quartz cells in order to prevent the drying of gels. Measurements times ranged between 20 min to 8 h. Neutrons with an average wavelength λ of 7 Å and a wavelength spread $\Delta\lambda/\lambda \leq 0.2$ were used. A two-dimensional array detector at three different sample-to-detector distances, 2, 8 and 20 m detected neutrons scattered from the samples. These configurations permitted to collect the scattered intensity in a range of scattering vectors between 0.002 and 0.12 $Å^{-1}$.

Raw data were corrected for electronic background and empty cell scattering. Detector sensitivity corrections and transformation to absolute scattering cross sections $\left(\mathrm{d}\,\sigma(q)/\mathrm{d}\,\Omega\right)$ were made with a secondary Lupolene standard. Raw data were also corrected for intensity of background I_{bck} and intensity of empty cell I_{EC}. Data were then radially averaged and absolute scattering cross sections were obtained.

Shear modulus measurements. Oscillatory dynamic mechanical measurements were performed on PVA hydrogels in as-formed and rehydrated state (24 hours and 14 days). All these rheological measurements were performed using a strain-controlled Rheometrics RFSII rheometer equipped with parallel plates geometry (diameter 25mm).

The experiments were carried out at 25°C. Disc shaped samples (diameter 26mm, thickness ~ 0.5-1mm) of PVA gel were placed between the tools. The samples were protected from drying by a homemade cover which prevented the water from evaporating. This protection ensured the sample stability over a time period long enough (i.e. 1 hour) to perform the measurements of the shear mechanical properties. In all experiments, a weak normal force was applied on the surface of the sample discs in order to avoid the sweeping of the gel from the tool plates. This force ensured a slight compression of the sample. In the frequency sweep experiments, the shear loss (G'') and elastic (G') moduli were measured in the linear viscoelastic regime, for frequencies ranging from 1 to 50 rad/s, at a maximum strain, γ_0, of 0.1-0.6%, depending on the sample. The γ_0 value was determined by preliminary strain sweep experiments, in which the storage and loss modulus were measured as a function of strain at a fixed frequency value of 1 Hz, to check if the deformation imposed to the gel structure by the rheological experiment is entirely reversible.

Each measurement was repeated at least twice, on two different disc specimens from the same sample. The relative error on the storage modulus was of the order of 15%.

Results and Discussion

The polymer concentration in as-formed PVA hydrogels slightly increases with increasing the number of freeze/thaw cycles and, for all gels, is higher than the polymer concentration of the mother solution. It ranges from 12.0 %w/w for GEL-1 to 14.9 %w/w for GEL-9 (see Table 1).

Dried PVA gels, dipped in water, do not dissolve and are able to swell and lead to rehydrated PVA hydrogels with a water content of about 77 - 83 %wt, depending on the number of freeze/thaw cycles (see Table 1).

The X-ray diffraction profile of as-prepared GEL-9 sample along with the X-ray diffraction profile of the dried GEL-9 sample and of the 14 days rehydrated GEL-9 sample are reported in Figure 1 as an example, after subtraction of a straight base line which approximates the background contribution. For comparison, the X-ray diffraction pattern of pure deuterated water, which is the major component of these gels in the as-formed and rehydrated state, is also indicated in Figure 1a,c (dashed line).

The X-ray diffraction profiles of dried PVA hydrogels exhibit the strong diffraction maximum centered around $2\theta = 19.4°$, corresponding to the $10\bar{1}$ reflection of PVA crystals (see Figure 1b, as an example).[8,23]

Table 1. Polymer concentration, total D_2O content, fractional amount of free D_2O, degree of crystallinity (x_c), fraction of swollen amorphous PVA, fraction of crystalline PVA with respect to the sum of the crystalline fraction and the swollen amorphous component (f_c) and apparent crystallite dimensions (along the $[10\bar{1}]$ lattice direction) of freshly prepared PVA/D_2O gels obtained by different numbers of freeze/thaw cycles (fresh) and of gels samples dried immediately after their preparation and dipped in D_2O for 14 days (rehydrated).

gel sample	n° freeze/thaw cycles	gel state	polymer concentration[a] (%w/w)	total D_2O[a] (%w/w)	free D_2O[b] (%)	x_c[b] (%)	swollen amorphous[b] (%)	f_c[b] (%)	Apparent dimensions of crystallites[b] (Å)
GEL-1	1	fresh	12.0	88.0	86.2	0.4	13.4	2.5	28
		rehydrated	17.2	82.8	69.6	1.1	29.0	3.7	39
GEL-3	3	fresh	12.7	87.3	84.2	0.8	15.0	4.8	34
		rehydrated	23.4	76.6	69.8	2.8	27.4	9.2	46
GEL-5	5	fresh	13.4	86.6	80.9	1.1	18.0	5.6	35
GEL-7	7	fresh	14.7	85.3	80.1	1.1	18.8	5.7	38
GEL-8	8	fresh	13.7	86.3	78.5	1.4	20.2	6.3	40
GEL-9	9	fresh	14.9	85.1	78.1	1.4	20.6	6.3	39
		rehydrated	23.3	76.7	72.6	2.9	24.5	10.6	50

[a]Determined by gravimetric measurements. [b]Determined by X-ray powder diffraction experiments.

The diffraction profiles of the as-formed and 14 days rehydrated GEL-9 samples (Figure 1a,c) exhibit two halos centered at 2θ ≈ 28 and 41°, as in the diffraction profile of pure water, and a weak peak in the 2θ range 18 - 21° which corresponds to the $10\bar{1}$ reflection of crystalline PVA (Figure1b).

This result demonstrates the presence of a low amount of small crystalline PVA aggregates in the gel samples in freshly prepared and rehydrated state.

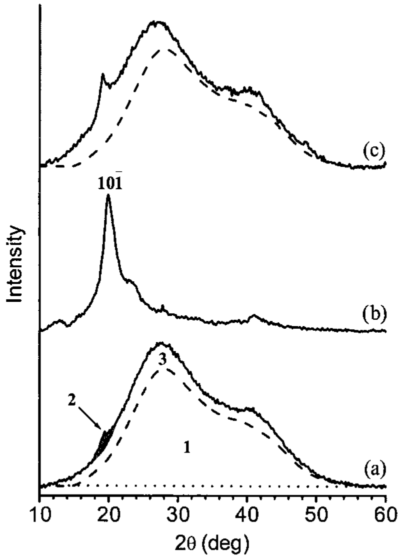

Figure 1. X-ray powder diffraction profiles of freshly prepared PVA hydrogel sample (a), of dried gel (b) and of 14 days rehydrated gel (c), obtained after 9 freeze/thaw cycles (continuous line). The X-ray diffraction profile of liquid D_2O is also reported (dashed line). The crystalline reflection 2 in 2θ range 18 - 21° in (a) is evidenced in grey, whereas the $10\bar{1}$ reflection of the crystalline PVA is indicated in (b).

The X-ray diffraction profile of GEL-n samples (Figure 1a) is considered as the sum of three contributions: a large contribution (area A1) due to the scattering of pure D_2O (dashed curves), a small diffraction component in the range from 18 to 21° due to the crystalline aggregates (area A2) and a third component, (area A3) due to the presence of amorphous PVA swollen by water molecules.

The relative amount of "free D_2O" in the gels, the degree of crystallinity (x_c), the fraction of the swollen amorphous PVA phase and the relative amount of crystalline PVA with respect to the sum of the crystalline and swollen amorphous portions, f_c, may be determined by measuring the areas A1, A2 and A3, as the ratios A1/(A1+A2+A3), A2/(A1+A2+A3), A3/(A1+A2+A3) and A2/(A2+A3), respectively. These values thus obtained and the apparent crystalline dimensions along the $[10\bar{1}]$ lattice direction, determined using the Scherrer formula[22], are reported in Table 1.

In freshly prepared gels, the relative amount of "free water" decreases with increasing the number of freeze/thaw cycles (n), whereas in rehydrated samples is nearly constant (≈70%). In both kinds of gels the degree of crystallinity increases with increasing n, up to a plateau for n = 5.

Moreover, for freshly prepared gels the apparent crystalline dimensions (along the $[10\bar{1}]$ lattice direction) increase with n, ranging from 28 Å for GEL-1 to 40 Å for GEL-9. It is worth noting that, going from GEL-1 to GEL-9, the degree of crystallinity, f_c, increases from 2.5 to 6.3% whereas the polymer concentration slightly increases from 12 to 15 %w/w (see Table 1).

The degrees of crystallinity, f_c, in rehydrated GEL-1, GEL-3 and GEL-9 are 1.1, 2.8 and 2.9 %, respectively; they are slightly higher than the degree of crystallinity of the corresponding as-formed gels (see Table 1). Moreover, the apparent dimensions of crystallites of rehydrated PVA hydrogels slightly increase during rehydration (see Table 1).

Our structural analysis supports previous models proposed in the literature,[2,12] which describe the structure of freeze/thaw PVA hydrogels in terms of a porous polymer network where the crystals act as knots, the polymer segments ensure the connectivity all over the macroscopic gel sample, while free water fills the pores. Water also acts as a swelling agent in the disordered zones of polymer matrix forming hydrogen bonds with the OH groups of PVA chains. The porous walls consist of swollen amorphous PVA. The crystalline knots ensures high dimensional stability and elastic properties of the gel.

The quenching at low temperature, during the freezing step, induces a liquid-liquid phase separation and the formation of ice crystals in polymer-poor phase. The ice crystals, in turn, expel amorphous polymer segments, increasing the polymer concentration in the surrounding environment. The size of ice crystals formed in the polymer-depleted pockets increases through the repeated freeze/thaw cycles. The size of phase-separated domains is of the order of microns, thus accounting for the opaque aspects of PVA hydrogels. Upon thawing, the ice crystals melt and leave the porous structure of the hydrogel unaltered. Practically, water works as a porosigen in PVA solution whereas the polymer network increases its stability during the freezing step.[1,3,24,25]

It is worth noting that, during rehydration of dried PVA gels in D_2O, a certain amount of polymer is released in the solvent. For GEL-1, during the swelling step of the dried PVA hydrogel, the percentage of PVA which is dissolved in D_2O after 2 weeks is of 21.5 %wt whereas for GEL-9, the amount of dissolved PVA does not exceed 4 %wt. This difference in behaviour could be due to the fact that an increase of the number of freeze/thaw cycles improves the stability of the whole gel structure and consequently induces a minor solubility of the PVA chains embedded in the network.

SANS profiles obtained for as-formed PVA GEL-1 and GEL-9 are reported in Figure 2. The SANS profile of the PVA/D_2O (11%w/w) starting solution used for gel preparation is also shown in Figure 2A.

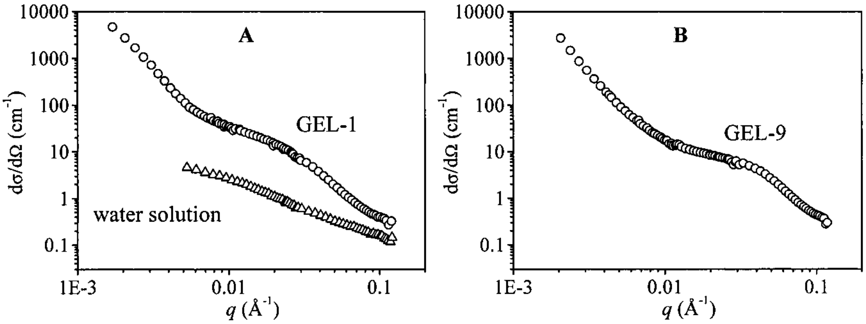

Figure 2. SANS data for (A) as-prepared GEL-1 (O) and PVA solution (Δ) and (B) as-prepared GEL-9 (O).

The scattering cross section profiles ($^{d\sigma}/_{d\Omega}$) of GEL-n sample present a background higher than the profile of mother solution, mainly due to incoherent scattering contribution. The differences of background are fully accounted for by the different polymer concentration of the various samples (see Table 1).

Inspection of the Figures 2 indicate that SANS data obtained for the PVA starting solution are consistent with the presence of small scattering objects, essentially the individual chains of polymer with a Gaussian coil conformation. Moreover, the scattering cross-section profile of the homogeneous starting solution appears quite different from those of PVA GEL-n samples confirming that, in the gels, PVA chains and solvent molecules are highly organized. In the SANS profiles of the gels we can distinguish three different regions:

1) A region at low q values ($q < 0.009$Å^{-1}). In this region the scattering cross section exhibits an upturn, which is clearly not present in the scattering cross section curve of the homogeneous starting solution. In this zone, the data could reflect a supramolecular organization, which we associate to the presence of two separated phases constituted by polymer-rich and polymer-poor regions.
2) A region at intermediate q values ($0.009 < q < 0.035$Å^{-1}). In this region an inflexion point is present which gives an indication of the average distance between the scattering crystallites (given approximately by $2\pi/q$). In all gel samples this inflexion point is at $q \approx 0.03$Å^{-1}, indicating a distance between crystallites of the order of 200 Å.
3) A region where $0.035 < q < 0.08$Å^{-1}. In this zone, scattering cross sections decrease with a power law $\mathrm{d}\sigma/\mathrm{d}\Omega \propto q^{-D}$, the value of D depending on the number of cycles. More precisely $D \cong 2$ for GEL-1, $D \cong 3$ for GEL-9. Provided we are looking in this region at the boundary structure between two phases, possibly the crystallites and the swollen amorphous in the polymer rich phase, we may apply the surface fractal concept to the function $\mathrm{d}\sigma/\mathrm{d}\Omega$. According to this concept, the exponent D is related to the surface fractal dimension d_s in a d-dimensional space, through $D = 2d - d_s$. For example, in 3-dimensional space, d_s ranges from 2 to 3, corresponding to a range of D from 4 to 3. If the boundary were smooth, Porod's law ($\mathrm{d}\sigma/\mathrm{d}\Omega \propto q^{-4}$) would be observed. For our PVA gels, values of D less than 4 suggest that the boundary is not very clear, or it would not be a boundary in the case of GEL-1.

Oscillatory dynamic mechanical measurements were performed on PVA GEL-n in the fresh[13] and rehydrated (24 hours and 14 days) states as a function of n.

The frequency sweep experiments show that both G' and G" moduli of rehydrated GEL-n do not depend on frequency in the range between 1 and 50 rad/s.[14] Moreover, the storage modulus (G') is always higher than the loss modulus (G"). This result indicates that the rehydrated PVA GEL-n samples behave like a highly elastic gel, in agreement with the existence of a network structure and with the results obtained for as-formed GEL-n samples in similar experiments.[13]

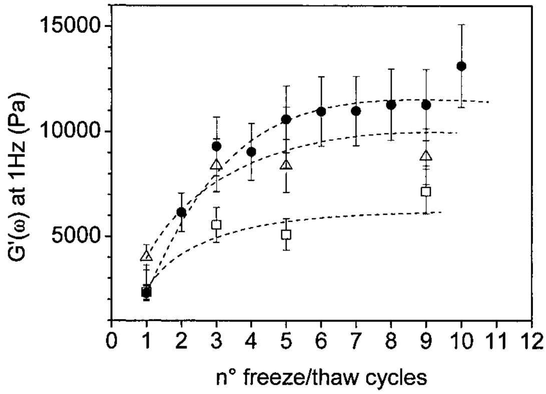

Figure 3. Storage modulus, *G'*, at 1Hz, as a function of the number of freeze/thaw cycles, for PVA hydrogels: as-formed (●); 24h hours rehydrated in D_2O (□); 14 days rehydrated in D_2O (△). Fixed strain amplitude of 0.3-0.6% for rehydrated gel samples and of 0.1% for as-formed samples.[13]

The values of the storage modulus, G', determined at 1 Hz for the 24 hours and 14 days rehydrated PVA GEL-n samples are reported in Figure 3 and compared to those of the corresponding as-formed PVA GEL-n,[13] as a function of the number of freeze/thaw cycles, n. The storage modulus, G', for all rehydrated GEL-n samples, increases with increasing the number of freeze/thaw cycles, tending to a plateau value after the first 3-5 freeze/thaw cycles. The storage modulus, G' for 24 hours rehydrated GEL-n samples is lower than that of freshly prepared samples, except for the GEL-1, for which the storage modulus in the freshly prepared state and after 24 hours rehydration of dried samples are similar. The dried GEL-n samples, at higher number of freeze/thaw cycles, n, need a 14 days rehydration to give G' values comparable to those in the as-prepared state. In the case of GEL-1 sample, 14 days rehydration, instead, results in higher G' values than the G' value in the freshly prepared state.

The drying/ 14 days rehydration procedure gives rise to rehydrated GEL-n samples with G' values comparable to (for n higher than 1) or higher (for n equal to 1) than those in as-formed state.

The relationships between the network structure of the PVA hydrogels and their rheological behavior can be better understood in Figure 4 where the storage modulus values at 1 Hz, G' are plotted as a function of the degree of crystallinity, f_c, determined from X-ray diffraction analysis.

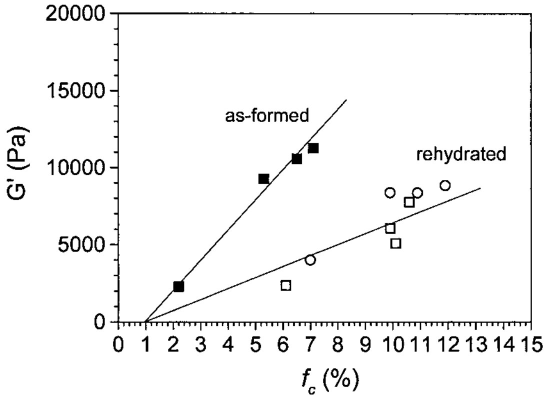

Figure 4. Storage modulus, G', at 1 Hz, as a function of the degree of crystallinity, f_c, for PVA hydrogels in the as-formed state (■), 24 hours rehydrated state (□) and 14 days rehydrated state (○).

For a given PVA concentration, the storage modulus, G', strongly increases as sample crystallinity increases. As a first approximation, the three data sets obtained for the PVA hydrogels (in the fresh, and 24h and 14 days rehydrated states) can be fitted to straight lines. Extrapolation of these lines to G' equal to zero leads to a common value of the sample crystallinity, on the order of 1%. This result indicates that a minimum crystallinity is required for these PVA samples to exhibit gel behavior.

In the hypothesis that our gels may be regarded as a classic network, in spite of the fact that they present a complex hierarchical structure, the storage modulus determined at low strain amplitude and low frequency of our gels (i.e. in the fully elastic, reversible regime), G', may be related to the average number of equivalent units in a "network strand", N, connecting two "ideal" junctions and to the approximate, average value of the network mesh size L_C by use of Equations 2 and 3:[26]

$$G' = \frac{RT}{N_{Av}a^3N}\phi^{1/3} \qquad (2)$$

$$L_c = (\phi)^{-1/3}(C_\infty N)^{1/2}a \qquad (3)$$

In Equation 2 N_{Av} is the Avogadro number, G' is the shear modulus of PVA gel samples, R is the gas constant, T the absolute temperature, ϕ the polymer volume fractions of gel in the swollen state, $N_{Av}\,a^3$ is the molar volume of the solvent, N is the average number of equivalent units with volume equal to the solvent volume (a^3), comprised between two junctions (in a "network strand").[27] For deuterated water $N_{Av}\,a^3$ is of the order of 18cm^{3}mol^{-1}. In Equation 3 C_∞ is the characteristic ratio of PVA, equal to 8.3.[28]

Values of L_c thus obtained are of the order of 150-200 nm. They indicate that the network mesh size of the gels are $\approx$10 times higher than the average correlation distance between the fringed micelle-like crystallites ($\approx$20 nm), determined in SANS analysis.[15,29] Such large values of L_c, indeed, reflect the microscopic heterogeneity of freeze/thaw PVA hydrogels originating from their phase-separated nature, including pores of various dimensions (from nanometers to µm[12,30,31]), highly interconnected through the network scaffolding ensured by the PVA chains.

Conclusions

The structural organization of poly(vinyl alcohol) (PVA) hydrogels obtained by repeatedly freezing and thawing dilute solutions of PVA in D_2O(11% w/w PVA) is investigated by use of X-Ray diffraction and small angle neutron scattering measurements (SANS), as a function of the number of cycles, n. The results of the structural analysis are related to the viscoelastic properties of freeze/thaw PVA hydrogels. The structural analysis techniques and the viscoelastic measurements have been also extended to PVA hydrogels obtained by rehydrating in D_2O the dried samples.

Our results indicate that in freeze/thaw PVA hydrogels the polymer chains and solvent molecules are organized at different hierarchical length scales. The structural organization on the length scales of the order of micron originates from the presence of two separated phases constituted by polymer-rich and polymer-poor regions. The organization on the medium length scales (nanometers) is provided by the presence of small crystallites, highly connected by swollen amorphous tie chains, within the polymer-rich phase. The presence of these tie chains ensures the connectivity of the macroscopic network. The structural organization on the short length scale (angstroms) is essentially provided by the relative arrangement of chains within the crystallites and in the swollen amorphous zones.

It is shown that drying the freeze/thaw hydrogels immediately after their preparation and rehydrating the dried gels for a time long enough (~ 2 weeks) the structure and properties of PVA hydrogels in the fresh state are restored almost completely avoiding the negative and unpredictable effects of aging.

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[1] Hassan, C.M.; Peppas, N.A., *Advances in Polymer Science*, **2000**, *153*, 37.
[2] Yokoyama, F.; Masada, I.; Shimamura, K.; Ikawa, T.; Monobe, K., *Colloid and Polymer Science*, **1986**, *264*, 595.
[3] Lozinsky, V.I., *Russian Chemical Reviews*, **1998**, *67*, 573.
[4] Urushizaki, F.; Yamaguchi, H.; Nakamura, K.; Numajiri, S.; Sugibayashi, K.; Morimoto, Y., *International Journal of Pharmaceutics*, **1990**, *58*, 135.
[5] Stauffer, S.R.; Peppas, N.A., *Polymer*, **1992**, *33*, 3932.
[6] Peppas, N.A.; Scott, J.E., *Journal of Controlled Release*, **1992**, *18*, 95.
[7] Guenet, J.M., in *Thermoreversible Gelation of Polymers and Biopolymers*, B. R, Editor. 1992: San Diego.
[8] Ricciardi, R.; Auriemma, F.; De Rosa, C.; Lauprêtre, F., *Macromolecules*, **2004**, *37*, 1921.
[9] Komatsu, M.; Inoue, T.; Miyasaka, K., *Journal of Polymer Science: Polymer Physics Edition*, **1986**, *24*, 303.
[10] Watase, M.; Nishinari, K., *Journal of Polymer Science:Part B: Polymer Physics Edition*, **1985**, *23*, 1803.
[11] Watase, M.; Nishinari, K., *Makromol. Chem.*, **1989**, *190*, 155.
[12] Willcox, P.J.; Howie, D.W., JR.; Schimdt-Rohr, K.; Hoagland, A.; Gido, S.P.; Pudjijanto, S.; Kleiner, L.W.; Venkatraman, S., *Journal of Polymer Science:Part B: Polymer Physics*, **1999**, *37*, 3438.
[13] Ricciardi, R.; Gaillet, C.; Ducouret, G.; Lafuma, F.; Lauprêtre, F., *Polymer*, **2003**, *44*, 3375.

[14] Ricciardi, R.; D'Errico, G.; Auriemma, F.; Ducouret, G.; Tedeschi, A.; De Rosa, C.; Lauprêtre, F.; Lafuma, F., *Chem. Mat. submitted.*

[15] Kanaya, T.; Ohkura, M.; Kaji, H.; Furusaka, M.; Misawa, M., *Macromolecules*, **1994**, *27*, 5609.

[16] Kanaya, T.; Ohkura, M.; Takeshita, H.; Kaji, H.; Furusaka, M.; Yamaoka, H.; Wignall, G.D., *Macromolecules*, **1995**, *28*, 3168.

[17] Kanaya, T.; Takeshita, H.; Nishikoji, Y.; Ohkura, M.; Nishida, K.; Kaji, K., *Supramolecular Science*, **1998**, *5*, 215.

[18] Takeshita, H.; Kanaya, T.; Nishida, K.; Kaji, K., *Macromolecules*, **1999**, *32*, 7815.

[19] Takeshita, H.; Kanaya, T.; Nishida, K.; Kaji, K., *Physica B*, **2002**, *311*, 78.

[20] Takeshita, H.; Kanaya, T.; Nishida, K.; Kaji, K.; Takahashi, T.; Hashimoto, M., *Physical Review E*, **2000**, *61*, 2125.

[21] Lozinsky, V.I., *Russian Chemical Reviews*, **2002**, *71*, 489.

[22] Klug, H.P.; Alexander, L.E., in *X-ray diffraction Procedures*, J.W. Sons, Editor. 1959: New York. p. 512.

[23] Bunn, C.W., *Nature*, **1948**, *161*, 929.

[24] Chen, J.; Park, H.; Park, K., *J. Biomed. Mater. Res.*, **1999**, *44*, 53.

[25] Oxley, H.R.; Corkhill, P.H.; Fitton, J.H.; Tighe, B.J., *Biomaterials*, **1993**, *14*, 1065.

[26] Flory, P.J., *Principles of Polymer Chemistry*, **1953**, *Cornell University Press*, Ithaca NY.

[27] Rubinstein, M.; Colby, R.H., *Polymer Physics*, **2003**, *Oxford University Press*,

[28] Brandrup, J.; Immergut, E.H.; Grulke, E.A., *Polymer Handbook*. 1999, fourth edition: John Wiley & Sons, Inc.

[29] Ricciardi, R.; Mangiapia, G.; Lo Celso, F.; Paduano, L.; Triolo, R.; Auriemma, F.; De Rosa, C.; Lauprêtre, F., *Chem. Mat.* **2005**, *17*, 1183.

[30] Lozinsky, V.I.; Plieva, F.M., *Enzyme and Microbial Technology*, **1998**, *23*, 227.

[31] Lozinsky, V.I.; Galaev, I.Y.; Plieva, F.M.; Savina, I.N.; Jungvid, H.; Mattiasson, B., *Trends in Biotechnology*, **2003**, *21*, 445.