Nanogels formation on theta solution using EB radiation

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Abstract

Nano-hydrogels made of bio-compatible hydrophilic polymers can be used in various medical applications such as drug delivery, imaging, etc. Preliminary results on the synthesis of poly(vinyl pyrrolidone) (PVP) nanogels in dilute theta solutions using electron beam, will be presented here. Radiation-induced synthesis of polymer nanogels, based on intra-molecular crosslinking process, was studied using dynamic light scattering. It was observed polymeric size reduction with radiation dose increase, indicating nanogel formation.

Structures in nanometer scale has gained interest due to their unique properties and performance resulting from their small size. Soft materials, such as polymers gels can also be prepared to form nano-structured products. Polymer gels may be categorized by their dimensions as either nano- or macroscopic gels, and each type may be selectively produced through the control of gel-forming crosslinking reactions used to produce them. According to Jung-Chul An[1] the inter-molecular crosslinking process which yields in macroscopic gels has been extensively studied and is relatively well understood. However, the radiation-induced synthesis of polymer nanogels, which is based on an intramolecular crosslinking process, is less explored. For example, pulsed high-energy electron irradiation has been demonstrated as one synthesis method for the intra-molecularly crosslinked PVP. A discrete decrease of the PVP gyration radius, R_g, was demonstrated in concert with the maintenance of a constant PVP molecular weight (Mw)[1]. Understanding the mechanism of crosslinking formation is very important in order to control the final, physical properties (size, conformation, molecular weight, crosslinking density) of the nanohydrogel produced. Soft materials, such as polymer gels that exhibit mutability and responsiveness to their surroundings can be engineered to form nanostructured products. The mutability and responsiveness of these materials have led to a great deal of research interest for potential applications including drug-delivery carriers, sensors, nanoreactors, and bio-mimetic mechanical devices (e.g., artificial muscle)[1-4]. Nano-hydrogels made of bio-compatible hydrophilic polymers can be used in various medical applications such as drug delivery and imaging. Intravenously introduced hydrogel-drug conjugate (10 - 200nm particles) can be effectively accumulated in tissues/organs by prolonged circulation and can be selectively transported into tumor tissues by the EPR (enhanced permeability and retention) effect. Al-Sheikhly[2] has investigated the radiation-induced synthesis of functionalized polymer nano-hydrogels that can serve as targeted nano-medicine carriers. PVP is one of the most broadly studied hydrophilic materials due to its biocompatibility and has been applied in pharmaceuticals, biomedical sciences, foods, and cosmetics[1].

The latest results on the synthesis of poly(vinyl-1-pyrrolidone) (PVP) nanogels in dilute theta solutions using γ-rays and electron beams, will be presented in this study.

2. Experimental
2.1. Material
Poly(vinyl-1-pirrolydone), poly-[1-(2-oxo-1-pyrrolidyl)-ethylene], PVP, molecular weight 1.2×10^6 (K-90) from GAF Co.

2.2. Solution preparation
PVP K-90 weight was measured in a Mettler Toledo AB 304-S. Solution was prepared by adding distilled water and rest one night to complete volume of analytical flask. Theta solution (acetone:water 66.7:33.3) was prepared adding acetone with agitation to the aliquot of PVP water solution corresponding to the concentration of 10mg of PVP in 1 mL of solution.
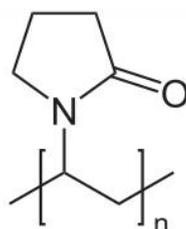Figure 1 shows poly(vinyl-1-pirrolydone) structural formula.

Figure 1. – Structural formula of poly(vinyl-1-pirrolydone), poly-[1-(2-oxo-1-pyrrolidyl)-ethylene].

2.3. Irradiation
Irradiation processing of samples was performed at IPEN. PVP theta solution was distributed in quartz flasks with tap to avoid acetone evaporation for gamma irradiation; and Petri dishes with 2mm solution height for EB irradiation.

2.3.1. Gamma
Multipurpose Co-60 Gamma Irradiation Facility. Radiation doses: 5kGy, 10kGy and 15 kGy; dose rate of 8 kGy h^{-1}.
Gammacell irradiator. Radiation doses: 5kGy, 10kGy and 15 kGy; dose rate of 2 kGy h^{-1}.

2.3.2. Electrons
Electron Beam Accelerator Dynamitron (E = 1.4 MeV, i=3mA) from Radiation Dynamics Inc. Radiation doses of 5kGy and 10 kGy; dose rate of 11 kGy·s^{-1}.

2.4. Dynamic Light Scattering
Hydrodynamic diameter of non irradiated and ionizing radiation irradiated PVP dilute samples were obtained using Zetasizer Nano ZS90 from Malvern Instruments of IF-USP; range 0.3nm to 5μm; measurement angle 90°; at room temperature (25^oC); 633nm He-Ne laser. It was used quartz cuvette. R_h was calculated dividing diameter value by two.

3. Results and discussion
According to Joanny[5], the mean field theory, implicitly assumes that polymer chains have a Gaussian structure at short length scales. This is true in a θ solvent or a poor solvent, and the, Flory-Huggins theory well describes the experimental results in these situations. In this study PVP dilute solution is θ solvent, so last theory would be applicable.

According to Abdel-Azim[6] et al using binary liquid mixtures as solvents for a polymer introduces theoretical and experimental problems of various kinds, some of them similar to those for

single solvents, and other specific of ternary systems, such as unperturbed dimension variations. The authors have studied the effect of solvent on the unperturbed dimensions of PVP in a series of mixed solvents. It has been established in the literature that water/acetone binary mixture having the volume fraction of acetone equal to 0.668 is θ-solvent for PVP at 298.15 K. In this study it is desirable that polymeric chains suffer alteration on dimension in order to favor intramolecular crosslinks by ionizing radiation processing.

According to Stevens[7] assuming polymer molecules of a given molecular weight are fully separated from one to another by solvent, the hydrodynamic volume will depend on a variety of factors, including interactions between solvent and polymer molecules, conformational effects arising from the polarity and steric bulk of the substituent groups, and restricted rotation caused by resonance. Because of Brownian motion, molecules are changing shape continuously. If a molecule were fully extended, its size could be computed from knowledge of bond lengths and bond angles. This is not the case with most of common polymers; therefore size is generally expressed in terms of the mean-square average distance between chain ends, for a linear polymer, or the square average radius of gyration about the center of gravity for a branched polymer. The average shape of the coiled molecule is spherical. The greater the affinity of solvent for polymer, the larger will be the sphere, that is, the hydrodynamic volume. As the solvent-polymer interaction decreases, intramolecular interactions become more important, leading to a contraction of the hydrodynamic volume. At the theta state the polymer is on the brink of becoming insoluble, where the solvent is having a minimal solvation effect on the dissolved molecules. Any further diminution of this effect causes the attractive forces between polymer molecules to predominate, and the polymer precipitates[7].

According to Joanny[8], as the concentration of an extremely dilute polymer solution increases, the volume fraction occupied by the chains in the solution becomes larger and interchain interactions are more and more relevant. Chains start to overlap and one cannot consider them individually, their behavior becomes cooperative[9]. Concerning the hydrodynamic properties of semi-dilute solutions, Rouse free-draining model is based on a local response of the monomers, however, it is well known that the motion of each monomer creates a backflow velocity field in the solvent that decays very slowly with the distance. The hydrodynamic interactions are taken into account in a modified version of the Rouse model, the Zimm model. The result is that because of the hydrodynamic interactions, the flow field does not penetrate the polymer chain and the polymer moves as a whole like an equivalent sphere with a hydrodynamic radius R_h, proportional to its radius of gyration R_G.

It was observed in the literature that at temperatures above 60°C PVP chains start to collapse decreasing its average hydrodynamic radius, R_h from 23 (at 20°C) to 15.6 nm (at 80°C) due to the disruption of polymer-water hydrogen bonds. The collapsed form of the PVP molecules enhances the intra-crosslinking reactions of the radiolytically produced free radicals leading to a further decrease in its average R_h to the value of 14 nm (γ-ray irradiation with 10 kGy)[3]. According to Jung-Chul An[3] the conformation of PVP chains in aqueous solution is sensitive to temperature. As temperature increases, polymer–water hydrogen bonds (e.g., between hydrogen atoms from water and the oxygen atoms of the carbonyl groups of PVP) are destroyed, thereby leading to collapsed chains with greater stiffness. A mixture of intra- and inter-molecular crosslinking reactions was observed to take place during irradiation of PVP in both its random coil (at room temperature) and thermally collapsed form (at high temperature) at concentrations of 0.9×10^2 mol L^{-1}. The interaction of ionizing radiation with matter results in the transfer of energy giving distinctive chemical products, often through the intermediate formation of free radicals[8]. Ionizing radiation can produce excited species in matter directly and also indirectly by ion neutralization. Produced species are concentrated along the tracks

of charged particles. Ionizing radiation is not selective as light and can excite any part of any molecule in the system.

Preliminary studies by the authors using temperature increase of PVP water solution instead of theta solution were not very successful, maybe due to the difficult on controlling temperature during irradiation processing, resulting in macrogel formation.

Figure 2 show hydrodynamic radius, R_h, variation with increasing radiation dose and dose rate, DR, of PVP K-90 samples irradiated with gamma and electron beam radiation using Multipurpose Irradiator (DR 8 kGy h^{-1}), Gammacell (DR 2 kGy h^{-1}), (DR 11 kGy s^{-1}). R_h of irradiated PVP decreased with 4 kGy radiation dose and slightly decresed with increasing radiation dose.

It is possible to observe that DR variation of around 5 times did not affect significantly R_h of gamma irradiated samples. Otherwise, R_h of electron beam irradiated samples are smaller compared to gamma irradiated ones. R_h seems to not be affected by increasing radiation dose of electron beam irradiated samples in the dose range studied.
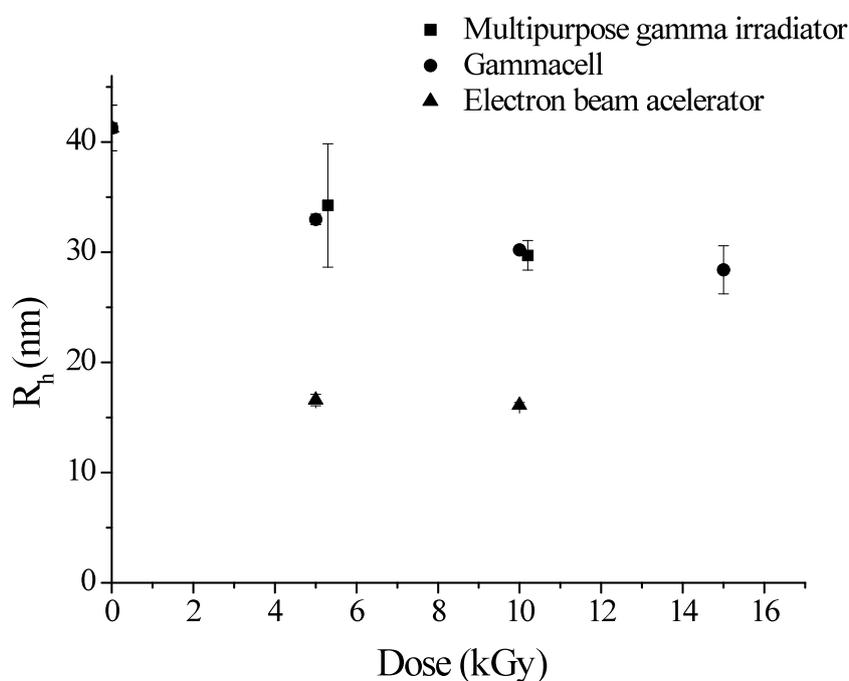

Figure 2 – PVP K-90 hydrodynamic radius, R_h, variation with radiation dose: ■ Multipurpose gamma irradiator (DR 8 kGy h^{-1}), ● Gammacell (DR 2 kGy h^{-1}); and ▲ electron beam accelerator (DR 11 kGy s^{-1}).

According to Spinks and Woods[9] the high-energy γ photons from radioactive sources produce energetic secondary electrons as they interact with matter so that both γ and electron-beam sources produce similar chemical changes in the absorbing material. Unlike charged particles, which generally lose energy continuously through a large number of small energy transfers as they pass through matter, photons that constitute electromagnetic radiation tend to lose a relatively large amount of energy whenever they interact with matter. However, not all incident photons will interact with any finite thickness of material and photons that do not interact will pass through the material without change of energy or direction. All ionizing radiations will give rise to qualitatively similar chemical effects. However, radiation of different types and energy will lose energy in matter at different rates, and consequently will form tracks that may be densely or sparsely populated with

active species. The differences observed in the chemical effect of different radiations, differences in the quantities and proportions of the chemical products, stem from the different densities of active species in the particle tracks. The difference observed on the Rh size decrease of the PVP dilute samples irradiated with gamma and EB radiation observed in Figure 2 seems to be due to fact exposed here.

A complete radiation-chemical mechanism includes all steps between the deposition of energy in the system by charged particles and the final formation of stable chemical products[8]. Products characteristic of the radical-radical reaction(s) will become more important as the absorbed dose rate is increased. And dose-effects are observed when radical-radical reactions are enhanced by high concentration in the bulk of the medium. When dilute aqueous solutions are irradiated practically all the energy absorbed is deposited in water molecules and the observed chemical changes are brought about indirectly by the molecular and, particularly, the radical products of water radiolysis. Radical and molecular yields (mol J^{-1}) in irradiated water, at pH 3-11, are: $g(-H_2O)$ 0.43; $g(H_2)$ 0.047; $g(H_2O_2)$ 0.081; $g(e^-_{aq})$ 0.28; $g(H)$ 0.062; $g(OH)$ 0.28; $g(HO_2)$ 0.0027. Liquid acetone rapidly scavenges thermal electrons. Acetic acid may be formed in irradiated acetone by addition of water, giving ketene[9]. Then all of those processes would affect intermediate species and further final product after chemical equilibrium. Solvent used in this study is acetone/water so it is necessary to consider yields of products from both solvents.

Spinks and Woods mentioned that direct action due to energy deposited directly in the solute is generally not important in dilute solutions (below 100 mol m^{-3}). At higher solute concentrations direct action may be significant, and there is evidence that high concentrations of solutes may react directly with the initial products of energy deposition, the unsolvated electron and H_2O^+; reaction with excited water molecules may also be possible.

According to Williams[10], from the results of radiation chemical experiments in gases, liquids and solids, most of the effective electron scavengers are molecules which are known to undergo thermal electron capture. Molecules that undergo to this process invariably possess low-lying orbitals to accommodate the additional electron. These include most compounds having functional groups such as carbonyl among others with π antibonding orbitals. There is no evidence to suggest that thermal electron attachment to molecules such as water, and the saturated hydrocarbons results either in dissociative electron capture or in the formation of a monomer radical anion. The reactions occurring in solution can affect the kinetic mechanism of intermediate and products obtained by radiation processing of PVP nanogel. This study shall add information on PVP nanogel formation.

Williams[10] mentioned that in a material of density 1 g cm^{-3}, the range of 1.25 MeV electrons is 0.5 cm and a beam of photons with the same energy is attenuated less than 5% in travelling 1 cm. Hence, X rays or γ rays can be used to produce high-energy electrons more or less uniformly throughout a large body of material, and the simplicity of this irradiation technique is suitable for much routine work when there is no restriction on time required to attain the desired dose. It should also be mentioned that, for photons and electrons of incident energies around 1-2 MeV the details of energy absorption are only modified in a very minor way by differences in the molecular structure of the irradiated object. Consequently, all the molecules are affected by high-energy radiation in contrast to the highly selective absorption of visible and ultraviolet radiation.

Jung-Chul An[1] has cited that much of the previous research on ionizing radiation synthesis of PVP hydrogels had concentrated on the synthesis of macroscopic gel systems which are inter-molecularly crosslinked. Just few studies on the polymer nanohydrogels had been reported. Pulsed high-energy electron irradiation had demonstrated as one synthesis method for the intra-molecularly crosslinked PVP. A discrete decrease of the PVP Rg had demonstrated in concert with the

maintenance of a constant PVP molecular weight (Mw). So, this result is similar to the decrease observed in this study, even though here we used continuous EB, meaning lower radiation doses.

Other study by Jung-Chul An[3], polymer nanohydrogels were prepared from dilute poly(vinylpyrrolidone) (PVP) aqueous solutions by means of irradiation with pulsed, high energy electrons. The author observed mixed crosslinking reaction in the thermally collapsed PVP. High temperature (77°C) introduced the collapsed-stiffer PVP chain conformation and produced smaller nanohydrogel upon irradiation due to the favorable intra-crosslinking reactions between closely located C-centered radicals on the same chain.

4. Conclusion

As preliminary results, it was possible to observe hydrodynamic radius decrease of PVP irradiated in theta solution. This behavior suggested that intramolecular crosslinks were promoted by ionizing radiation of PVP polymeric molecules.

Acknowledgements

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