In vitro dynamic swelling behaviors of radiation synthesized polyacrylamide with crosslinkers in the simulated physiological body fluids

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Abstract

Acrylamide hydrogels, containing different amounts and types of crosslinkers, were synthesized via γ-irradiation technique. Their swellings in simulated body fluids, such as physiological saline (0.89% NaCl) isoosmotic phosphate buffer at pH 7.4, gastric fluid at pH 1.1 (glycine–HCl), protein (aqueous solution of bovine serum albumin), urine (aqueous solution of urea), glucose and distilled water, were studied. Equilibrium swellings of the hydrogels were changed in the range 27–85 depending upon the fluids, type and amount of crosslinkers. The diffusion exponents were found over half for all hydrogels. © 2002 Elsevier Science B.V. All rights reserved.

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1. Introduction

Among the many biomaterials, synthetic hydrogels has recently been gaining a lot of attention for their resemblance to living tissues. Hydrogels are macromolecular networks that swell, but do not dissolve, in water. Synthetic hydrogel networks are useful for applications that require a material, which has good compatibility with aqueous fluids, yet will not dissolve. Such applications include biomaterials, controlled release devices and electrophoresis gels. Many properties of hydrogels make them suitable for biomedical applications that require contact with living tissue. The ability to absorb and retain aqueous media not only gives hydrogels a strong superficial resemblance to living tissue, but also makes them permeable to small molecules such as oxygen, nutrients and metabolites [1]. That is why hydrogels are better as implants, as long as their mechanical properties are acceptable, when compared to other materials such as ceramic and metals [2].

Hydrogels can be synthesized by the γ-irradiation technique [3]. A large number of papers have

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been published on the characterization of the hydrogels prepared by crosslinking of a homo- or copolymer in solution in the presence of crosslinkers with γ-irradiation. It is argued that more homogeneous network structures can be synthesized, if crosslinking is accomplished with γ-irradiation in the absence of an initiator and a crosslinking agent. The structural homogeneity of the network affects the swelling behavior and mechanical properties.

The water content of the hydrogels at equilibrium is one of their basic properties. A hydrogel with higher water content is generally more advantageous in increasing permeability and biocompatibility. A lot of work was dedicated to various aspects such as the swelling and shrinking of non-ionic and ionic gels in aqueous solution, phase transition in gels, the elastic properties of gels and the structure of solvents inside a gel [4,5]. Particularly, hydrogels based on polyacrylamide gels have been investigated by many research groups [6]. Polyacrylamide hydrogels exhibit a very high capability to absorb water, are permeable to oxygen and possess good biocompatibility.

In our previous works, radiation synthesized acrylamide based copolymeric hydrogels have been studied in the adsorption of protein [7,8], biocompatibility with human sera [9,10], the influence of amino acids and simulated body fluids to the swelling behavior [11–13] and in vivo biocompatibility with subcutaneous tissues of rats [14]. The objective of this work is the influence of simulated physiological body fluids on the swelling behavior of radiation synthesized polyacrylamide (AAm) hydrogels containing different types and amounts of crosslinkers.

2. Materials and methods

2.1. Materials

Acrylamide (AAm) monomer was purchased from BDH (Poole, UK) while crosslinkers (N, N' methylenebisacrylamide (N), 1,4 butanediol dimethacrylate (B) and ethylene glycol dimethacrylate (E)) were purchased from Sigma (St. Louis, MO, US) and used as received.

2.2. Preparation of radiation synthesized hydrogels

An aqueous solution of AAm monomer and crosslinkers such as N, B and E (100:0, 99.5:0.5, 99.0:1.0 and 97.0:3.0 in mole ratio in 1 ml of distilled water) were mixed. These solutions were placed in PVC straws of 3 mm diameter and irradiated to 2.6 kGy in air at ambient temperature in a ^{60}Co Gammacell 220 type γ irradiator source at a fixed dose rate of 0.72 kGy h^{-1}. Freshly obtained hydrogel rods were cut into pieces of 3–4 mm length. They were washed with distilled water and dried first in air and vacuum and stored for further use.

2.3. In vitro swelling studies in simulated physiological body fluids

The swellings of radiation crosslinked AAm hydrogels in distilled water (DW) and simulated body fluids [15] such as physiological saline solution (0.89% NaCl), (PS), isoosmotic phosphate buffer in pH 7.4, (PB), simulated gastric fluid, pH 1.1 (gylicine–HCl buffer), (GF), aqueous solution of protein (1% bovine serum albumin), (BSA), urine (urea) (UR) and the aqueous solution of glucose (1% glucose) (GL) were studied at 37 ± 0.1 °C to determine the swelling parameters and diffusion. Swollen gels, removed from the water-thermostated bath at regular time intervals, were dried superficially with filter paper, weighed and placed in the same bath. The radii of cylindrical gels were measured by a micrometer. The degree of swelling, S, was calculated as [16]

$$S = \frac{m_t - m_0}{m_0},$$

where m_0 is the mass of the dry gel at time 0 and m_t is the mass of the swollen gel at time t.

3. Results and discussion

3.1. Preparation of radiation crosslinked hydrogels

When monomers of AAm with or without crosslinker have been irradiated in water with

ionization rays such as γ-rays, free radicals from water, monomers and crosslinkers if any are generated. Random reactions of these radicals cause polymerization and formation of network. When irradiation dose has been increased beyond a certain value, the polymer chains crosslink and then gel is obtained.

It has been reported that acrylamide needs 2.0 kGy of γ-ray irradiation to obtain crosslinked polyacrylamide at ambient temperature [17]. So, 2.6 kGy of γ-ray irradiation were chosen for the preparation of the AAm hydrogels. During polymerization and crosslinking reactions, all monomers reacted together by applied γ-ray irradiation. The radiation technique is used for sterilization hydrogel systems at the same time. We assumed that there is no monomer (such as toxic acrylamide) at the end of the polymerization and crosslinking reactions, since 2.0 kGy is sufficient dose for 100% gelation of acrylamide without crosslinker [17]. In dry state, hydrogel gels were hard and glassy, but in swollen state, gels were soft, tender and easy to handle. The hydrogels are obtained in the form of cylinders. Upon swelling the hydrogels retained their integrity.

3.2. In vitro swelling

A fundamental relation exists between the swelling of a polymer in a solvent and the nature of the polymer and the solvent. The phosphate buffer at pH 7.4 (pH of cell fluid, plasma, edema fluid, synovial fluid, cerebrospinal fluid, aqueous humor, tears, gastric mucus and jejunal fluid), glycine–HCl buffer at pH 1.1 (pH of gastric juice), physiological saline, albumin, urea, glucose and distilled water intake of initially dry hydrogels were followed until reaching a constant swelling (equilibrium swelling, S_{eq}). Some representative swelling curves of AAm hydrogels are shown in Fig. 1(a)–(c).

Fig. 1(a) shows the swelling of AAm hydrogel containing 1% E crosslinker in physiological body fluids. The S_{eq} changes in the interval 4.94–5.69. This is expected due to different interaction parameters of physiological body fluid and AAm hydrogels. Due to the non-ionic character of AAm, the effect of pH and ionic strength of the
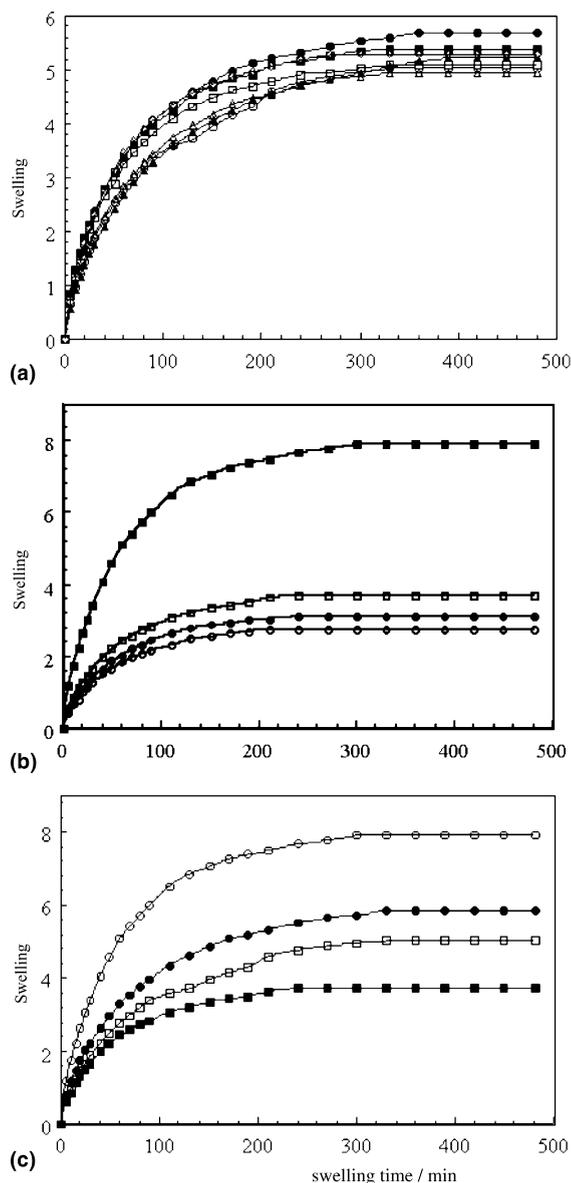

Fig. 1. (a) The effects of simulated physiological body fluids on the swelling behaviors of AAm hydrogel containing 1.0% E: (○) PS, (●) UR, (■) PB, (□) GF, (△) BSA, (▲) GL, (◇) DW. (b) The effects of types of crosslinker on the swelling behaviors of AAm hydrogel containing 3.0% crosslinker in the physiological saline solution: (○) N, (●) B, (□) E, (■) no crosslinker. (c) The effects of concentrations of crosslinker on the swelling behaviors of AAm hydrogel containing E crosslinker in the physiological saline solution: (○) no crosslinker, (●) 0.5%, (□) 1.0%, (■) 3.0%.

solutions has little effect on S_{eq} (equilibrium swelling, $t \rightarrow \infty$).

As can be seen in Fig. 1(b), the type of cross-linkers immensely effective on the swelling behaviors of AAm hydrogels in the physiological saline solution.

Again Fig. 1(c) illustrates the concentration effect of crosslinker type-E on AAm hydrogel swellings.

S_{eq} of the hydrogels for all physiologically fluids can be estimated from Fig. 1. It can be deduced that the swellings of AAm hydrogels in the simulated physiological body fluids are in the order of

no crosslinker > E > B > N.

It is also noteworthy to mention the concentration effect of the crosslinkers on the swelling behavior of AAm hydrogels. From Fig. 1(c), the increase in the concentration of crosslinker reduces the swellings for the same crosslinker in the same simulated physiological body fluid. The reason for this behavior is obvious that the crosslink density increases parallel to an increase in the crosslinker concentration leading to a reduction in the swelling behavior.

And all S_{eq} values of the hydrogels were greater than the percent water content values of human body. Thus, the radiation synthesized AAm hydrogels exhibit similarity with respect to the fluid contents with those of living tissues.

For extensive swelling of polymers, it can be written following second-order kinetics relation [18],

$$\frac{t}{S} = A + Bt, \tag{1}$$

where $B = 1/S_{max}$ is the inverse of the maximum or equilibrium swelling, $A = 1/(dS/dt)_0$ is the reciprocal of the initial swelling rate (r_0) of the gel.

Fig. 2 shows the linear regression of the swelling curves obtained by means of Eq. (1) for radiation synthesized AAm hydrogels containing different types of crosslinkers in physiological saline solution. The values of r_0 and S_{max} of the hydrogels can be calculated from the slope and the intersection of the lines.

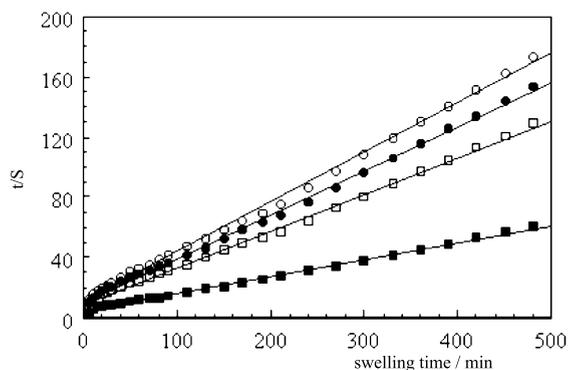Swelling processes of AAm hydrogel are quicker than the swelling rate of AAm hydrogels

Fig. 2. Swelling kinetics graphs of AAm hydrogels containing 3.0% crosslinker in the physiological saline solution: (○) N, (●) B, (□) E, (■) no crosslinker.

formed in the presence of crosslinker. This is conceivable; because with the increase in the amount of crosslinker, the crosslink density increases resulting in shrinkage of the pore of AAm hydrogels. Thus, diffusion of the solute gets harder leading to a decrease in the initial swelling rate. Same explanation is valid for other simulated physiological body fluids unless there is a specific interaction (i.e. chemical bonding) between functional groups on the polymer backbone and solute molecules or different relation surfaces/volumes.

3.3. Diffusion

The following equation was used to determine the nature of diffusion of water and fluids into hydrogels [16,19],

$$F = kt^n, \tag{2}$$

where F denotes the amount of solvent fraction at time t in the gel, k is a constant related to the structure of the network and the exponential n is a number indicative of the type of diffusion. This equation is applied to the initial stages of swelling and plots of $\ln(F)$ versus $\ln(t)$ are shown in Fig. 3 for radiation synthesized AAm hydrogels containing different types of crosslinker in physiological saline solution. The exponents n can be calculated from the slope of the lines, and it is clear that the values of diffusional exponents range between 0.5 and 0.7. For the hydrogels studied here,
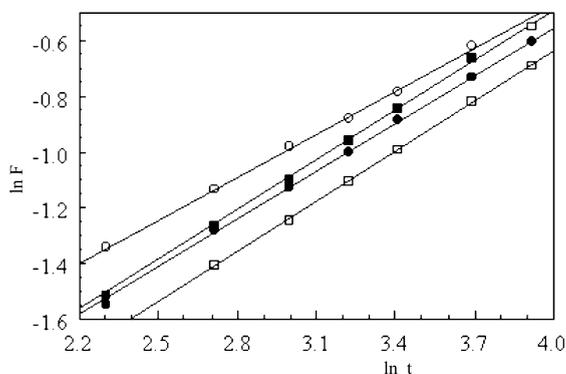
Fig. 3. $\ln F$–$\ln t$ graphs of AAm hydrogels containing 1.0% crosslinker in the physiological saline solution: (○) N, (●) B, (□) E, (■) no crosslinker.

the n values, indicating the type of diffusion, are found to be over 1/2. Hence the diffusion of the fluids into the hydrogels is non-Fickian in character. In this type of diffusion, fluid diffusion rate and polymer relaxation rate are about the same order of the magnitude [20].

4. Conclusion

In vitro dynamic swelling study of radiation synthesized acrylamide hydrogels, containing various amounts and types of crosslinker, has shown that swellings depend upon the type of fluids as well as the nature and the extent of used cross-linkers. Depending on used specific composition, hydrogels can exhibit different swellings, absorptive capacities, etc. in various media. However, their medical properties are very close to each other. The technique used in this work known since many years as "clean method" can also be used for synthesis of other hydrogels for biomedical materials. It is possible to obtain hydrogels with a controllable mesh and/or porosity depending on the required properties and places where they are used.

The preliminary in vitro study in the simulated physiological body fluids is very important for the application of hydrogels as biomaterials. Thus, the prediction of behaviors of hydrogels provides great advantage to a designer from a scientific point of view. And it can be concluded that the use of radiation for hydrogel synthesis is very useful and promising.

Acknowledgements

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