# Adsorption of Bovine Serum Albumin onto Radiation-crosslinked Poly(acrylamide/acrylic acid)

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**ABSTRACT:** Poly(acrylamide/acrylic acid) (AAm/AAc) hydrogels were prepared at initial acrylic acid compositions of 70, 80 and 85 mol%, respectively. Mixtures of AAm and AAc monomers were irradiated in a $^{60}$Co γ-ray source at a dosage of 8 kGy. These hydrogels were used in experiments associated with the swelling, diffusion and adsorption of bovine serum albumin (BSA) from aqueous solution. The data obtained allowed the swelling and diffusion parameters for the hydrogels to be calculated. In the BSA adsorption experiments, the adsorption kinetics together with the influence of the pH of the medium, the initial BSA concentration and the composition of the hydrogels on the adsorption efficiency of the AAm/AAc hydrogels were all studied.

The rates of BSA adsorption were found to conform to pseudo-first-order kinetics and a kinetic model was used to calculate the corresponding rate constant for the adsorption processes. The adsorption of BSA onto AAm/AAc hydrogels decreased with increasing pH, with the maximum adsorption being observed at a pH value of 3.7. In terms of the Giles classification, the adsorption was of type C. BSA adsorption increased as the AAc content of the hydrogels increased. Significant amounts of adsorbed BSA (up to 95%) were eluted when an elution medium containing 1.0 M NaSCN was employed at a pH value of 8.0.

## INTRODUCTION

Synthetic polymers used in adsorbent materials are designed to be both strongly adsorbent towards water and durable. These materials are generally considered to be resistant to attack by microorganisms and their enzymes. Different types of polymer have been used as adsorbents for adsorption purification and the chromatographic separation of biomolecules.

Hydrogels are crosslinked polymeric networks that can imbibe quantities of water without network dissolution. The hydrophilicity of these materials is due to the presence of water-compatible groups such as –OH, –COOH, –$CONH_2$, –$SO_3H$, etc. The water content of hydrogels at equilibrium is one of their basic properties. A hydrogel with a higher water content is generally more advantageous in increasing permeability and biocompatibility (Rosiak and Yoshii 1999).

Radiation chemical studies of polymeric materials, especially water-soluble monomers that can be crosslinked by ionizing radiation to form hydrogels, are of current interest. The use of ionizing radiation can result in dramatic changes in such properties as mechanical behaviour, solubility

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and swelling. Acrylamides and their derivatives are known to form three-dimensional network structures when polymerized in aqueous solution by irradiation. Ionizing radiation has been recognized as a suitable tool for the formation of hydrogels. Thus, the ready process control, high purity, sterilization, the need for no initiators or crosslinking agents and the relatively low running costs all make irradiation the method of choice in the synthesis of hydrogels. In recent years, much attention has been given to "stimuli responsive hydrogels" that undergo controllable volume changes in response to a small variation in solution conditions, such as the solvent composition, temperature and pH. Gels with these properties have been considered for controlled drug delivery devices, wound dressings, superabsorbents, soft contact lenses and separation processes (Rosiak *et al.* 1995).

Polyacrylamide hydrogels have a high capability for water absorption. They are also permeable towards oxygen and possess good biocompatibility. Acrylic acid exhibits a structural similarity with acrylamide and has a carboxylic group that makes it highly hydrophilic. The aim of the present work was to use hydrogels based on copolymers of acrylamide and acrylic acid (AAm/AAc) prepared via radiation techniques for the adsorption of proteins. The protein chosen for in vitro adsorption study was BSA, i.e. a major component of serum proteins. This shows various functions in vivo, e.g. osmotic pressure control as well as the transport and storage of nutrients and drugs. Moreover, it plays an important role in the interaction between a biomedical polymer surface and biocomponents (Peters 1996).

In our previous studies, radiation-induced acrylamide-based hydrogels (Saraydın *et al.* 2001; Karadağ *et al.* 1994; Şolpan *et al.* 2002) have been studied for the adsorption of proteins (Saraydın *et al.* 1994; Karadağ *et al.* 1994). The current work is an extension of these studies.

## EXPERIMENTAL

### Materials

The acrylamide and acrylic acid monomers used in this study were obtained from BDH Chemicals Ltd. (Poole, UK) while BSA and Coomassie Brilliant Blue G250 were purchased from Sigma (St. Louis, MO, USA).

### Preparation of radiation-induced hydrogels

Aqueous solutions of monomeric AAm and AAc were prepared at three different compositions (AAm/AAc mole ratios of 30:70, 20:80 and 15:85, respectively). These solutions were placed in PVC straws of 3-mm diameter and irradiated in air at ambient temperature to 8 kGy in a $^{60}$Co Gammacell 220 type γ-irradiator at a fixed dose rate of 2.67 Gy/min. The resulting hydrogels were obtained as long cylindrical shapes that were cut into pieces of 4–5 mm length, washed and dried first in air and then in vacuum.

### Swelling experiments

To correlate the swelling tendency of poly(AAm/AAc) hydrogels with their BSA adsorption capacity, the behaviour of these gels was studied at 25°C in distilled water, in physiological saline (0.9 w/v% NaCl), in a 10 mg/l BSA solution in distilled water and in a 10 mg/l BSA solution in physiological saline, respectively. The swollen gels removed from the water bath at regular

intervals were dried superficially with filter paper, weighed and placed back into the same bath. A micrometer was used to measure the radii of the cylindrical swollen gels.

### Effect of pH on BSA adsorption

To investigate the effect of pH on the adsorption of BSA, known weights (0.1 g) of the poly(AAm/AAc) hydrogel sample containing 85 mol% AAc irradiated to 8 kGy were added to 20 ml of 10 mg/l BSA solutions maintained at various pH values. The pH of the experimental medium was then changed to values between 3.7 and 7.4 through the use of a buffer system (0.2 M $CH_3COONa/CH_3COOH$ for pH values of 3.7–5.6 and 0.2 M $Na_2HPO_4/NaH_2PO_4$ for pH values of 6.0–7.4). The samples were maintained in the solution for 1 d at 25°C in a rotary shaker and their BSA concentrations determined by the method of Bradford at ambient temperature using a Shimadzu A160 double-beam spectrophotometer (Scopes 1984).

### Adsorption kinetics

To investigate the adsorption kinetics, ca. 500 mg of the poly(AAm/AAc) hydrogels containing 70, 80 and 85 mol% AAc subject to irradiation to 8 kGy were each transferred into 100 ml of a 10 mg/l BSA solution. The adsorption kinetics were determined at a controlled temperature (25°C) employing a high stirring rate, with the BSA concentration in the solution being measured at known time intervals by spectrophotometry.

### Effect of initial BSA concentration on BSA adsorption

To investigate the effect of the initial BSA concentration on its adsorption, 0.1-g samples of poly(AAm/AAc) hydrogel containing 70, 80 and 85 mol% AAc and subject to 8 kGy irradiation were transferred into 20 ml of solutions containing 2–20 mg BSA per litre. These solutions were incubated in a rotary shaker for 24 h at 25°C, an aliquot of protein solution being then removed and its concentration determined.

### Desorption of BSA

Desorption was investigated using hydrogels loaded with BSA. These were left in distilled water for 3 d at 25°C and 1 M NaSCN solution (pH 8.0) then added to the system. Analysis of the BSA generated in the solution was then undertaken.

## RESULTS AND DISCUSSION

### Preparation of hydrogels

A radiation technique appeared to be promising for hydrogel preparation since a polymer in aqueous solution or in a water-swollen state readily undergoes crosslinking on irradiation to yield a gel-like material. Since this hydrogel is not contaminated with foreign additives, the crosslinked hydrogel must be composed of stable –C=C– bonds (Şolpan *et al.* 2002; Rosiak *et al.* 1983). For this reason, ionizing radiation was employed for the preparation of the AAm/AAc hydrogels studied. When a solution containing AAm and AAc was irradiated with γ-rays, one of the bonds

in the double bonds of both AAm and AAc was broken and monomer radicals were formed. These monomer radicals then combined together to form the AAm/AAc copolymer. On continuing the irradiation, the crosslinking reaction commenced, crosslinking the individual chains with each other and allowing gelation to occur in the system.

To prepare the AAm/AAc hydrogels, AAm and AAc monomer mixtures containing an initial AAm composition of 15, 20 or 30 mol% were exposed to an 8 kGy dose of γ-radiation. After preparation, the dried gels were glassy and very hard; however, the swollen gels were very soft. The dried poly(AAm/AAc) hydrogels exhibited a cylindrical shape. If the dried hydrogel was placed in water, the swollen hydrogels exhibited the same geometrical shape as their dried counterparts. No dissolution, deformation, dispersion or damage was observed under such conditions.

**Swelling and diffusion**

The degree of swelling (S) of the poly(AAm/AAc) hydrogels was calculated from the following relationship:

$$S = \frac{m_s(t) - m_0}{m_0} \quad (1)$$

where $m_s(t)$ is the mass of swollen gel at time t and $m_0$ is the initial mass of dry gel.

The swelling curves of the hydrogels in distilled water (DW), physiological saline solution (PS), albumin in distilled water (BSA) and albumin in physiological saline solution (PS + BSA) are depicted in Figure 1. In all cases, the degree of swelling (S) increased with time but finally attained a constant value. This swelling value may be referred to as the equilibrium degree of

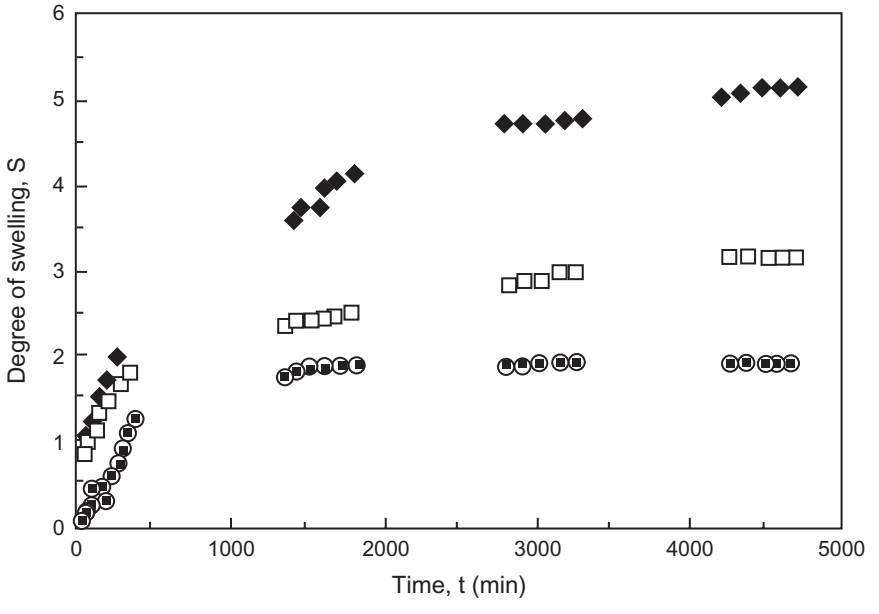



**Figure 1.** Swelling curves for AAm/AAc hydrogels in the various solutions studied. Data points: ◆, DW; □, BSA; ○, PS; ■, BSA + PS.

**TABLE 1.** Swelling and Diffusion Parameters of AAm/AAc Hydrogels (15:85 mol ratio)

| Solution | $S_{eq}$ (g fluid/(g gel) | $S_{max}$ (g fluid/g gel) | $r_0 \times 10^3$ (g fluid/g gel min) | n | $k \times 10^2$ ($min^{-0.5}$) | $D \times 10^7$ ($cm^2/s$) |
|---|---|---|---|---|---|---|
| DW | 5.15 | 5.45 | 14.30 | 0.5 | 2.14 | 1.41 |
| BSA | 3.18 | 3.27 | 12.28 | 0.5 | 3.30 | 1.93 |
| PS | 1.90 | 1.98 | 14.48 | 0.5 | 3.55 | 2.58 |
| PS + BSA | 1.90 | 2.00 | 13.97 | 0.5 | 4.23 | 2.26 |

swelling ($S_{eq}$). The $S_{eq}$ values for the hydrogels are presented in Table 1 from which it will be seen that hydrogel swelling in the various solutions followed the order DW > BSA > PS = PS + BSA.

A simple kinetic analysis could be undertaken using the second-order rate equation (Ho and McKay 1999) in the form:

$$\frac{dS}{dt} = k_S(S_{max} - S)^2 \tag{2}$$

where $k_S$ is the swelling rate constant and $S_{max}$ denotes the maximum swelling or the degree of equilibrium. After integration applying the conditions S = 0 at t = 0 and S = S at t = t, equation (2) becomes:

$$\frac{t}{S} = \alpha + \beta t \tag{3}$$

where $\alpha$ and $\beta$ are coefficient whose physical sense may be interpreted as follows. After treatment for an extended time span, $\beta t >> \alpha$ and, according to equation (3), $\beta = 1/S_{max}$, i.e. the reciprocal of the theoretical equilibrium or maximum swelling. In contrast, after a very short treatment time, $\alpha >> \beta t$. At the limit, equation (2) becomes:

$$\lim_{t \to 0}\left(\frac{dS}{dt}\right) = \frac{1}{\alpha}$$

Hence, the intercept $\alpha$ is the reciprocal of the initial swelling rate $r_0$ or $1/k_S S_{max}^2$.

Figure 2 shows representative graphs obtained by the application of equation (3) to the swelling data. Straight lines with excellent correlation coefficients were obtained in all cases, thereby demonstrating that the swelling behaviour of these systems followed second-order kinetics. The calculated kinetic parameters are tabulated in Table 1. As seen from the data listed in this table, the results of the kinetic model agreed with the swelling experiment data.

The following equation may be used to determine the nature of the diffusion process:

$$F = kt^n \tag{4}$$

where F denotes the value of the solvent fraction at time t and k is a constant related to the structure of the network. The exponent n is indicative of the type of diffusion involved. The fluid adsorption characteristics of a gel exhibit anomalous behaviour, ranging between Fickian and case II extremes depending on the experimental temperature and thermodynamic compatibility of

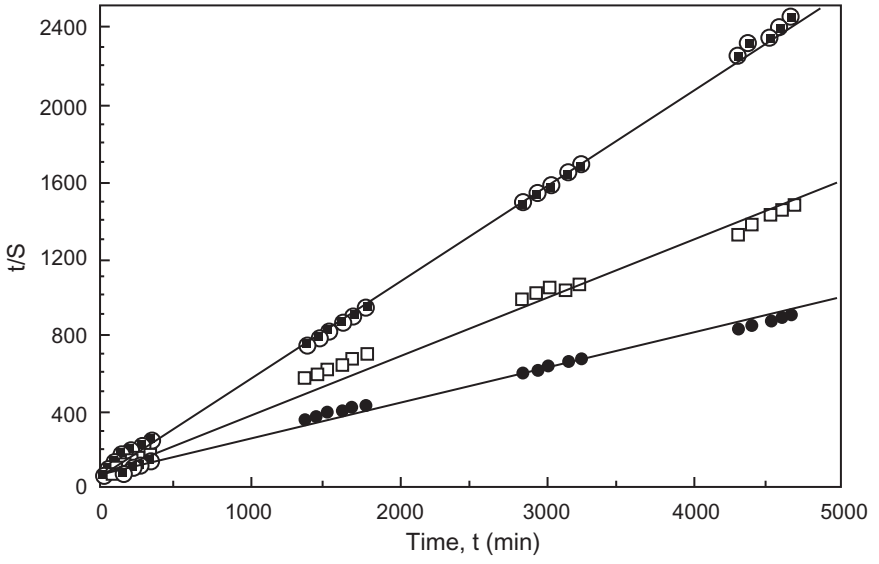



**Figure 2.** Kinetic swelling curves for AAm/AAc hydrogels in the various solutions studied. Data points: ●, DW; □, BSA; ○, PS; ■, BSA + PS.

the penetrant and the gel. Typically, both the diffused amount and the penetrating swelling front position in case II transport are completely time-dependent in a linear fashion, whereas Fickian diffusion is dependent on the square root of the time. An intermediate situation, known as non-Fickian or anomalous diffusion, occurs whenever the rates of Fickian diffusion and polymer relaxation are comparable (Masaro and Zhu 1999).

Equation (4) has been applied to the initial stages of swelling and some plots of ln F versus ln t are presented in Figure 3(a). The values of the exponents calculated from the slopes of the linear plots are presented in Table 1.

Table 1 show that the number (n) determining the type of diffusion was 0.50. Hence the diffusion of fluids into the AAm/AAc hydrogels was taken to be of a Fickian character. In Fickian-type diffusion, the system is controlled by the diffusion step, as the fluid mobility is very low in comparison to the relaxation rate of the hydrogel.

Diffusion through swollen polymers in thermodynamic equilibrium is of the Fickian type. The kinetics of swelling where the diffusion mechanism follows Fick's law may be described by the following equation:

$$F = 4\left[\frac{D}{\pi r^2}\right]^{0.5}\sqrt{t} \qquad (5)$$

where D is the apparent diffusion coefficient in $cm^2/s$, t is the time in seconds and r is the radius of the cylindrical polymer sample.

The fractional fluid uptake as a function of the square root of time for different amounts of AAc in the AAm/AAc hydrogels is shown in Figure 3(b). At small swelling times ($F < 0.6$), a linear relation was observed between both parameters, confirming that the swelling of hydrogels in thermodynamic equilibrium followed a Fickian diffusion mechanism. This fact allowed D to be

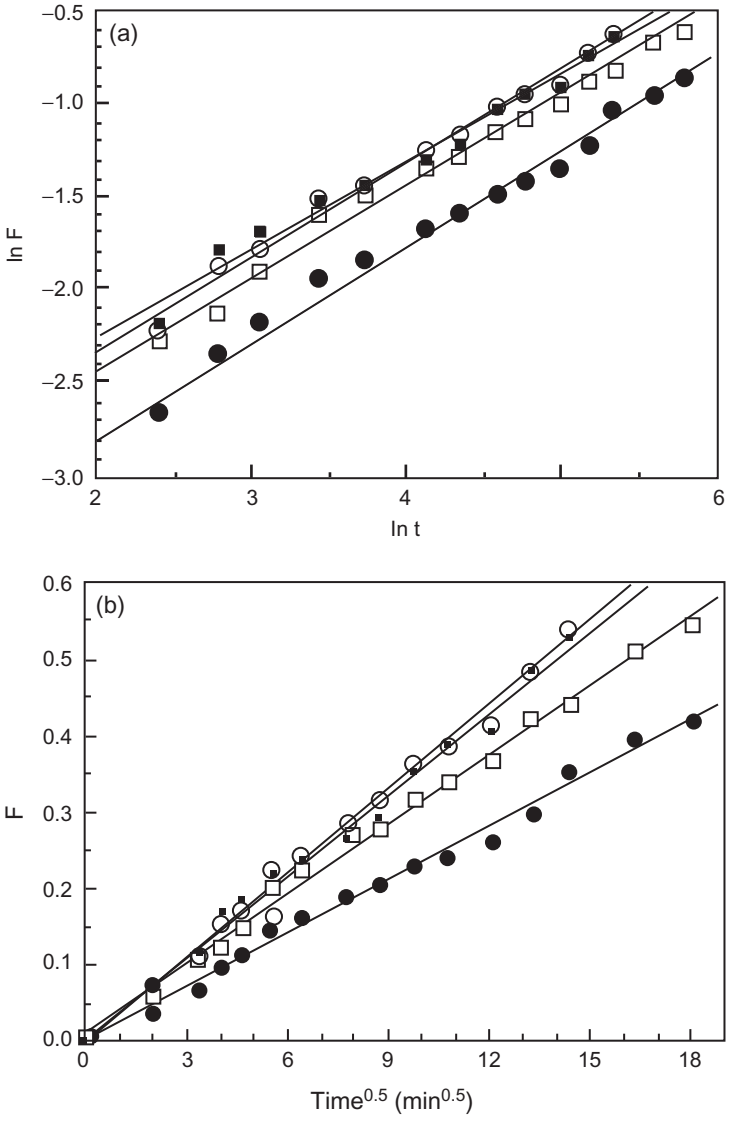



**Figure 3.** Plots of (a) ln F versus ln t and (b) F versus $\sqrt{t}$ for diffusion into the AAm/AAc hydrogels in the various solutions studied. Data points: ●, DW; □, BSA; ○, PS; ■, BSA + PS.

calculated directly from the slope of the linear plot. Table 1 lists the values of the diffusion coefficient for the AAm/AAc hydrogels thus obtained.

## Adsorption of BSA

This stage was devoted to a study of the uptake of BSA by the radiation-crosslinked AAm/AAc hydrogels and thereby investigating their suitability as sorbents for BSA.

When an adsorption system is at equilibrium, the total solute concentration ($C_0$ g/l) may be written as:

$$C_0 = C_B + C \quad (6)$$

where $C_B$ is the equilibrium concentration (g/l) of the solute on the adsorbent (bound solute concentration) and C is the equilibrium concentration (g/l) of the solute in the solution (free solute concentration). The binding ratio, Q, may be defined as:

$$Q = \frac{(C_0 - C)}{m} \times V \quad (7)$$

where V is the total volume of the BSA solution and m is the mass of the hydrogel.

*Effect of pH on BSA adsorption*

To explain the effect of pH on BSA adsorption, a graph of Q versus pH was plotted as depicted in Figure 4. The maximum adsorption of BSA was observed at pH 3.7. The pH range employed would have a considerably effect on the net charges on BSA. At a pH of 3.7, the adsorption process occurred via the positively charged sites of the amino acids of BSA. Anionic ligands bind in a hydrophobic pocket that is adaptable to the ligand, with its negative charge being bound electrostatically by the positive charge of a neighbouring lysyl or arginyl residue of BSA. Since poly(AAm) is non-ionic, the number of ionizable groups on the copolymer were increased by adding AAc to the AAm monomer. Hence, these hydrogels possessed a considerable number of carboxyl groups that led to an increase in the interaction between BSA and the hydrogel carboxyl groups in the hydrophobic pocket of BSA. The decrease in the adsorption capacity of the hydrogel

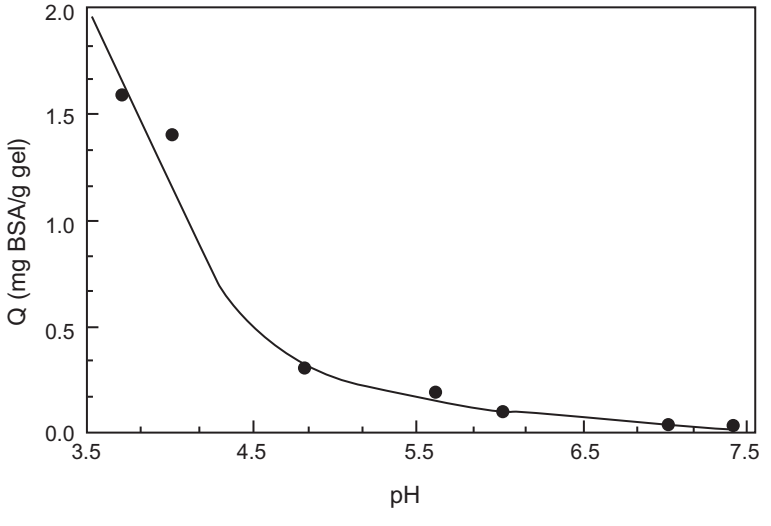
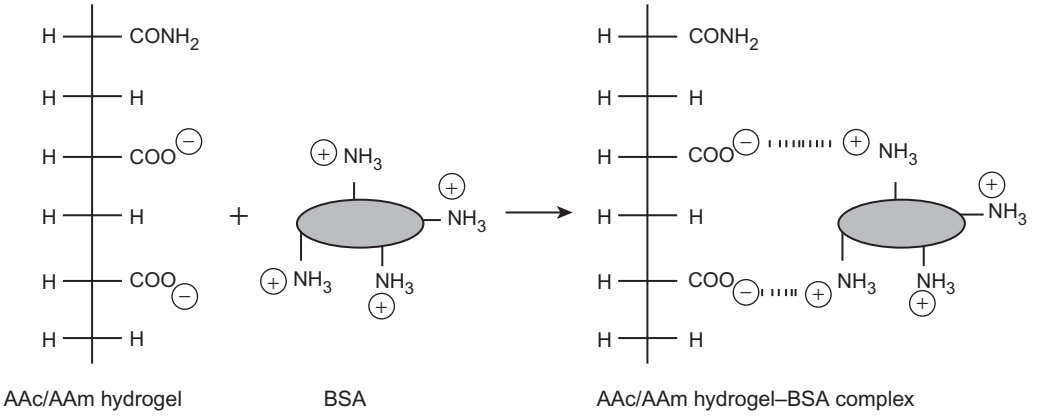



**Figure 4.** The influence of pH on the adsorption of BSA within AAm/AAc hydrogels.




**Scheme 1.** Possible interactions between BSA and AAm/AAc hydrogel.

towards BSA with increasing pH may be attributed to electrostatic repulsion between the negatively charged carboxyl groups of BSA and those of the AAm/AAc hydrogel.

It is well known that BSA adapts its conformation readily and often reversibly to variations in the environmental conditions. Thus, as a consequence of pH change, albumin undergoes reversible conformational isomerization; at pH values lower than 4.0, albumin undergoes expansion with a loss of the intra-domain helices. This expanded form is known as the E-form. The native form–E-form transition involves a decrease in the content of ordered (secondary) structure, thus allowing the ready occurrence of interactions between the polymer and the ionizable amino acid residues on the expanded form of BSA (Norde and Giacomelli 2000). The possible interactions between BSA and AAm/AAc hydrogel are presented in Scheme 1.

*Adsorption kinetics*

In kinetic studies, the adsorption experiment was carried out as a function of time using AAm/AAc hydrogels with mole ratios of 30:70 (1), 20:80 (2) and 15:85 (3), respectively. As seen from Figure 5, all the curves corresponding to the adsorption of BSA by AAm/AAc hydrogels attained a stationary state within 24 h.

Kinetic models were used to test the experimental data in order to examine the controlling mechanism of the adsorption process such as mass transfer and the chemical reaction involved. Different chemical groups on the adsorbent's surface were created during the preparation of AAm/AAc hydrogels (i.e. $–NH_2$, $–CONH_2$, –OH, –COOH). These groups imply that many different types of interactions were possible between bovine serum albumin molecules and the hydrogels. In this case, kinetic models (pseudo-first-order and second-order equations) can be used if it is assumed that the measured concentrations are equal to the surface concentrations on the adsorbent.

Kinetic analysis of the adsorption process may be effected via an equation of the form (Wu *et al.* 2001):

$$\frac{dQ}{dt} = k(Q_e - Q)^n \qquad (8)$$

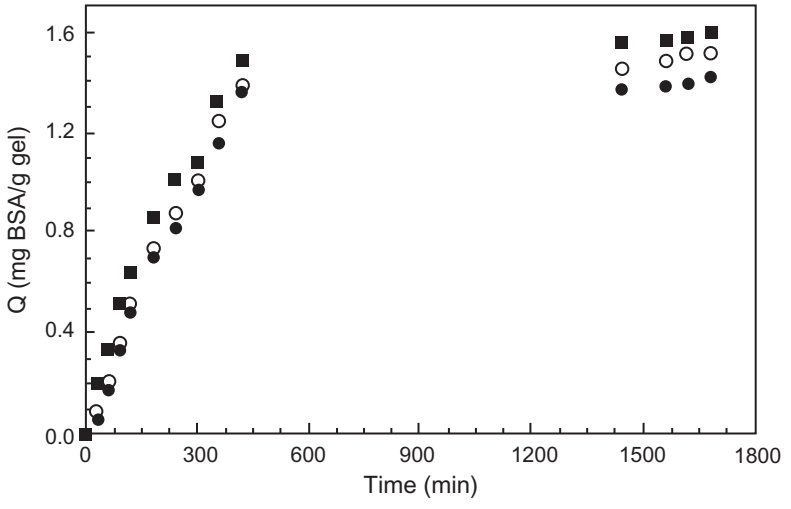



**Figure 5.** Adsorption curves of BSA by AAm/AAc hydrogels with mole ratios of 30:70 (●), 20:80 (○) and 15:85 (■), respectively.

where k is the rate constant for the adsorption process, n is the kinetic order and $Q_e$ denotes the amount of adsorption at equilibrium. After integration applying the initial conditions Q = 0 at t = 0 and Q = Q at t = t, equation (8) becomes

$$\ln(Q_e - Q) = \ln Q - k_1 t \qquad \text{for } n = 1 \tag{9}$$

or

$$\frac{t}{Q} = \frac{1}{k_2 Q_e^2} + \frac{1}{Q_e} t \qquad \text{for } n = 2 \tag{10}$$

Plots of $\ln(Q_e - Q)$ versus t and of t/Q versus t are depicted in Figure 6(a) and (b), respectively. In order to compare the applicability of each model quantitatively, a normalized standard deviation ΔQ was calculated:

$$\Delta Q\,(\%) = \sqrt{\frac{\sum \left[(Q_{exp} - Q_{cal})/Q_{exp}\right]^2}{n_d}} \times 100 \tag{11}$$

where $n_d$ is the number of data points. Table 2 lists the calculated results. As shown in Figure 6(a) and (b), and from the correlation coefficients ($r^2$) of the graphs and the values ΔQ listed in Table 2, the adsorption of BSA was best fitted by the pseudo-first-order equation. Thus, the adsorption processes were diffusion-controlled. The calculated values of k are also listed in Table 2.

*Adsorption isotherms*

Adsorption isotherms were used to evaluate adsorption properties during the batch experiments. An isotherm plot for BSA binding is shown in Figure 7. The adsorption of BSA onto

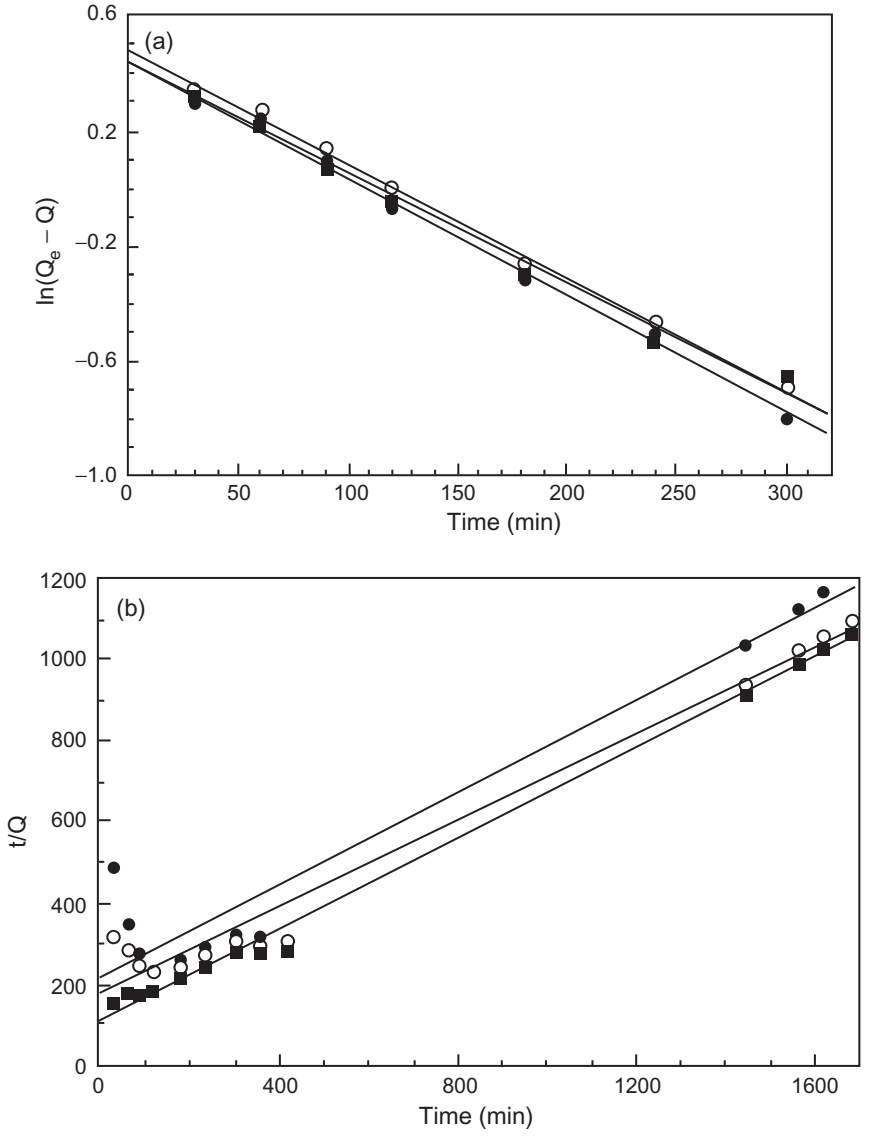



**Figure 6.** (a) First-order and (b) second-order kinetic curves of BSA adsorption by AAm/AAc hydrogels with mole ratios of 30:70 (●), 20:80 (○) and 15:85 (■), respectively.

poly(AAm/AAc) hydrogels of different composition all corresponded to type C1 adsorption isotherms as listed in the Giles classification system for the adsorption of a solute from its solution (Giles *et al.* 1974). This type of isotherm is characterized by the constant partition of co-solute between the solution and the substrate (i.e. the polymer). Conditions favouring the C-type curve appear to be a porous substrate with flexible molecules and a co-solute with a higher affinity for the substrate and better penetrating power than the solvent.

**TABLE 2.** Kinetic Parameters, Correlation Coefficients and Normalized Standard Deviations for BSA Adsorption onto AAc/AAm Hydrogels at 25°C

| Pseudo-first order | | | | Pseudo-second order | | |
|---|---|---|---|---|---|---|
| AAm/AAc mol ratio | $k_1 \times 10^3$ ($min^{-1}$) | $r^2$ | ΔQ (%) | $k_2 \times 10^3$ ($min^{-1}$) | $r^2$ | ΔQ (%) |
| 30:70 | 4.08 | 0.99 | 1.13 | 1.56 | 0.94 | 2.23 |
| 20:80 | 3.94 | 0.99 | 1.41 | 1.62 | 0.97 | 1.43 |
| 15:85 | 3.82 | 0.99 | 3.20 | 1.74 | 0.99 | 4.71 |

Fundamentally, the linearity observed demonstrates that the number of adsorption sites remained constant (i.e. as more co-solute was adsorbed, more sites were created). Such situations could arise when the co-solute has a higher attraction for the substrate molecules than the solvent itself. The co-solute could then rupture inter-substrate bonds more readily than the solvent and, if its molecular dimensions were suitable, could penetrate into the structure of the substrate in regions not already penetrated by the solvent. This action has been compared with the opening of a zipper, the fastenings representing the intermolecular bonds of the substrate and the slider the first molecule or group of molecules of co-solute to penetrate; this opens up the structure and allows more co-solute molecules to enter.

Where a linear relationship exists between the binding ratio and the equilibrium concentration of the solute, the isotherm can be also be classified as a Nernst isotherm, i.e.

$$Q = K_C C \tag{12}$$

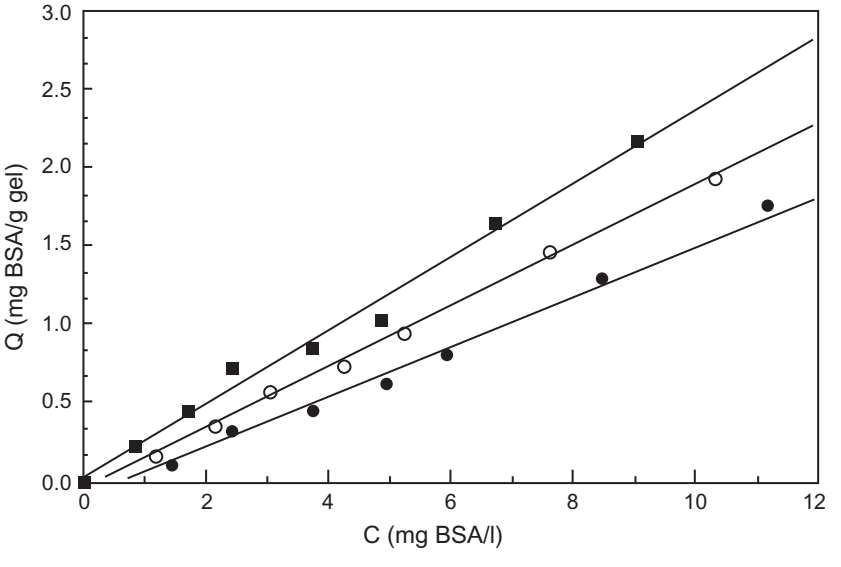



**Figure 7.** Isotherms for BSA adsorption within AAm/AAc hydrogels with mole ratios of 30:70 (●), 20:80 (○) and 15:85 (■), respectively.

**TABLE 3.** Equilibrium Parameters for BSA Adsorption

| AAm/AAc mol ratio | $K_C$ [ml/(g gel)] | ΔG (kJ/mol) |
|---|---|---|
| 30:70 | 161.86 | –12.60 |
| 20:80 | 192.94 | –13.04 |
| 15:85 | 234.82 | –13.52 |

The angular coefficient of this isotherm ($K_C$) corresponds to the ratio between the protein in water and in the solid phase and can be defined as a 'partition coefficient' or 'equilibrium constant'. Calculated values of $K_C$ are listed in Table 3.

The free energy of adsorption (ΔG) of the protein–hydrogel binding systems was calculated from the equation:

$$\Delta G = - RT \ln K_C \quad (13)$$

Calculated values of ΔG are listed in Table 3. The negative value of ΔG indicates that the binding of the protein to the hydrogel occurred spontaneously.

*The effect of composition of hydrogel on BSA adsorption*

The changes in BSA adsorption with AAc content are illustrated in Figure 8. The adsorption of BSA onto AAm/AAc hydrogels increased linearly with increasing AAc content in the AAm/AAc hydrogels. It has been observed that the addition of AAc to AAm increases AAm hydrogel

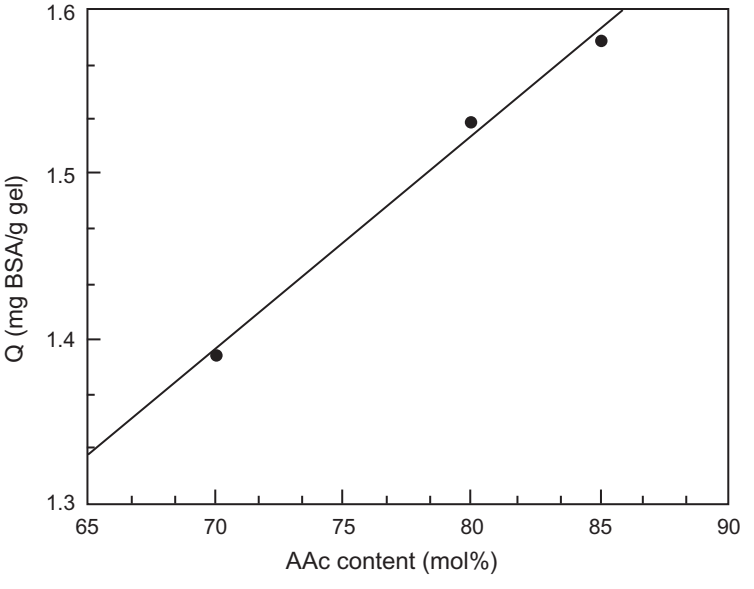



**Figure 8.** The effect of AAc content in AAm/AAc hydrogels on the adsorption of BSA.

swelling (Şolpan *et al.* 2002). Such AAm/AAc hydrogels swelled extensively during the early stages of BSA adsorption, the higher swelling of the hydrogels allowing the presence of more BSA molecules and water inside the hydrogel. Some BSA molecules were adsorbed onto the surface of the hydrogel via electrostatic effects while others infiltrated into the hydrogel itself.

### Desorption

Hydrogels loaded with BSA were allowed to stand for 3 d at 25ºC in either distilled water or 1 M NaSCN solution (pH 8.0) to investigate possible desorption processes. No desorption of BSA occurred in distilled water whereas more than 90% of the adsorbed BSA was desorbed when NaSCN was used for elution. The $SCN^-$ is a chaotropic anion, i.e. one that prevents non-ionic interaction by ordering the structure of water (Roe 1989). For this reason, NaSCN was used to elute BSA molecules adsorbed on the hydrogel.

## CONCLUSIONS

AAm/AAc hydrogels were prepared via γ-irradiation. The swelling of such hydrogels in solution [distilled water (DW), physiological saline solution (PS), albumin in distilled water (BSA) and albumin in physiological saline solution (PS + BSA)] followed the order: DW > BSA > PS = PS + BSA. The highest swelling value for the hydrogel was found in distilled water. The cationic molecules interact with the carboxyl groups of AAc in the poly(AAm/AAc) hydrogel, with the result that the hydrophilic groups in the hydrogel were not bound with water. Consequently, hydrogel swelling decreased. The diffusion of fluids into the AAm/AAc hydrogels was assumed to be of a Fickian nature. It was observed that the pH of the medium had an important effect on the adsorption equilibrium of BSA, with preferential interaction between BSA and the hydrogel occurring at pH 3.7. The isoelectric point (pI) of BSA is 4.7 and hence BSA molecules would be cationic at lower pH values. AAm/AAc hydrogels possess many carboxyl groups that are dissociated in aqueous medium. The resulting negative charge would interact with the positive charge of BSA.

The adsorption kinetics of BSA within AAm/AAc hydrogels could best be described by the pseudo-first-order rate equation. Adsorption of BSA within AAm/AAc hydrogels of different composition corresponded to type C1 isotherms in the Giles classification for the adsorption of a solute from its solution. The adsorption of BSA onto AAm/AAc hydrogels increased with an increase in the AAc content in the AAm/AAc hydrogels. The adsorbed BSA molecules could be eluted with a 1.0 M NaSCN solution (pH 8).

The results presented are of fundamental importance to the application of radiation-crosslinked poly(AAm/AAc) for the separation of biological molecules and the development of AAm/AAc hydrogel-based biomaterials.

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