Interaction of Nicotine and Its Pharmaceutical Derivatives with Acrylamide/Itaconic Acid Hydrogels

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ABSTRACT: Acrylamide/itaconic acid hydrogels (AAm/IA) containing different amounts of itaconic acid prepared by irradiating with γ radiation were used in experiments on swelling, diffusion, and interactions of the pharmaceuticals nicotine, nicotinic acid, nicotinamide, and nikethamide. AAm/IA hydrogel containing 60 mg itaconic acid and irradiated at 4.65 kGy has been used for swelling and diffusion studies in water and aqueous solutions of the pharmaceuticals. For this hydrogel, swelling studies indicated that swelling increased with the following order: water > nicotine > nikethamide > nicotinamide > nicotinic acid. Diffusions of water and the pharmaceuticals within hydrogels were found to be non-Fickian in character. The uptake of the pharmaceuticals to AAm/IA hydrogels was studied by batch adsorption technique at 25°C. In the experiments of the adsorption, C-type adsorption in Giles's classification system was found. Some binding and thermodynamic parameters for AAm/IA hydrogel-pharmaceutical systems were calculated by using the Klotz method. The values of adsorption heat, free energy, and entropy of this system were found as negative values. © 1997 John Wiley & Sons, Inc. J Appl Polym Sci **66:** 733–739, 1997

Key words: Hydrogel; poly(acrylamide/itaconic acid); nicotine; sorption; interaction

INTRODUCTION

Synthetic polymer hydrogels have found a wide range of biomedical applications, including controlled drug delivery systems, replacement blood vessels, wound dressing, coatings for biosensors, soft tissue substitution, contact lenses, and a variety of other related and potential uses. As a family of polymeric materials, synthetic hydrogels are generally well tolerated when implanted *in vivo* and can be tailored to suit many potential functions of prosthetic devices in contact with blood or soft tissue. This success of hydrogels as biomaterials lies partially in their superficial resemblance to living tissue, a property attributable to their relatively high water content (20–99%), which immediately results in minimal frictional irritation of surrounding tissues. In addition, hydrogels can be nontoxic and chemically stable and (owing to their water content) can exhibit a low interfacial tension with aqueous environments. In a general sense then, the most important concept here is that synthetic hydrogel as biomaterials, show favourable interfacial properties in biological environments.[1–4]

The interactions between polymers and cosolutes are both biochemically significant and scientifically interesting, although they are also technologically important in several respects.[5] There are several techniques for the immobilization of biomolecules on and within hydrogels, including physical entrapment, electrostatic attraction, physical adsorption with or without crosslinking, and chemical bonding. Physical adsorption via

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secondary molecular forces is an important way to immobilize a biological species on a hydrogel.[6]

In our previous articles, acrylamide-based hydrogels have been studied in adsorption of protein and biocompatibility with human sera.[7–10] In this study, the interactions between such pharmaceuticals as nicotine, nicotinic acid, nicotinamide (Vitamin PP), and nikethamide with novel hydrogels prepared with acrylamide and itaconic acid were investigated.

EXPERIMENTAL

Acrylamide (AAm) and itaconic acid (IA) monomers were obtained from B.D.H. (Poole, UK), and nicotine, nicotinic acid, nicotinamide, and nikethamide were purchased from Sigma (St. Louis, MO).

A suitable mass of itaconic acid and irradiation doses for acrylamide and itaconic acid hydrogels was selected by taking previous experiments into consideration.[11,12]

Acrylamide weighing 1 g was dissolved in 1 mL of aqueous solutions of 0, 20, 30, 40, 50, and 60 mg itaconic acid. These solutions were placed in PVC straws of 3 mm diameter and irradiated to 2.60, 3.73, 4.65, 5.20 and 5.71 kGy in air at ambient temperature in a Gammacell 220 type γ irradiator at a fixed dose rate of 0.72 kGy h^{-1}. Hydrogels obtained in long cylindrical shapes were dried in air and under vacuum. These samples were then used in experiments involving swelling, diffusion, and binding.

Acrylamide and AAm/IA hydrogel containing 60 mg IA and irradiated to 4.65 kGy were swollen in the distilled water and 100 mg/L nicotine, nicotinic acid, nicotinamide, and nikethamide solution at 25°C to determine the parameters of diffusion and swelling. Swollen gels removed from the water bath at regular intervals were dried superficially with filter paper, weighed, and placed in the same bath. The radii of cylindrical swollen gels were measured by a micrometer.

The synthetic aqueous solutions of nicotine were prepared in the concentration range of 20 to 100 mg/L. Acrylamide–itaconic acid (AAm/IA) hydrogels containing 60 mg IA and irradiated to 4.65 kGy weighing 0.1 g were transferred into 50 mL of aqueous solutions of nicotine and allowed to equilibrate for 24 h at 25°C. These solutions were separated by decantation from hydrogels. Spectrophotometric measurements of these nicotine solutions were carried out using a Shimadzu A160 model UV-VIS double-beam spectrophotometer at ambient temperature. The absorbance of these solutions was recorded at a wavelength of 261 nm. Distilled water was chosen as a reference. The equilibrium concentrations of nicotine were determined by means of a precalibrated scale.

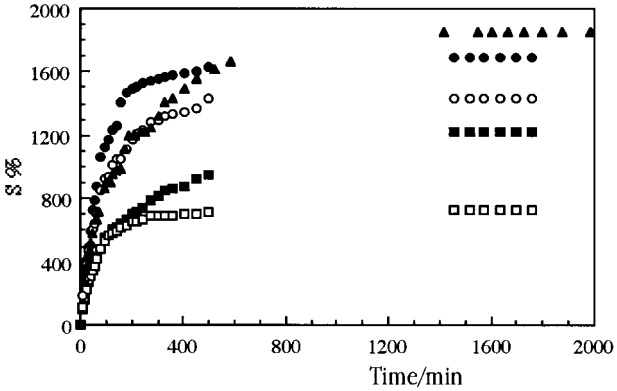


Figure 1 The swelling curves of AAm/IA hydrogel in the solutions; (▲) water; (○) nicotine; (●) nikethamide; (■) nicotinamide; (□) nicotinic acid.

The effects of irradiation dose and IA content were investigated for adsorption of nicotine within AAm/IA hydrogels. Hydrogel samples weighing 0.1 g and prepared with different concentrations of IA and irradiation doses were added to 50 mL of 50 mg/L nicotine and incubated for 1 day at 25°C. The spectrophotometric method was applied to these solutions.

RESULTS AND DISCUSSION

Swelling and Diffusion

Analysis of the mechanisms of diffusion in swellable polymeric systems has received considerable attention in recent years because of important applications of swellable polymers in biomedical, pharmaceutical, environmental, and agricultural engineering.[13,14]

The swelling of AAm and AAm/IA containing 60 mg IA irradiated at 4.65 kGy in water and the pharmaceuticals was calculated from the following relation:

$$\%S = [(m_t - m_0)/m_0] \times 100 \qquad (1)$$

where m_t is the mass of swollen gel at time t and m_0 is the initial mass of the swollen gel.[15] Swelling curves of AAm and AAm/IA in water and the solutions of the pharmaceuticals are shown in Figure 1.

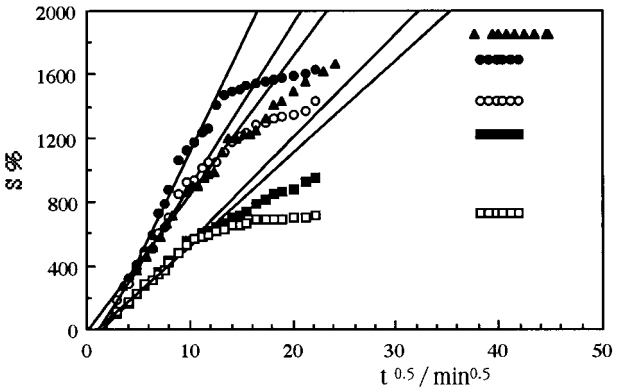


Figure 2 The plots of swelling versus square roots of immersion time; (▲) water; (○) nicotine; (●) nikethamide; (■) nicotinamide; (□) nicotinic acid.

The equilibrium swelling of AAm/IA hydrogel prepared from 60 mg IA and irradiated at 4.65 kGy in water is greater (1845%) than the equilibrium swelling of AAm/IA hydrogel in the aqueous solutions of nicotine and its derivatives (1690–730%). The molecules of nicotine and its derivatives are larger than the molecules of water; hence, molecules of water can diffuse into gel pores more easily than molecules of nicotine and its derivatives.

The polar pyrrolidine ring (due to second dissociation) in the nicotine molecule can cause greater swelling of the hydrogel than the other pharmaceutical molecules, because the hydrogel in the solution of the nicotine is greater than in the other solutions. Nicotinamide and nikethamide molecules contain amide groups like acrylamide molecules. Thus, amide groups in nicotinamide, nikethamide, and acrylamide can interact, which results in more pronounced swelling of the hydrogel in these solutions than is encountered in the solution of nicotinic acid. On the other hand, there will be anionic repulsion between carboxylic groups of nicotinic acid and itaconic acid in the hydrogel, and thus the swelling degree of the hydrogels is decreased sharply to 730%. The AAm/IA hydrogel in the aqueous solutions of the pharmaceuticals is swollen in the following order: water > nicotine > nikethamide > nicotinamide > nicotinic acid. Swelling rate constants of the hydrogels were calculated from the following relation[16]:

$$S = k_s t^{0.5} \tag{2}$$

where k_s is the swelling rate constant. Swelling rate curves for water and other solutions are shown in Figure 2. Swelling rate constants of the hydrogel–pharmaceutical systems suggest similar swelling behavior (Table I).

The following equation was used to determine the nature of the diffusion of water, nicotine, nicotinic acid, nicotinamide, and nikethamide into hydrogels[7–10,15,17]:

$$F = \frac{M_t}{M_\infty} = kt^n \tag{3}$$

Here F is the fractional uptake, M_t/M_∞, where M_t is the amount of diffusant sorbed at time t, M_∞ is the maximum amount absorbed, k is a constant incorporating characteristics of the macromolecular network system and the penetrant, and n is the diffusional exponent, which is indicative of the transport mechanism. Eq. (3) is valid for the first 60% of the normalized solvent uptake. Fickian diffusion and Case II transport are defined by n equal to $\frac{1}{2}$ and n equal to 1, respectively. Anomalous transport behavior (non-Fickian diffusion) is intermediate between Fickian and Case II, which is reflected by the fact that anomalous behavior is defined by values of n between $\frac{1}{2}$ and 1.[17] This equation is applied to the initial stages of swelling, and plots of ln F versus ln t are shown in Figure 3. The exponents n and k values were calculated from the slope and intercept of the lines, respectively, and are presented in Table I.

In the experiments, the number to determine the type of diffusion (n) was found to be over 0.50 (Table I). Hence the diffusion of the pharmaceuti-

Table I The Parameters of Swelling and Diffusion of the Hydrogel

Solution	S %	k_s	$k \times 10^2$	n	$D \times 10^6$/cm^2 sec^{-1}	$\mathcal{D} \times 10^6$/cm^2 sec^{-1}
Water	1845	0.86	3.99	0.53	4.94	7.67
Nicotine	1690	1.34	2.41	0.73	6.44	8.15
Nikethamide	1435	1.01	3.17	0.66	5.64	7.21
Nicotinamide	1225	0.65	2.81	0.68	2.33	2.88
Nicotinic acid	730	0.58	4.26	0.63	4.96	5.80

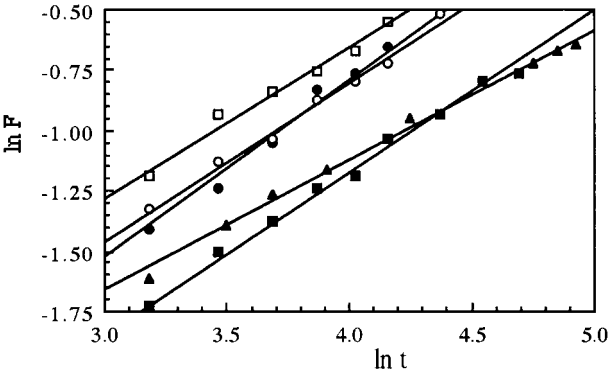


Figure 3 The curves of swelling kinetics of hydrogel; (▲) water; (○) nicotine; (●) nikethamide; (■) nicotinamide; (□) nicotinic acid.

cals into AAm/IA hydrogels was taken to be of non-Fickian character. This is generally explained as being a consequence of the slow relaxation rate of the hydrogel.

Diffusion coefficients are important penetration parameters of some chemical species to polymeric systems. The diffusion coefficient (D) gives a measure of diffusion and mass flow of penetrant to the system (bulk diffusion), but the intrinsic diffusion coefficient ($\mathcal{D}$) gives only diffusion (pore diffusion). Diffusion coefficients were calculated from the following relation:[18]

$$D = 0.049/(t/4l^2)_{1/2} \qquad (4)$$

where D is in $cm^2\ sec^{-1}$, t is the time at which the swelling is one half the equilibrium value ($V/V_0 = \frac{1}{2}$), and l is one half of the cylindrical sample radius. The intrinsic diffusion coefficient, $\mathcal{D}$, may be expressed as

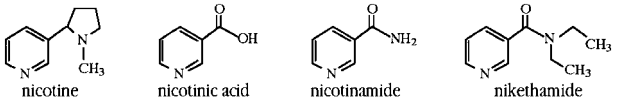
$$\mathcal{D} = D(1 - V)^{-3} \qquad (5)$$

where V is volume fraction of solvent penetrating the polymer by the time t defined above.[18]
The swelling parameters of diffusion coefficients of the hydrogel are listed in Table 1.

If Table I is examined, it can be seen that the values of the intrinsic diffusion coefficient of the hydrogel in the water and the aqueous solutions of pharmaceuticals are bigger than the values of their diffusion coefficients.

Interactions of the Pharmaceuticals with AAm/IA Hydrogel

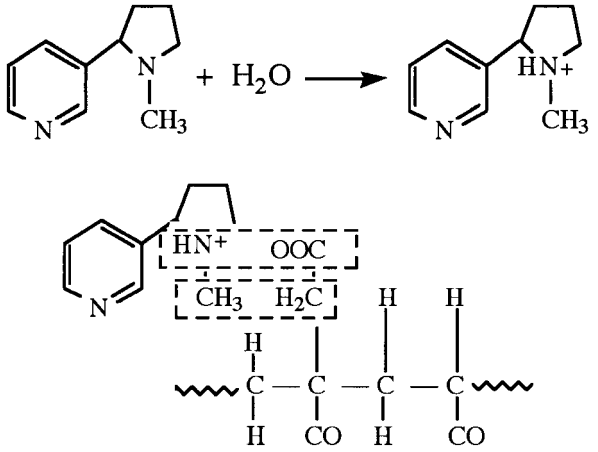
To observe binding of nicotine and its pharmaceutical derivatives to AAm and AAm/IA, hydrogels



Scheme 1 Structure of nicotine and its pharmaceutical derivatives.

were placed in aqueous solutions of nicotine and its pharmaceutical derivatives nicotinic acid, nicotinamide, and nikethamide and allowed to equilibrate for 2 days. At the end of this time, AAm did not sorb nicotine or its pharmaceutical derivatives from the solutions, and AAm/IA sorbed only nicotine. Nicotine was therefore selected for binding studies to AAm/IA hydrogels.

AAm hydrogel did not sorb any pharmaceutical molecules, whereas AAm/IA hydrogel sorbed only nicotine molecules. AAm is a nonionic polymer,[19] and thus acrylamide hydrogel generally did not interact with many small molecules.[20–25] By the addition of IA to acrylamide, acrylamide copolymers contain ionizable groups. These groups can interact with the small molecules, and these molecules bind to acrylamide copolymers with noncovalent or covalent bonds.[20–25] In the nicotine–AAm/IA hydrogel system, nicotine has a weak basic character, and the polar pyrrolidine ring in the nicotine molecule, when nicotine has a positive charge, can interact by electrostatic forces with the carboxyl groups of itaconic acid in the hydrogel. At the same time, the methyl group of nicotine and the methylene group of itaconic acid in the hydrogel can interact with hydrophobic forces (Scheme **2**). Amide groups of nikethamide and



Scheme 2 Second dissociation of nicotine and possible interactions between nicotine and AAm/IA hydrogel.

nicotinamide can not interact with itaconic acid because amide groups are not ionic or polar in character. On the other hand, there will be anionic repulsion between carboxyl groups of nicotinic acid and itaconic acid in the hydrogel. Thus, nicotinic acid does not bind to the AAm/IA hydrogel.

In a batch adsorption system at equilibrium, total cosolute concentration (C_I, mol/L) is

$$C_I = C_B + C \tag{6}$$

where C_B is the equilibrium concentration of the cosolute on the adsorbent in moles per liter (bound cosolute concentration), and C is the equilibrium concentration of the cosolute in the solution in mol/L (free cosolute concentration). The amount bound can therefore be conveniently expressed as the binding ratio, r, defined by

$$r = C_B/P. \tag{7}$$

Thus with C_B in mol/L and P in base mol (moles of monomer units) per liter, r represents the average number of molecules of solute bound to each monomer unit at that free solute concentration.

The binding data were interpreted on the basis of the uniform site-binding model (u.s.b.), which in statistical-thermodynamic terms corresponds to the formation of an ideal localized one-dimensional monolayer of cosolute on the polymer chains.[26] This leads to the linear form of binding isotherm, which applies to many aqueous polymer/cosolute binding systems[5]:

$$r = \frac{nKC}{1 + KC} \tag{8}$$

where K is the binding constant, that is, the equilibrium constant for the attachment of a molecule of nicotine N onto a site S by a specific combination of noncovalent forces

$$N + S = N \sim S,$$

and n is the site density (i.e., the limiting value of r for "monolayer" coverage, which is thus a measure of the density of the sites S along the polymer chain). The reciprocal of n is the site size, u, which may be taken to represent either the average number of monomer units occupied by the bound cosolute molecule or, more generally, the average spacing of cosolute molecules when the chain is saturated. The initial binding constant, K_i, is the initial slope of the binding isotherm and thus represents the average binding strength of a cosolute molecule by a single monomer unit on an occupied chain. In the u.s.b. model it is equal to the product Kn.

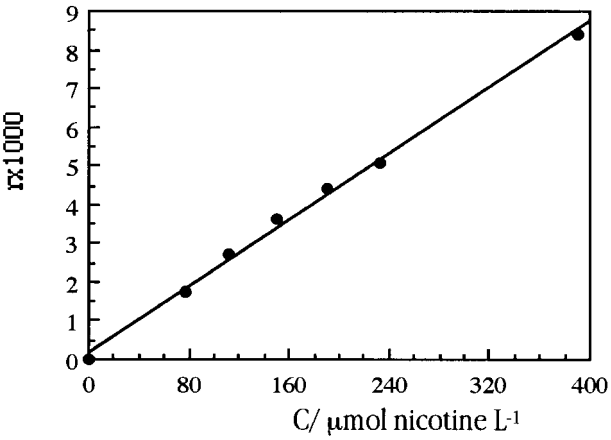


Figure 4 The binding isotherm of nicotine: AAm/IA hydrogel system.

A plot of the binding isotherm of nicotine is shown in Figure 4. Figure 4 shows that adsorption of the nicotine within the AAm/IA hydrogel corresponds to Type C adsorption isotherms in the Giles classification system for adsorption of a cosolute from its solution.[27,28]

The type C adsorption isotherm is characterized by the constant partition of cosolute between solution and substrate (i.e., polymer). The conditions favoring the C curve appear to be a porous substrate with flexible molecules and a cosolute with higher affinity for the substrate than the solvent has and with better penetrating power. Fundamentally, the linearity shows that the number of sites for adsorption remains constant (i.e., as more cosolute is adsorbed more sites must be created). Such situations could arise when the cosolute has a higher attraction for the substrate molecules than the solvent itself has. The cosolute could then break intersubstrate bonds more readily than the solvent could 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 cosolute to penetrate; this opens up the structure and allows more cosolute molecule to enter.[27,28]

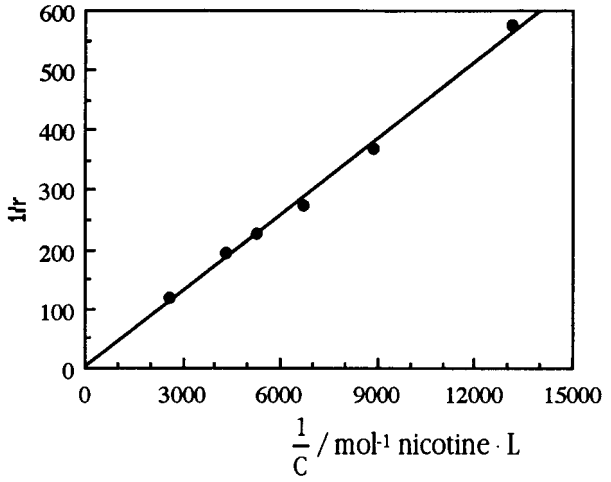


Figure 5 Double reciprocal graph of nicotine: AAm/IA hydrogel system.

The Klotz equation derived on the basis of a uniform site-binding model is[5]

$$\frac{1}{r} = \frac{1}{n} + \frac{1}{nK}\frac{1}{C} \tag{9}$$

where r, C, n, and K are defined above. The Klotz plot of an AAm/IA–nicotine system is shown in Figure 5.

Binding parameters of the nicotine–hydrogel system were calculated from the intercept and slope of Klotz plot. The derived values of the binding parameters K and n are listed in Table II for nicotine with AAm/IA hydrogel. The binding parameters of the nicotine–hydrogel system were calculated from the intercept and slope of Klotz plots and tabulated in Table II.

To determine the thermodynamic parameters of the nicotine–hydrogel binding system, an adsorption experiment was made at 37°C. Adsorption heat, adsorption free energy, and adsorption entropy of this system were calculated from the following equations:[29]

$$\ln \frac{C_2}{C_1} = \frac{\Delta H}{R}\left(\frac{1}{T_1} - \frac{1}{T_2}\right) \tag{10}$$

$$\Delta G = -RT \ln K \tag{11}$$

$$\Delta G = \Delta H - T\Delta S \tag{12}$$

where C_1 and C_2 are the free nicotine concentrations at the absolute temperatures T_1 and T_2, respectively; R is the universal gas constant; and ΔH, ΔG, and ΔS are heat of adsorption, free energy of adsorption, and entropy of adsorption, respectively.

As already pointed out, physical adsorption is an exothermic process. That this should be so can be seen by reference to the well-known thermodynamic equation $\Delta G = \Delta H - T\Delta S$ relating the changes ΔG, ΔH, and $T\Delta S$ in the free energy, the heat, and the entropy, respectively, that occur during a physicochemical process such as adsorption. If adsorption is to take place spontaneously, the free energy must diminish during the process so that ΔG must have a negative value; moreover, ΔS will be negative, for the molecules of cosolute must have less freedom when adsorbed than in the solution. Consequently, ΔH will have to be negative (i.e., the process is exothermic).[29] The thermodynamic parameters of the nicotine–hydrogel binding system are tabulated in Table II.

In the nicotine–AAm/IA binding system, all of the thermodynamic parameters are found to have negative values. Thus, adsorption is an exothermic and spontaneous process. The adsorption of nicotine onto AAm/IA is physical adsorption because the adsorption heat is about -20 kJ/mol^{-1} (Table II).

In later experiments, the binding ratios of nicotine–hydrogel versus content of IA and irradiation doses were plotted, as presented in Figure 6. The binding ratios of AAm hydrogels increased after adding IA. The binding of nicotine was approximately constant with the increased IA content in AAm/IA hydrogels. The binding ratio of nicotine gradually increased with increasing irradiation. Increasing the IA content and irradiation dose does not affect the binding of nicotine onto hydrogels, although the addition of IA to acrylamide does affect the binding of nicotine onto hydrogels.

Table II Binding and Thermodynamic Parameters of the Nicotine–AAm/IA Hydrogel System

K_i (L/mol)	K (L/mol)	u	n	ΔG (kJ/mol)	ΔH (kJ/mol)	ΔS (e.u.)
23.45	79.14	3.375	0.30	−10.83	−21.85	−109.60

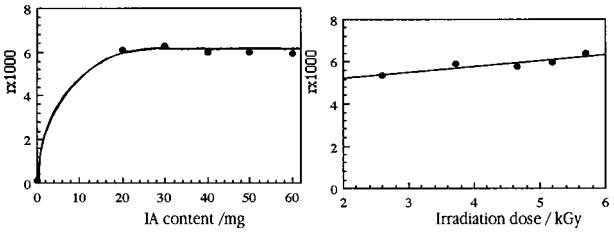


Figure 6 The effect of IA content and irradiation dose on the binding of nicotine onto AAm/IA hydrogel.

CONCLUSION

The study and application of immobilized biologically active molecules have been of increasing interest in both medicine and industry. One of the advantages of using immobilized biomolecules, particularly enzymes and antibodies, for chemical analysis and preparation is their high substrate specificity. Immobilization of biomolecules and cells on and within synthetic polymeric hydrogel is used in the areas of column techniques, biosensors, and immunoassays. There are many medical and industrial applications of hydrogel-immobilized biomolecules and cells.

Nicotine binding is thought to be one of the more important initial events that occurs upon exposure of solid surfaces to the biological environment. In this study, adsorption of nicotine was found to result from physical adsorption. Binding of nicotine onto AAm hydrogels was found to increase with the addition of IA into the hydrogels, and this increase was approximately constant with increasing amounts of IA in the AAm/IA hydrogels. Adsorption of nicotine within AAm/IA hydrogels prepared with the same amount of IA gradually increased with increasing irradiation doses. As a result, acrylamide/itaconic acid hydrogels can be used as adsorbents for immobilization of pharmaceuticals such as nicotine.

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