Water Uptake and Urea Release of Hydrogels Based on Acrylamide, Methyl Methacrylate and N,N’-Methylene-bis-Acrylamide

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In Memoriam to Prof. Dr. Evaristo Riande

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Abstract

Water swelling kinetics, equilibrium water content, urea release kinetics, and freezable and non–freezable water of hydrogels based on acrylamide (AM), methyl methacrylate (MMA) and N,N’-methylen-bis-acrylamide (NMBA) were studied. A series of copolymeric gels of AM, MMA and NMBA as a crosslinker agent were prepared by free radical solution copolymerization using a mixture of ethanol and water as a solvent and benzoyl peroxide (BPO) as initiator. Equilibrium water content (Wω) was strongly influenced by copolymer composition. For hydrogels richer in AM, EWC increased as crosslinking decreased. Instead, for high content methacrylate hydrogels, the hydrophilicity of NMBA decreased Wω. Swelling process in distilled water was studied by dynamic swelling measurements (gravimetric method). It was found that synthesized hydrogels fits second–order swelling. Urea release studies were carried out in distilled water and the urea concentration was determined by High Performance Liquid Chromatography (HPLC). The hydrogels delivered urea very rapidly, for all the studied compositions most of the urea was released in less than 200 minutes. Differential Scanning Calorimetry (DSC) was employed to determine the relative proportions of non–freezing and freezing water. DSC traces shown multiple endotherms that were related with different states of association between water and the polymeric matrix.

Keywords: Hydrogel; Acrylamide; Methyl methacrylate; Release; Urea; DSC; HPLC

1. Introduction

Hydrogels are chemically crosslinked polymers that in presence of water can swell to

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an equilibrium value but remain insoluble in water due to the presence of a three-dimensional network (Chandy and Pillai, 1995; Buckley et al., 1962; Pedley et al., 1980). This water absorption capacity is due to hydrophilic functional groups such as alcohol, carboxylic acids, and amides, which are attached to the polymer chain. Their maximum level of swelling is given by the equilibrium between an osmotic driving force that swells the network and a retractile elastic force caused by the chemical crosslinking (Ratner and Hoffman, 1976). Due to their high water absorption capacity hydrogels have been extensively used as superabsorbent materials in applications such as baby diapers, water reservoir for plants, medicine and food industry (Blanco et al., 2008). The physical properties of a swollen hydrogel (rubbery consistency, low interfacial tension, and softness) resemble those of natural tissues. In addition they are in general biocompatible, nontoxic, chemically stable and well tolerated when implanted in living beings. All those properties make hydrogels very useful as biomaterials. A hydrogel with high equilibrium water content is in general more permeable and biocompatible (Pedley et al., 1980). However, those factors that improve water absorption, i.e. low crosslinking density and highly hydrophilic monomers, generally result in low mechanical properties. In order to improve the mechanical behavior of swollen gels, hydrophobic monomers such as methyl methacrylate, ethyl acrylate and styrene, are usually included into the polymerization recipe.

Hydrogels are increasingly used in controlled delivery systems of drugs, herbicides, etc (Sahiner and Ilgin, 2011; Francis and Kumar, 2012). The delivery of drugs has been studied more than the release of any other solute, and it is possible to find a lot of scientific papers and patents, as well as diffusional models, about the release of a great variety of pharmaceuticals (Katime et al., 1999; Liu et al., 2012).

Hydrogels based on acrylamide and its copolymers have been widely studied. These hydrogels absorb water to a high equilibrium value, present good biocompatibility and are capable of absorb and release drugs and other compounds. In their synthesis, in order to improve its behavior, acrylamide is usually combined with hydrophilic and hydrophobic monomers. Karadag and coworkers have studied hydrogels of acrylamide and itaconic acid (Karadag et al., 1997). They showed that these hydrogels have a better absorption of nicotine and nicotine derivatives, compared with hydrogels prepared with only acrylamide. Methyl methacrylate is combined with more hydrophilic monomers in order to improve mechanical properties and for inducing to the polymer the ability to swell in organic liquids (Davies and Huglin, 1990) The hydrogels that are able to swell in both water and organic are known as amphygels. Methyl methacrylate is a hydrophobic monomer with many applications in polymer industry but of limited use in hydrogels synthesis due to its low water absorption, however MMA has been copolymerized with acrylic acid, methacrylic acid and other monomers in order to produce hydrogels with good mechanical properties.

Urea is an organic compound that is produced by live organisms and also is one of the most important sources of nitrogen for plants, so the study of urea release from hydrogels is useful for both pharmaceutical and agriculture applications. The aim of this work was to synthesize hydrogels of AM and MMA crosslinked with N,N,N',N'-tetraakis(2-hydroxyethyl)ethylene diamine (TMEDA) and to study their swelling kinetics in water as well as their urea release in distilled water. The states of water in the synthesized hydrogels were characterized by DSC and their compression modulus were determined by DMA.
2. Experimental

2.1 Materials
MMA, NMBA, benzoyl peroxide (BPO) and urea (UR) were purchased from Merck, AM was obtained from J.T. Baker and methanol from EM Industries–Germany. All the reagents were analytical grade and were used as received. Distilled water was used as polymerization solvent and in the swelling experiments and urea release studies.

2.2 Polymerization
Free radical polymerization was carried out in solution. The monomers AM and MMA were dosed by weight in eight different ratios (AM/MMA = 20/80, 30/70, 40/60, 50/50, 60/40, 70/30, 80/20 and 90/10). 2.8 g of the monomer mixture was poured into a 1.4 cm diameter glass test tube. NMBA was used as crosslinking agent, and was added to the monomer solution in proportions of 1, 3 and 5% by weight respect to the AM/MMA mixture. 0.11 g of initiator (BPO) were added to the test tube; the initiator concentration was held constant in all synthesis. A methanol – water solution (78% vol) was used as the polymerization solvent. 4.4 mL of the methanolic solution were added to the polymerization tubes. The mixture was homogenized and then was degassed by nitrogen bubbling for two minutes. The tubes were sealed and submerged in a water bath. Polymerization was carried out at 60°C for 6 h. At the end of this time, no liquid component remained in the glass tubes. Tubes were then broken and the polymer gels were removed. Hydrogels were submerged in methanol for one week to extract any methanol soluble oligomer, then they were dried at room temperature and after that they were submerged in water for a week to extract any water soluble component and dried to constant weight at room temperature. During the extraction procedures, methanol and water were changed several times to improve the extraction process. Finally, washed gels were dried to constant weight at room temperature. The drying time was settled for about three weeks to avoid material breaking.

2.3 Water swelling
Hydrogels were cut in discs of 1 mm thick (10–12 mm diameter). For the swelling kinetics study, the discs were immersed in distilled water at 30°C until swelling equilibrium was attained. At regular time intervals, gels were removed from the water, and lightly surface dried with absorbent paper to eliminate excess water, then were weighted in an analytical balance and finally, placed again in water (Baker et al., 1992). Water content was calculated from the following relation:

\[ W = \left( \frac{\text{weight hydrogel} - \text{weight xerogel}}{\text{weight xerogel}} \right) \]

where hydrogel and xerogel are the water swollen gel and the dehydrated gel, respectively. Even though equilibrium water content was attained in less than five hours for all the compositions studied, hydrogels remained in water for a period no less than a week before doing any further characterization.

2.4 Differential Scanning Calorimetry (DSC)
A differential scanning calorimeter (TA Instruments 2910) connected to a liquid nitrogen cooling system (LNCS) accessory was used for the DSC study. Samples of swollen gels (about 10 mg) were
lightly surface dried and encapsulated in a hermetic pan. After cooling at −40°C, samples were heated at a rate of 5 per minute to 40°C under nitrogen atmosphere (50 mL·min⁻¹). The instrument was previously calibrated with water and the content of freezing water in a swollen sample was determined from the melting endotherm area at 0°C. This area, when compared with the heat of fusion of water (previously determined) gives the mass of freezing or bound water. The difference between \( W_\omega \) and freezing water was taken as the content of non–freezing water.

2.5 Urea release
In order to load urea into the gels, water swollen gels were immersed in an aqueous urea solution (10% weight) at room temperature for a week. The release experiments were carried out transferring urea–loaded gels into 10 mL of distilled water. The gels were periodically removed and submerged into a new volume of fresh water at fixed time intervals (Liang and Liu, 2007). Urea content in the solutions was determined by High Performance Liquid Chromatography HPLC. The equipment used for this purpose was a Waters 410 equipped with a differential refractometer detector (Waters). The mobile phase was fresh filtered HPLC water, at a rate of 0.5 mL per minute; and the stationary phase was a HPLC column Novapack C18. The chromatograph was previously calibrated with Urea standards between 0.0012 and 1 g·L⁻¹. The fractional release, mass of urea released at time \( t \)/total of urea released, was reported as function of time.

3. Results and discussion
Dry polymer discs were transparent, rigid and brittle over all the composition range studied. However, in the swollen state the increase in acrylamide content, make hydrogels more opaque, whiter and more flexible. Hydrated gels with high content of MMA were transparent and rigid. Hydrogels with composition AM/MMA/NMBA = 90/10/5, presented many fissures after the polymerization process, this fractures hindered any subsequent characterization of this materials

3.1 Swelling in water
Figure 1 shows dynamic swelling behavior for all the samples prepared with 1% of crosslinker. Hydrogels initially absorb water very fast and then absorption rate decreases to obtain the equilibrium swelling \( (W_\omega) \) at around 200 minutes. The water absorption capacity increases with increasing the polyacrylamide, content similar results were obtained for the other two crosslinker concentrations used. Due to the difference in hydrophilicity of the two comonomers, \( W_\omega \), spans in a wide range of values. \( W_\omega \) for all the compositions studied are reported in table 1. As expected, increasing acrylamide content increases \( W_\omega \) and increasing NMBA decreases \( W_\omega \). Swelling kinetics was adjusted with a second order kinetics proposed elsewhere (Katime et al., 2004):

\[
\frac{dW}{dt} = K(W_\omega - W)^2
\]  

where \( K \) is a constant. Equation (2) integrated and rearranged yields:

\[
\frac{t}{W} = \frac{1}{KW_\omega^2} + \frac{t}{W_\omega}
\]  

(3)
Fig 1. Effect of monomer ratio on absorption capacity of AM/MMA hydrogels crosslinked with 1% w/w of NMBA. (A) AM90/MMA10; (B) AM80/MMA20; (C) AM70/MMA30; (D) AM60/MMA40; (E) AM50/MMA50; (F) AM40/MMA60; (G) AM30/MMA70; (H) AM20/MMA80.

Table 1. Equilibrium water content of AM/MMA copolymers.

<table>
<thead>
<tr>
<th>Sample</th>
<th>% Acrylamide</th>
<th>% Crosslinker 1</th>
<th>% Crosslinker 3</th>
<th>% Crosslinker 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>AM20</td>
<td>20</td>
<td>31.61</td>
<td>20.09</td>
<td>22.16</td>
</tr>
<tr>
<td>AM30</td>
<td>30</td>
<td>33.87</td>
<td>32.64</td>
<td>31.93</td>
</tr>
<tr>
<td>AM40</td>
<td>40</td>
<td>49.61</td>
<td>44.62</td>
<td>41.84</td>
</tr>
<tr>
<td>AM50</td>
<td>50</td>
<td>65.15</td>
<td>56.71</td>
<td>53.32</td>
</tr>
<tr>
<td>AM60</td>
<td>60</td>
<td>73.94</td>
<td>67.24</td>
<td>64.35</td>
</tr>
<tr>
<td>AM70</td>
<td>70</td>
<td>81.55</td>
<td>75.16</td>
<td>71.63</td>
</tr>
<tr>
<td>AM80</td>
<td>80</td>
<td>85.18</td>
<td>79.95</td>
<td>75.80</td>
</tr>
<tr>
<td>AM90</td>
<td>90</td>
<td>87.10</td>
<td>83.18</td>
<td>¿</td>
</tr>
</tbody>
</table>

Figure 2 shows that swelling kinetics for the hydrogels synthetized using 1% of crosslinker regardless of their composition follows a second–order kinetics. Hydrogels synthetized with 3 and 5% of crosslinker also followed second–order swelling kinetics.

Experimental data can be analyzed by representing the fractional release in function of time as:

\[
\frac{M_f}{M_\infty} = K t^n
\]
In this case K represents a kinetic parameter and n is related to the diffusional mechanism.

For the series studied, the swelling experimental data were analyzed using eq. (4), and the n values are shown in table 2. The n values ranged from 0.55 to 0.76, which indicates that diffusion and polymer relaxation rates are comparables (case II, Non–Fickian or anomalous diffusion) (Katime et al., 2001).

**Table 2. Values for n of AM/MMA hydrogels.**

<table>
<thead>
<tr>
<th>Sample</th>
<th>% Crosslinker</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>AM20</td>
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</tr>
<tr>
<td>AM30</td>
<td>0.55</td>
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<tr>
<td>AM40</td>
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<td>AM50</td>
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</tr>
<tr>
<td>AM60</td>
<td>0.71</td>
</tr>
<tr>
<td>AM70</td>
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</tr>
<tr>
<td>AM80</td>
<td>0.72</td>
</tr>
<tr>
<td>AM90</td>
<td>0.76</td>
</tr>
</tbody>
</table>

**Fig 2.** Linear regression according to second order kinetics of AM/MMA/NMBA (20/80) hydrogels.
3.2 DSC studies
Water is present in hydrogels in several states which range from free water whose behavior is similar to pure water, to associate water where water molecules are hydrogen bonded to the polymer. When swollen hydrogels are frozen, only free water can freeze and melt (Baker et al., 1992; Liang and Liu, 2007). Quantitative determination of free water (or freezing water) is obtained by DSC, and bound water (or non-freezing water) is obtained by the subtraction of freezing water from EWC (Katime et al., 2004; Katime et al., 2001)

Figure 3 shows DSC heating curves of water swollen AM/MMA hydrogels with a content of 5% of NMBA at a heating rate of 5 degree·min⁻¹. All the hydrogels show overlapped and/or split peaks at approximately 0°C. This suggests that there is more than one state of freezing water (i.e. free water and freezable bound water) present in the hydrogels. Multiple melting endotherms have been reported for systems where a high hydrophilic monomer is copolymerized with a hydrophobic one. Hydrogels with medium and high amounts of AM gave broad overlapped peaks. Similar results were obtained for the other hydrogels with contents of 1 and 3% of NMBA. DSC Hydrogels with compositions AM/MMA (60/40) and (50/50) and 5% of NMBA (Figure 3) show peaks at around −12°C which indicate tendency of melting recrystallization and melting, as it is usually seen in semi crystalline polymers and has also been reported for hydrogels. As NMBA concentration increases the network becomes more tightly and there is a substantial decrease in free water content. However, when the NMBA content increases there is more notorious the presence of multiple peaks.

![Fig 3. DSC thermograms of AM/MMA hydrogels containing 5% w/w of NMBA. (a) AM90/MMA10; (b) AM80/MMA20; (c) AM70/MMA30; (d) AM60/MMA40; (e) AM50/MMA50; (f) AM40/MMA60; (g) AM30/MMA70; (h) AM20/MMA80.](image-url)
For high MMA content hydrogels, an increment in NMBA concentration, leads to the reduction of peak splitting tendency as could be seen in Figure 4. Hydrogels with 80% of MMA gave only one endothermic peak for the three-crosslinking levels studied. The appearance of a second endothermic peak depends of both acrylamide and NMBA levels. For 1% NMBA second peak appeared at 30% AM, for 3% NMBA at 40% and for 5% NMBA at 50%. This tendency supports the idea that for the gels rich in MMA synthesized in this work, NMBA plays an important role as a hydrophilic component and induce different states for water association.

**Fig 4.** Effect of crosslinker on peak splitting for hydrogels with high content of MMA. (A) AM/MMA hydrogels with 1% of crosslinking agent, (B) AM/MMA hydrogels with 3% of crosslinking agent and (C) AM/MMA hydrogels with 5% of crosslinking agent.

For computational purposes, the area of all the peaks in thermograms is assumed as freezing water. The distribution of water in the studied hydrogels is schematized in figures 5–7, as function of acrylamide content. Respect to AM content at constant NMBA, for the hydrogels richer in
acrylamide, most water is freezable water. However, in low AM hydrogels most of the water is bound to the polymer and freezing water content falls to a low value. The water absorption capacity for these highly hydrophobic hydrogels is reduced due to the hydrophobic nature of MMA and water should be strongly bound to the few amide groups. The strong binding of water molecules causes that the majority of the water present low mobility and cannot freeze and melt. Bound water content increases as acrylamide content increases up to a maximum at about 40 or 50% AM. This indicates the bound water is dependent on the hydrophilic component, when AM predominates in the copolymer composition, water molecules are more mobile and their behavior is more similar to free water.

Fig 5. Effect of monomer ratio on the water contents of AM/MMA hydrogels crosslinked with 1% w/w of NMBA. (A) EWC; (B) freezing water; (C) non-freezing water.
Fig 6. Effect of monomer ratio on the water contents of AM/MMA hydrogels crosslinked with 3% w/w of NMBA. (A) EWC; (B) freezing water; (C) non-freezing water.

Fig 7. Effect of monomer ratio on the water contents of AM/MMA hydrogels crosslinked with 5% w/w of NMBA. (A) EWC; (B) freezing water; (C) non-freezing water.

Figure 4 shows the thermograms obtained for different samples of MM/AM hydrogels with different molar compositions and different crosslinking degrees. A can be observed that when the content of crosslinker is at 1% the equilibrium water content increases significantly with the
increasing of a hydrophilic monomer (AM) in the copolymeric hydrogel composition, presenting a
transition near to 0°C. On the other hand, on thermogram B can be appreciate that when there is
present a higher degree of crosslinking in the polymer network (3%) the displayed transition of
hydrogels moves towards higher values near 0.3°C, this is due that by increasing the crosslinking
density of polymer network this acquires a greater sense of rigidity and thus the transition will be
presented with greater temperature, this behavior can also be explained in terms of content of
hydrophilic monomer in the gel, as observed in this thermogram a higher content of AM causes an
increase in the equilibrium water content (W_e) whose peak is below the value presented by the
hydrogel with 1% crosslinking (see table 1). This tendency to increase the transition temperature is
checked with thermogram C. The presence of hydrophilic groups as well as a greater crosslinking
degree show that the EWC has a lower value for those AM/MMA hydrogels synthesized with 1 and
3% of crosslinking agent.

Upon initial absorption of water, water molecule disrupted the intermolecular hydrogen bonds and
bounded with hydrophilic sites and uniformly distributed to the whole network with restricted
mobility, (bound or non-freezable water). Increasing water content caused sufficient increase in non–
freezable water content up to 30% and eventually reached almost saturated value. When water content
reached above this critical value, a part of water began to crystallize at 50% water content, leading to
appearance of freezable water. It indicates that hydrophilic groups interact with 30–40% water
(water–polymer interaction), either directly or via other water molecule. Otherwise, increasing water
content tends to increase freezable water content, confirming the existence of bulk–like water or
water cluster in water–saturated hydrogel.

In the table 1 it is shown how the W_e increases with higher amounts of acrylamide. This effect is
due to the hydrophilic groups from the acrylamide in the hydrogel. As a consequence of this, more
secondary interactions with the water appear. The electrostatic interactions from free electron par
from the amide stabilize the polymeric matrix. This produces a less collapsed matrix that make the
system more suitable to swell. Referring to the effect of the crosslinker agent, its increment makes
the W_e decreases. This indicates that the increase of the feeding of the crosslinker agent in the
polymer has been effective. The NMBA has been incorporated as other comonomer in the polymer
chains, generating a decrease of the average number of repetitives units between crosslinking
points and restricting the free volume available among the hydrogel chains, due to the increase of
elastically effective chains concentration. This global effect is noticeable in the hydrogels with more
quantity of crosslinker agent and less quantity of acrylamide.

3.3 Urea release
One example of the behavior of the urea released experiments is shown in figure 8, where it can be
seen that release rate is fast, 50% of the urea was released in approximately 50 minutes and around
200 minutes all the urea was released. Similar results were obtained for the release of urea from the
other hydrogels.
Absolute quantities of released urea are shown in figure 8. Since practically all of the urea present in the hydrogels was released, from this plot it is concluded that the urea captation capacity is higher as the acrylamide content increases. When using a high proportion of acrylamide to synthesize the hydrogel (90 and 80% AM), the water and urea content diminishes as the amount of crosslinker (NMBA) increases because there is less available space inside the polymeric network. Figure 9 also shows that at lower AM content (≤ 70% AM) total urea released presents an u–shaped curve because by increasing crosslinker content from 1 to 3% being the network more tight the capacity of the hydrogel to absorb water and urea decreases, however, by a further increase in NMBA, though the net becomes more tight, the hydrophylicity of the NMBA overcomes the decrease on free volume and the net effect is a slightly larger capacity of urea absorption.

4. Conclusions

In this paper we have examined the effect chemical structure on the water absorption and urea release behavior of AM/MMA hydrogels crosslinked with NMBA. The equilibrium water content is clearly related to a hydrophilic—hydrophobic balance in the copolymer; thus EWC increases as hydrophilicity of the hydrogel increases. As expected the absorption capacity decreases with increasing crosslink density, although hydrogels with the lowest acrylamide content were affected by crosslinker’s hydrophilicity, increasing the absorption capacity as NMBA was increased in the polymerization recipe. According to these results, NMBA acts not only as a multifunctional monomer but also as a hydrophilic component, and the effect in the hydrogel behavior depends on the balance between crosslinking and hydrophilicity.

The experimental data indicated that the swelling process follows second order kinetic for all the studied hydrogels. The kinetic parameters showed dependence with chemical composition. Data
could not be analyzed by Fick's equation, because the n exponent was below 0.5 for all the hydrogels.

The release of urea from AM/NMBA copolymers presented anomalous behavior respect to crosslinking ratio; hydrogels with 3% of crosslinker released the lowest urea amount. This result is in agreement with the behavior of water swelling. At the beginning of the urea release process, the velocity was very high. Then the release of the urea became very slow and complete release was over in 60 minutes.

DSC experiments showed the free water contents follow the same behavior that the total water content (EWC) respects to the chemical structure. The percent of non–freezing water decreased as hydrophilicity of the hydrogel increased. This clearly shows that in hydrophilic networks the majority of the imbibed water molecules only fills up the vacant spaces and don’t form chemical bonds with the polymer. In contrast, for hydrophobic gels most of the water is strongly associated to the polymer. Multiple peaks show that more than one state of association is present for this hydrogels.

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References


