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# Synthesis and characterization of new hydrogels through copolymerization of *N*-acryloyl-tris-(hydroxymethyl) aminomethane and different crosslinking agents

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## ABSTRACT

In this study, new hydrogels in rod shape were prepared from *N*-acryloyl-tris-(hydroxymethyl)aminomethane (NAT) using three different crosslinking agents: poly(2-methyl-2-oxazoline) bismacromonomer (BM), ethylene glycol dimethacrylate (EGDMA) and *N,N'*-methylenebisacrylamide (BIS). Dimethylformamide (DMF) was used as solvent and 2,2'-azobisisobutyronitrile (AIBN) as initiator. Polymeric matrices with different properties were obtained by free radical polymerization by changing the crosslinker (BM, EGDMA or BIS) or the concentration of BM. The hydrogel structures were characterized by high resolution magic angle spinning (HRMAS) NMR technique. Swelling experiments and rheological studies were used to test the water absorption capacity and viscoelastic properties of the hydrogels, respectively. For a given NAT/crosslinking agent molar ratio, the hydrogel synthesized with BM displays higher water absorptive capacity and larger range of linear viscoelasticity than those synthesized with BIS or EGDMA. The relatively larger hydrophilic character of the former and the lower crosslinking density generated by the longer molecules of BM might be the cause of this behavior. The results also reveal that water diffuses into the network following a non-Fickian mechanism. This is concluded from the value of the diffusion exponent  $n$ , which is higher than 0.50. The elastic modulus and the equilibrium water content (EWC) measurements suggest that these materials may have potential application as biomaterials.

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## 1. Introduction

Hydrogels, which are three-dimensional water-swella-ble crosslinked structures with physical integrity, have many potential applications [1–4]. It is well known that these applications are closely related to the intrinsic properties of the polymeric networks [5]. In this sense two main characteristics of hydrogels should be specially considered, the chemical structure of the components and the morphology of the network. The former affects the hydrophilic/

hydrophobic ratio of the gel and, consequently, the water retention and absorption capacity of them. In general, monomers and crosslinking agents with polar functional groups increase the hydrophilic nature of the gel. Therefore, monomers such as 2-hydroxyethylmethacrylate (HEMA), acrylamide (AA), acrylic acid (AAc), and crosslinkers such as ethylene glycol dimethacrylate (EGDMA) and *N,N'*-methylenebisacrylamide (BIS) are commonly used for the synthesis of hydrogels with high water retention capacity. The other characteristic, the morphology of the network, exerts influence on the mechanical and rheological properties of the network since these properties are affected by

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the stiffness/flexibility of segments between crosslinks and crosslink density. Consequently, considering the possible applications of hydrogels are important to analyze both, the swelling index and the elastic modulus. Hydrogels with high water retention capacity and different elastic modulus may have diverse macroscopic physical properties related to damping of vibrations, deformability, mechanical strength, and so forth which may give place to several applications [6–12]. However, hydrogels rarely possess high values of both parameters simultaneously [4].

Different chemical and physical crosslinking methods have been reported for the production of hydrogels [3]. Several works have also described the synthesis of new macromonomers [13–19] and crosslinkers [20–22] to be used in the manufacture of polymeric networks. Different swelling and mechanical properties are observed in hydrogels according to the nature and concentration of the crosslinking agent and to the nature of the swelling medium used in the preparation [4].

*N*-acryloyl-tris-(hydroxymethyl)aminomethane (NAT) is a hydrophilic monomer rarely used in the synthesis of hydrogels. Only few papers report the employ of this monomer. NAT has been utilized as a component of multifunctional polyacrylamides [23] and in gels for electrophoresis and electrofocusing applications where BIS was used as crosslinking monomer [24]. A quasi-interpenetrating network formed by poly(NAT) and polyvinylpyrrolidone was used as separation matrix for double-stranded DNA and single-stranded DNA fragments by capillary electrophoresis with UV detection [25]. Additionally, hydrogels in rod shape have been prepared utilizing NAT and HEMA or AAc as comonomers. Some of the final products of these polymerizations were selected to perform urea release assays, conducted through swelling-controlled release [6].

Moreover, the polymerization of 2-substituted 2-oxazolines has been intensively studied by several research groups since the 1960s and reviewed in many references [26,27]. Different structures can be easily synthesized using these monomers, for example, block and graft copolymers, telechelics and hydrogels [28–32]. In particular, Rueda et al. have reported non-ionic hydrogels that were synthesized by the radical homopolymerization of vinylterminated poly(2-methyloxazoline) bismacromonomer (BM) and by radical copolymerization of this telechelic with 1-vinyl-2-pyrrolidone [30].

In this work, new hydrogels synthesized with NAT as the hydrophilic monomer and three different crosslinking agents: BM, BIS and EGDMA, are discussed. The matrices were characterized by high resolution magic angle spinning (HRMAS) NMR and infrared spectroscopy (FT-IR). Swelling behavior in water and linear viscoelasticity were compared and analyzed to gain insights into the influence of the crosslinker agent on these hydrogel properties.

## 2. Experimental section

### 2.1. Reagents and equipment

The following chemicals were used as purchased: *N*-acryloyl-tris-(hydroxymethyl) aminomethane (Aldrich),

*N,N'*-methylenebisacrylamide (Mallinckrodt), ethylene glycol dimethacrylate (90%, Fluka), 2,2'-azobisisobutyronitrile (AIBN) (Aldrich) and sodium iodide (Merck). Dimethylformamide (DMF) (p.a., Cicarelli) was purified through vacuum distillation (84 °C/90 mm Hg); diethyl ether (Aldrich) and chloroform (Merck) were used as received. Chloromethylstyrene (CMS) [mixture of *meta*- (70 mol-%) and *para*- (30 mol-%) isomers] was distilled before use. 2-methyl-2-oxazoline (MeOXA) and *N*-(4-vinylbenzyl)piperazine (VBP) were synthesized according to literature procedures [33,34]. Acetonitrile was distilled twice from calcium hydride (CaH<sub>2</sub>) and stored under a dry nitrogen atmosphere. Fourier transform infrared spectra (FT-IR) were obtained on a Nicolet 5-SXC FT-IR spectrometer on potassium bromide (KBr) discs. A Bruker DRX 500 spectrometer and a rotational rheometer RDA-II from Rheometrics were used to characterize the hydrogels. The NMR spectrum of poly (NAT) was obtained using a Bruker 200 MHz NMR spectrometer.

### 2.2. Synthesis of vinylterminated poly(2-methyl-2-oxazoline) bismacromonomer

Poly(2-methyl-2-oxazoline) bismacromonomer (BM) was synthesized according to reference [30]. 2-Methyl-2-oxazoline (28.20 mmol) and sodium iodide (11.26 mmol) were dissolved in acetonitrile (15 ml) and chloromethylstyrene (5.63 mmol) was added to the mixture. The system was closed and stirred under dry nitrogen atmosphere at 85 °C for 7 h. The system was then cooled and *N*-(4-vinylbenzyl)piperazine (VBP) (11.26 mmol) was added. The reaction mixture was stirred at 25 °C for 12 h. The acetonitrile was then evaporated and the excess of VBP in the polymer was extracted exhaustively with diethyl ether. The synthesized polymer was dissolved in chloroform, and potassium carbonate was added to the solution and stirred for 12 h at room temperature. The mixture was filtered, the chloroform evaporated and the polymer was finally dried under vacuum. A 95% monomer conversion was attained. The BM was characterized by NMR and its  $M_n$  was determined from the <sup>1</sup>H NMR spectrum by end group analysis.

### 2.3. Synthesis of products

Five hydrogels were prepared by free radical crosslinking polymerization: H1, H2, H3, H4 and H5. The procedure used during the copolymerizations can be described as follows: the NAT monomer and the crosslinking agent (BM, EGDMA or BIS) were dissolved in DMF in glass tubes (14 mm internal diameter and 15 cm long) used as polymerization reactors. Then, the initiator (AIBN) was added to each solution which was stirred during 75 min in an ultrasonic bath and left to react for 2.5 h at 65 °C. The experimental conditions used to prepare all the synthesized materials are summarized in Table 1.

After breaking the tubes, the yielded matrices of long cylindrical shape were cut into pieces (3 mm thick and 14 mm diameter) and immersed twice in a large excess of distilled DMF at room temperature for 24 h. Finally the discs were exhaustively washed with water and methanol,

**Table 1**

Experimental reaction conditions used in the synthesis of the products

Product <sup>a</sup>	Cross-linking agent	Crosslinking agent(g)	Crosslinking agent (mmol)	R <sup>b</sup>	Type of product
Poly(NAT)	–	–	–	–	Soluble
H1	BM	0.0960	0.137	0.030	Insoluble
H2	BM	0.1465	0.209	0.046	Insoluble
H3	BM	0.1954	0.279	0.061	Insoluble
H4	EGDMA	0.0552	0.278	0.061	Insoluble
H5	BIS	0.0420	0.273	0.060	Insoluble

<sup>a</sup> All products were obtained using 0.8 g (4.57 mmol) of NAT and 0.066 g (0.40 mmol) of AIBN. Reaction conditions: 2.5 h at 65 °C in 2.5 mL DMF.

<sup>b</sup> R = total moles of crosslinking agent/total moles of monomer.

dried under vacuum to constant weight, and stored at room temperature.

Besides, NAT was also homopolymerized without cross-linking agent (Table 1). In this case, NAT was dissolved in DMF in a glass tube. Then AIBN was then added and the solution was stirred for 75 min in an ultrasonic bath and left to react for 2.5 h at 65 °C. A soluble poly(NAT) product was attained, which was later purified, dried and later characterized by <sup>1</sup>H NMR.

#### 2.4. HRMAS NMR and FT-IR characterization

For the FT-IR characterization, the polymers were powdered and mixed with KBr. Because the hydrogels are insoluble but swellable, they were characterized by high resolution magic angle spinning (HRMAS) technique [29,30,35]. The samples (ca. 3 mg) were packed into a 4-mm HRMAS rotor and D<sub>2</sub>O was directly added to the hydrogels inside the rotor. The samples were spun with 4.5 kHz at 303 K. The 500 MHz <sup>1</sup>H NMR spectrum of the BM was recorded with the same spectrometer using CDCl<sub>3</sub>/DMSO-*d*<sub>6</sub> (4:1 v/v) as solvent. The spectrum was calibrated on the DMSO-*d*<sub>5</sub> signal (2.5 ppm).

#### 2.5. Swelling experiments

Dried samples of hydrogels were placed in distilled water and kept at room temperature in order to determine their swelling behavior. Swollen samples removed from the water bath at regular intervals were superficially dried with tissue paper, weighed at an electronic balance and placed back in the water bath. The measurements were repeated until a constant weight was achieved for each sample. The weight swelling ratio at time,  $q_w$ , the weight equilibrium swelling ratio,  $q_{w,e}$ , the degree of swelling, DS, and the equilibrium water content, EWC, were calculated according to the following equations:

$$q_w = \text{swollen mass at time/dry mass} \quad (1)$$

$$q_{w,e} = \text{swollen equilibrium mass/dry mass} \quad (2)$$

$$\text{DS} = (\text{swollen equilibrium mass-dry mass})/\text{dry mass} \quad (3)$$

$$\text{EWC} = (\text{swollen equilibrium mass-dry mass})/\text{swollen equilibrium mass} \quad (4)$$

To determine the mechanism of diffusion of water into the matrices, the data were fitted with Fick's law [36]:

$$F = M_t/M_\infty = kt^n \quad (5)$$

where  $M_t$  denotes the amount of water diffused into the matrix at time  $t$  (swollen mass at time  $t$  – dry mass),  $M_\infty$  corresponds to the amount of water that has diffused into the matrix at equilibrium (swollen mass at equilibrium – dry mass) and  $k$  is a constant related to the structure of the network. The exponent  $n$  is the number that determines the type of diffusion mechanism. When Eq. (5) was applied to the initial stages of swelling, the plots of  $\ln F$  as a function of  $\ln t$  yielded straight lines up to values of  $q_w$  of approximately 0.6. From the fitting of these curves to Fick's law,  $n$  and  $k$  values were determined from the slope and the intersection of the lines, respectively.

#### 2.6. Rheological characterization

The elastic,  $G'$ , and the viscous,  $G''$ , moduli of the matrices were measured in a rotational rheometer using parallel plates of 25 mm in diameter and gaps of approximately 2 mm. The dynamic moduli were measured in small-amplitude oscillatory shear flow at room temperature. Dynamic strain sweeps at a frequency of 10 s<sup>-1</sup> were performed on each sample to determine the strain range of linear response of each material. Then, dynamic frequency sweeps were applied to the samples in a 0.1–400 s<sup>-1</sup> range. No evidence of dehydration during the tests was found when a second frequency sweep was applied to some of the samples.

### 3. Results and discussion

In this study, free radical polymerization process was used to synthesize new gel-like materials derived from NAT monomer and three different crosslinking agents: BM, BIS and EGDMA. Fig. 1 shows the chemical structure of the start materials.

#### 3.1. Synthesis of vinylterminated poly(2-methyl-2-oxazoline) bismacromonomer

The bismacromonomer BM was synthesized by the “living” cationic ring-opening polymerization of MeOXA, through the successive application of “initiating” and “end-capping” methods. Iodomethylstyrene and *N*-(4-vinylbenzyl)piperazine were used as polymerization initiator and termination agent as described in the literature [30]. Due to the “living” character of the polymerization of the 2-methyl-2-oxazoline, it is possible to adjust the polymer molecular weight by the molar ratio between

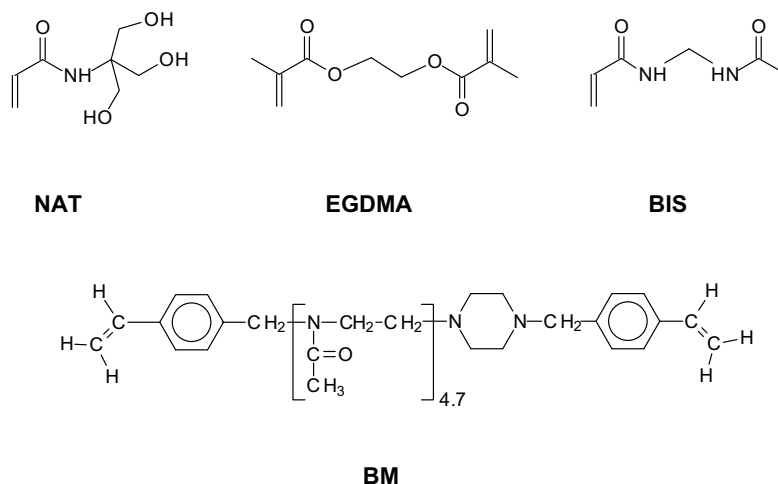


Fig. 1. Chemical structure of NAT and crosslinking agents.

monomer to initiator. The synthesized BM, with a molecular weight of  $M_n = 750$  g/mol, was obtained with a monomer conversion of 95% and a polymerization degree of  $x = 4.7$ . The value of  $x$  is in agreement with the theoretical value of 5 calculated from the ratio  $[\text{MeOXA}]/[\text{CMC}]$  with 100% conversion.

In agreement with its hydrophilic nature, BM is soluble in polar solvents.

The structure of BM was confirmed by its  $^1\text{H}$  NMR spectrum, which is depicted in Fig. 2.

### 3.2. Synthesis of products

As a first step, NAT monomer was homopolymerized under the experimental reaction conditions listed in Table 1. Poly(NAT) remained soluble in the medium of reaction. Its  $^1\text{H}$  NMR spectrum in  $\text{DMSO}-d_6$  showed the  $-\text{CH}_2-\text{OH}$  meth-

ylene group signal of NAT at 3.80 ppm and  $-\text{CH}_2-$  and  $-\text{CH}-$  of the backbone structure as broad signals at 1.64 and 2.26 ppm, respectively.

Afterwards, hydrogels from NAT monomer and different crosslinking agents were synthesized by free radical copolymerization. Due to the double vinyl functionality and the oligomeric character of the bismacromonomer BM, this element played the role of a “macro-crosslinker” in the polymerization reaction. Three hydrogels (H1–H3) were prepared using different concentrations of BM (see Table 1). A crosslinking agent/NAT molar ratio ( $R$ ) larger than 0.045 was necessary to obtain a stable three-dimensional network. Lower ratios led to soluble or unstable networks (H1). The other two synthesized matrices, H4 and H5, were prepared using EGDMA and BIS as crosslinking agents, respectively.

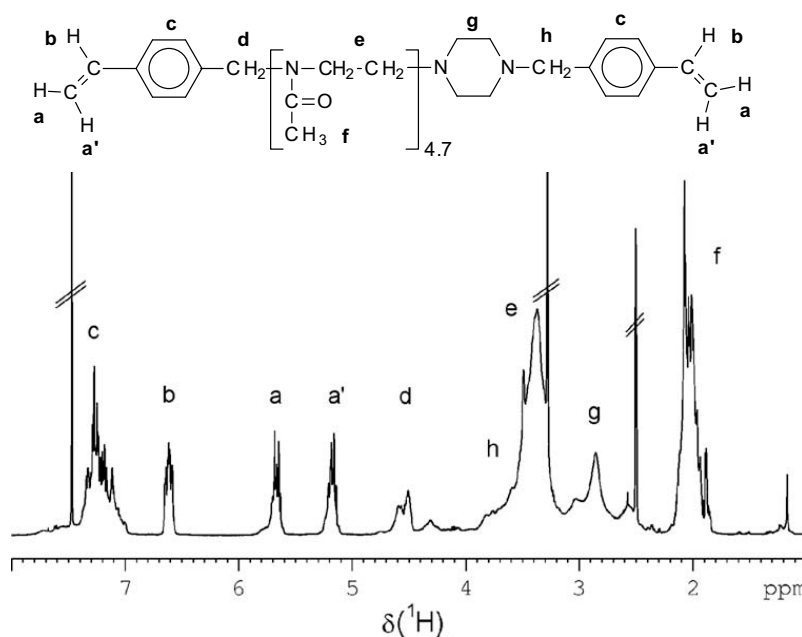


Fig. 2.  $^1\text{H}$  NMR spectrum of the synthesized poly(2-methyl-2-oxazoline) bismacromonomer in  $\text{CDCl}_3/\text{DMSO}-d_6$  (4:1 v/v).

### 3.3. HRMAS NMR and FT-IR characterization

The chemical structural characterization of the hydrogels was done by FT-IR and  $^1\text{H}$  HRMAS NMR. The FT-IR spectra showed the bands confirming the presence of the functional groups of NAT (polymeric association O–H: 3230–3400  $\text{cm}^{-1}$ ; amide C=O: 1660  $\text{cm}^{-1}$ ; and primary alcohol C–O: 1049 and 1380  $\text{cm}^{-1}$ ). However, the bands associated to the crosslinking agents were not noticeable in the spectra, surely due to the low content of these components in the structure of the networks. No evidence of the absorption bands of vinyl groups, present in both, the monomer and the crosslinkers (905 and 990  $\text{cm}^{-1}$ ), was found in all spectra, which suggest that complete reaction of polymerization has occurred in all cases.

From the analysis of the  $^1\text{H}$  NMR spectra of all samples swollen in  $\text{D}_2\text{O}$ , it can be observed that the signals of the poly(NAT) structure are well resolved (see Fig. 3). In accordance with the  $^1\text{H}$  NMR spectrum of NAT homopolymer, the  $-\text{CH}_2-\text{OH}$  methylene group signal at 3.80 ppm is the strongest one. The  $-\text{CH}_2-$  and  $-\text{CH}-$  signals of the backbone structure appear in the region of 1.4–2.5 ppm as indicated in Fig. 3b. The corresponding  $^{13}\text{C}$  NMR signals appear at 36–40 ( $\text{CH}_2$ ), 44–46 ( $\text{CH}$ ), 63.3 ( $\text{CH}_2\text{OH}$ ), 65.2 (C) and 180.1 ppm ( $\text{C}(\text{O})\text{NH}$ ). The proof of the crosslinking agents by their NMR signals is hampered by the low content of crosslinker and by the restricted mobility at the crosslinking points resulting in larger signal broadening. For H1, H2 and H3 the incorporation of BM in the gels is confirmed by

the  $^1\text{H}$  NMR signals of the polyoxazoline backbone at 2.15 ppm ( $\text{CH}_3\text{C}(\text{O})-$ ) and 3.62 ppm ( $\text{NCH}_2$ ) (Fig. 3a) and also by  $^{13}\text{C}$  NMR spectra. Compared to the intensity of the  $-\text{CH}_2-\text{OH}$  methylene group signal of NAT the intensity of the assigned BM signals increase from H1 to H3 as expected from the crosslinker feed concentration. However, a quantification of both components was not possible due to signal overlap. Vinyl group signals, which would indicate dangling chain ends, could not be detected. A phenyl signal corresponding to the relatively more hydrophobic styrene of the crosslinks could be observed at  $\sim 7.3$  ppm, showing the mobilizing effect of the well solvated and mobile polyol backbone units on the isolated crosslinking points.

In H4 and H5, the crosslinking points introduced by EGDMA and BIS are separated by just 7 and 6 bonds, respectively. This separation, apparently, is not large enough to achieve sufficient chains mobility to obtain detectable NMR signals by swelling and MAS. So, whether neither the  $\text{OCH}_2$  nor the  $\text{NCH}_2\text{N}$  moiety was proven. However, a signal of the free rotating methyl group of EGDMA appeared both in the  $^1\text{H}$  NMR (1.12 ppm, Fig. 3b) and in the  $^{13}\text{C}$  NMR ( $\sim 21.3$  ppm) spectra.

### 3.4. Swelling experiments

The swelling behavior of the hydrogels constitutes one of the most important characteristics when evaluating the ability of a polymeric gel to function in a particular application [3,5,37]. As already explained, the swelling behavior of the prepared hydrogels in water was gravimetrically determined.

Table 2 lists the measured values of the equilibrium parameters,  $q_{w,e}$ , DS and EWC, of the matrices. As expected, in the case of the hydrogels based on BM, the increase in crosslinking density, produced by the increase in concentration of the crosslinking agent, results in a reduction of the amount of water retained by the hydrogel. Thus, the three parameters, DS,  $q_{w,e}$  and EWC, decrease when R increases. In addition, EWC values of the hydrogels (0.81–0.92) are higher than the value of water content found in most human tissues (about 0.6) [7]. This result suggests that the analyzed NAT hydrogels may have biomedical applications.

The swelling behavior of the hydrogels is affected by several characteristics of the crosslinker agents, such as hydrophilicity, length and reactivity. Higher hydrophilicity and larger distance between crosslinking points formed lead to the increase of water retention capacity, while

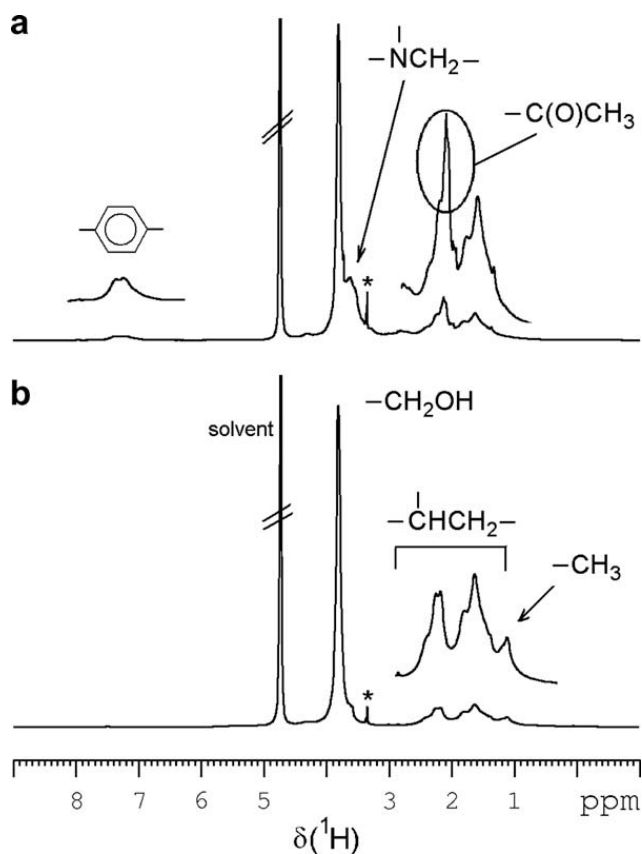


Fig. 3.  $^1\text{H}$  HRMAS NMR spectra of hydrogels H3 (a) and H4 (b) in  $\text{D}_2\text{O}$ . (\*) Residual methanol from synthesis.

Table 2  
Results of swelling tests

Hydrogel	R	$q_{w,e}$	DS	EWC	n
H1	0.030	a	a	a	a
H2	0.046	12.74	11.74	0.92	–
H3	0.061	9.03	8.03	0.89	0.85
H4	0.061	6.65	5.65	0.85	0.70
H5	0.060	5.15	4.15	0.81	0.66

<sup>a</sup> H1 was very swellable in water. It was not possible to determine with precision the parameters  $q_{w,e}$ , DS and n was impossible.

higher reactivity of the crosslinker produce the contrary effect due to the increase of crosslinking degree. Consequently, for equal monomer and crosslinker concentrations, it should be expected that the hydrogels synthesized with bismacromonomer BM ( $DP = 4.7$ ) must have larger absorptive capacity than those synthesized with BIS or EGDMA. Effectively, BM, the largest crosslinker, which also contains methyl oxazoline groups (polar groups), leads to the hydrogel with the largest water retention capacity (see Table 2).

Fig. 4a shows the time-dependent swelling behavior curves of materials H3, H4 and H5 while Fig. 4b displays the same curves in the first stages of swelling up to 120 min. This figure depicts the difference in hydrogel swelling speed. The larger the final swelling capacity of the hydrogels, the higher the swelling speed was reached. However, for the three samples, the equilibrium weight swelling ratio  $q_w$  was reached after approximately 6 h.

The fitting of Eq. (5) to the data of the initial stages of swelling was carried out to determine the mechanism of diffusion of water into the hydrogels. For samples of cylindrical shape the diffusion is Fickian if  $n$  is in the range of 0.45–0.50, whereas the diffusion is non-Fickian type if

$0.50 < n < 1.00$ . In Table 2 are listed the values of the diffusion exponents  $n$  calculated for H3, H4 and H5. These values are larger than 0.50, indicating that in the three cases the diffusion of water into the hydrogels follows a non-Fickian mechanism (diffusion-relaxation controlled process). This behavior is caused by the relatively slow relaxation rate of the polymeric matrix, which is similar to the diffusion rate of the solute [38]. This means that the chains need relatively large times to adapt themselves to the penetration of water molecules and to adopt new equilibrium conformations according to the swelling pressure [36].

### 3.5. Rheological characterization

Fig. 5a displays the elastic modulus of the five hydrogels obtained during dynamic strain sweeps at ambient temperature using a frequency of  $10 \text{ s}^{-1}$ . This test determines the strain range of linear response of polymeric materials [39]. The data show that the family of hydrogels based on BM present larger range of linear viscoelastic behavior. Moreover, H3, prepared with practically the same concentration of crosslinking agent than H4 and H5, has a critical strain (limit of linear response)  $\sim 4$  times larger than the

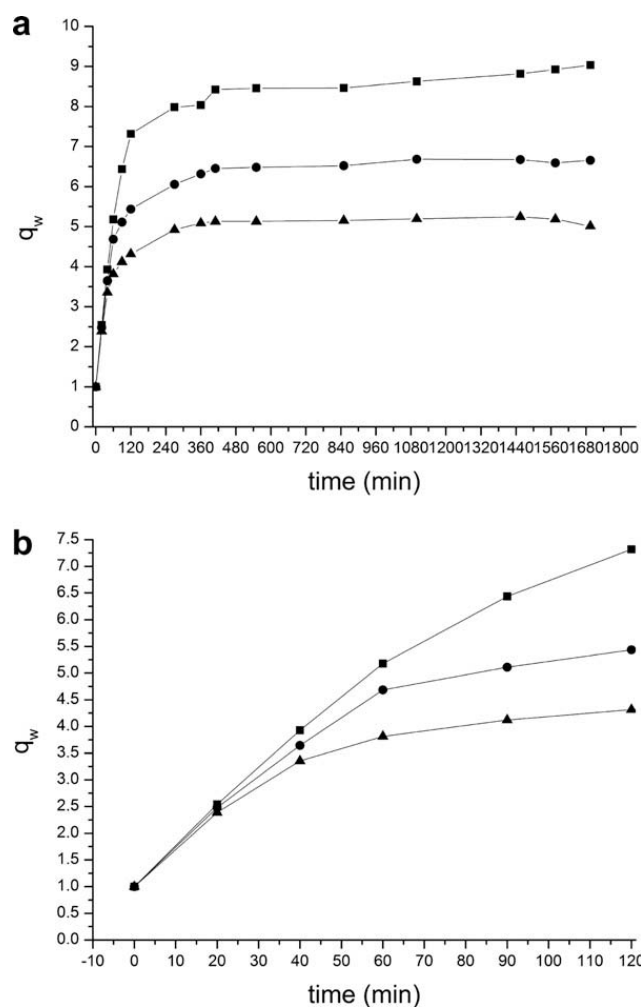


Fig. 4. (a) Time-dependent swelling behavior of hydrogels H3 (—■—), H4 (—●—) and H5 (—▲—). (b) Time-dependent swelling showing the data corresponding to the first 120 min.

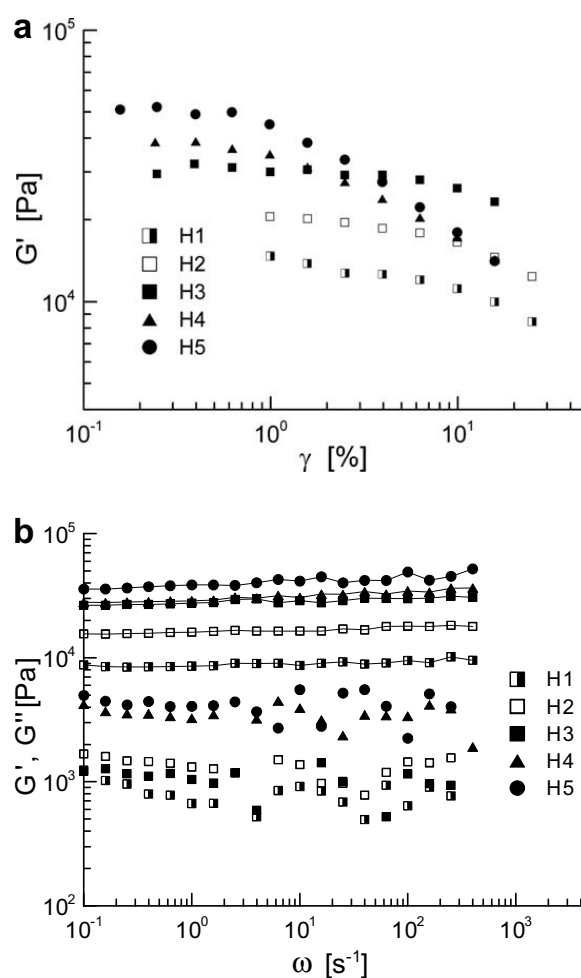


Fig. 5. (a) Elastic modulus as a function of strain measured using a frequency of  $10 \text{ s}^{-1}$ . (b) Elastic (symbols linked by full lines) and viscous (symbols with no lines) moduli of all hydrated matrices as a function of frequency.

critical strain of these two hydrogels. At the light of the diffusion results, the smaller size of the region of linear viscoelasticity of H4 and H5 might be related to the smaller size of the network pores and consequent larger crosslinking density of the matrices prepared with BIS and EGDMA.

The rheological characterization was completed with dynamic frequency sweeps performed at ambient temperature using strains smaller than the critical one for each material (in the range 0.1–4% depending on the polymer). The elastic and viscous moduli are displayed in Fig. 5b as a function of frequency. The rheological behavior of all samples is characteristic of lightly crosslinked polymer [39]. The elastic modulus is larger than the viscous one at low frequencies and has a value practically constant. As expected, the elastic modulus of the hydrogels based on BM increases as the concentration of crosslinking agent increases. The viscous modulus is much less affected by R, therefore, the phase angle ( $G''/G' = \tan \delta$ ) decreases as the crosslinking density increases. Interestingly, the elastic moduli of the three swollen matrices prepared with similar amounts of different crosslinking agents are quite similar. However, the viscous modulus of H3 is smaller than those of H4 and H5 giving place to a much lower  $\tan \delta$  for this material. The results suggest that, even though the matrix based on BM might have smaller crosslinking density, the stretched state of its water-swollen pores produces a crosslinked system that shows an elastic behavior similar to that of H4 and H5 but with a smaller amount of energy dissipated during the flow [39].

#### 4. Conclusions

In this work, hydrogels based on NAT monomer and different crosslinking agents, BM, BIS and EGDMA were synthesized. The structure of the hydrogels was analyzed using HRMAS NMR and FT-IR spectroscopy. Swelling and rheological measurements were used to characterize the synthesized materials. For a given monomer and crosslinker concentration the hydrogel synthesized with bismacromonomer BM has larger water absorptive capacity than those synthesized with BIS or EGDMA. The relatively higher hydrophilicity of BM and the relatively larger size of the network pores due to the greater length of BM may be the reason of this.

The water diffuses into the networks following a non-Fickian mechanism. This is concluded from the value of the diffusion exponents  $n$  which are larger than 0.50.

The hydrogels based on BM present a larger range of linear viscoelasticity than those prepared with BIS and EGDMA. The frequency dependence of the dynamic moduli is typical of lightly crosslinked polymer, i.e. the elastic modulus is larger than the viscous one at low frequencies and has a value practically constant. As expected, the elastic modulus of the hydrogels based on BM is more affected than the viscous one by increasing of the concentration of the crosslinking agent. Additionally, the elastic moduli of the three swollen matrices prepared with the same concentration of different crosslinking agents are quite similar whereas the viscous modulus of H3 is smaller than that of H4 and H5. This behavior may result from the stretched

state of the water-swollen pores of the more hydrophilic H3 that, according to the  $G''$  results, dissipates less amount of energy than the other two matrices in an equivalent flow.

Furthermore, the hydrogels developed in this study exhibit fluid contents (EWC) and viscoelastic properties similar to those of soft body tissues, suggesting that they have potential biomedical applications.

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#### References

- [1] Langer R, Peppas NA. Advances in biomaterials, drug delivery and bionanotechnology. *AIChE J* 2003;49:2990–3006.
- [2] Hennink WE, Van Nostum CF. Novel crosslinking methods to design hydrogels. *Adv Drug Deliv Rev* 2002;54:13–36.
- [3] Galaev I, Mattiasson B. Smart polymers: applications in biotechnology and biomedicine. Boca Raton, FL: CRC Press; 2008.
- [4] Katime I, Katime O, Katime D. Los materiales inteligentes de este milenio: los hidrogeles macromoleculares. Síntesis, propiedades y aplicaciones. Bilbao, Spain: Servicio Editorial de la Universidad del País Vasco; 2003.
- [5] Gupta P, Varmani K, Garg S. Hydrogels; from controlled release to pH-responsive drug delivery. *Drug Discov Today* 2002;7:569–79.
- [6] Alvarez CI, Arrua RD. Crosslinking reactions and swelling behavior of matrices based on *N*-acryloyl-tris-(hydroxymethyl)aminomethane. *Polym Bull* 2005;55:19–29.
- [7] Karadag E, Üsum OB, Saraydin D. Swelling equilibria and dye adsorption studies of chemically crosslinked superabsorbent acrylamide/maleic acid hydrogels. *Eur Polym J* 2002;38:2133–41.
- [8] Hoffman AS. Hydrogels for biomedical applications. *Adv Drug Deliv Rev* 2002;43:3–12.
- [9] Jeon O, Song S, Lee K, Hyang Park M, Lee S, Kwan Hahn S, et al. Mechanical properties and degradation behaviors of hyaluronic acid hydrogels crosslinked at various crosslinking densities. *Carbohydr Polym* 2007;70:251–7.
- [10] Chang J, Oh Y, Choi M, Kim Y, Kim C. Rheological evaluation of thermosensitive and mucoadhesive vaginal gels in physiological conditions. *Int J Pharm* 2002;241:155–63.
- [11] Loizov E, Weisser J, Dundigalla A, Dorrar L, Schmidt G, Wilker J. Structural effects of crosslinking a biopolymer hydrogels derived from marine Mussel adhesive protein. *Macromol Biosci* 2006;6:711–8.
- [12] Ahearne M, Jang Y, El Haj A, Then K, Liu K. Characterizing the viscoelastic properties of thin hydrogel based constructs for tissue engineering applications. *J R Soc Interface* 2005;2:455–63.
- [13] He S, Timmer MD, Yaszemsky MJ, Yasko AW, Engel PS, Mikos AG. Synthesis of biodegradable poly(propylene fumarate) networks with poly(propylene fumarate)-diacrylate macromers as crosslinking agents and characterization of their degradation products. *Polymer* 2001;42:1251–60.
- [14] Atta AM, El-Ghazawy RAM, Farag RK, Abdel-Aziz AA. Crosslinked reactive macromonomers based on polyisobutylene and octadecyl acrylate copolymers as crude oil sorbers. *React Funct Polym* 2006;66:931–43.
- [15] Se K, Inoue N, Yamashita M. Anionic living polymerization of macromonomers: preparation of a particle-like purging reagent and characterization of poly(macromonomer)s. *Polymer* 2005;46:9753–61.
- [16] Martens P, Anseth KS. Characterization of hydrogels formed from acrylate modified poly(vinyl alcohol) macromers. *Polymer* 2000;41:7715–22.
- [17] David G, Alupeı V, Simionescu BC. End-capping of living poly(2-methyl-2-oxazoline) with maleic acid. *Eur Polym J* 2001;37:1353–8.



- [18] Yildiz S, Hepuzer Y, Yagci Y, Peckan O. Photon transmission technique for studying swelling and drying kinetics of heterogeneous gels formed with various cross-linker contents. *Eur Polym J* 2002;38:1591–9.
- [19] Laukkanen A, Valtola L, Winnik FM, Tenhu H. Thermosensitive graft copolymers of an amphiphilic macromonomer and *N*-vinylcaprolactam: synthesis and solution properties in dilute aqueous solutions below and above the LCST. *Polymer* 2005;46:7055–65.
- [20] Arun A, Reddy BSR. In vitro drug release studies from the polymeric hydrogels based on HEA and HPMA using 4-((E)-[(3Z)-3-(4-(acryloyloxy)benzylidene)-2-hexylidene]methyl} phenyl acrylate as a crosslinker. *Biomaterials* 2005;26:1185–93.
- [21] Elliot JE, Macdonald M, Nie J, Bowman CN. Structure and swelling of poly(acrylic acid) hydrogels: effect of pH, ionic strength, and dilution on the crosslinked polymer structure. *Polymer* 2004;45:1503–10.
- [22] Gomez C, Strumia MC. Synthesis of a triacrylate crosslinking agent and its use in the preparation of macroporous polymers. *Polym Bull* 2001;47:261–8.
- [23] Saito N, Sugawara T, Matsuda T. Synthesis and hydrophilicity of multifunctionally hydroxylated poly(acrylamides). *Macromolecules* 1996;29:313–9.
- [24] Kozulic M, Kozulic B, Mosbach K. Poly-*N*-acryloyl-tris gels as anticonvection media for electrophoresis and isoelectric focusing. *Anal Chem* 1987;163:506–12.
- [25] Wang Q, Dr XX, Dai L. A new quasi-interpenetrating network formed by poly(NAT) and polyvinylpyrrolidone: separation matrix for double-stranded DNA and single-stranded DNA fragments by capillary electrophoresis with UV detection. *Electrophoresis* 2006;27(9):1749–57.
- [26] Aoi K, Okada M. Polymerization of oxazolines. *Prog Polym Sci* 1996;21:151–208.
- [27] Hoogenboom R, Fitjen MWM, Schubert US. Parallel kinetic investigation of 2-oxazoline polymerizations with different initiators as basis for designed copolymer synthesis. *J Polym Sci A: Polym Chem* 2004;42:1830–40.
- [28] Nuyken O, Rueda JC, Voit B. Synthesis of amphiphilic graft copolymers by ring-opening polymerization of 2-methyl-2-oxazoline initiated by poly (isobutene-co-(*m,p*) chloromethylstyrene) macroinitiators. *Macromol Rapid Commun* 1997;18:125–31.
- [29] Rueda JC, Suica R, Komber H, Voit B. Synthesis of new polymethyloxazoline hydrogels by the macroinitiator method. *Macromol Chem Phys* 2003;204:954–63.
- [30] Rueda JC, Komber H, Cedrón JC, Voit B, Shevtsova G. Synthesis of new hydrogels by copolymerization of polymethyloxazoline telechelics and *N*-vinylpyrrolidone. *Macromol Chem Phys* 2003;204:947–60.
- [31] Rueda JC, Zschoche S, Komber H, Schmaljohann D, Voit B. Synthesis and characterization of thermoresponsive graft copolymers of NIPAAm and 2-alkyl-2-oxazolines by the grafting from method. *Macromolecules* 2005;38:7330–6.
- [32] Kobayashi S, Uyama H. Polymerization of cyclic imino ethers: from its discovery to the present state of the art. *J Polym Sci A: Polym Chem* 2002;40:192–209.
- [33] Witte H, Seeliger W. Cyclische Imidsauereester aus Nitrilen und Aminoalkoholen. *Liebigs Ann Chem* 1974:996–1009.
- [34] Gross A, Maier G, Nuyken O. Synthesis and copolymerization of macromonomers based on 2-nonyl- and 2-phenyl-2-oxazolines. *Macromol Chem Phys* 1996;197:2811–26.
- [35] Annunziata R, Franchini J, Ranucci E, Ferruti P. Structural characterization of poly(amidoamine) networks via high-resolution magic angle spinning NMR. *Magn Res Chem* 2007;45:51–8.
- [36] Peppas N, Bures P, Leobandung W, Ichikawa H. Hydrogels in pharmaceutical formulations. *Eur J Pharm Biopharm* 2000;50:27–46.
- [37] Gomez C, Alvarez CI, Strumia MC, Rivas BL, Reyes P. Effects of experimental variables on the synthesis of porous matrices. *J Appl Polym Sci* 2001;79:920–7.
- [38] Masaro L, Zhu XX. Physical models of diffusion for polymer solutions, gels and solids. *Prog Polym Sci* 1999;24:731–75.
- [39] Ferry JD. *Viscoelastic properties of polymers*. 3rd ed. NY: Wiley; 1980.