

SYNTHESIS AND CHARACTERIZATION OF PHEMA HYDROGELS BY INFRARED LASER¹

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Abstract

Poly (2-hydroxyethyl methacrylate) (PHEMA) hydrogels are very versatile in medicine. They are biocompatible and hydrophilic and show similarities to the soft tissues of the body. Chemical agents, ultraviolet radiation and thermal curing can be used as synthesis techniques. However, depending polymerization type, PHEMA hydrogels may have different characteristics. The final properties are related to the crosslinking density present in structure and define the application. 2-hydroxyethyl methacrylate (HEMA) monomer was varied at the 40, 60, 80% w/w and diethylene glycol dimethacrylate (DEGDMA) curing agent at the 1, 2, 3% w/w levels. Taking this into consideration, this paper presents the synthesis of PHEMA hydrogels by infrared laser. The characterization was performed by degree of swelling (EWC), gel fraction, scanning electron microscopy (SEM), FTIR and thermal analysis (DSC). Results demonstrated that it is possible to obtain PHEMA hydrogels by infrared laser through a short process, cheap and uniform heating.

Keywords: PHEMA; Synthesis; Infrared laser; Hydrogels; Biomaterials.

¹ *Technical contribution to 68th ABM International Congress, July, 30th to August 2nd, 2012, Belo Horizonte, MG, Brazil.*

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1 INTRODUCTION

The synthesis and characterization of novel materials with improved and desired properties as well as specific performance for several different applications is an expanding field, which includes subjects ranging material science, chemistry, physics to biology.⁽¹⁻⁴⁾ Polymeric hydrogels are getting more interest among the researchers, due to their excellent properties and similarities with living tissues. They may be natural or artificial, with the ability to swell without dissolving through a three-dimensional network and have shock-absorbing effect variable, depending on the degree of crosslinking. One of the most studied hydrogels in biomedical applications is poly (2-hydroxyethylmethacrylate) (PHEMA),^(1,5-14) due to their properties of biocompatibility, high permeability (also to macromolecules), high hydrophilicity, solubility, chemical stability, and its relatively easiness to be obtained.^(1, 15)

Depending on the chosen synthesis technique (chemical agents, ultraviolet radiation or thermal curing), of initial concentrations of reagents and operating parameters, PHEMA hydrogels with different physicochemical properties, porosity or density can be obtained. They are considered attractive for artificial articular cartilage, contact lenses, scaffolds, systems delivery, vascular grafts, tissue engineering.^(16,17)

The final properties of hydrogels are extremely important to the viability of the biomaterial and reproduction of the conditions in vivo. One of the basic requirements for the polymeric biomaterials to be in contact with body fluids is the absence of toxicity. The radiation technique becomes an interesting alternative. It is an efficient tool for synthesis and sterilization, allowing both processes to be completed in a single step.⁽¹⁸⁾ The technique also allows for easy control of the process conditions. It is possible to modify the physical and chemical properties of the hydrogel through adjustment laser beam intensity as well as the time of exposure of the material to the radiation. This allows simplification of the technology and reduces production costs. In this study, infrared laser radiation source was used. The use of an infrared laser, or rather, the coupling of a fiber laser in the system, has the advantage of being flexible and enables the control of the temperature and the irradiation location. The consequence is the development of material with specific geometries for medical applications.

According to Burns,⁽¹⁹⁾ the infrared energy emitted by the laser is absorbed by the sample as heat and this heat leads to rotational-vibrational motion of the molecules, which induces the polymerization reaction. Energy absorbing agent transfers the energy of infrared beam to the medium, which contains thermal initiator and/or cross-linking agents and are responsible by formation of free radicals. The process of obtainment of three-dimensional polymeric network can be described by three stages: the induction stage, gelation and vitrification. In the first stage, there is a system diffusion of the polymer molecules and cross-linking agent. Then, the gelation occurs, i.e. the polymer is partially reacted. Coexisting unreacted molecules and molecules linked. The system then goes from a viscous liquid to an elastic gel. After gelation, the cross-linking process continues, but more slowly, since the mobility of the molecules becomes restricted due to the increase in crosslinking density. Crosslinking occurs as a phenomenon associated to solid state diffusion, increasing the glass transition temperature and mechanical properties of the material (vitrification).^(2,19-21)

The main advantage of using the laser resides in obtaining faster and localized cross-linking. Once the flow of energy deposited in the sample is controlled, and the cross-linking polymer is in a defined volume, it is possible to obtain specific geometries, with the desired characteristics for use in tissue engineering. This scientific field is one of the most fascinating fields of medical technology,⁽²²⁾ focusing mainly on the development of cartilage and bone structures, and even skin, with anatomical features.⁽²³⁻²⁹⁾ It will require the development of devices with the complexity of the geometric shapes of organs and tissues. Furthermore, the use of lasers is presented as a less expensive technique due to the lower producing costs when compared to conventional photo-curable polymer techniques.

The main objective of this work was to evaluate the feasibility of the infrared laser, with specific wavelength, in the synthesis of PHEMA hydrogels and to obtain controlled physico-chemical properties for different applications, especially for use in Tissue Engineering. Porous and dense PHEMA hydrogels were synthesized with different concentrations of the 2-hydroxy ethyl methacrylate (HEMA) monomer. Diethylene glycol dimethacrylate (DEGDMA) was used as cross-linking agent. The hydrogels were characterized by swelling behavior, gel fraction, morphology through scanning electron microscopy (SEM), glass transition temperatures using differential scanning calorimetry (DSC) and Fourier transform infrared spectroscopy (FTIR). Results showed that it is possible to obtain pHEMA hydrogels with different physico-chemical properties using infrared laser, in short periods of processing. It was varied the concentration of the crosslinking agent and of the monomer. The gel fraction, as predicted, increased with the concentration of reagents, presenting porous and dense structures. The thermal properties of these polymers presented glass transition temperature values around 112°C and the FTIR analysis provided evidence of the presence of bonds and specific functional groups of pHEMA.

2 MATERIALS AND METHODS

2.1 Materials

2-hydroxyethyl methacrylate (HEMA)(monomer), the cross-link agent - diethylene glycol dimethacrylate (DEGDMA) and potassium persulfate (KPS) were supplied by Aldrich. Dibenzoyl peroxide (BPO) was supplied by Peroxid-Chemie. All reagents were of analytical grade and were used without further purification, as received.

2.2 Synthesis of Porous and Dense PHEMA Hydrogels

Porous and dense poly (2-hydroxy ethyl methacrylate) hydrogels were obtained by the radical polymerization of solutions of the 2-hydroxy ethyl methacrylate (HEMA) monomer. The polymerization and cross-linking of PHEMA hydrogels were performed in a single step. The procedure used for the preparation of porous hydrogels and dense hydrogels can be described as follows. Porous PHEMA were synthesized at 40, 60 e 80 wt% HEMA relative to weight of distilled water solution. Diethylene glycol dimethacrylate (DEGDMA) at a concentration of 2wt% and potassium persulfate (KPS) at a concentration of 1wt%, both relative to weight monomer HEMA, were added to the

HEMA/distilled water solution. The mixture was stirred at room temperature for 30 minutes. Then, 2 mL of the mixture was poured into glass vials cylindrical with dimensions of 1.5 cm diameter x 4 cm x 0.1 cm thick. Finally the mixture was cured by infrared laser for 3 ± 0.5 minutes. The wavelength of the source infrared laser and diameter were 1070 nm and 0.8 cm respectively. The laser power was held constant at 30 ± 0.5 W. The distance from the focus of the laser to the center point of the solution was 9.5 cm tall. The synthesis of dense PHEMA hydrogels followed a similar procedure, but without the addition of distilled water. Diethylene glycol dimethacrylate (DEGDMA) was added at concentrations of 1, 2 and 3 wt% relative to weight monomer HEMA. The free radical initiator, in this case, was benzyl peroxide (BPO) at concentration of 1 wt% based on the weight of the monomer. Then, 2 mL of the mixture were poured into petri dishes 50 mm in diameter and cured by infrared laser radiation. Finally, the formation of hydrogel was observed by complete gelation.

2.3 Characterization

After curing, all obtained hydrogels were weighed and submitted to extraction sol-gel with distilled water at a temperature of 85 ± 5 °C for 24 hours and in methanol at 40 ± 5 °C for another 12 hours. The objective was to remove unreacted chemicals and low molecules weight chains. The gel fraction (insoluble fraction) was determined by Equation 1.

$$F_g = m_s / m_{gel} \quad (1)$$

Where:

m_s = initial mass of dry hydrogel (in this case, includes water, unreacted monomer and initiators).

m_{gel} = mass of dry hydrogel after extraction

F_g = gel fraction

The equilibrium water content (EWC) in all hydrogels was determined through gravimetric measurements at a temperature of 37 °C by 24 hours, in thermostatic bath, using Equation (2). The EWC of samples were measured at constant mass by 24 hours.

$$EWC = \frac{W_s - W_d}{W_s} \cdot 100 \% \quad (2)$$

W_d and W_s are, respectively, the mass of dried and swollen hydrogel. The excess water in the swollen state was removed with paper towels. Averaged values for dense or porous hydrogels were calculated from triplicates.

SEM images of the hydrogels were obtained by LEO Electron Microscopy SEM / Oxford, Leo model 440i, with an increase from 30 to 300.000 times. The dried hydrogels samples were fractured in liquid nitrogen, then, fixed in metallic support and coated with gold layer for 120 seconds at Sputer Coater equipment polaron SC762.

Fourier transform infrared (FTIR) transmittance spectra were recorded in the 400-4000 cm^{-1} using a Thermo Scientific Nicolet IR 100 FTIR at resolution 4 cm^{-1} .

DSC measurements were performed in a Mettler Toledo DSC 823e to determine the thermal properties of the PHEMA hydrogels. Each test specimen consisted of 10-15 mg of material accurately weighed in a standard aluminum pan, stuck upside down. All the

hydrogels were dried in an oven for 24 hours at a temperature of 40 ± 5 °C for later thermal analysis. The pans were scanned from room temperature of 25 a 250 °C at a heating rate of 20 °C.min⁻¹ to eliminate the thermal history of the materials, cooled to 25 °C and reheated to 250 °C at 20 °C.minutes⁻¹, under nitrogen atmosphere. Calibration was made with indium standards.

3 RESULTS AND DISCUSSION

The synthesis resulted in the fabrication of opaque and transparent materials with low volume shrinkage during polymerization. Figure 1 shows PHEMA hydrogel at 60 wt % monomer HEMA in the (a) swollen state and (b) dry. The samples with 40 and 80 wt % HEMA also showed similar appearance and good dimensional stability. The increase in the monomer rate allowed a higher reaction rate with formation of the white polymer (hydrogel) and insoluble in water.

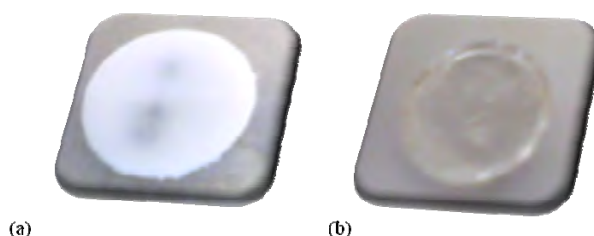


Figure 1. PHEMA hydrogels: (a) Swollen; (b) Dry.

Figure 2 shows a PHEMA hydrogel at 1 wt % DEGDMA. Samples obtained with 1, 2 and 3 wt % of cross-linking agent presented transparency both in the swollen and the dry state. The increase in the rate of DEGDMA allowed a higher reaction but lead to the formation of hydrogels with more rigid and brittle structures.

All hydrogels show to be flexible when hydrated and vitreous when dry, as expected.



Figure 2. PHEMA hydrogels at 1 wt % DEGDMA in swollen state.

Figure 3 shows the gel fraction (F_g) of the PHEMA hydrogels after extraction sol-gel in methanol. As can be seen, a higher content of the monomer provides higher gel fraction. The same happens with the increase of cross-linking agent. The increase of the DEGDMA provides an increase of intramolecular bond strength and shortening of the bonds, decreasing the capacity of moving between the chains, which leads to less elastic hydrogels when swollen.

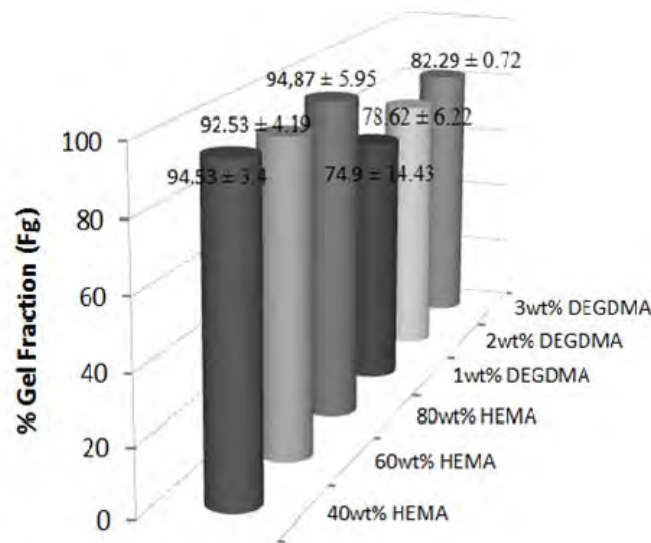


Figure 3. Gel Fraction of PHEMA hydrogels

The equilibrium water content (EWC) of the polymer network also characterizes the crosslink density; more cross-linked hydrogels tend to absorb less fluid. This amount of fluid present in the polymer network is an important feature in the application of hydrogels as substituent of natural articular cartilage. This is because the human cartilage contains large amounts of water present in an extracellular matrix, in fact, around 65-85 %. Two thirds of the water, however, is poorly connected to their three-dimensional network, being exuded after application of loading during movement cycles.^(30,31) Figure 3 displays the equilibrium water content (EWC) of the PHEMA hydrogels. As can be observed, lower concentration of HEMA (40 wt %) and DEGDM (1 wt %) provided structures with higher degrees of swelling. The swelling ratio of hydrogels depends on their free volume, degree of the chain flexibility, cross-link density and hydrophilicity.^(32,33)

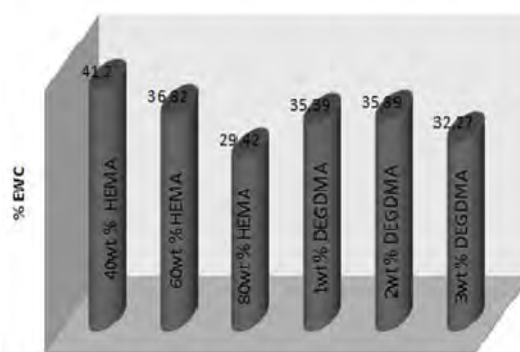


Figure 4. Equilibrium water content of PHEMA hydrogels.

The morphologies of PHEMA hydrogels at different concentration of HEMA are shown in Figure 5. PHEMA hydrogels at a concentration HEMA 40 wt % (Fig. 5a) and 60 wt % (Fig.5b) show a significant amount of pores relative to the PHEMA at 80 wt % (Fig. 5c)

Typically, samples with higher content of HEMA show surface micrographs denser because of the higher degree of cross-linking. The porous diameter was between 32.12 and 53.70 μm at a concentration 60 wt % HEMA, as depicted in Fig. 5d.

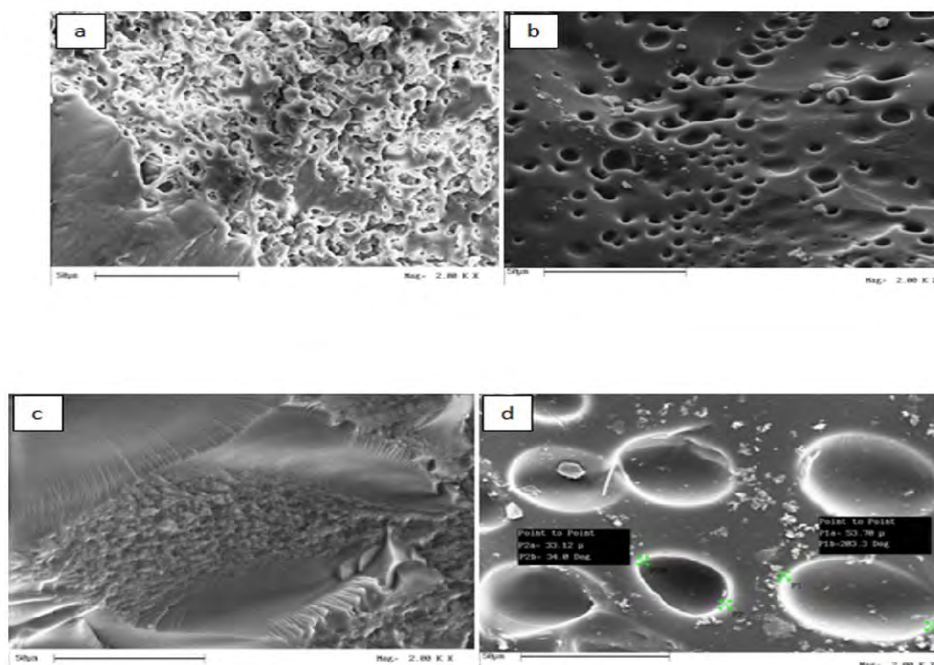


Figure 5. SEM micrographs of fracture of porous PHEMA hydrogels: (a) 40 wt % HEMA; (b) 60 wt % HEMA; (c) 80 wt % HEMA; (d) pores diameter at 60 wt % HEMA.

Figure 6 shows the morphologies of PHEMA hydrogels at different concentrations of DEGDMA. As can be seen in Figures 6 (a), 6 (b) and 6 (c) there are no pores or significant difference between the morphologies, performing smooth and dense as expected.

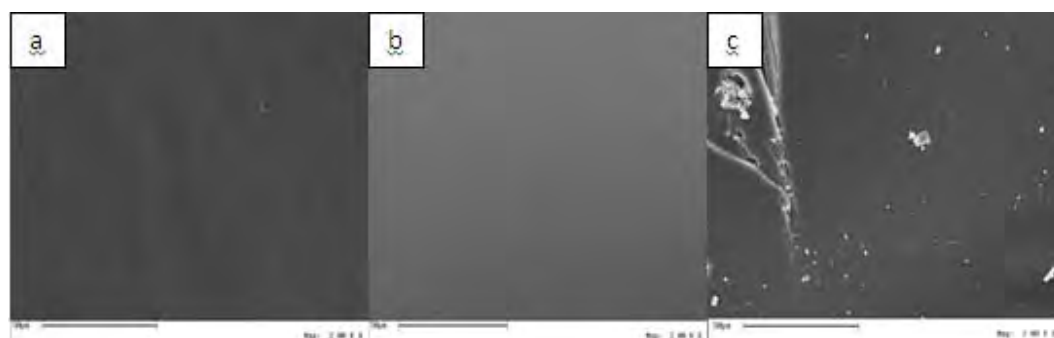


Figure 6. SEM micrographs of fracture of dense PHEMA hydrogels: (a) 1 wt % DEGDMA; (b) 2 wt % DEGDMA; (c) 3 wt % DEGDMA.

FTIR spectra obtained for PHEMA hydrogels relative at a commercial PHEMA hydrogel (Aldrich) are reported in Figure 7. Broad band at $3700\text{-}3000\text{ cm}^{-1}$ can be attributed to O-H stretching, vibrations of side groups; at $3000\text{-}2800\text{ cm}^{-1}$ corresponding to C-H stretching and at $1770\text{-}1640\text{ cm}^{-1}$ attributed to C=O stretching.⁽³⁴⁾

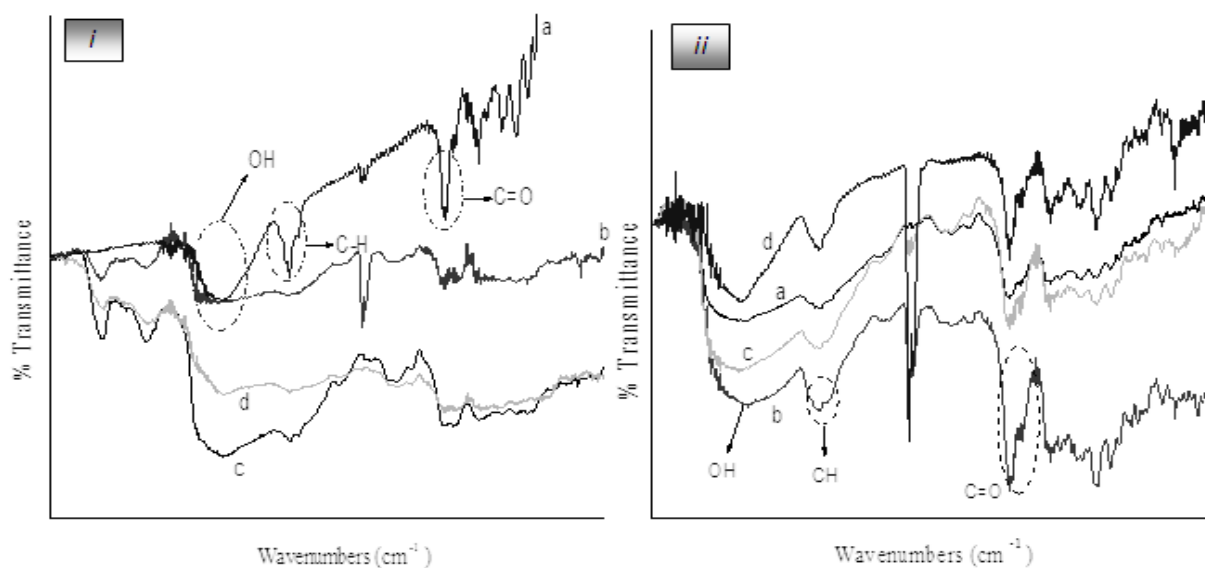


Figure 7. FTIR spectra of: *i*(a) Commercial PHEMA; *i*(b) 40 wt % HEMA; *i*(c) 60 wt % HEMA; *i*(d) 80 wt % HEMA; *ii*(a) commercial PHEMA; *ii*(b) 1 wt % DEGDM; *ii*(c) 2 wt % DEGDM; *ii*(d) 3 wt % DEGDM.

For all the obtained PHEMA hydrogels, the shape of the band in the OH stretch region should be clear in hydrogen bonds due to the hydroxyl groups on the chain terminal PHEMA as OH...OH.⁽³⁴⁾ Similar features in the stretch region due to hydrogen bonds which have been found in a variety of systems, such as water, alcohol, and polymers.⁽³⁴⁻³⁷⁾ Furthermore in all the PHEMA hydrogels spectra, porous and dense, the presence of two overlapping bands in the wavelength range 1712-1641 cm⁻¹ was observed. These bands can be attributed to hydrogen bonding in the structure of PHEMA of the type C=O...HO. These results provide evidence of the presence of molecular inter and intra bonds in the obtained PHEMA hydrogels by infrared laser irradiation.

Table 1 shows the assignments of the PHEMA hydrogels obtained by infrared radiation in comparison to the commercial PHEMA hydrogel.

Table 1. Assignments PHEMA hydrogels by infrared laser

	Commercial PHEMA	Obtained PHEMA hydrogels	Authors ^(32,34,38-42)
OH stretching	3424-3395	3445-3371	3700-3000
CH stretching	2934-2870	2976-2856	2962-2888
C=O stretching	1712-1641	1712-1641	1700-1640

A further characterization of the PHEMA hydrogels obtained by infrared laser was the DSC curve reports (Fig.8). The slope changes in the temperature represents the glass transition temperature (T_g) obtained in the second heating. The T_g was determined using the software tool Glass Transition Mettler-Toledo STARe. The T_g of the hydrogels at different concentrations of HEMA (Figure 8i) was approximately 112 °C. Both materials, porous (Fig.8i) and dense (Fig.8ii), showed no crystallinity (amorphous

hydrogels) and the results in DSC are in agreement with values reported in the literature.⁽⁴³⁻⁴⁶⁾

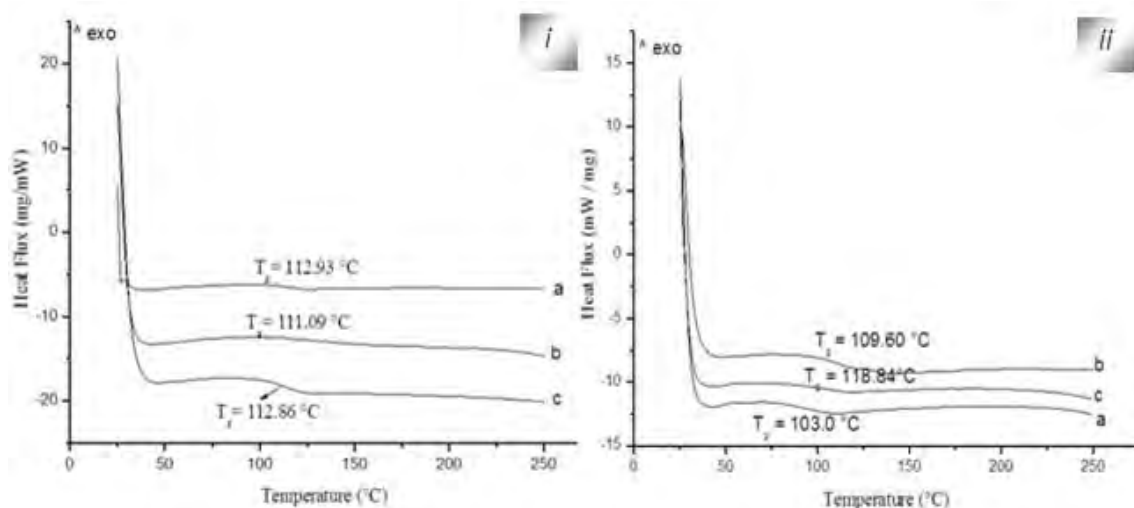


Figure 8. DSC curves of: *i*(a) 40 wt % HEMA; *i*(b) 60wt % HEMA; *i*(c) 80 wt % HEMA; *ii*(a) 1 wt % DEGDMa; *ii*(b) 2 wt % DEGDMa; *ii*(c) 3 wt % DEGDMa.

3 CONCLUSIONS

This paper describes the synthesis of PHEMA hydrogels obtained by infrared laser and the polymers characterization by several methods. It was possible to obtain PHEMA hydrogels with different physico-chemical properties with the modification of HEMA and DEGDMa contents, in a relatively short processing time. The synthesis and sterilization of the hydrogel was done in only one step by crosslinking localized in the diameter of the laser beam.

The effect of the gel fraction on the swelling was investigated by varying the concentration of HEMA and DEGDMa. The data obtained indicated that the swelling ratio decreases with increasing concentration of both, HEMA and DEGDMa. The results may be explained by the fact that increasing the monomer concentration and the curing agent, there is an increase in the density of crosslinking present in the sample. In its turn, this reduces the free volume in the polymer, accessible to water molecules. SEM micrographs show that PHEMA hydrogels with water as diluent present a proportional rate of pore water / HEMA. FTIR results provide evidence of the presence of connections and specific functional groups of the PHEMA by infrared laser. DSC curves showed no significant differences in T_g materials at different HEMA content and the T_g values of all the synthesized hydrogels are consistent with other studies reported in the literature.⁽⁴³⁻⁴⁶⁾ However, the system should be better studied. Mechanical tests and surfaces properties should be further explored. Still the determination of molecular weight of hydrogels should be evaluated by techniques that do not require solubilizing the material, since the materials presented chemical crosslinks, becoming insoluble and infusible.

Acknowledgements

The authors gratefully acknowledge FAPESP for the financial support.

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