

## Design, Synthesis and Characterization of a pH-Sensitive DS/GM Hydrogel

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**Abstract:** Dextran sulfate/glycidyl methacrylate (DS/GM) hydrogels were synthesized by intermolecular side-chain reaction of DS hydroxyl groups with monomeric crosslinking agent, GM in aqueous solutions at 75 °C in the basic medium in this study. The optimum conditions of effective crosslinking for DS/GM system were determined with changing amounts of crosslinker and initiator (benzoyl peroxide, BPO). The swelling abilities of the hydrogels were explained with determining of certain structural parameters for the hydrogel such as equilibrium water content, percentages of gelation, and swelling ratio. pH dependent swelling behaviors of hydrogels were investigated and swelling ability at basic medium was higher than that at acidic medium. Surface morphologies of DS and its hydrogels were examined by scanning electron microscope (SEM) and they compared. Structures and thermal stabilities of DS/GM hydrogels were characterized by infrared (FTIR), thermal gravimetric analysis (TGA), differential thermal analysis (DTA), and Differential Scanning Calorimeter (DSC); effect of GM on crosslinking was investigated. The viscosities of DS/GM reaction products were recorded continuously as a function of time with rotary viscometer and gelation points of binary system reaction between DS and GM were determined via observed viscosity numbers, changes during reaction.

**Keywords:** dextran sulfate, glycidyl methacrylate, hydrogel, swelling, characterization.

### Introduction

In recent years, synthesis of polymeric gels on hydrophilic hydrogels has played an important role in a great variety of applications. Because of their several desirable characteristics such as controllable dimensions; networks absorb and retain large amounts of water, large surface area and momentary response time.<sup>1,2</sup> Hydrogels can change their shape and some properties in response to environmental stimulant (temperature, magnetic field, pH, electricity *e.g.*) Due to the properties of these materials, they are found widespread application in different technological areas, such as food packaging;<sup>3</sup> wound dressing;<sup>4</sup> lenses;<sup>5</sup> controlled protein release;<sup>6</sup> drug release;<sup>7</sup> micro reactors;<sup>8</sup> and dye removal.<sup>9</sup>

Many researchers<sup>10-14</sup> have recently published biopolymers which are good candidate to prepared hydrogels for many applications such as pharmaceutical, food, biomedical, chemical, and delivery systems, because of their biocompatibility, low toxicity, and high content of functional groups.<sup>15,16</sup> Among the known biopolymers, dextran have been proposed for biomedical, biotechnological, and nano-biotechnological fields.<sup>17</sup> In addition, dextran polymer contains a great number of hydroxyl groups which entered conjugation and has high water solu-

bility. Pahimanolis *et al.*, 2010; Maina *et al.*, 2008; Leathers, 2002 have also reported that dextran ideal molecular model for scientific studies because the degree of branching of them can be as low as 0.5% and dextran which has varied molecular weights is commercially findable.<sup>18-20</sup> Dextran sulfate (DS) is glycosaminoglycan which prepared by sulfating a selected fraction of dextran. It is composed of major  $\alpha$ -(1,6) linkages and a minor  $\alpha$ -(1,3) linkage of sulfated glucose. DS is a semisynthetic, hydrophilic, biocompatible and biodegradable polyanionic polymer. The special properties make DS a quite versatile material, with extensive applications such as pharmaceutical, environmental fields, biomedical.<sup>21-23</sup>

The present paper focused on the hydrogel which is composed of anionic dextran sulfate and glycidyl methacrylate. DS/GM hydrogels were characterized using FTIR and SEM techniques. The thermal behaviors of monomer, DS and its hydrogel were studied with by TGA, DSC, and DTA. Furthermore, the pH-dependent swelling of the hydrogel were measured gravimetrically. For the defined gel point, the viscosity of crosslinking reaction of DS and GM was noted nonstop as a function of time.

### Experimental

**Materials.** DS (from *Leuconostoc mesenteroides*) which had

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**Table I. Summary of Gel Compositions and Percentages of Gellation**

	Gellation (%)	DS (g)	GM (mL)	BPO (g)
H1	97	0.11	6	0.04
H2	89	0.11	6	0.02
H3	98	0.11	12	0.04

an average molecular weight ( $M_w$ ) of approximately 500,000 g/mol (supplier information) was supplied by Sigma-Aldrich. GM (crosslinker),  $N,N,N',N'$ -tetramethylethylenediamine (TEMED; accelerator) ammonium persulfate (APS; initiator), and BPO (radical initiator) were supplied by Merck. All reagents were used without further purification.

**Preparation of DS/GM Hydrogels.** DS/GM hydrogels were prepared by radical reaction of aqueous DS/GM solutions (solvent: 0.2 M  $\text{Na}_2\text{HPO}_4$ , pH 8.5) as a function of the DS/GM concentration. with modified for DS of procedure described for dextran by De Smedt *et al.* 1995.<sup>24</sup> 0.2 g of DS powder was dissolved and wait for two days in the buffer solution. Two different solutions (Solution A and Solution B) were prepared by using of DS solution. Solution A was obtained by adding 6 mL of TEMED to 6 mL of a DS solution, while Solution B was a mixture of 5 mL of a DS solution and 10 mL of APS (80 mg/mL). BPO and GM were added to mixture in different molar rates in Table I after Solution A was added to Solution B. The mixture was stirred with magnetic stirrer until complete homogenization at 75 °C. Approximately 7 min after adding Solution A to Solution B, gelation started. The fresh hydrogel was held in water at room temperature for 7 days, in order to remove unreacted monomer, crosslinker, initiator and the accelerator. The water was replaced with fresh water twice a day. DS/GM hydrogel was dried with vacuum at 40 °C, until stable weight was reached. The dried hydrogel was characterized with swelling test, FTIR, TGA, DSC, DTA, and SEM. The compositions of DS/GM hydrogels were referred as H1, H2, and H3.

**Instrumentation.** The FTIR spectra were performed at the frequency range of 4000-400  $\text{cm}^{-1}$  with a Nicolet 6700 FTIR spectrometer. Morphological analysis of DS and DS/GM hydrogel were carried out by SEM JEOL 5500/OXFORD Inca-X. The TGA was studied using Shimadzu TGA 50. The DTA measurements were recorded Shimadzu i70453700237 DSC60. DSC analysis was examined with a METTLER TOLEDO/DSC 1 STARE and system temperature between 0 and 500 °C and under  $\text{N}_2$  atmosphere at a heating rate of 10 °C/min.

**Viscosity Measurements.** The gelation behavior between DS and GM was carried out at 75 °C by using a Fungilab Rotary Cylinder Viscometer (Premium Series) equipped with low viscosity adapter (LCP) supplied with spindle and 180 rpm (rotational spindle speed). The experiments were performed three times.

**Swelling Experiments.** The dry hydrogel ( $w_i$ ) was immersed

in water at 25 °C until it swelled to equilibrium with the purpose of determining the equilibrium water content of DS/GM hydrogel. The swelling equilibrium of the hydrogel sample was reached in approximately 3 h. The swollen sample was instantly weighed (after the external water was wiped off using absorbent paper) ( $w_e$ ). The equilibrium water content of DS/GM hydrogel was calculated from the following equation:

$$\text{Equilibrium water content (\%)} = \frac{w_e - w_i}{w_i} \times 100$$

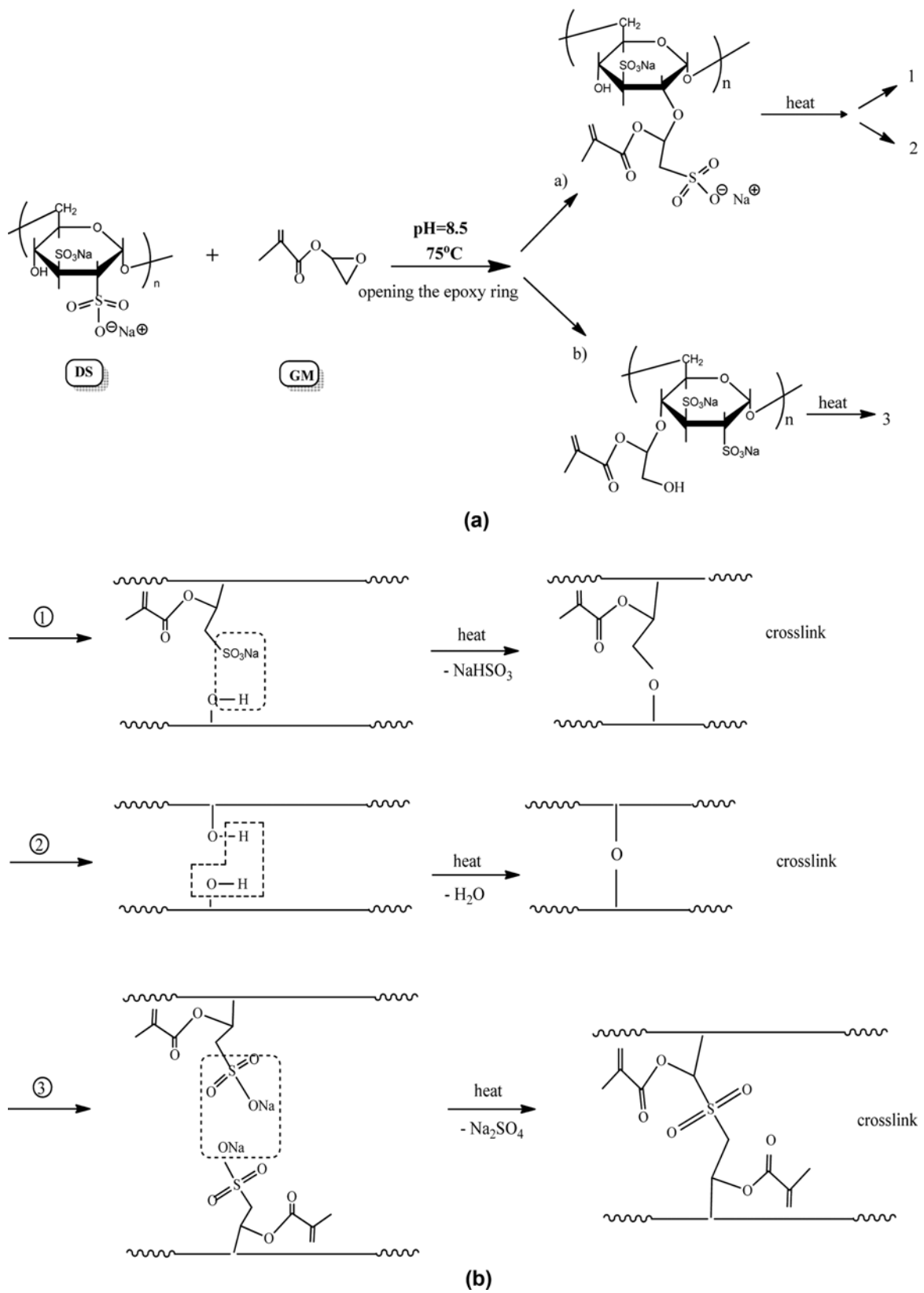
**Swelling of the DS/GM Hydrogel in Different pH Mediums.** The swelling studies on DS/GM hydrogels at different pH values (pH=2.0, pH=7.0, and pH=9.0) were studied. The swelling ratio in different pH medium values were determined by using of mentioned method in Viscosity Measurements Section. The experiments were performed three times. The time-swelling graphs were drawn at different pH values and temperatures for DS/GM hydrogels.

## Results and Discussion

**Synthesis of the DS/GM Hydrogel.** The crosslinking reaction of DS was accomplished by coupling of GM to DS in the presence APS, TEMED and BPO as radical initiators at 75 °C.<sup>25,26</sup> The polysaccharide could react with GM by opening the epoxy ring when using protic solvent, *e.g.* aqueous solution.<sup>27</sup> It thought that crosslinking was easily formed between highly reactive free epoxide groups in GM and OH groups of DS.<sup>28-30</sup>



The proposed mechanism for side-chain reaction between DS and GM was illustrated in Figure 1(a) and (b). Each repeating unit of DS chain contains both  $\text{SO}_3\text{Na}$  and OH groups. And so "a" and "b" structures can form with opening of ring of GM in reaction mechanism were shown in Figure 1(a). It was thought that if "a" was proceed to heat, there are probably form hydrogel chain structures with "1" and "2" numbers. However if "b" was proceed to heat, it will probably form chain structure with "3" number. Probably crosslink kinds were given in Figure 1(b).<sup>24,25,29</sup>

**FTIR Spectroscopy.** The changes in position and intensities of main absorption bands were summarized in Table II. Intensities of bands of GM were considerable decreased with gel formation. FTIR spectroscopy results designate that crosslinking reaction occurs through the side-chain reactions of DS hydroxyl groups with epoxy group of GM. Decreasing intensity of 2932  $\text{cm}^{-1}$  and 815-909  $\text{cm}^{-1}$  (epoxy ring) was showed opening of ring at structure of hydrogel.<sup>31,32</sup> In the FTIR spectroscopy of DS and DS/GM gel showed the characteristics absorption bands, at 1259  $\text{cm}^{-1}$  describing an asymmetrical S=O vibration and at 988-970  $\text{cm}^{-1}$  indicating a symmetrical C-O-S vibration associated to a C-O-SO<sub>3</sub>- group.<sup>33</sup> In addition, the absorption bands at about 1160 represented C-O-C bending vibration. The peaks at 930-970  $\text{cm}^{-1}$  at DS and hydrogel respectively, the characteristic band for C-O-S from the pyranosic ring or



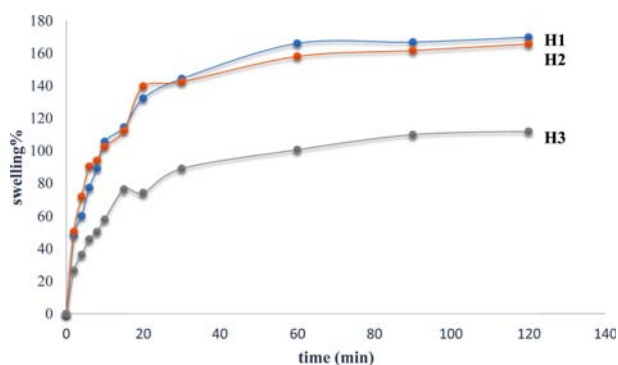
**Figure 1.** (a) The proposed mechanism for crosslinking reaction and (b) schematic representation of the DS hydrogel-crosslinked with GM.

**Table II. The Changes in Position and Intensities of Main Absorption Bands of GM, DS, and DS/GM Hydrogel**

Wavenumber (cm <sup>-1</sup> )						Functional Group
DS	GM	H1	H2	H3		
3470	-	3446	3415	3414	free and H-bonded O-H	
2950	2959	2929	2923	2920	C-H stretching	
-	2932-3002 3062	-	-	-	Epoxy ring 	
-	1720	1722	1725	1719	C=O	
-	1637	1630	1646	1640	C=C	
1462	1455	1462	1474	1474	C-C stretching	
1160	1170	1166	1167	1163	C-O-C stretching	
1022-1247	-	1056-1259	1050-1259	1054-1232	S=O symmetric S=O asymmetric	
930-987	944	971	971	970	C-O stretching	
825	-	845	848	837	asymmetric O-S-O stretching	
-	815	-	-	-	Epoxy ring 	
584	-	550	520	595	symmetric O-S-O stretching	

$\alpha$ -(1-6) linkage was observed.<sup>34</sup> The band of the hydroxyl stretching vibration of the polysaccharide was observed in the region of 3470 cm<sup>-1</sup>.<sup>35</sup> Those bands are proof of the occurrence of crosslinking reaction DS with GM.

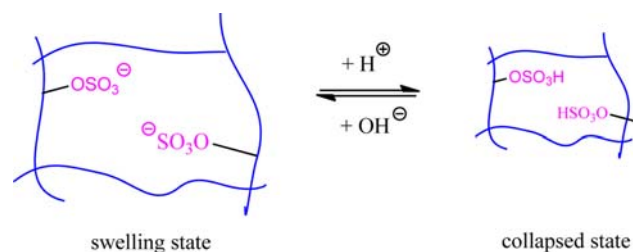
**Swelling Studies.** The physical properties of the DS/GM hydrogels were utilized with measuring the degree of swelling in water. The swelling ratios are 170%, 165%, and 112% for H1, H2, and H3 respectively. Percent equilibrium water content was measured from swelling of the hydrogels are 63%, 62%, and 52% for H1, H2, and H3 respectively. Figure 2 shows swelling curve for H1, H2, and H3 in water. Figure 2 also shows equilibrium swelling was reached approximately 3 h. Equilibrium water content and swelling ratio of H1, H2, and H3 were summarized in Table III. The DS hydrogel with the highest crosslink agent, GM showed the lowest swelling ratio. If the molar percent of crosslinker increase the number of efficient crosslink per unit volume increase therefore

**Figure 2.** Kinetic swelling curves of DS/GM hydrogel in water at 25 °C.**Table III. Equilibrium Water Content and Swelling Ratio of H1, H2, and H3 at 25 °C**

	Equilibrium Water Content (%)	Swelling Ratio (%)
H1	62	170
H2	62	165
H3	52	112

crosslink density and network chain density increase. The ionic strength increases, free volume decrease with the parameter increase. As a result the swelling ratio decreases.<sup>2,36,37</sup>

**Poly-Electrolyte Behavior of the DS/GM Hydrogel in Different pH.** pH-Reversibility of DS/GM hydrogels were investigated at 25 °C as a function of time. Swelling ratios at the different pH values (pH 2.0, pH 7.0 and pH 9.0) of DS/GM hydrogels were given in Table IV. Maximum swelling (187%) was obtained at pH 9.0 for H1. As it is seen in Figure 3, at a low pH mediums (pH=2.0), -SO<sub>3</sub>O<sup>-</sup> groups are non-ionized; therefore, the network is in a collapsed state. At higher pH

**Figure 3.** The main mechanism of swelling DS/GM hydrogel in different pH mediums.

**Table IV. Swelling Ratios in Different pH Mediums (pH 2.0, pH 7.0, and pH 9.0) at 25 °C**

	Swelling Ratio (%)		
	H1	H2	H3
pH 2.0	152	162	103
pH 7.0	170	165	112
pH 9.0	187	216	127

(pH=7.0), some  $-\text{SO}_3\text{O}^-$  groups are ionized and the electrostatic interaction between groups causes an enhancement of the swelling capacity. At high pH values (pH=9.0), the  $-\text{SO}_3\text{O}^-$  groups repel each other, causing the swelling of the system due to anion-anion repulsive electrostatic forces.<sup>38-40</sup> Water uptake at higher pH depends upon the available free volume of the expanded polymer matrix, polymer chain relaxation.<sup>41</sup> That resulted in a more hydrophilic polymer network and contributed to the higher water absorption.<sup>35</sup> Water-uptake values are presented in Table IV. The same trend is observed in H2, and H3.

**Viscosity Measurement.** The reaction mixture was determined with dilute solution viscometer as a function of time to determine the gel point during the crosslinking reaction between DS and GM, for H1, H2, and H3 at 75 °C. As seen as the Figure 4, during the crosslinking reaction progresses increased the molecular weight of the polymers and the viscosity by time, but after a certain period, it was monitored stable viscosity number.<sup>42,43</sup> The maximum viscosity are 80.71  $t=32$  min for H1; 79.96  $t=23$  min for H2; 24.2  $t=9$  min for H3. It was observed partially gelatinized towards gel point at the crosslink reaction between DS and GM. The sudden increase in the viscosity which is gelation point was shown completed of gelation reaction.

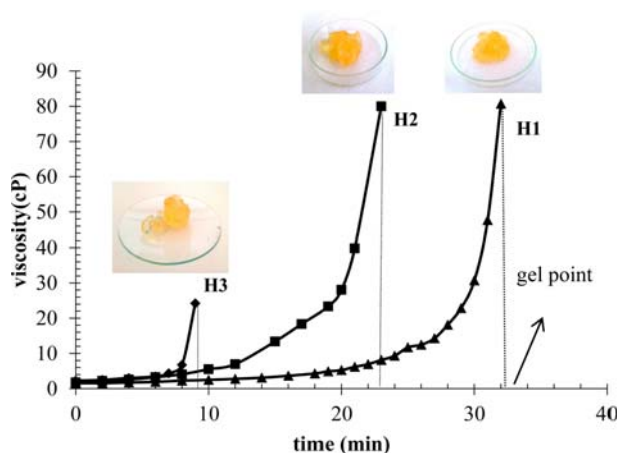
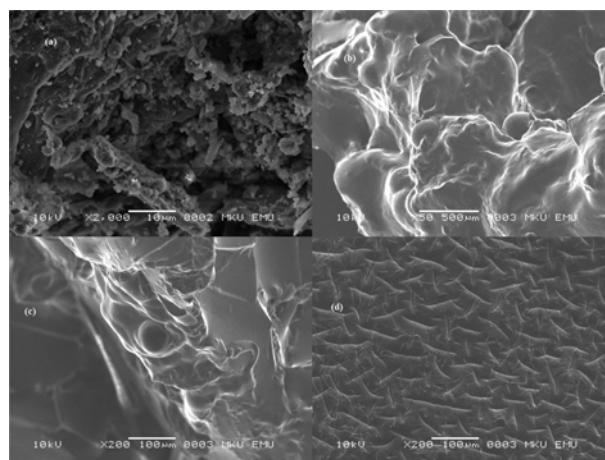
**Thermal Stability.** The thermogravimetric analysis (TGA) was carried out dynamically to pursue the thermal stability and crosslinking of DS. The mass loss occurring of 100 °C is due to the loss of water molecules from the DS and DS-GM hydrogels. It was observed that beyond 227 °C, the DS con-

tinues to absorb heat and decomposes finally at 730 °C. In the first stage the mass loss of about 33% occurs at 227 °C, and the second stage the mass loss of about 87% occurs at 730 °C. The thermal degradation of hydrogels essentially takes place in three stages. The decomposition of hydrogels started 210 °C and completed at nearly 411 °C, 218 °C and completed at nearly 406 °C, 214 °C and completed at nearly 418 °C respectively H1, H2, H3. Weight loss is approximately 87%, 75%, 75%, and 82% for DS, H1, H2, and H3 respectively. The maximum rate degradation temperature ( $T_{\text{max}}$ , first derivative peak temperature) is 730, 411, 406, and 418 °C for DS, H1, H2, and H3 respectively. According to TGA, the DS/GM hydrogels were appeared to be thermally stable up to 400 °C. It is emphasized that the thermal stability was not improved after crosslinking of the methacrylate substitution of hydroxyl groups on DS.<sup>44,45</sup>

DSC analysis and DTA were used to determine glass transition temperature ( $T_g$ ), melting temperature ( $T_m$ ). DS and hydrogel samples have shown a broad endothermic peak at 100 °C corresponding to their dehydration. The data ( $T_g$  and  $T_m$ ) of DS and DS/GM hydrogel determined from DSC were summarized in Table V. As it is seen from the Table V, DS showed peaks due to  $T_g$  (64 °C),  $T_m$  (226 °C).<sup>46</sup> In other words DS indicates crystalline in nature. The  $T_g$  peaks for H1, 50 °C; for H2 42 °C; for H3 56 °C. The  $T_m$  peaks for H1, 300 °C; for H2 288 °C; for H3 171 °C. A decrease in the  $T_g$  values compared to that of DS (64 °C) was observed for all of hydrogel samples suggesting that the crosslink with GM

**Table V. The Data ( $T_g$  and  $T_m$ ) of DS and DS/GM Hydrogel Determined from DSC**

	$T_g$ (°C)	$T_m$ (°C)
DS	64	226
H1	42	300
H2	42	288
H3	56	171

**Figure 4.** Gel points of gellation reaction between DS and GM.**Figure 5.** SEM images of DS (a), H1 (b), H2 (c), and H3 (d).

played an effective role as an internal plasticizer for DS.<sup>44</sup>

**Morphology of DS and Its Hydrogels.** The microphotographs of DS and hydrogel samples were examined by SEM. Figure 5 illustrates the surface morphology of the DS and its hydrogels. As it is seen from the SEM image of DS, DS is relatively smooth surface. The pore structure simplifies the diffusion of the water molecules in and out through hydrogel.<sup>47</sup> The presence of a porous morphology in the DS hydrogels based on GM has been also showed from Figure 5. As shown in these figures, while H1 and H2 exhibit porous and smooth matric, H3 has porous honeycomb-like structure.<sup>35</sup>

## Conclusions

In conclusion, a novel pH-sensitive DS/GM hydrogel was successfully synthesized by the crosslinking polymerization reaction method, and it was characterized by FTIR, DSC, TGA, DTA, and SEM. The swelling ratio of the DS/GM hydrogel was increased with the pH value. It was clearly suggested that H1 and H2 exhibit porous and smooth matric and H3 has porous honeycomb-like structure from SEM micrographs. The results of swelling measurement showed that the hydrophilic DS/GM polymer network contributed to the high water absorption.

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