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# Preparation and NMR characterization of superporous hydrogels (SPH) and SPH composites

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## Abstract

In this investigation the mechanism of synthesis and characterization of Superporous Hydrogel (SPH) and SPH composites were studied. Solid-state $^{13}$C NMR was used for elucidating the structure of the polymers. Scanning electron microscopy (SEM), apparent density and swelling ratio studies were used to characterize these polymers. The results showed the SPH polymer was made of the sequence of acrylamide and acrylic acid monomers as a backbone chain, which was cross-linked with *N*,*N*′-methylenebis acrylamide. Sodium bicarbonate added in the last step of the synthesis was used to generate $CO_2$ formation, causing a large number of interconnecting pores within the polymer structure. SPH composite polymers were made in the same way, except for changing the amount of monomers and using Ac-Di-Sol as a stabilizer. The cellulosic fibres of Ac-Di-Sol will overlap the backbone chain, so the mechanical stability of these polymers will be increased. Characterization of the prepared polymers by SEM, measurement of the apparent density together with swelling ratio studies showed that SPH polymers have more pores and higher swelling ratio but less mechanical stability compared to SPH composite polymers, which have less pores and lower swelling ratio but a higher mechanical stability.

*Keywords*: Superporous hydrogel (SPH); Superporous hydrogel composites; Solid-state NMR

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## 1. Introduction

Hydrogels are cross-linked hydrophilic polymers with a network structure consisting of acidic, basic, or neutral monomers which are able to imbibe large amounts of water. Because of their network structures and the possibility of rearrangements of hydrophobic/hydrophilic domains during the swelling process, including entanglements and crystalline regions, these polymers are water insoluble [1–3]. A variety of hydrogels has been employed for different biomedical applications. Due to their biocompatibility and biodegradability, they are used among other possibilities, as bio-absorbable materials in surgeries and biotechnology, and also in other medical, agricultural and pharmaceutical applications for delivery of medicines. For the latter application they are unique carriers for controlled drug delivery. Release control can be governed by both swelling and biodegrading properties [4–6].

Basically, hydrogels are divided into two major groups with respect to swelling: conventional hydrogels and new generation of hydrogels. The most important difference between these two groups is their swelling characteristics. The swelling properties of hydrogels are mainly related to the elasticity of the network, the presence of hydrophilic functional groups (such as -OH, -COOH, -$CONH_2$, -$SO_3H$) in the polymer chains, the extent of cross-linking, and porosity of the polymer. Additionally, the physical characteristics of hydrogels including their swelling ratio also depend on the balance between attractive and repulsive ionic interactions and solvent mediated effects [7,8]. Because of their rigid crystalline structure and low elasticity in the polymer chains, conventional hydrogels swell very slowly and correspondingly the time for absorbing water is long, ranging from a few hours to even days. Although such slow swelling is beneficial for many applications, there are many situations where a fast swelling of the polymer is more desirable. Therefore, a new generation of hydrogels, which swell and absorb water very rapidly, has been developed. Examples of this new generation are Superporous Hydrogels (SPH) and SPH composites, which swell to equilibrium size in a short period of time [9]. The fast swelling of these polymers cannot be related only to the above-mentioned

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factors, but also to capillary wetting of interconnected open pores. In the case the swelling ratio of this category of polymers exceeds 100, it is obvious that during the synthesis of these polymers a large internal surface by a large number of interconnected pores is formed; therefore, water can be rapidly absorbed by capillary attraction forces within the pores, and these polymers swell to their maximum volume very quickly [9–13].

In this study, SPH and SPH composites were synthesized in order to make these polymers appropriate for intestinal delivery of hydrophilic macromolecular drugs. For this purpose, the structure of the synthesized polymers was investigated by solid-state NMR to evaluate which free functional groups exist in the structure of this category of polymers in order to be able to predict their swelling properties and drug release characteristics in the gut.

## 2. Experimental

### *2.1. Materials*

Acrylic acid, containing 0.02% hydroquinone monomethyl ether as a scavenger, was purchased from Aldrich Chemie (Zwijndrecht, The Netherlands). Since the difference between the boiling point of acrylic acid (139°C) and hydroquinone monomethyl ether (243°C) is high enough, pure acrylic acid was obtained by distillation in vacuo at 50°C and used for synthesis purposes. Pluronic F127 (Poloxamer) and sodium bicarbonate were obtained from Sigma Chemie (Zwijndrecht, The Netherlands) and Ac-Di-Sol (cross-linked sodium carboxymethylcellulose) was obtained from FMC Corp. (Philadelphia, PA, USA). The water (MQ) used was filtered by a Milli-Q UF plus ultrapure water system from Millipore (Etten-Leur, The Netherlands). All other compounds used were from Aldrich Chemie (Zwijndrecht, The Netherlands) and used as obtained.

### *2.2. Synthesis of SPH and SPH composites*

The method reported by Chen et al. [9] was used as a basis to prepare SPH and SPH composites, and adapted as described below. All ingredients except for sodium bicarbonate and Ac-Di-Sol were used as solution in MQ water in concentrations as percent (W/V) as mentioned below. The pH of the monomer solutions has to be higher than 5, and in case of initial acidic solutions the pH was adjusted to 5.5 with 10 M NaOH. When the pH is lower than 5, $CO_2$ formation takes place before the start of polymerization and no pores are formed inside the synthesized polymers.

For the synthesis of SPH the following substances were added subsequently into a test tube at ambient temperature: 300 μl acrylamide (AM) 50%; 200 μl acrylic acid (AA) 50%; 70 μl *N*,*N*′-methylenebis acrylamide (Bis) 2.5%; 300 μl MQ water; 30 μl Pluronic F127 10%; 25 μl ammonium persulphate (APS) 20%; 25 μl *N*,*N*,*N*′,*N*′-tetramethylethylenediamine (TMEDA) 20%; and 100 mg of sodium bicarbonate. In this procedure the amount of Bis as a cross-linker and sodium bicarbonate as a foaming agent were reduced from 100 μl as given by Chen et al. [9] to 70 μl for Bis and from 120 to 100 mg for sodium bicarbonate. These reductions remarkably influence the swelling ratio of the SPH polymer. In addition, the amounts of APS as an initiator and TMEDA as a catalyst were increased from 20 to 25 μl for both compounds, which facilitates the initiation of the reaction. After adding each substance to the test tube, the reaction mixture was vigorously shaken. Finally, sodium bicarbonate was added very quickly to the solution and mixed with a spatula. If sodium bicarbonate was not added quickly enough, the polymerization had already been started by APS as initiator. Under this condition, some clamps are formed additionally and the prepared SPH polymer is not homogenous.

Two different SPH composites have been synthesized: The first was the same as for SPH; however, 150 mg of Ac-Di-Sol was added to the mixture after adding APS and before adding TMEDA. For the synthesis of the second SPH composite the following substances were added one by one to a test tube and after adding of each ingredient the tube was vigorously shaken: 1.2 ml AM 50%; 0.9 ml potassium salt of 3-sulfopropyl acrylate (SPAK) 50%; 0.3 ml Bis 2.5%; 90 μl Pluronic F127 10%; 30 μl AA 50%; 45 μl APS 20%; 120 mg of crosscarmellose sodium (Ac-Di-Sol); 45 μl *N*,*N*,*N*′,*N*′-tetramethylethylenediamine 20%; 120 mg of sodium bicarbonate. As modifications, the amounts of Bis were reduced from 0.45 to 0.3 ml and Ac-Di-Sol from 270 to 120 mg. On the other hand, the amount of sodium bicarbonate was increased from 100 to 120 mg. These changes appeared to influence both the mechanical stability and the swelling properties of the SPH composites. It should be noted that also for the optimal synthesis of SPH composites sodium bicarbonate must be added very quickly in order to obtain a homogenous polymer and to prevent formation of clamps. The reason for using SPAK as an alternative to AA monomer for the synthesis of the second SPH composite, was because the mixture of SPAK/AM in comparison to AA/AM monomers allowed for a somewhat higher variability in synthesis procedure and the synthesis was practically more applicable. Therefore, using SPAK/AM as a monomer mixture facilitates the preparation of more homogenous polymers.

In both procedures (SPH and SPH composites), polymerization was allowed to continue for approximately 10 min. Then the polymers were taken from the reaction tubes and dialysed with a dialysis membrane (molecular weight cut off 6000 Da) against deionized water for at least 5 days to remove possible residual monomers.

### *2.3. Effect of drying process on the swelling ratio of the polymers*

The synthesized polymers were subjected to various drying procedures in order to study the effect of the drying

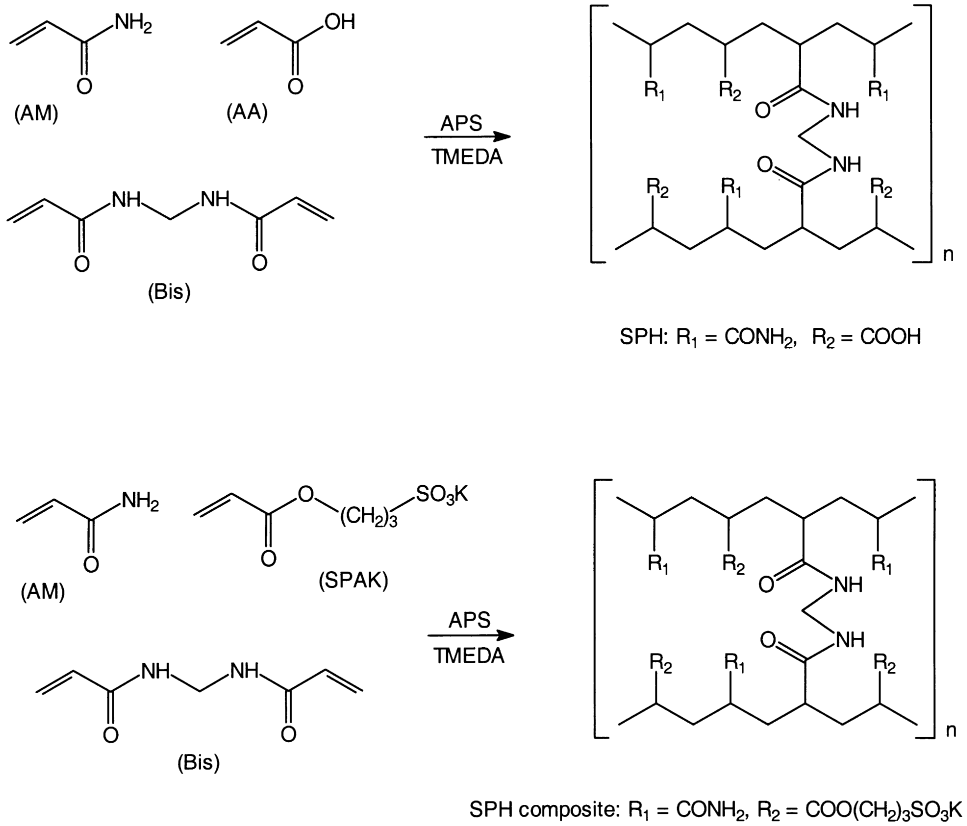

Fig. 1. Synthesis of SPH and SPH composites (schematic view): Acrylic acid (AA); Acrylamide (AM); *N*,*N*′-methylenebis acrylamide (Bis); Ammonium persulphate (APS); *N*,*N*,*N*′,*N*′-tetramethylethylenediamine (TMEDA); Potassium salt of 3-sulfopropyl acrylate (SPAK); Superporous Hydrogel (SPH).

process on the swelling ratio of the polymers. Two methods of drying were used for SPH: (1) freeze-drying, and (2) using different organic solvents to dehydrate the polymers quickly. For further drying the polymers were placed in an oven at 60°C for 1 day. The SPH composites were dried either at ambient temperature or under reduced pressure in an oven at 60°C.

In the case of freeze-drying, the polymers were shock-frozen in liquid nitrogen for 10 min and then put in the freeze-dryer (Christ Alpha1-2, Osterode, Germany) for 1–2 days until drying was completed. In the second drying method, SPH was submerged three times in one of the following organic solvents for 10 min: absolute ethanol, acetone and diethyl ether. Thereafter, the polymers were put in an oven at 60°C for 1 day in order to be sure that the polymers have been dried completely.

The dried polymers were used to determine their swelling ratio in milli-Q water. For calculation of the swelling ratio, the following equation is used:

$$Q = \frac{M_s - M_d}{M_d} \qquad (1)$$

where $Q$ is the swelling ratio, $M_s$ the mass in the swollen state and $M_d$ the mass in the dried state. At the beginning of each experiment, $M_d$ of a piece of polymer was measured by weight and then it was immersed in an excess of deionized water for swelling. At specific time intervals the polymer was removed from the water and weighed in order to measure $M_s$ [9]. Since the swollen polymers appeared to be fragile, they were put on a grid boat with a mesh size of 1 mm. This technique allows to put the polymer in water and to weigh it without breaking. Each time the grid boat with the polymer was removed from water, it was gently dried by paper tissue in order to remove adhering water.

### 2.4. Density measurements of the polymers

Dried polymers were used for density measurements, which actually show the apparent densities of the polymers. Pieces of polymers were taken and weighed in order to determine the mass of each piece. For measuring their volumes, the solvent replacement method was used. By this method the solvent penetrates into the pores of the polymer; therefore, the volume can be related to the porosity of the polymer, which results in the apparent density instead of actual density of the polymer. For this purpose, a hydrophobic solvent such as hexane that is not absorbed by the polymers was used. By the use of forceps, a piece of the polymer was immersed in a predetermined volume of hexane in a graduated cylinder, and the increase in the hexane volume was measured as the volume of the polymer.

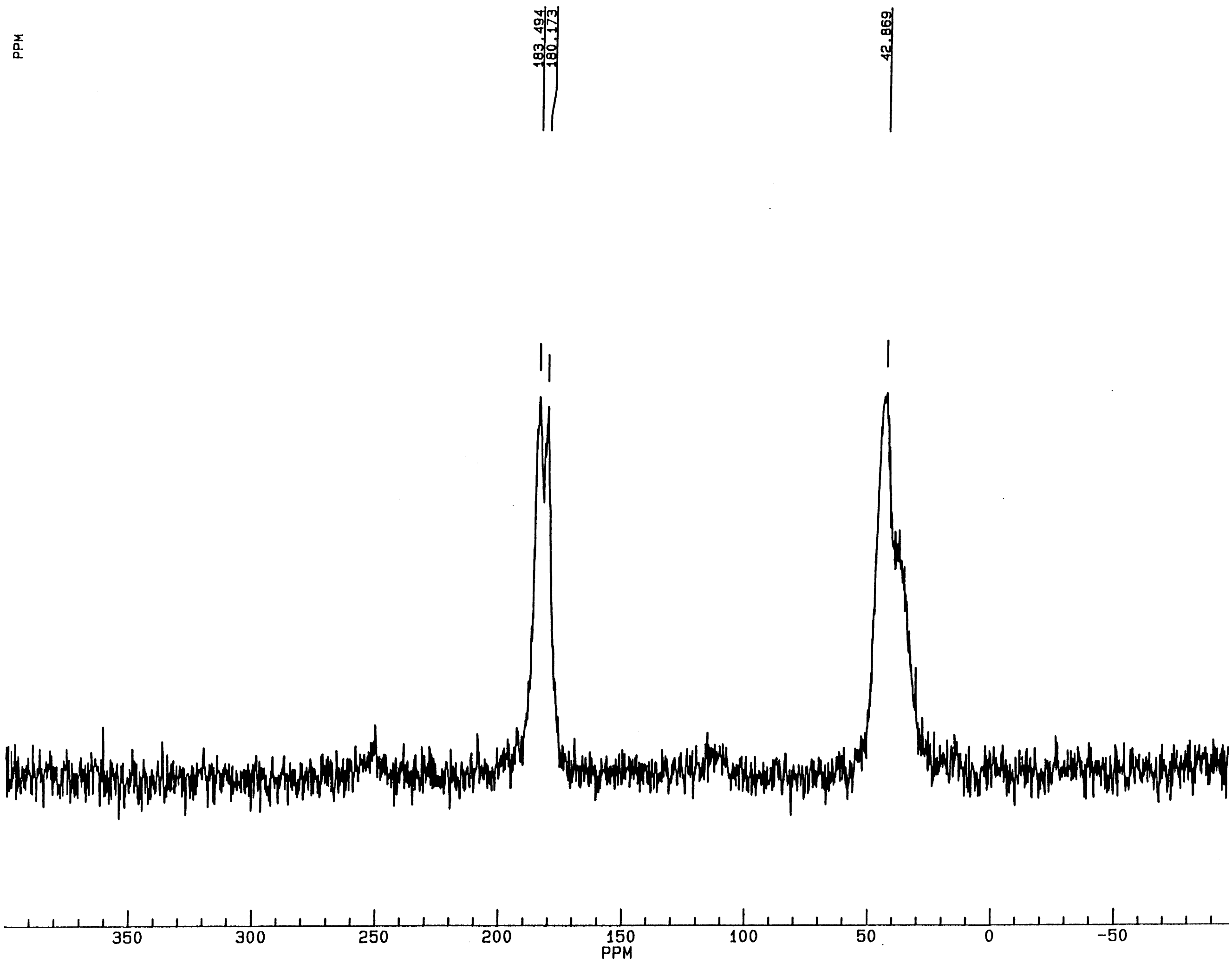

Fig. 2. $^{13}$C-NMR spectrum of SPH, as obtained by solid-state NMR.

### 2.5. NMR measurements

$^{13}$C solid-state CP/MAS NMR was used for the characterization of the structure of the water-insoluble SPH and SPH composite polymers. The measurements were carried out with a BRUKER MSL 400 MHz spectrometer. A 4-mm MAS probe was used with a proton channel and a broadband channel tuned to 100 MHz, the resonance frequency of $^{13}$C. A typical value of the 90° $^{1}$H pulse was 5 μs. The contact time was 2 ms and a relaxation delay of 4 s was used. The spinning speed of the sample, 7 kHz, was regulated with a home-built spinning speed controller. The spectra were calibrated using $^{13}$C labelled glycine with a chemical shift value of 176.04 ppm at the carbonyl position. The same sample was used to find the Hartmann–Hahn matching condition. All measurements were done at room temperature.

### 2.6. Scanning electron microscopy

The dried polymers were used for scanning electron microscopy (SEM) studies. In order to keep the pores of the polymers intact for imaging, the dried polymers were placed in liquid nitrogen and then broken. The inner surfaces of the polymers were sputter-coated with gold for 3 min, and thereafter the pictures were obtained using a Philips SEM 525 electron microscope (Eindhoven, The Netherlands).

## 3. Results and discussion

### 3.1. Synthesis and structure of SPH and SPH composites

In order to study the structure of SPH and SPH composites, the role of each component in the synthesis of these polymers will be discussed first. In the synthesis procedure of SPH (Fig. 1), AA and AM are the monomers. Bis is used as a cross-linker, and Pluronic®F127 is used as a foam stabilizer of the foam which is formed by carbon dioxide originating from sodium bicarbonate. Pluronic®F127 does

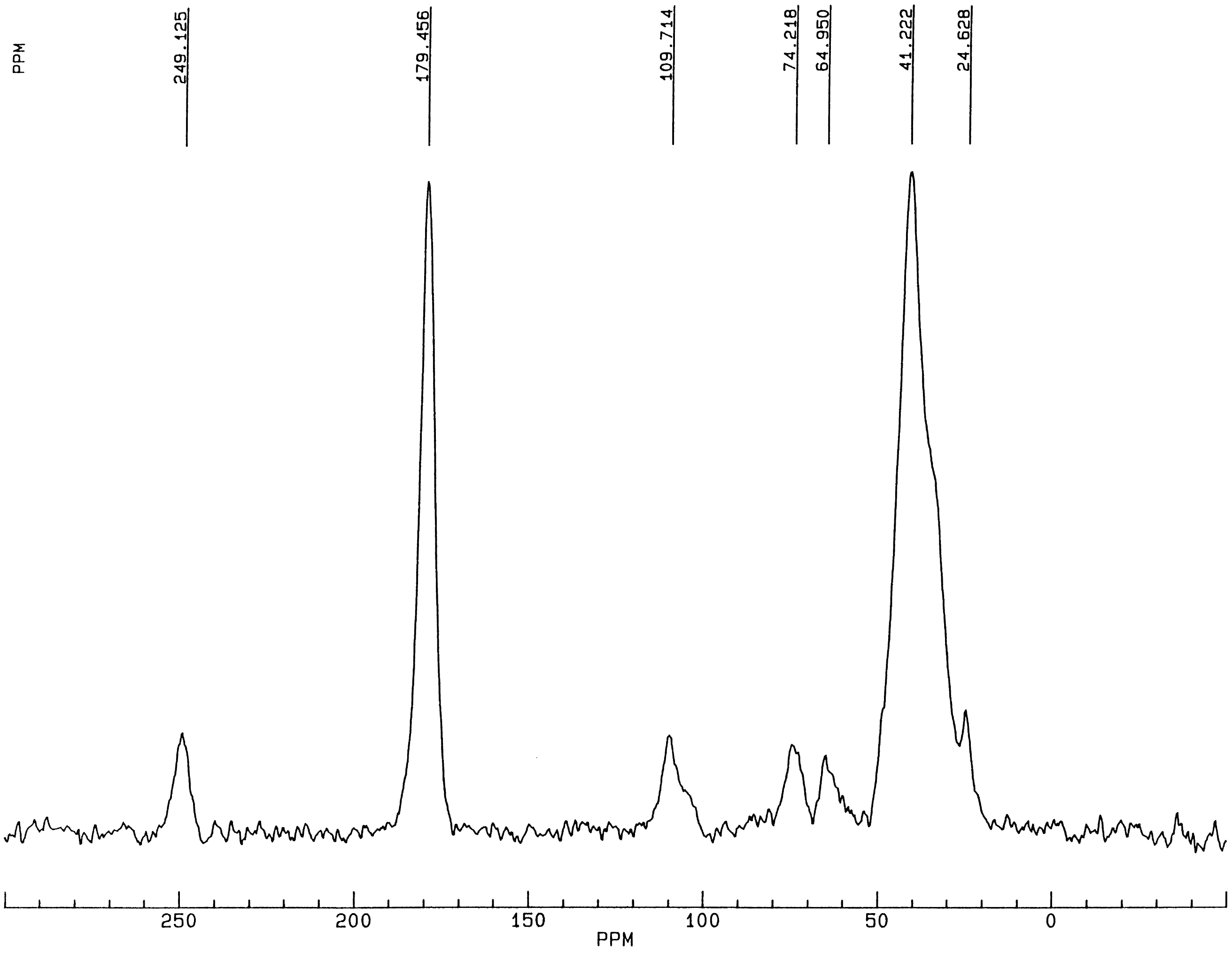

Fig. 3. $^{13}$C-NMR spectrum of SPH composites, as obtained by solid-state NMR.

not contribute to the chemical structure of the polymer, but is very important as a surface active agent to create the highly porous polymer structure. After the foam formation has started the foam should be stable for a few minutes in order to introduce the desired large number of pores during the synthesis of the SPH polymer. APS is used as a polymerization initiator and TMEDA as a catalyst.

In the synthesis of SPH composites all above-mentioned compounds have the same role. However, in one of the SPH composites the potassium salt of 3-sulfopropyl acrylate is used as a monomer instead of AA and Ac-Di-Sol (a stabilizer) is used for introducing additional mechanical stability of the polymer by physical entanglement of the polymer chains with Ac-Di-Sol fibres. Ac-Di-Sol does not contribute to the chemical structure of the polymer, but is applied to enhance mechanical stability of the polymer.

In Figs. 2 and 3 the solid-state $^{13}$C NMR spectra of SPH and SPH composites are depicted. In order to elucidate the spectra and define each peak, these spectra were compared with previously published reference spectra [14]. When the spectrum of the SPH polymer prepared from AA and AM (Fig. 2) is compared with the reference spectra of the polymers of AA and AM [14], it becomes clear that the large peak with a shoulder at 42.869 ppm represents the main carbon backbone. The two split peaks at 180.173 and 183.494 ppm depict the carbon atoms of the amide and carboxylic groups, respectively. These last two peaks overlap with each other, because the range of the appearing carbon peak for both monomers (in polyacrylic acid and polyacrylamide) is approximately the same [14]. In Fig. 3, which shows the spectrum of the SPH composite, the large peak with a shoulder at 41.222 ppm represents the carbon chain of the polymer. Since the SPH composite does not contain AA as a monomer, the single peak at 179.456 ppm is representative for the carbon of the amide group. The two parallel peaks on both sides of the amide group (peaks at 109.714 and 249.125 ppm) are called satellite peaks, which represent the spinning side band that generally appears in solid-state NMR. These peaks can be distinguished easily, because the distance of these satellite peaks from the main peak is approximately the same (70 ppm; see Fig. 3). Since for the synthesis of SPH

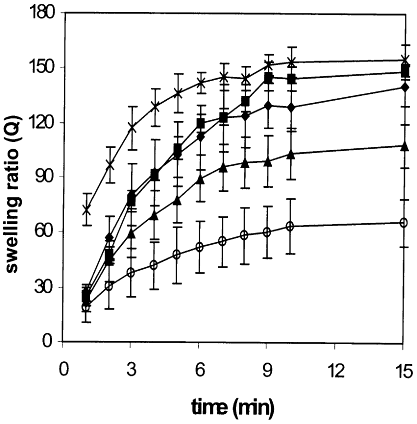

Fig. 4. Swelling ratio curves for SPH polymers dried with various methods: (◆) drying with absolute ethanol; (■) drying with acetone; (▲) drying with diethyl ether; (×) freeze-drying for 2 days; (○) freeze-drying for 1 day. Data are depicted as the mean ± SD of three experiments.

composites the potassium salt of 3-sulfopropyl acrylate instead of AA was used as monomer, the appearance of three other peaks related to the carbon atoms of SPAK is also evident in Fig. 3. When the carbon atom is attached to a more electronegative group, it shifts to the left (higher ppm value). Consequently, the peak at 74.218 ppm is related to the carbon attached to $-SO_3K$ of SPAK which is the most electronegative group, and the peak at 64.950 ppm represents the carbon atom attached to the oxygen of SPAK which is less electronegative. The peak at 24.628 ppm relates to the middle carbon (attached to the other two carbons) of SPAK which has the least electronegativity, and this peak overlaps with the peak related to the main carbon backbone. Because the number of carbon atoms in the side chain is much less than in the main chain, this peak is much smaller.

According to the obtained NMR spectra, the following mechanisms of reaction are proposed for the synthesis of SPH and SPH composites (Fig. 1).

The combination of APS/TMEDA will initiate the radical polymerization. The radicals formed will then attack the double bonds of AA and AM, and also to a less extent to the double bond of Bis. Subsequently, the double bonds will be opened and the monomers will covalently bind to each other and form a long aliphatic chain. These chains are subsequently cross-linked by the added cross-linker. The frequency of cross-linking between the polymer backbone chains is related to the concentration of the cross-linker used during polymerization. The amount of cross-linker also influences the swelling ratio of the polymer, because with increasing amounts of cross-linker the two polymer chains will attach to each other more strongly and the size of pores during the foam formation will be smaller. On the other hand, when the amount of cross-linker is high, the flexibility of the chains will be less, resulting in a reduced swelling capacity of the polymer.

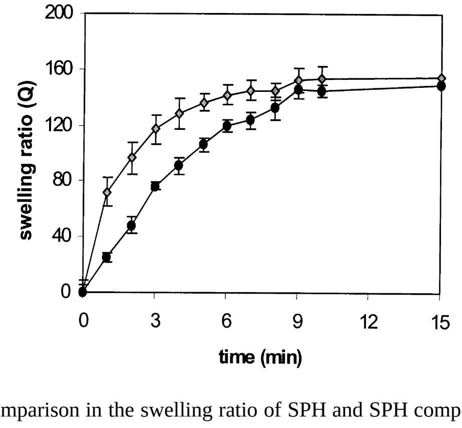

Fig. 5. Comparison in the swelling ratio of SPH and SPH composites: (◇) SPH freeze-dried for 2 days; (●) SPH composite. Data are depicted as the mean ± SD of three experiments.

For the synthesis of the SPH composites the procedure was the same as for SPH. Only SPAK instead of AA was used as a monomer. Therefore, in the proposed mechanism the sequence of AM and SPAK monomers (instead of the AM and AA monomers) is indicated with the same cross-linker (Fig. 1).

### 3.2. Swelling ratio studies

The SPH polymers dried by different procedures were used for the swelling ratio studies. As shown in Fig. 4, the polymers dried by freeze-drying for 1 day, have the lowest swelling ratio, indicating that these polymers were not dried completely. However, when the polymers were freeze-dried for 2 days, the swelling ratio was much higher than observed for all the other methods of drying (150 times after approximately 9 min; Fig. 4). Apparently, after 2 days of freeze-drying, these polymers possess a minimum amount of water that can absorb very quickly a large amount of water and thus the polymers swell very rapidly. When the polymers were dried with organic solvents and subsequently dried at 60°C, the swelling ratios were found to be between those after 1 and 2 days freeze-drying. There was no difference between drying with acetone and absolute ethanol. With diethyl ether, however, the swelling ratio was less, probably due to its fast evaporation and reduced water miscibility that resulted in closing of some of the pores in the polymer.

The reason for the higher swelling ratio of SPH after 2 days of freeze-drying in comparison with polymer dried with organic solvents is probably due to the fact that no changes in the porous structure of the polymers occur during the freeze-drying process. On the contrary, when polymers are placed in organic solvents, the water will be deprived from the polymers very quickly and some of the pores of the polymer may be closed. Capillary related swelling of these polymers is directly related to capillary wetting, and hence dependent on the amount of capillaries.

There was no difference between the two drying methods of SPH composites on their swelling ratio (data not shown). Fig. 5 depicts the difference between the swelling ratio of

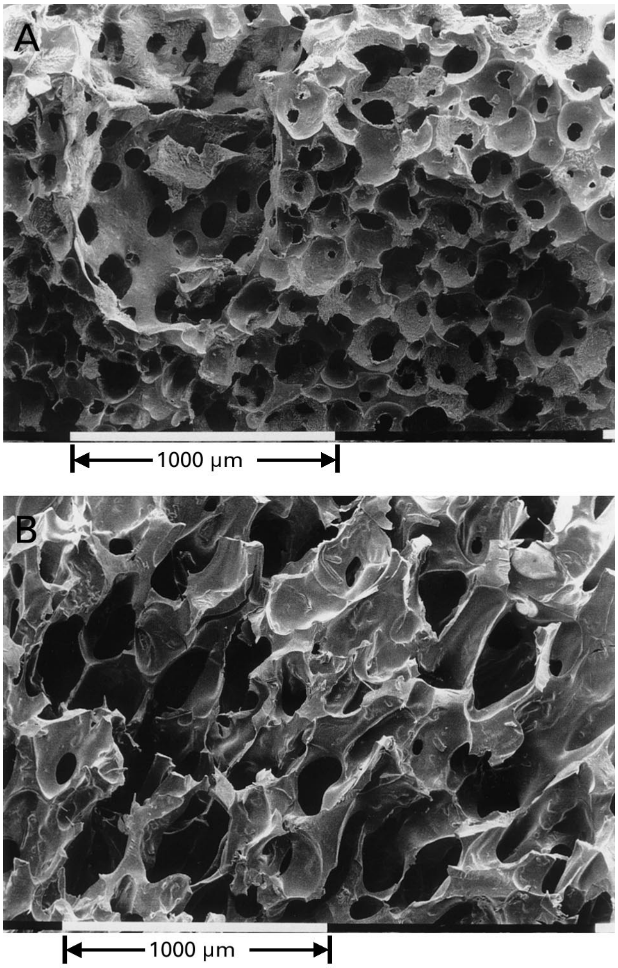

Fig. 6. Scanning electron microscopy pictures of: (A) SPH and (B) SPH composite.

SPH and SPH composites. SPH composites contain Ac-Di-Sol to increase the mechanical stability of these polymers. The cellulosic fibres of Ac-Di-Sol will occupy some space of the pores inside the polymer, and will reduce the flexibility of the polymer chain. Therefore, at the start of the swelling SPH composites cannot absorb water very quickly, but after some initial swelling the pores are opened enough to absorb additional water. Thus, the final swelling of SPH composite after 9–15 min was about the same as observed for SPH after freeze-drying for 2 days.

Table 1
Apparent density of SPH after different drying procedures

| Drying procedure | Apparent density[a] (g/cm$^3$) |
|---|---|
| Absolute ethanol | 0.53 ± 0.08 |
| Acetone | 0.49 ± 0.07 |
| Diethyl ether | 0.65 ± 0.09 |
| Freeze-drying for 1 day | 0.74 ± 0.07 |
| Freeze-drying for 2 days | 0.39 ± 0.05 |

[a] Data are expressed as the mean ± SD of three experiments.

### 3.3. Measurements of apparent density

Since the SPH polymers are very porous (as observed by SEM; Fig. 6), the measured density is related to the porosity of these polymers and can be defined as apparent density. The actual densities of the polymers are the same but when the polymer has less pores (such as SPH composites), the occupied volume will be less; therefore, this will result in a high apparent density. As shown in Table 1, the apparent density of SPH polymers obtained by freeze-drying for 2 days is substantially lower than SPH dried by organic solvents or by freeze-drying for 1 day. Therefore, it is very likely that SPH freeze-dried for 2 days contains more pores and can swell more rapidly as already shown in the swelling ratio studies (Fig. 4). The apparent densities of the polymers dried by organic solvents and by 1 day freeze-drying are higher, indicating a reduced amount of free space of pores. These results are in accordance with the present swelling ratio studies. The apparent density of the SPH composite is $0.91 \pm 0.12$ g/cm$^3$ and is higher in comparison to the SPH polymer due to the incorporation of the cellulosic fibres within the polymer structure.

### 3.4. SEM studies

Fig. 6 shows the scanning electron microscopic pictures of SPH and SPH composites. The SPH polymer (Fig. 6A) has a higher porosity, which is in accordance with apparent density results. In the structure of SPH composites (Fig. 6B) the pores are less, which explains the lower swelling ratio and higher apparent density for SPH composites.

## 4. Conclusions

From the present $^{13}$C NMR studies, it is concluded that the structure of SPH consists of a sequence of acrylic acid (AA) and acrylamide (AM) as long aliphatic chains, which are cross-linked with *N*,*N*′-methylenebis acrylamide (Bis). The high porosity of the polymer is achieved by the $CO_2$ formation during the polymerization process, resulting in a capillary structure of interconnecting pores. The high polar internal surface of these pores within the SPH is responsible for the very fast swelling rate and resulting high swelling ratio of the polymers. SPH composites consist of a sequence of AM and potassium salt of 3-sulfopropyl acrylate (SPAK), cross-linked by Bis. The superporous structure, also induced by $CO_2$, is additionally stabilized by cellulosic fibres (Ac-Di-Sol) which are responsible for a delayed but also complete swelling of these SPH composites. The unique characteristics of these polymers open a new field of application in controlled drug delivery. Because the swelling properties of both SPH and SPH composites are pH independent, these polymers can be used as drug delivery systems for drug release in the stomach and in the intestine. The drug release can easily be related to the swelling properties of these polymers. Hence, both types of polymers can be used as novel carriers for controlled drug delivery.

## References

[1] English AE, Tanaka T, Edelman ER. Polymer 1998;39:5893–7.

[2] Guzmán J, Iglesias M, Riande E, Compañ V, Andrio A. Polymer 1997;38:5227–32.

[3] Sen M, Güven O. Polymer 1998;39:1165–72.

[4] Peppas NA, Nikos AG. Hydrogels in medicine and pharmacy, vol. 2. Boca Raton, FL: CRC Press, 1986 (chap 3; p. 65–95).

[5] Park H, Park K, Hwang SJ. Crit Rev Ther Drug Carrier Syst 1998;15:243–84.

[6] Montheard JP, Chatzopoulos M, Chappard D. J Macromol Sci—Rev Macromol Chem Phys 1992;C32:1.

[7] Barbieri R, Quaglia M, Delfini M, Brosio E. Polymer 1998;39:1059–66.

[8] Bhalerao VS, Varghese S, Lele AK, Badiger MV. Polymer 1998;39:2255–60.

[9] Chen J, Park H, Park K. J Biomed Mater Res 1999;44:53–62.

[10] Van Dijk-Wolthuis WNE, Tsang SKY, Kettenes-van den Bosch JJ, Hennink WE. Polymer 1997;38:6235–42.

[11] Peniche C, Cohen M, Vazquez B, San Roman J. Polymer 1997;38:5977–82.

[12] Gudeman LF, Peppas NA. J Appl Polym Sci 1995;55:919.

[13] Lee YM, Soo Kim S. Polymer 1997;38:2415–20.

[14] Mathias LJ, Colletti RF, Halley RJ, Jarrett WL, Johnson CG, Powell DG, Warren SC. Solid-state NMR, polymer spectra, Collected volume I. Hattiesburg, MS, USA: MRG Polymer Press, 1990 (p. 1 and 39).