Inulin Hydrogels as Carriers for Colonic Drug Targeting. Rheological Characterization of the Hydrogel Formation and the Hydrogel Network

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Abstract □ Free radical polymerization converts aqueous solutions of methacrylated inulin into cross-linked hydrogels. The purpose of this work was to study the hydrogel formation and to characterize the fully cured hydrogels. The gelation process of aqueous solutions of methacrylated inulin was monitored as a function of time by means of linear oscillatory shear measurements, at a fixed frequency and amplitude. The fully cured inulin hydrogels were characterized by measurement of the frequency-dependency of the linear elastic modulus *G′*. The effects of the degree of substitution and feed concentration of methacrylated inulin on both the gelation kinetics and the rigidity of the obtained hydrogels were determined. The effect of the concentration of the initiators of the radical polymerization reaction has been studied as well. The weight fraction of polymer which was not incorporated in the hydrogel networks was determined using the anthrone reaction, and physical chain entanglements were determined by solution viscosity measurements. The gelation kinetics and the elastic modulus were proportional to the degree of substitution and feed concentration of methacrylated inulin. Increasing concentrations of radical-forming compounds also accelerated the hydrogel formation, but lowered the elastic modulus of the obtained hydrogels. The amount of polymer chains incorporated in the hydrogel network seemed to be especially influenced by the degree of substitution of the derivatized inulin, and for a feed concentration of 27% w/w of methacrylated inulin, entanglements have to be accounted for. The gelation kinetics and the elastic modulus of inulin hydrogels are not only affected by the degree of substitution and the feed concentration of methacrylated inulin, but also by the concentration of the initiators of the free radical polymerization reaction.

Introduction

Inulin hydrogels were developed as carriers for colonic drug targeting. Inulin is a storage polysaccharide found in many plants such as garlic, onion, artichoke, and chicory.[1] It consists of β 2−1 linked D-fructose molecules, and the majority of the fructose chains have a glucose unit at the reducing end. The β 2−1 glycosidic bond is not significantly hydrolyzed by the endogenous secretions of the human digestive tract,[2] but bacteria residing in the colon, and more specifically *Bifidobacteria*, are able to ferment inulin.[3−5] *Bifidobacteria* constitute up to 25% of the normal gut flora of man and animals.[6] Hence, inulin was selected as candidate polymer for the development of a colon-specific drug delivery systems.

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Inulin hydrogel formation has been previously described by Vervoort et al.[7] Vinyl groups were incorporated in the fructose chains by reaction of inulin with glycidyl methacrylate at room temperature using 4-(dimethylamino)-pyridine as catalyst. Transesterification occurred during the reaction of inulin with glycidyl methacrylate, since the obtained reaction product was characterized as methacrylated inulin (MA-IN) instead of glyceryl methacrylated inulin. This confirmed the findings which have been published previously by van Dijk-Wolthuis et al.[8] for the derivatization of dextran with glycidyl methacrylate. By varying the molar ratio of glycidyl methacrylate to inulin, the degree of substitution (DS), i.e., the amount of methacryloyl groups per 100 fructose units, of MA-IN could be tuned. Aqueous solutions of MA-IN were subsequently converted into cross-linked three-dimensional networks by free radical polymerization using ammonium persulfate (APS) and *N,N,N′,N′*-tetramethylethylenediamine (TMEDA) as radical generating compounds. Figure 1 gives a schematic representation of the derivatization reaction of inulin with glycidyl methacrylate and MA-IN hydrogel formation.

This work describes the rheological characterization of the synthesized inulin hydrogels as well as the hydrogel formation process. The influence of the degree of substitution and feed concentration of MA-IN, and the concentration of radical-generating compounds, were studied. The rheological data are correlated with previously obtained results with respect to the equilibrium-swelling degree of the inulin hydrogels and the in vitro enzymatic degradation of the devices.

Experimental Section

Materials—Chicory inulin (Raftiline HP; average degree of polymerization between 22 and 25) was kindly provided by Orafti (Tienen, Belgium), and was used for the synthesis of methacrylated inulin. APS was supplied by UCB (Leuven, Belgium) and TMEDA by Sigma (St. Louis, MO). Anthrone was obtained from ICN Biomedicals (Ohio) and D(−)-fructose from Merck (Darmstadt, Germany). For details about the materials used for the synthesis of MA-IN, reference is made to Vervoort et al.[7] For the rheological experiments, MA-IN solutions with feed concentration of 16%, 22%, and 27% w/w were prepared in 0.5 M phosphate buffer pH 6.5. MA-IN with three different degrees of substitution was used: DS = 4.4, 12.1, and 22.3.

Rheological Experiments—Dynamic oscillatory measurements were performed on a strain-controlled rheometer (Rheometrics Mechanical Spectrometer RMS705), equipped with a parallel plate geometry (diameter 25 mm). To avoid slippage of the sample, the plates were covered with sand-paper. The temperature was kept constant at 20 °C by a fluids bath. MA-IN solutions were prepared and immediately after adding the radical-generating compounds, the solutions were poured onto the lower plate (cup) of the rheometer, after which the upper plate was positioned at a gap of 1 mm. The free surface of the sample was

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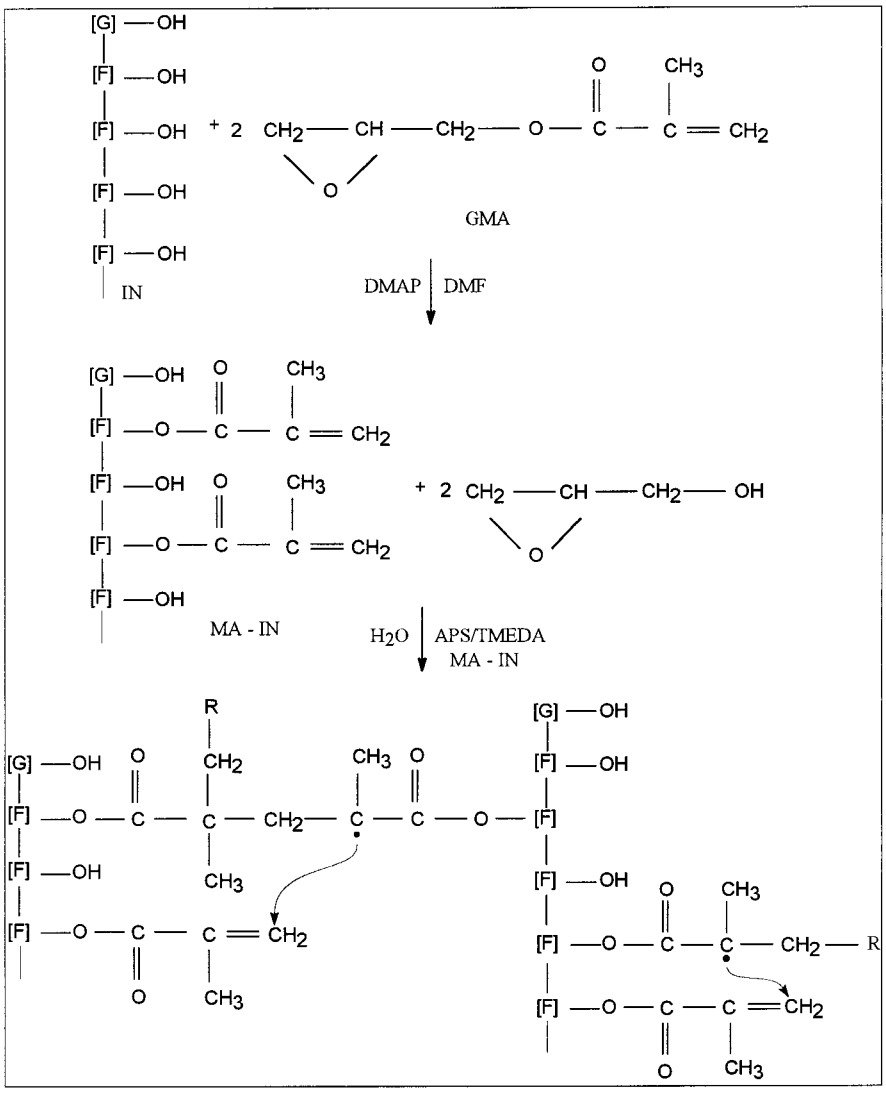

Figure 1—Schematic representation of the synthesis of MA-IN and the formation of inulin hydrogels. ([F]-OH = fructosyl unit; [G]-OH = glucosyl unit; IN = inulin; GMA = glycidyl methacrylate; DMAP = 4-(dimethylamino)pyridine; DMF = *N,N*-dimethylformamide; MA-IN = methacrylated inulin; APS = ammonium persulfate; TMEDA = *N,N,N′,N′*-tetramethylethylenediamine; R = initiator.)

covered with a mineral oil of low viscosity to prevent evaporation of the solvent during the experiment. The loading procedure took 160 s, after which the dynamic measurements started.

Small strain oscillations at a fixed frequency (4 rad/s) were applied to the samples to monitor the hydrogel formation. The strain was kept low enough to avoid interference with the formation of the MA-IN networks. After completion of the gelation process, a frequency sweep experiment was performed to characterize the fully cured MA-IN hydrogels. Before the latter experiment, the excess of gel at the rim of the geometry was removed to minimize errors due to edge effects. The dynamic behavior could be recorded over a frequency range from 0.01 to 10 rad/s. A linearity check was performed on every hydrogel to ensure that all measurements were probing the linear behavior of the hydrogels (results not shown).

Determination of Weight Fraction of Soluble Material (sol fraction)—The weight fraction of soluble material of a hydrogel (sol fraction) is defined as the amount of polymer chains, which are not incorporated in the hydrogel network and which can consequently be extracted and separated from the hydrogel.[9] To determine this sol fraction, MA-IN hydrogels of known dry weight were prepared and washed in demineralized water to extract the polymer chains, which are not attached to the hydrogel network. The sol fraction was calculated as the weight ratio of extracted material to initial material. The amount of extracted sugars was determined by means of the anthrone method,[10,11] and for the calculations, the degree of substitution of MA-IN used to prepare the respective hydrogels was taken into account.

The anthrone reagent was prepared by dissolving 50 mg of anthrone in a mixture of 28 mL of water and 72 mL of concentrated sulfuric acid. This freshly prepared reagent (2.5 mL) was mixed with 0.3 mL of collected samples of the hydrogel washing water containing the polymer chains, which were not incorporated in the hydrogel network. The samples were incubated for 10 min at 100 °C followed by cooling to room temperature. Finally, the absorbance of the samples was read spectrophotometrically with a HP 8452A spectrophotometer (Hewlett-Packard, Santa Clara, CA) at 625 nm. Since the same absorption is given by a sugar compound as if it was first hydrolyzed and then determined,[12] fructose solutions with concentrations ranging from 0.08 to 0.2 mg fructose/mL were used to construct a calibration line. Beside fructose, inulin also contains glucose units, but this does not compromise the anthrone method for sol fraction determination since equal amounts of glucose and fructose give identical absorption values.[12]

Determination of the Intrinsic Viscosity—To investigate whether chain entanglements, which can act as additional intermolecular cross-links, occur in the MA-IN solutions of different feed concentration, the intrinsic viscosity of MA-IN DS 12.1 was determined by solution viscosity measurements using an Ubbelohde suspended-level capillary viscometer (Schott Geräte, Hofheim, Germany). Efflux times of MA-IN solutions in 0.5 M phosphate buffer pH 6.5 were determined at 20 °C, and the respective reduced viscosities were calculated. The intrinsic viscosity was obtained by extrapolating the plot of the obtained values of reduced viscosity against the MA-IN concentration to infinite

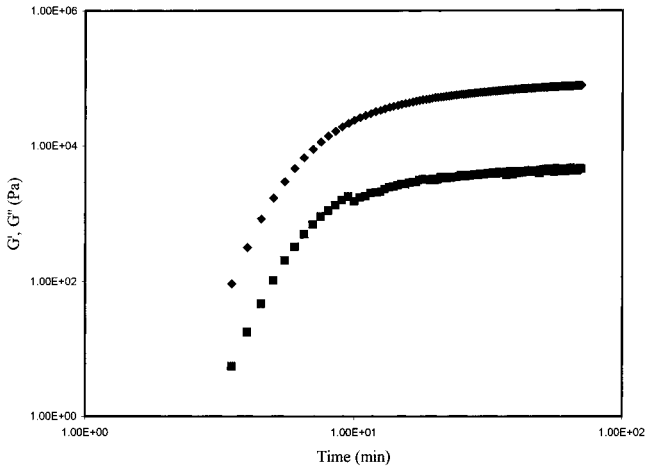

Figure 2—G' (◆) and G'' (■) as a function of polymerization time for hydrogel formation of a 27% w/w MA-IN DS 12.1 solution. (Concentration of APS = 17.5 μmol/mL buffer and TMEDA = 39.4 μmol/mL buffer.)

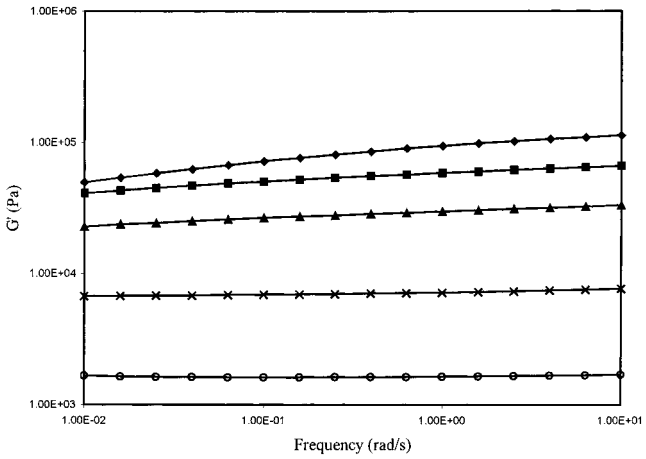

Figure 3—G' as a function of frequency for fully cured MA-IN hydrogels with different degree of substitution and feed concentration (○ = DS 4.4; 27%, × = DS 12.1; 16%, ▲ = DS 12.1; 22%, ■ = DS 12.1; 27%, ◆ = DS 22.3; 27%). (Concentration of APS = 17.5 μmol/mL buffer and TMEDA = 39.4 μmol/mL buffer.)

dilution according to the Huggins equation:

$$\eta_{red} = [\eta] + k[\eta]^2 c \quad (1)$$

with η_{red} the reduced viscosity, $[\eta]$ the intrinsic viscosity expressed in dL/g, k the Huggins constant, and c the concentration of the MA-IN solutions expressed in g/dL.

Results and Discussion

The addition of the radical-generating compounds APS and TMEDA to aqueous MA-IN solutions results in free radical polymerization. Dynamic rheological measurements at a fixed frequency and amplitude were performed during this process in order to follow the gradual formation of a three-dimensional network. The formation of intermolecular cross-links should show up in an increase of the elastic modulus G' as a function of time. G' is a measure of the energy stored and recovered per cycle of oscillatory deformation and represents the elastic behavior of the material.[13] Beside intermolecular cross-links, the radical polymerization reaction probably also introduces loops or intramolecular cross-links, loose ends or chains incorporated in the network by a single bond, unattached material or sol fraction, and physical chain entanglements in the network. These structures hardly contribute to an increase in G'.[14−16] The viscous modulus G'', on the other hand, is a measure of energy dissipation per cycle of sinusoidal deformation[13] and represents the viscous behavior of the system under investigation. The viscosity is known to be sensitive to the molecular weight of polymer chains. Figure 2 shows a plot of G' and G'' versus time, representing the gelation process of a 27% w/w MA-IN DS 12.1 solution. From the point on where the moduli have increased sufficiently to be measurable, G'' is an order of magnitude smaller than G'. This was the case for all hydrogel formulations tested. Since the fully cured hydrogels exhibited no or only a limited frequency dependence of G' (Figure 3), it can be assumed that a network has indeed been formed by the radical reaction. The profile of G' versus time (Figure 2) can thus be interpreted as the gradual formation of a hydrogel network from a MA-IN solution. The three stages commonly encountered in hydrogel formation can be distinguished from the evolution of G' in time: a pregelation period, in which no measurable increase in G' is seen, a gelation period showing a rapid increase in G', and a postgelation period in which G' hardly changes anymore and reaches a constant value. During the pregelation period free radicals are formed and reaction within and between chains starts. The incorporation of the MA-IN polymers in a three-dimensional network takes place during the gelation period while gelation is ending during the post-gelation period.

Table 1—Time (min, loading procedure included) To Reach 50% of G' end for Hydrogels of Different Composition ([APS] = 17.5 μmol/mL buffer, [TMEDA] = 39.4 μmol/mL buffer)

hydrogel composition		
DS	feed concn (% w/w)	time (min)
4.4	27	60.5
12.1	16	21.3
12.1	22	18.8
12.1	27	16.8
22.3	27	15.8

Table 2—Time (min, loading procedure included) To Reach 50% of G' end for Hydrogels Prepared from 16% w/w Solutions of MA-IN DS 12.1 as a Function of the Concentrations of APS and TMEDA (μmol/mL buffer)

[APS]	[TMEDA]	time (min)
17.5	39.4	21.3
17.5	78.7	13.1
17.5	157.4	4.6
70.1	39.4	3.8

To quantify a characteristic time for network formation, the time needed to reach 50% of the final value of G' (G' end) has been determined. Table 1 lists the results as a function of the degree of substitution and the feed concentration of MA-IN. It can be concluded that increasing the degree of substitution or the feed concentration accelerates the gelation process, implying that the radical cross-linking reaction is promoted by increasing concentrations of reactive vinyl groups. The characteristic time for network formation is also influenced by the concentration of the initiators of the polymerization reaction as is shown in Table 2. TMEDA is believed to exert a promoting effect on the vinyl polymerization initiated by persulfate attributed to an acceleration of the homolytic scission of the latter compound.[17,18] Increasing the concentration of the radical-generating compound APS, and thus introducing more free radicals in the reaction solution, also decreases the gelation time. To verify whether the accelerating effect of the

Table 3—Pregelation Period (min, loading procedure included) and Slope (Pa/min) of the Curves of G' versus Time ($0.1 < G'/G'$ end < 0.6), Indicative for the Gelation Period, for Hydrogels Prepared from 16% w/w Solutions of MA-IN DS 12.1 as a Function of Varying Concentrations of APS and TMEDA (μmol/mL buffer)

[APS]	[TMEDA]	pregelation period (min)	slope (Pa/min)
17.5	39.4	6.57	265.70
17.5	78.7	3.86	267.45
17.5	157.4	1.03	709.14
70.1	39.4	0.29	780.00

initiators was caused by shortening either the pregelation or the gelation period, both periods were separately defined, and the effect of the initiator concentration was studied. The pregelation period was defined as the time needed to reach 1% of G' end. The slope of the curves of G' versus time ($0.1 < G'/G'$ end <0.6) was considered as an indication of the rate of gelation. Table 3 illustrates that increasing initiator concentrations accelerate both stages of hydrogel formation: using high concentrations of APS and TMEDA, intermolecular cross-link formation is detectable almost immediately after mixing the two compounds with the MA-IN solutions, and the gelation process is characterized by a steep rise in G'.

From this kinetic study, it was concluded that the formation of MA-IN hydrogels can be performed within 2.5 h, because cross-linking of MA-IN chains will have ended in this period of time, at least for the range of degree of substitution and feed concentration studied.

If an ideal three-dimensional network is formed by the radical polymerization, the elastic modulus G' is independent of frequency. This frequency-independent elastic modulus is called the equilibrium elastic modulus G_e and is a measure of the structure that has been formed by the cross-linking reaction. Several theories, such as the affine theory, the phantom theory, the constrained junction model, or the Langley−Graessley model,[19−21] have been developed to relate the macroscopic elastic modulus of a polymeric network to its molecular structure. From these theories, it can generally be concluded that G_e is proportional to the number of moles of elastic chains, i.e., chains attached to the network with both ends, per volume unit of the network (ν) according to the following equation:

$$G_e \sim \nu RT \qquad (2)$$

with R the gas constant and T the absolute temperature.

Determination of G_e can thus give an idea about the concentration of elastic network strands and hence about the amount of intermolecular cross-links formed.

For the cross-linked materials under investigation, the frequency dependence of the elastic modulus is shown in Figure 3 over three decades in frequency. For hydrogels prepared from MA-IN with low degree of substitution (27% w/w solution with DS = 4.4) or low feed concentration (16% w/w solution with DS = 12.1), the values of G' appear to be almost independent of frequency. To a first approximation, these hydrogels can thus be considered as ideal networks, and the constant value of G' consequently represents the equilibrium elastic modulus G_e. However, the other hydrogels tested show a slight decrease of G' with decreasing frequency, indicating that relaxation (rearrangements) can still occur in the network. For these systems, G_e could not be determined. Nevertheless, it may be concluded that the amount of elastic chains per volume unit of the network increases with increasing degree of substitution and feed concentration, since both parameters definitely have an increasing effect on G'. These observations support the results of the equilibrium degree of swelling study and the in vitro enzymatic degradation study of the MA-IN hydrogels.[22,23] The decreased equilibrium swelling and the decreased enzymatic degradation of the hydrogels with increasing degree of substitution or feed concentration of MA-IN can be attributed to the increased intermolecular cross-linking density (higher G' values) of the networks, which increases the contractility of the hydrogels, resulting in a restriction of expansion upon swelling, and which reduces the permeability of the networks toward degrading enzymes.

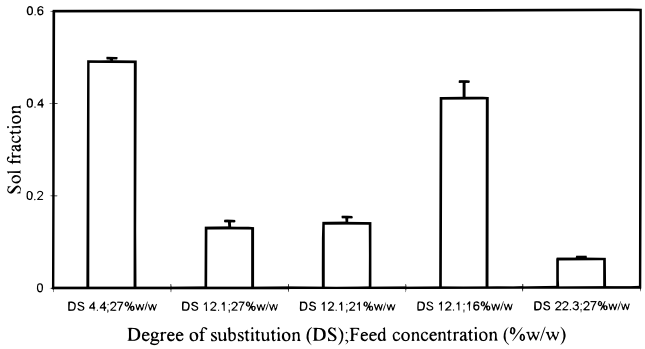

Figure 4—Weight fraction of soluble material (sol fraction) for MA-IN hydrogels with different compositions.

The observed frequency dependence of G' can possibly be attributed to imperfections in the structure of the network.[19] Loose ends, loops, and MA-IN chains which are not incorporated in the network, can relax, causing a decrease in G' at low frequencies.

It was first checked whether the presence of a sol fraction can explain the relaxation in G'. As gelation of the MA-IN solutions took place between the plates of the rheometer, the chains which are not incorporated in the hydrogel network, have not been removed. Figure 4 shows the sol fractions of MA-IN hydrogels of different feed composition. As expected, the sol fraction is reduced with an increasing amount of reactive groups per chain (DS): more chains will participate in the network when there are more groups present which can link them to the network. The effect of concentration on sol fraction can be understood from the promoting effect of dilution on intramolecular cross-linking, which does not incorporate chains in the network. The probability that bonds will be formed between different chains of the system is proportional to the probability that these chains lie in the same small volume-element[14] and therefore increases with concentration. Hence, the hydrogels prepared from a 16% w/w solution (MA-IN DS = 12.1) exhibited a higher sol fraction when compared with the more concentrated solutions (22 and 27% w/w). However, comparison of the data from Figure 3 and Figure 4 clearly indicates that the sol fraction data cannot explain the frequency dependence of G': the samples with the higher sol fraction do not correspond to the samples with the more pronounced frequency dependence of G'.

A second possible explanation for the observed decrease of G' is the relaxation of loose ends and loops. MA-IN solutions of high feed concentration form dense networks in a short period of time compared with more diluted solutions (Figure 3 and Table 1). As mentioned above, intermolecular cross-links are predominantly formed starting from a concentrated solution, whereas intramolecular cross-link formation is promoted by a diluted solution. Due to the higher extent of intermolecular cross-linking in case of a concentrated solution, the mobility of the polymer chains can be restricted to such an extent that full conversion of the vinyl groups can be impeded. Consequently, more loose ends may remain in the hydrogel network, and these are able to relax on the time scales

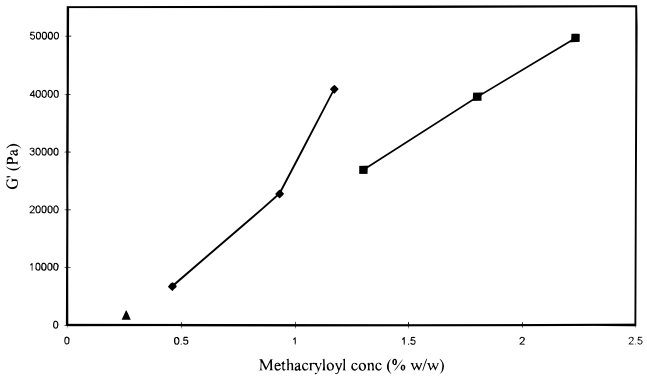

Figure 5—*G′* as a function of the real methacryloyl concentration in the hydrogel network prepared from MA-IN DS 4.4 (▲), DS 12.1 (◆), and DS 22.3 (■).

probed by the oscillatory experiments. In addition, when the feed concentration of MA-IN exceeds the critical concentration at which MA-IN chains start to overlap, physical chain entanglements are formed which promote intermolecular cross-linking even more. Moreover, the entanglements themselves can be considered as additional intermolecular cross-links, contributing to an increase in *G′* and additionally limiting chain mobility thereby restricting full conversion of the vinyl groups.

A rule of thumb for predicting when concentration effects will become important is when the coil overlap parameter $c[\eta]$, with c the concentration of the polymer solution (g/dL) and $[\eta]$ the intrinsic viscosity of the polymer (dL/g), is near unity.[24] Consequently, it can be postulated that:

$c[\eta] < 1$: the polymer coils are isolated one from another, and the solution can be considered as a particle solution;

$c[\eta] = 1$: the polymer coils begin to overlap;

$c[\eta] > 1$: the polymer coils form entanglements, and the solution can be considered as a network solution.

The intrinsic viscosity of MA-IN DS 12.1 is 0.038 dL/g as determined by solution viscosity measurements. This implies that for the highest feed concentration (density = 1.14 g/mL), entanglements have to be accounted for, which may increase the amount of loose ends.

Unfortunately, the degree of conversion of MA-IN during hydrogel formation could not be determined, neither by FT-IR nor by solid state ^{13}C NMR spectroscopy.[7] But van Dijk-Wolthuis et al.[18] described indeed a low final percentage conversion of the methacryloyl groups of methacrylated dextran upon free radical polymerization at a high initial concentration of methacrylated dextran compared with a low initial concentration.

With respect to the effect of degree of substitution on the observed frequency dependence of *G′*, the same remark can be made as for the effect of the feed concentration. Solutions of MA-IN with a high degree of substitution are converted rather fast in rigid networks (high value of *G′*), implying again that full conversion of the double bonds may be impeded and loose ends may remain. A high degree of substitution additionally promotes cyclization or the formation of loops,[25] which can also relax during the rheological experiments.

To investigate the relative amount of inter- to intramolecular cross-links, G_e, being a measure of the intermolecular cross-links, was plotted in Figure 5 as a function of the methacryloyl concentration, a measure of the amount of reactive sites. For the perfect networks, G_e was considered whereas for the imperfect networks *G′* at a frequency of 0.01 rad/s was taken. Since the sol fraction of the systems under investigation is not negligible (Figure 4), the real methacryloyl concentration in the network, i.e., the concentration corrected for the not incorporated polymer

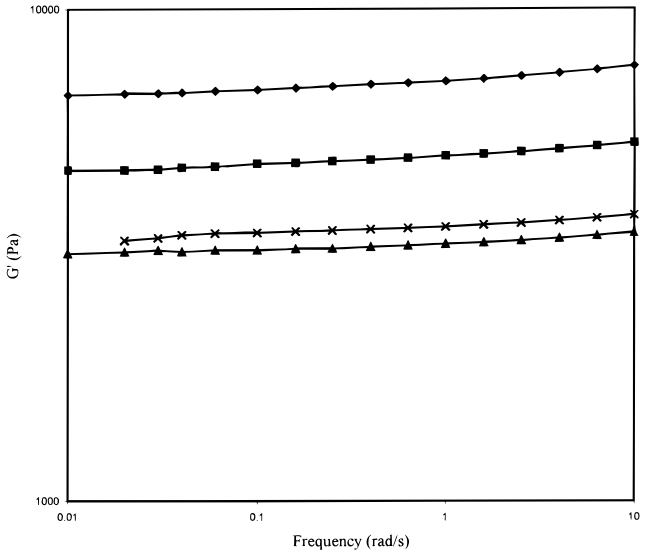

Figure 6—*G′* as a function of frequency for MA-IN hydrogels with DS 12.1 and feed concentration 16% w/w prepared with different concentrations of APS and TMEDA (μmol/mL buffer). ([APS]; [TMEDA] = 17.5; 39.4 (◆), 17.5; 78.7 (■), 17.5; 157.4 (▲), and 70.1; 39.4 (×).)

chains, has been considered. Determination of the sol fraction of hydrogels prepared from 16% and 22% w/w solutions of MA-IN DS 4.4 could not be performed, as these hydrogels were too weak to manipulate. Hence, only one value for DS 4.4 is depicted in Figure 5. Figure 5 clearly illustrates that increasing amounts of reactive vinyl groups in the polymerization reaction (increased feed concentration) results in the increased formation of intermolecular cross-links, confirming the conclusions from Figure 3. In addition, this figure suggests that for the same methacryloyl concentration, hydrogels prepared from MA-IN with a low degree of substitution exhibit higher values of *G′* than hydrogels from MA-IN with a higher degree of substitution. In other words, to form an equal amount of intermolecular cross-links, higher substituted MA-IN may need a higher amount of polymerizable groups, since reactive sites may be lost by intramolecular cross-linking or perhaps may not have reacted at all (lower degree of conversion). These observations agree with the remarks made in order to explain the frequency dependence of *G′*.

Finally, the effect of the concentration of the initiating species APS and TMEDA on the resulting structure of the hydrogel has been investigated. Figure 6 represents the dynamic behavior of fully cured networks prepared from 16% w/w solutions of MA-IN DS 12.1 with various initiator concentrations. Apparently, hydrogels prepared with higher initiator concentrations exhibit lower values of *G′*, at least for the range of concentrations studied. This can be explained by the fact that the higher concentrations of initiators result in the formation of an increased number of radicals in the initiation step of the radical polymerization reaction. Consequently, the number of active centers formed on the inulin chains is also increased, implying that less unreacted vinyl groups are left to cross-link the chains, which results in lower *G′* values.

Conclusions

Oscillatory shear measurements, performed on inulin solutions during the gelation process as well as on fully cured hydrogels, showed that the degree of substitution and feed concentration of MA-IN, and the concentration of the initiating species of the free radical polymerization reaction (APS and TMEDA), all affect the gelation process and the

rigidity of the obtained hydrogels. A higher degree of substitution or feed concentration cause an acceleration of the network formation. The resulting hydrogels are characterized by a higher mechanical strength (higher G' values), arising from the increased amount of intermolecular cross-links formed. Increasing concentrations of APS and TMEDA also shorten the process of hydrogel formation, but result in hydrogels with lower G' values.

References and Notes

1. Van Loo, J.; Coussement, P.; De Leenheer, L.; Hoebregs, H.; Smits, G. On the presence of inulin and oligofructose as natural ingredients in the western diet. *Crit. Rev. Food Sci. Nutr.* **1996**, *36*, 525−552.
2. Dysseler, P.; Hoffem, D. Inulin, an alternative dietary fiber. Properties and quantitative analysis. *Eur. J. Clin. Nutr.* **1995**, *49*, S145−S152.
3. Gibson, G. R.; Roberfroid, M. B. Dietary modulation of the human colonic microbiota: introducing the concept of prebiotics. *J. Nutr.* **1995**, *125*, 1401−1414.
4. Roberfroid, M. B. Dietary fiber, inulin, and oligofructose: a review comparing their physiological effects. *Crit. Rev. Food Sci. Nutr.* **1993**, *33*, 103−148.
5. Wang, X.; Gibson, G. R. Effects of the in vitro fermentation of oligofructose and inulin by bacteria growing in the human large intestine. *J. Appl. Bacteriol.* **1993**, *75*, 373−380.
6. McKellar, R. C.; Modler, H. W. Metabolism of fructooligosaccharides by *Bifidobacterium spp*. *Apl. Microbiol. Biotechnol.* **1989**, *31*, 537−541.
7. Vervoort, L.; Van den Mooter, G.; Augustijns, P.; Busson, R.; Toppet, S.; Kinget, R. Inulin hydrogels as carriers for colonic drug targeting. I. Synthesis and characterization of methacrylated inulin, and hydrogel formation. *Pharm. Res.* **1997**, *14*, 1730−1737.
8. van Dijk-Wolthuis, W. N. E.; Kettenes-van den Bosch, J. J.; van der Kerk-van Hoof, A.; Hennink, W. E. Reaction of dextran with glycidyl methacrylate: An unexpected transesterification. *Macromolecules* **1997**, *30*, 3411−3413.
9. Peppas, N. A.; Barr-Howell, B. D. Characterization of the cross-linked structure of hydrogels. In *Hydrogels in medicine and pharmacy*; Peppas, N. A., Ed.; CRC Press: Boca Raton, FL, 1986; Vol. I, pp 27−56.
10. White, R. P.; Samson, F. E.; Kan, L. Determination of inulin in plasma and urine by use of anthrone. *J. Lab. Clin. Med.* **1954**, *March*, 475−478.
11. Summerfield, A. L.; Hortin, G. L.; Smith, C. H.; Wilhite, T. R.; Landt, M. Automated enzymatic analysis of inulin. *Clin. Chem.* **1993**, *39*, 2333−2337.
12. Morris, D. L. Quantitative determination of carbohydrates with Dreywood's anthrone reagent. *Science* **1948**, *107*, 254−255.
13. Ferry, J. D. Illustrations of viscoelastic behavior of polymeric systems. In *Viscoelastic properties of polymers*; Ferry, J. D., Ed.; John Wiley & Sons: New York, 1961; pp 21−40.
14. James, H. M.; Guth, E. Theory of the increase in rigidity of rubbers during cure. *J. Chem. Phys.* **1947**, *15*, 669−683.
15. Langley, N. R. Elastically effective strand density in polymer networks. *Macromolecules* **1968**, *1*, 348−352.
16. Tonelli, A. E.; Helfand, E. Elastically ineffective cross-links in rubbers. *Macromolecules* **1974**, *7*, 59−63.
17. Feng, X. D.; Guo, X. Q.; Qiu, K. Y. Study of the initiation mechanism of the vinyl polymerization with the system persulphate/N,N,N′,N′-tetramethylethylenediamine. *Makromol. Chem.* **1988**, *189*, 77−83.
18. van Dijk-Wolthuis, W. N. E.; Franssen, O.; Talsma, H.; van Steenbergen, M. J.; Kettenes-van den Bosch, J. J.; Hennink, W. E. Synthesis, characterization, and polymerization of glycidyl methacrylate derivatized dextran. *Macromolecules* **1995**, *28*, 6317−6322.
19. Patel, S. K.; Malone, S.; Cohen, C.; Gillmor, J. R.; Colby, R. H. Elastic modulus and equilibrium swelling of poly(dimethylsiloxane) networks. *Macromolecules* **1992**, *25*, 5241−5251.
20. Gottlieb, M.; Macosko, C. W.; Benjamin, G. S.; Meyers, K. O.; Merrill, E. W. Equilibrium modulus of model poly(dimethylsiloxane) networks. *Macromolecules* **1981**, *14*, 1039−1046.
21. Gnanou, Y.; Hild, G.; Rempp, P. Molecular structure and elastic behavior of poly(ethyleneoxide) networks swollen to equilibrium. *Macromolecules* **1987**, *20*, 1662−1671.
22. Vervoort, L.; Van den Mooter, G.; Augustijns, P.; Kinget, R. Inulin hydrogels. I. Dynamic and equilibrium swelling properties. *Int. J. Pharm*., accepted for publication.
23. Vervoort, L.; Rombaut, P.; Van den Mooter, G.; Augustijns, P.; Kinget, R. Inulin hydrogels. II. In vitro degradation study. *Int. J. Pharm*., in press.
24. M. Tirrel. Rheology of polymeric liquids. In *Rheology. Principles, measurements and applications*; Macosko, C. W., Ed.; VCH Publishers: New York, 1994; pp 475−514.
25. De Smedt, S. C.; Lauwers, A.; Demeester, J.; Van Steenbergen, M. J.; Hennink, W. E.; Roefs, S. P. F. M. Characterization of the network structure of dextran glycidyl methacrylate hydrogels by studying the reological and swelling behavior. *Macromolecules* **1995**, *25*, 5082−5088.

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