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Synthesis enhancements for generating highly soluble tetrabutylammonium alginates in organic solvents

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ABSTRACT

The adaptation of alginates to food and pharmaceutical specifications is limited to aqueous chemistry due to the insolubility of sodium alginate (Na-Alg) and the insufficient solubility of tetrabutylammonium alginate (TBA-Alg) in organic solvents. In the present investigation, these restrictions were resolved by optimizing the solubility of TBA-Alg by improving its synthesis from Na-Alg via heterogeneous acidification with hydrochloric and formic acid, followed by neutralization with tetrabutylammonium hydroxide. The best acidification results were achieved with formic acid, because the reaction controlling solubility of the by-product in the acidic solvent was improved in comparison to hydrochloric acid. The solubility of TBA-Alg in polar aprotic organic solvents improved by increasing the degree of TBA substitution (DS_{TBA}), decreasing the molecular weight of TBA-Alg and increasing the relative permittivity of the solvent. The best TBA-Algs, with DS_{TBA} = 0.95 and relative high molecular weights, gave optically clear solutions with a turbidity of about 1 NTU.

1. Introduction

Brown algae and some bacteria produce alginates (Hay, Ur Rehman, Ghafoor, & Rehm, 2010), an unbranched polysaccharide consisting of blocks of (1 → 4) linked ß-D-mannuronic acid (M) and its C5 epimer α-L-guluronic acid (G). Their excellent biocompatibility and gelling properties have resulted in numerous food and pharmaceutical applications, such as thickening or gelling agents (Saha & Bhattacharya, 2010), wound dressing materials (Schultz et al., 2003) and drug delivery systems (Anal & Singh, 2007; Tonnesen & Karlsen, 2002), and assures that they remain interesting for the development of new applications and the improvement of existing applications (Draget, Skjak-Braek, & Smidsrod, 1997).

Alginate chemistry, however, is limited to modifications in aqueous solvent systems due to the insolubility of sodium alginate (Na-Alg) and alginic acid (H-Alg) in organic solvents. The introduction of water-insoluble functional groups is therefore very difficult. This can be overcome by converting Na-Alg into tetrabutylammonium alginate (TBA-Alg), which is supposed to be soluble in polar aprotic organic solvents such as dimethylformamide (DMF) and dimethylsulfoxide (DMSO). Two common syntheses of TBA-Alg in laboratories are the direct conversion of Na-Alg using an ion exchange resin (Della Valle & Aurelio, 1992; Della Valle & Aurelio, 1994) and the heterogeneous acidification of Na-Alg followed by neutralization with tetrabutylammonium hydroxide (TBAOH) (Babak et al., 2000; De Boisseson et al., 2004; Pelletier, Hubert, Lapicque, Payan, & Dellacherie, 2000; Yang, Xie, & He, 2011). To execute further reactions, TBA-Alg is generally dissolved in an organic solvent by agitating overnight. However, Pawar and Edgar (2011) recently reported that TBA-Alg did not completely dissolve in organic solvents and that it was necessary to add tetrabutylammonium fluoride (TBAF) to obtain a homogeneous solution. In our own pre-tests, we also observed considerable turbidity and incompletely dissolved TBA-Alg in DMF (unpublished results). In general, part of the TBA-Alg dissolved rather quickly, in less than 1 h, whereas the remaining TBA-Alg did not dissolve completely, even overnight and the solutions obtained always appeared to be inhomogeneous. Neither TBA-Alg solubility nor the homogeneity of the solution was reproducible. A possible explanation for these observations could be an incomplete acidification of Na-Alg, which could be represented by a degree of substitution with TBA (DS_{TBA}) of less than 1 (Fig. 1). The solubility of TBA-Alg could therefore depend on the chosen solvent and on the ratio of its TBA, sodium and hydrogen counter ions. This paper investigates the robustness and optimization of the conversion of Na-Alg via H-Alg into

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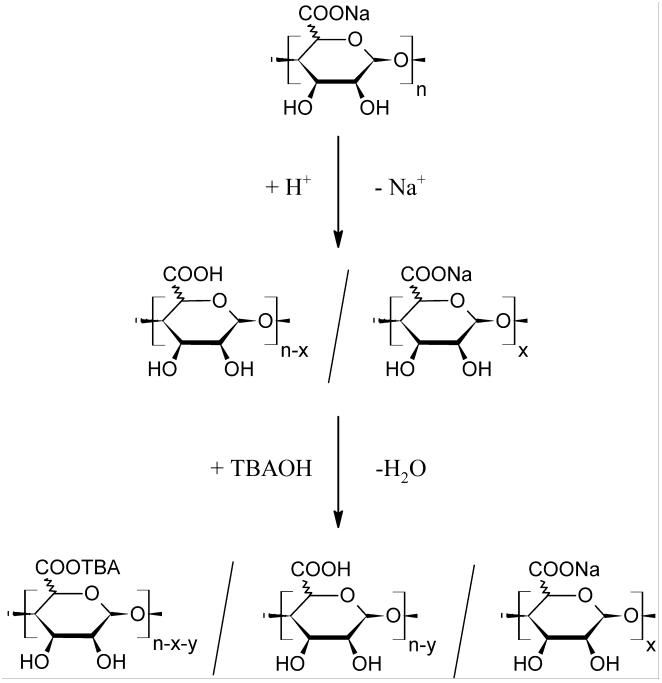

Fig. 1. Incomplete transformation of sodium alginate (Na-Alg) into tetrabutylammonium alginate (TBA-Alg) via alginic acid (H-Alg).

TBA-Alg, and the solubility of TBA-Algs in polar aprotic organic solvents depending on the counter ion ratios (TBA^+:H^+:Na^+) and molecular weight.

2. Experimental

2.1. Materials

Na-Algs from brown algae with low (12 cP, 1% in water, 25 °C) and medium viscosity (≥2000 cP, 2% in water, 25 °C), and H-Alg from brown algae were purchased from Sigma (St. Louis, MO, USA), dimethylacetamide (DMA (water ≤0.1%)), TBAOH 30-hydrate, tetrabutylammonium chloride (TBACl), sodium chloride (NaCl) for chromatography from Sigma-Aldrich, ~40% TBAOH solution from Fluka (Buchs, Switzerland), hydrochloric acid (HCl, 37%), formic acid, ethanol (99%), acetone, dimethylformamide (DMF (water ≤0.01%)) and dimethylsulfoxide (DMSO (water ≤0.05%)) from VWR (Radnor, PA, USA), acetonitrile (water ≤0.02%) from Biosolve (Valkenswaard, The Netherlands), potassium bromide (KBr) from ACROS (Geel, Belgium) sodium hydroxide solution (0.1 N, titrisol) and Certipur buffers (pH 4.00 and 6.86) from Merck (Darmstadt, Germany), ethylenediaminetetraacetic acid (EDTA) standard from the Leco Corporation (St. Joseph, MI, USA) and StablCal formazin standards from Hach (Loveland, CO, USA). In general, Milli-Q water was used. All materials were used without any further purification. The Na-Algs Manucol DM (M/G ratio: 0.61/0.39), Protonal GP 5450 (M/G ratio: 0.45/0.55), Protonal SF 120 RB (M/G ratio: 0.45/0.55) and Protonal GP 1740 (M/G ratio: 0.37/0.63) from FMC BioPolymer were received as a gift from IMCD Benelux N.V. (Mechelen, Belgium).

2.2. Chemistry

The standard procedures for the synthesis of H-Alg and TBA-Alg were defined with deviations according to the instructions for the heterogeneous acidification of Na-Alg followed by neutralization with TBAOH (Babak et al., 2000; De Boisseson et al., 2004; Pelletier et al., 2000; Yang et al., 2011). Different aspects of the reactions were studied for optimization by a systematic deviation of one parameter of the reaction conditions at a time.

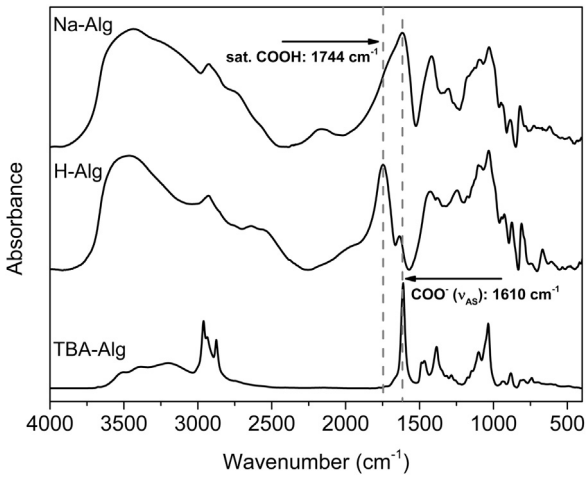

Fig. 2. FTIR spectra of sodium alginate (Na-Alg), alginic acid (H-Alg) and tetrabutylammonium alginate (TBA-Alg). Na-Alg and H-Alg were investigated in the form of KBr pellets and TBA-Alg in the form of a cast film. Quantitative analysis of the band intensity of carboxylic acid (1744 cm^{-1}) to carboxylate ion (1610 cm^{-1}) was possible for TBA-Alg, due to the separation of both bands in the TBA-Alg spectrum.

2.2.1. H-Alg standard synthesis

Na-Alg (2 g, 10 mmol) was dispersed in aqueous ethanolic HCl (100 mL, 0.6 N HCl; ethanol/water: 70% (v)/30% (v)) at 4 °C. The dispersion was stirred at 4 °C for 1 h before the ice bath was removed. The reaction mixture was then allowed to warm up whilst being stirred overnight (approximately 13 h). The product was filtrated, washed first with aqueous ethanol (300 mL, ethanol/water: 70% (v)/30% (v)) and then with acetone (100 mL), dried under vacuum and then kept in a desiccator over a drying agent until further use. FTIR (KBr pellet; Fig. 2): υ(OH) 3900–3064 cm^{-1}, υ(CH) 3064–2787 cm^{-1}, (sat COOH) 1744 cm^{-1}, $\upsilon_{as}(COO^-)$ 1636 cm^{-1}, $\upsilon_s(COO^-)$ 1412 cm^{-1}, (C—O) 1323 cm^{-1}, (C—O—C) 1180 cm^{-1} and 1101 cm^{-1}, (C—OH) 1032 cm^{-1}. The M/G ratio was between 0.61/0.39 (808 cm^{-1}/787 cm^{-1}) and 0.54/0.46 (1030 cm^{-1}/1080 cm^{-1}). Elemental analysis: C: 40.1%; H: 4.7%; degree of acidification (DS_H): 0.80 (titration); yield: 1.42 g (78%). The M/G ratio of commercially available H-Alg was determined to be between 0.61/0.39 and 0.54/0.46 and for medium viscosity sodium alginate between 0.60/0.40 and 0.53/0.47.

2.2.2. Variations of the heterogeneous acidification

This reaction was systematically optimized by varying the warm-up period (0–20 h), the temperature during the first hour (0–50 °C), the water fraction in ethanol (0–100% (v)), the Na-Alg concentration (0.5–2% (w)), the HCl concentration (0.1–1 N), the acid system (aqueous ethanolic HCl and aqueous formic acid (formic acid: 10–70% (v)) and the alginate viscosity (12 cP, 1% in water and ≥2000 cP, 2% in water, 25 °C). The variation of the warm-up period required the batch size to be upscaled by up to 6 times to be able to take samples from the same batch at different times.

2.2.3. TBA-Alg synthesis

H-Alg (1 g, 5.7 mmol) was dispersed in 200 mL Milli-Q water, then carefully adjusted to pH 8 ± 1 using TBAOH solution (approximately 0.5 M), before being immediately frozen to avoid any carbon dioxide absorption. Finally it was lyophilized and kept in a desiccator over a drying agent until further

use. FTIR (casted film): υ(OH) 3685–3041 cm^{-1} (with maxima at: 3501 cm^{-1}, 3383 cm^{-1} and 3200 cm^{-1}), υ(CH) 2961 cm^{-1}, 2937 cm^{-1} and 2879 cm^{-1}, $\upsilon_{as}(COO^-)$ 1610 cm^{-1}, $\delta(CH_2$ and $CH_3)$ 1485 cm^{-1} and 1468 cm^{-1}, $\delta_s(CH_3)$ 1385, (C—O) 1317 cm^{-1} and 1285 cm^{-1}, (C—O—C) 1173 cm^{-1}, 1148 cm^{-1} and 1101 cm^{-1}, (C—OH) 1036 cm^{-1} and (sat COOH) 1744 cm^{-1} (very low intensity) for acid traces if present. Elemental analysis: C 61.9%, H 10.3%, N 3.3%, composition: Na-Alg: 6%, H-Alg: 2%, TBA-Alg: 92% (DS_{TBA}: 0.92), yield: 2.39 g (94%).

2.3. Methods

2.3.1. CHN analyses

The samples were dried at 70 °C and the analyses were performed on a True Spec instrument from the Leco Corporation. EDTA was used for calibration. The furnace and afterburner temperatures were set to 950 °C and 850 °C.

2.3.2. Titration

The samples were dried in a high vacuum prior to analysis. Afterwards, three solutions, dispersions of each sample (0.12 g in 25 mL water), respectively, were prepared and titrated with 0.05 N titrisol sodium hydroxide solutions using a Titro Line Alpha instrument from Schott AG (Mainz, Germany) to determine the DS_H. The degree of substitution with sodium ions (DS_{Na}) was deduced from the DS_H (as $DS_{Na} + DS_H = 1$).

2.3.3. FTIR

256 spectra were recorded for each sample with a Vertex 70 FTIR from Bruker (Billerica, MA, USA) in a wavenumber range between 4000 and 400 cm^{-1} with a resolution of 4 cm^{-1} in transmission mode, using a sample holder with an aperture diameter of 5 mm. KBr pellets were prepared for Na-Alg and H-Alg samples and films were cast for TBA-Alg samples. Humidity and carbon dioxide compensation, rubber band base line correction and a normalization of the spectra were done before signal evaluation. Signal assignment of FTIR-averaged spectra was done according to Hesse, Meier, and Zeh, (2008). The complete separation of the FTIR bands of the carboxylic acid and carboxylate groups (Na-Alg and TBA-Alg) at 1744 cm^{-1} and 1610 cm^{-1} in the FTIR spectra of TBA-Alg (Fig. 2) enabled its quantitative determination. Despite the perfect overlap of the carboxylate bands of TBA-Alg and Na-Alg at 1610 cm^{-1}, it was possible to determine the DS_{TBA} using the DS_{Na} previously determined by the titration of alginic acid. The DS_{TBA} was then calculated according to the formula: $DS_{TBA} = 1 - DS_H$ (FTIR) $- DS_{Na}$ (titration). The M/G ratio was determined by FTIR spectroscopy by employing the two characteristic signal pairs of mannuronic and guluronic acid at 808 cm^{-1} (M)/787 cm^{-1} (G) and at 1030 cm^{-1} (M)/1080 cm^{-1} (G), respectively (Gómez-Ordóñez & Rupérez, 2011). This was done for all materials which were not characterized by the supplier.

2.3.4. pH measurements

were taken with a FiveEasyFE 20 pH-meter equipped with a pH-electrode type LE438 from Mettler Toledo (Greifensee, Switzerland). Calibrations were performed with Certipur buffers at pH 4.01 and 7.00.

2.3.5. Turbidity studies

Turbidity measurements were taken with the 2100 portable turbidimeter from Hach (Loveland, CO, USA). The instrument was calibrated with formazin standards (<0.1, 20, 100 and 800 NTU (nephelometric turbidity unit)). Blank measurements were taken to check the optical transparency of each selected solvent (they all had values of less than 0.1 NTU). The upper limit of the scale was 1000 NTU (poor solubility). 1.5% (w/v) TBA-Alg solutions were prepared by dissolving approximately 225 mg of TBA-Alg in 15 mL of the desired solvent (DMF, DMA, DMSO or acetonitrile) in the cell and agitating for between 15 min and 24 h to obtain a homogenous solution. Trapped air bubbles were removed in an ultra-sonic bath treatment which lasted about 15 min. Finally, turbidity values were normalized for a TBA-Alg concentration of 15 g/L. Additionally, a 7% (w/v) TBA-Alg/DMF solution was prepared to test the solubility of a TBA-Alg with a high DS_{TBA} at higher concentration.

2.3.6. Size exclusion chromatography–multi angle light scattering (SEC–MALS)

The chromatographic system consisted of a bio-inert pump (Agilent G5611A, Santa Clara, CA, USA) with an integrated online degasser, and a temperature controlled bio-inert autosampler (Agilent G5667A) (Agilent G1330B) kept at 22 °C. One 7.8 mm OH pack SEC column (Shodex SB-806 M, SHOWA DENKO EUROPE GmbH, Munich, Germany) was employed. A DAWN HELEOS II multi-angle light scattering detector (Wyatt Technology Corporation, Santa Barbara, CA, USA) and an Optilab T-rEX (Wyatt Technology Corporation) differential refractive index detector were connected sequentially to the SEC column. The aqueous mobile phase consisted of 100 mM TBACl with 0.02% sodium azide or 100 mM NaCl with 0.02% sodium azide, at a flow rate of 0.5 mL/min using isocratic mode at room temperature. All reagents for SEC were HPLC grade and the mobile phase was filtered through Durapore VVPP 0.1 μm membrane filters (Millipore). The injection volume was 20 μL. Data collection and processing were performed using the ASTRA software, Version 6.1.1.17 (Wyatt Technology Corporation). Zimm formalism with first order regression was used for the processing of light scattering data.

3. Results and discussion

3.1. Heterogeneous acidification of Na-Alg

The heterogeneous reaction of Na-Alg under standard conditions resulted in a DS_H in the range of 0.80 to 0.86 for 7 samples with an average of 0.83 ± 0.02. Changing the duration of the warm-up period showed that the reaction equilibrium was reached after 5 h (6 h total reaction time), because no further DS_H increase was observed. Temperature variations between 0 and 50 °C for the first hour followed by equilibration to room temperature did not change the DS_H. At 50 °C, however, the reaction solution became slightly yellow. This was interpreted as a result of polymer degradation.

DS_H values of up to 0.95 were obtained by increasing the water ratio in the solvent system up to the total ethanol exchange (Fig. 3). The influence of water on the DS_H was interpreted as a result of the changing capacity of the acidic solvent system to dissolve NaCl, the by-product of the reaction. NaCl dissolves well in water (36.0 g/100 mL = 6.153 mol/L (Lide & Haynes, 2009)) but only slightly in ethanol (Lide & Haynes, 2009). Therefore, additional water in the acidic solvent system improved the ability of NaCl to dissolve, which resulted in a better removal of NaCl from the alginate particles. This shifted the reaction equilibrium to the product side and materials with a higher DS_H were synthesized. In contrast, the reduction of the water content in the solvent system finally resulted in materials with a poor DS_H, because the dissolving capacity of the solvent system for the by-product was diminished. Drawbacks of using large water ratios were the occurring agglomeration of the alginate particles and the poor reaction yields.

The optimization of the reaction conditions by decreasing the Na-Alg concentration to make more water available for the removal of the by-product was not successful. The reduction to 0.5% (w) Na-Alg in the dispersion led to H-Alg with a $DS_H = 0.83$, which was in the range of the standard procedure.

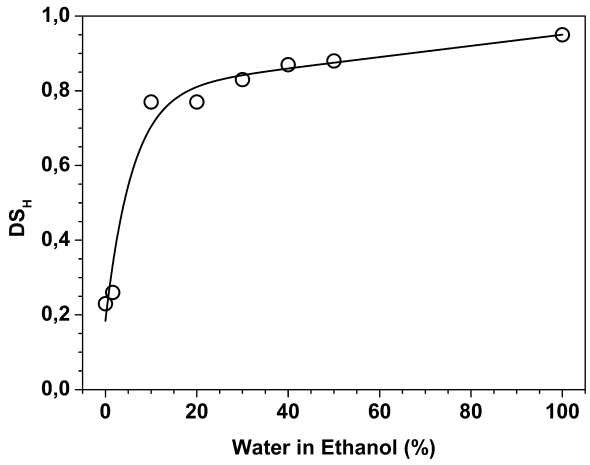

Fig. 3. Degree of acidification (DS_H) evolution depending on the water ratio in the 0.6 N aqueous ethanolic HCl. The fitting was found to follow the formula: $y = a + bx + c\rho^x$ with $a = 0.80$, $b = 0.0015$, $c = -0.61$ and $\rho = 0.84$.

Increasing the acid concentration is in general a good approach for optimizing such reactions. However, the effect of this was expected to be limited. Although increasing acid concentrations certainly increases pressure on the equilibrium towards the product side, the equilibrium will remain almost where it is, as long as the removal of the by-product is not improved. The results obtained (Fig. 4) confirmed our understanding and even the largest DS_H was still in the range of the standard deviation of the DS_H of the standard conditions.

Further reaction improvements required the use of an acid yielding an improved solubility of the by-product (sodium salt), such as formic acid (Table 1). Sodium formate, the new by-product, is more water soluble (94.9 g/100 mL = 13.95 mol/L (Lide & Haynes, 2009)) than NaCl, and also dissolves well in a 70% (w) formic acid solution (40.51 g/100 mL = 5.96 mol/L, (Elöd & Tremmel, 1927)). A critical point, however, was the small difference of the pK_a values of formic acid (3.75 (Lide & Haynes, 2009)) and H-Algs (3.42–4.36 (Haug, 1961)), which may not allow sufficient acidification. An excess of formic acid (26.5–186.7 mmol formic acid to 1 mmol Na-Alg) was used to overcome this obstacle. Varying the acid strength of formic acid, which in this case also corresponded to the variation of the available water, showed a similar behavior as observed for aqueous ethanolic HCl (Fig. 4). The larger formic acid concentration necessary for acidification and the slower increase of DS_H with increasing acid concentration compared to aqueous ethanolic HCl correlated well with the different nature of both acids. However, finally much higher DS_H values were achieved with the weaker formic acid, due to the better by-product removal. The highest DS_H values were even higher than that of the H-Alg commercially available (Table 1). In summary, it can be said that the acid strength determines the gradient of the DS_H increase, but the solubility of the by-product defines the achievable DS_H. This finding was not altered by using Na-Algs of medium viscosity (Table 1) or by changing the duration of the warm-up period as described for aqueous ethanolic HCl.

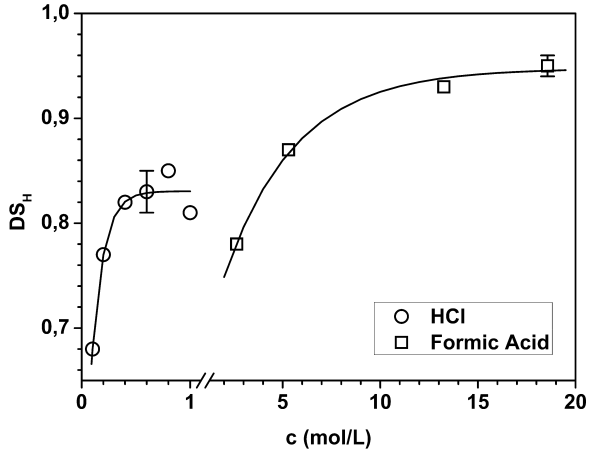

Fig. 4. Degree of acidification (DS_H) values depending on the hydrochloric and formic acid concentration and their fitting. Error bars ($n = 7$ (HCl) and $n = 6$ (formic acid)) are plotted for the values of 0.6 mol/L HCl and for 18.6 mol/L formic acid (70% (v)). The asymptotic fitting of HCl and formic acid followed the equation: $y = a - bc^x$ with $a = 0.83$, $b = 0.37$ and $c = 1.13 \times 10^{-4}$ for HCl and with $a = 0.95$, $b = 0.34$ and $c = 0.76$ for formic acid.

3.2. TBA-Alg

3.2.1. Synthesis

The TBA-Alg synthesis was not modified because the reaction of H-Alg and TBAOH was stopped at a pH between 7 and 9, which corresponded to a quantitative conversion of H-Alg. The upper pH limit during the TBA-Alg synthesis kept the potential substitution of Na by TBA ions within a quantitative insignificant range, as well as keeping the risk of polymer degradation low. The FTIR spectra, however, occasionally contained a very small signal of carboxylic acid residues at 1740 cm^{-1}. These small amounts of H-Alg most probably resulted from the carbon dioxide absorption of the solution from the air while freezing for lyophilization.

3.2.2. Molecular weight analysis

This was carried out in order to clarify the impact of the acid type on molecular mass and to reveal the relation between molecular mass and the DS_H. Therefore, aqueous TBACl solutions were used rather than aqueous NaCl solutions as the mobile phase for the analyses of the original Na-Alg and selected TBA-Algs, in order to avoid the formation of any mixed alginate salt. All TBA-Algs investigated had a lower molecular weight and slightly lower PD value, independent of their DS_H, in comparison to the original Na-Alg. The lowest molecular weight was found for TBA-Alg made from Na-Alg via acidification with aqueous ethanolic HCl, in which a reduced Na-Alg concentration (type 1) was used (Table 1). The highest molecular weights were found for TBA-Algs made from Na-Alg under the same reaction conditions but using a Na-Alg concentration of 2% (w) (type 2). This Na-Alg concentration difference corresponded to a difference in the amount of water and acid available per alginate unit in the reaction. Thus, a higher amount of Na-Alg was dissolved and more polymer degradation took place in the case of type 1 in comparison to type 2. Further, it was concluded that H-Alg precipitates faster with an increasing concentration of Na-Alg due to decreasing solubility, which led to a higher molecular weight in the case of type 2 TBA-Algs. The acidification was not improved because the solubility of the by-product was not sufficiently improved due to the availability of slightly larger amounts of water. The molecular weight of TBA-Alg made from Na-Alg via acidification with formic acid (type 3) was a little lower than the molecular weight of type 2, but much higher than that of type 1. The type 2 and 3 TBA-Algs for the molecular weight analysis were made from H-Algs, which were received in the experiments investigating the variation in the duration of the warm-up period. Interestingly, in each series, the polymer degradation was not time-dependent. The degradation observed was the same for the samples drawn after 30 min as for those drawn after a few hours. The final pH value (7 to 9) of the aqueous solution at the end of the TBA-Alg synthesis did not have an effect on the molecular weight. Furthermore, no relationship was identified between polymer degradation and the final DS_H and DS_{TBA}. Based on our results, the Na-Alg concentration and the acid system of the heterogeneous acidification were

Table 1
Overview of the relation between starting materials, conditions of the heterogeneous acidification of sodium alginate (Na-Alg) and tetrabutylammonium alginate (TBA-Alg) composition, solubility and molecular weight. Values are given as a mean ± standard deviation of individual values (n = 2–3 replicates).

Starting material	Type	H-Alg reaction			TBA-Alg Reaction			Molecular weight					Turbidity				
		Acid	$c_{Na\text{-}Alg}$ % (w)	t (h)	DS_H	pH	DS_{TBA}	Mn (kDa)	Mw (kDa)	Mz (kDa)	PD	Salt of mobile phase	$c_{TBA\text{-}Alg}$ (% (w))	DMF[c] (NTU)	DMA[c] (NTU)	DMSO[c] (NTU)	Acetonitrile[c] (NTU)
Low viscosity Na-Alg[a]	–	–	–	–	–	–	–	30.1 ± 0.5	87.4 ± 0.5	301.7 ± 4.4	2.90 ± 0.06	NaCl	–	–	–	–	–
Commercial H-Alg	4	–	–	–	0.89	9	0.84	–	–	–	–	–	1.5	30	–	–	–
Low viscosity Na-Alg	1	0.6 N aq. eth. HCl (30% water)	0.5	14	0.86[d]	9	~0.86[e]	23.4 ± 0.6	56.0 ± 0.4	190.7 ± 2.8	2.40 ± 0.05	TBACl	1.5	21	–	–	–
Low viscosity Na-Alg	1	0.6 N aq. eth. HCl (30% water)	0.5	14	0.83	8	0.81	–	–	–	–	–	1.5	5	–	–	–
Low viscosity Na-Alg	2	0.6 N aq. eth. HCl (30% water)	2.0	0.17	0.66	9	0.64	34.4 ± 0.1	80.4 ± 0.2	216.9 ± 2.9	2.34 ± 0.01	TBACl	1.5	861	828	216	≥1000
Low viscosity Na-Alg	2	0.6 N aq. eth. HCl (30% water)	2.0	14	0.81	9	0.78	–	–	–	–	–	1.5	167	–	–	–
Low viscosity Na-Alg	2	0.6 N aq. eth. HCl (30% water)	2.0	~20	0.85	7	0.81	34.5 ± 0.2	81.9 ± 2.2	233.5 ± 10.7	2.40 ± 0.10	TBACl	1.5	297	435	31	≥1000
Low viscosity Na-Alg	2	0.6 N aq. eth. HCl (30% water)	2.0	~20	0.85	9	0.81	34.2 ± 0.6	80.1 ± 0.0	225,4 ± 4.2	2.35 ± 0.03	TBACl	1.5	305	306	36	≥1000
Low viscosity Na-Alg	3	HCOOH (70%)	2.0	0.17	0.93	9	0.92	34.3 ± 1.1	75.6 ± 0.6	208.0 ± 2.5	2.21 ± 0.05	TBACl	1.5	3	6	1	32
Low viscosity Na-Alg	3	HCOOH (70%)	2.0	6	0.96	9	0.95	34.8 ± 0.7	77.8 ± 1.3	220.9 ± 4.0	2.24 ± 0.02	TBACl	1.5	3	3	1	18
Low viscosity Na-Alg	3	HCOOH (70%)	2.0	6	0.94	9	0.92	–	–	–	–	–	1.5	3	–	–	–
Low viscosity Na-Alg	3	HCOOH (70%)	2.0	6	0.94	9	0.92	–	–	–	–	–	7	9	–	–	–
Low viscosity Na-Alg	3	HCOOH (70%)	2.0	14	0.95	8	0.92	–	–	–	–	–	1.5	3	–	–	–
Low viscosity Na-Alg	6	HCOOH (50%)	2.0	14	0.93	9	0.91	–	–	–	–	–	1.5	7	–	–	–
Low viscosity Na-Alg	6	HCOOH (20%)	2.0	14	0.87	8	0.86	–	–	–	–	–	1.5	186	–	–	≥1000
Low viscosity Na-Alg	6	HCOOH (10%)	2.0	14	0.78	9	0.77	–	–	–	–	–	1.5	529	–	–	≥1000
Medium viscosity Na-Alg[b]	5	HCOOH (70%)	2.0	14	0.96	9	0.92	–	–	–	–	–	1.5	11	–	–	–

[a] Low viscosity sodium alginate: 12 cP, 1% in water, 25 °C.
[b] Medium viscosity sodium alginate: ≥2000 cP, 2% in water, 25 °C.
[c] Relative permittivity's: DMF: ε = 38.25; DMA: ε = 38.85; DMSO: ε = 47.24 and acetonitrile: ε = 36.64 (Lide & Haynes, 2009).
[d] Value was determined via IR spectroscopy.
[e] DS_H value was taken for the DS_{TBA} because no material was left for the direct DS_{TBA} determination.

Fig. 5. Laser beam passing through a 1.5% (w) tetrabutylammonium alginate (TBA-Alg) dimethylformamide (DMF) solution with a turbidity of 4 NTU.

identified as the main controlling factors of the polymer degradation. Therefore, the acidification of Na-Alg with aqueous formic acid is a good method for alginate acidification in a laboratory. Its advantages are the very high DS_H, which can be even higher than the DS_H of the H-Alg commercially available, and the relatively low polymer degradation.

3.2.3. Solubility

At very best, low turbidity solutions (Fig. 5) were obtained that were highly fluid and, on visual inspection, clear, as required for potable water (< 1 NTU (International Organization for Standardization, 2000)). At worst, solutions were inhomogeneous, jelly-like and very turbid. Highly soluble materials always dissolved in less than an hour. Poorly soluble materials did not give homogeneous solutions, even after 24 h.

The relative permittivity of the solvent, the DS_{TBA} and the molecular weight of TBA-Alg were identified as the main controlling parameters of the solubility on the basis of turbidity measurements (Table 1). The solubility capacity of the nearly dry solvents (water content ≤0.1%) almost followed the order of their relative permittivity. Thus, in our study, DMSO was the best solvent, DMF and DMA almost equal and acetonitrile the worst solvent (Fig. 6). Inhomogeneous solutions are expected for solvents with a lower relative permittivity than acetonitrile. The turbidity data of the best acetonitrile solutions were slightly higher than the other solvents. Lower quality TBA-Algs (DS_{TBA} < 0.9) that had been dissolved in acetonitrile gave solutions almost immediately, containing gel structures with turbidity values above the measurement range of the instrument. These findings correspond well to the formulated assumption that the solubility of TBA-Alg strongly depends on the ratio of its counter ions TBA^+:H^+:Na^+, because the only soluble parts of TBA-Alg are the TBA-Alg moieties. Na-Alg and H-Alg moieties are insoluble. Single or very small Na-Alg or H-Alg moieties may be kept in the solution by the surrounding TBA-Alg. Larger moieties will increase the turbidity due to precipitation. According to the present data, a DS_{TBA} of at least 0.9 was necessary to obtain a homogeneous solution, and this was independent of the solvent. Such a high DS_{TBA} value even allowed the preparation of homogeneous solutions with a much higher TBA-Alg concentration. The turbidity of the more concentrated TBA-Alg solution increased only very little and was still below that of one of the TBA-Alg solutions that was prepared from commercially available H-Alg. The DS_{TBA} of TBA-Alg made from commercially available H-Alg was just below the threshold and gave solutions with a slightly higher turbidity. It must be stated that these solutions still appeared to be homogeneous on visual inspection. Similar turbidity values were also observed for the type 1 TBA-Algs (Na-Alg: c = 0.5% (w)), which were characterized by lower molecular mass and a DS_{TBA} clearly below 0.9. This led to the conclusion that the solubility threshold depended on the molecular weight of the TBA-Alg. In other words, the influence of Na-Alg or H-Alg moieties on the solubility declined with decreasing molecular weight.

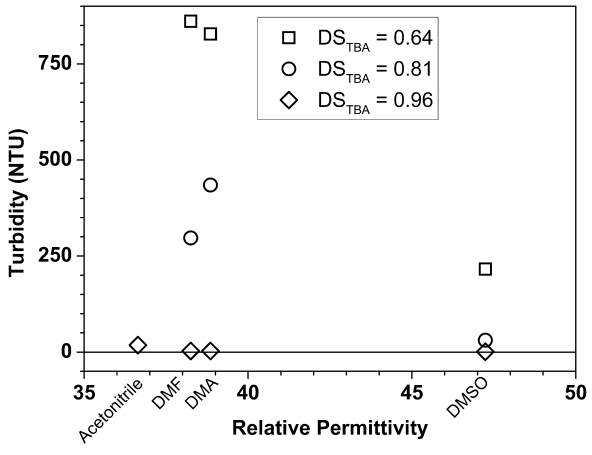

Fig. 6. Turbidity of TBA-Alg solutions as a function of the relative permittivity of the solvent for three TBA-Algs with different DS_{TBA}s. The turbidity values of the acetonitrile solutions from the TBA-Algs with a DS_{TBA} of 0.64 and 0.81 were larger than 1000 NTU, which is the upper measurement limit of the turbidimeter, and were therefore not plotted. The concentration of all TBA-Alg solutions was 1.5% (w).

Type 3-like TBA-Algs made from sodium alginates with an M/G ratio between 0.37/0.63 and 61/39 gave DMF solutions with turbidity values from 2 to 7 NTU, which is within the observed range of 3—9 NTU for type 3 TBA-Algs. Solutions of TBA-Algs made from medium viscosity Na-Alg (type 5) were slightly more turbid (11 NTU). From this observation the conclusion was drawn that the M/G ratio and the viscosity were not important for the solubility of TBA-Algs with a high DS_{TBA} in a polar aprotic solvent, because both saccharides are highly substituted. This, however, will most probably change for TBA-Algs with a lower DS_{TBA}.

4. Conclusion

The solubility of TBA-Alg impacts the success and the selectivity of chemical reactions such as esterification or amidation. It was proven that its solubility in polar aprotic organic solvents depended on the relative permittivity of the solvent, the DS_{TBA} and the molecular weight of the TBA-Alg. A DS_{TBA} larger than 0.9 was necessary to dissolve TBA-Algs with relative high molecular weights homogeneously in polar aprotic organic solvents with a relative permittivity of at least that of acetonitrile. For TBA-Alg with lower molecular weights, a DS_{TBA} of 0.83 was already sufficient to receive a homogenous solution almost as good as a solution resulting from TBA-Alg with high DSTBA values (<0.9). Synthesizing such TBA-Alg with DS_{TBA}'s larger than 0.9 from Na-Alg via heterogeneous acidification required aqueous formic acid (70% (v)), followed by neutralization with TBAOH. Using formic acid instead of hydrochloric acid considerably improved the removal of the by-product (sodium salt) from the alginate particle and enabled very high DS_H's and DS_{TBA}'s, which were even higher than those of commercially available H-Alg. Another advantage of aqueous formic acid over aqueous ethanolic HCl was the comparably low polymer degradation. Therefore, the heterogeneous acidification of Na-Alg with aqueous formic acid is an interesting synthesis for H-Alg with a low Na-Alg residue content and relatively high molecular weight.

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