ELSEVIER

PII: S0144-8617(96)00060-4

Carbohydrate Polymers **30** (1996) 25–30
Published by Elsevier Science Ltd
Printed in Great Britain.
0144-8617/96/$15.00 + .00

Preparation of a weakly basic ion exchanger by crosslinking starch with epichlorohydrin in the presence of NH_4OH[1]

Ivan Šimkovic[a], **Joseph A. Laszlo***[b] & **Arthur R. Thompson**[c]

[a]*Institute of Chemistry, Slovak Academy of Sciences, 84238 Bratislava, Slovak Republic*
[b]*USDA-ARS, National Center for Agricultural Utilization Research, Biomaterials Processing Research, 1815 N. University St., Peoria, Illinois 61064, USA*
[c]*USDA-ARS, National Center for Agricultural Utilization Research, Analytical Chemistry Support Unit, 1815 N. University St., Peoria, Illinois 61064 USA*

(Received 2 November 1995; revised version received 15 April 1996; accepted 17 April 1996)

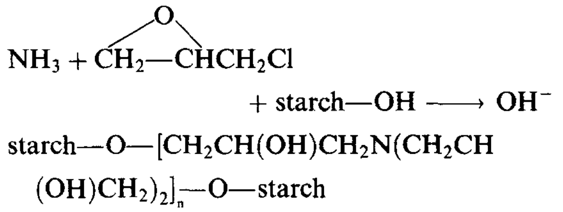
Crosslinking of starch with epichlorohydrin in the presence of NH_4OH, NaOH and water was studied. The reaction was optimized and the products analysed by solid-state ^{13}C-NMR in the presence of water, acid-base titration and dye-binding measurements. The presence of different types of hydroxypropyl groups was confirmed by NMR, but their linkage to polysaccharide could not be confirmed because of their minor abundance relative to all other CH_2 groups, and because the motion of the sidechains is much greater than the starch component, which remains fairly constant despite the presence of water. Yield of crosslinked, water-insoluble product is increased as a greater amount of NH_4OH and a lower amount of water is used in the reaction. Product yield using water-insoluble starch is higher than with a water-soluble starch. Closed-vessel (pressurized) reaction conditions increases product exchange capacity. Potentiometric titration of products in Cl^--form or in non-ionized form indicated smaller exchange capacity values than expected from product nitrogen contents, reflecting the difficulty in fully protonating the products. Dye binding by the crosslinked-starch exchangers is pH-dependent, with an increased quantity of dye bound at lower pH, which is consistent with the weak-amine nature of the products. Hydrolyzed Remazol Brilliant Red F3B, a trivalent reactive dye, binds less effectively under acidic conditions, on a molar basis, to the starch-based exchangers than does Alizarin Red S, a monovalent mordant dye. Dye-binding capacities are somewhat lower than the total ion-exchange capacities determined by potentiometric titration, suggesting that a portion of the exchange sites are unavailable to the dyes. Published by Elsevier Science Ltd

INTRODUCTION

The industrial uses of chemically modified starches are numerous. One of the potential applications arises in biodegradable anion exchangers suitable for decolorization of textile wastewaters (Laszlo, 1994). For many applications, starch needs to be crosslinked to prevent or retard degradation and to improve certain physical properties. Crosslinking starch with epichlorohydrin (E) is the most common method used in polysaccharide chemistry (Kuniak & Marchessault, 1972). Reacting E with NH_4OH is a convenient and inexpensive way of introducing weakly basic anion-exchange groups into polymer substrates (Vorchheimer, 1988). In this way, water-insoluble polyamines having predominantly tertiary amine groups are produced. The reaction of E with NH_4OH has not been applied previously for the crosslinking of starch. Under appropriate reaction conditions, a poly(hydroxypropylamine)-modified starch product may be produced, as suggested by the reaction scheme:

---

[1] Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by USDA implies no approval of the product to the exclusion of others that may also be suitable.
*To whom correspondence should be addressed.



$NH_3 + CH_2—CHCH_2Cl + starch—OH \longrightarrow OH^-$
$starch—O—[CH_2CH(OH)CH_2N(CH_2CH(OH)CH_2)_2]_n—O—starch$

The resulting material may be more biodegradable, more accessible to counter ions, and substantially less expensive than synthetic, petroleum-based, anion-exchange resins. The purpose of the present study was to prepare weakly basic ion exchangers by crosslinking starch with E in presence of NH_4OH, and to characterize the exchangers by solid-state NMR, acid-base titrations and dye-binding tests.

EXPERIMENTAL

Materials

Native, water-insoluble wheat starch (WS, Sigma, S-5127) and partially-hydrolyzed, water-soluble potato starch (WSS, ZŠ, Dolná Krupá, Slovakia; 19,060 Da (determined osmometrically); $[\alpha]^{20} = +154.0°$ (Cl; H_2O)) were used without further treatment. All chemicals used were reagent grade. The dyes used were: Alizarin Red S Monohydrate (Aldrich) and hydrolysed Remazol Brilliant Red F3B (C.I. Reactive Red 180, Hoechst Celanese Corp.) prepared as described previously (Laszlo, 1995).

Methods

^{13}C solid-state NMR spectra were obtained at 295 K on a Bruker MSL 300 instrument using cross polarization (CP) times (C–H contact time) of 0.5 ms, sweep width of 50,000 Hertz, with/without rotation rate of 3500 Hz and pulse width of 0.007 ms. The high power decoupled (Bloch decay) experiment was run with a pulse width of 0.004 ms, rotation rate of 3000 Hz and recycle time of 0.5 s. Adequate signal-to-noise ratios were obtained after 1,500 transients for CP and 5,000 transients for Bloch decay experiments on dry samples and 50,000 to 100,000 transitions for hydrated samples. The accuracy of the chemical shifts is dependent upon the peaks width and shape. Therefore, the reported solid-state NMR resonance chemical shift values are accurate approximately to ±0.3 ppm. Water was added to the sample directly and allowed to equilibrate for a day in order to allow the water to evenly disperse throughout the sample.

Proton and ^{13}C-NMR spectroscopy measurements on samples dissolved or dispersed in D_2O were run on a Bruker ARX spectrometer equipped with a 5-mm $^1H/^{13}C$ dual probe, at 400.13 MHz (^1H) and 100.61 MHz (^{13}C).

Potentiometric titrations were conducted using a Metrohm 702 SM Titrino (Herisau, Switzerland) titrator with samples in aqueous solutions under constant nitrogen flow. Total exchange capacities (Q values) were determined by the residual base method in which 0.1 to 0.2 g of sample, in H^+-form, is mixed with 10 ml of 0.0975 N NaOH (i.e., an excess of base) and rotated overnight. Samples were centrifuged (3,000 g, 15 min) and, after several hours of equilibrium, the sample packed volume (PV, ml/g dry weight of ion exchanger) was recorded. The whole mixture of each sample (solid portion and solution) was washed with water into the flask and diluted to 150 ml and titrated under vigorous stirring until the endpoint was reached by using 1 min intervals and 0.1 ml titrant additions. The exchange capacity (Q) was then calculated according to formula: $Q = (V_1 * f_{NaOH} - V_2 * f_{HCl})/w$, where V_1, V_2 and f_{NaOH}, f_{HCl} are the volumes and concentrations of NaOH and HCl, respectively, and w is the weight of dry ion exchanger. Direct titration of samples in Cl^- form or free-base form (obtained by cycling with 1% NaOH) was performed using equilibrium intervals of 1 min to 1 h, with 0.05 or 0.1 ml individually added titrant (0.0974 N) volumes.

Dye-binding tests were performed as follows: sample (0.015 or 0.05 g) was mixed with 100 ml of 1 mM Alizarin Red S (ARS) or 'F3B' and the solution pH was adjusted from 3 to 11 with solid NaOH or 2 M HCl. Sets of nine samples prepared this way were covered and stirred overnight, after which the pH and absorbance was determined for each sample. For ARS and F3B, the absorbance was determined at 594 nm and 540 nm, respectively after 1:50 dilution into 0.1 M Na_2CO_3. The concentration of dye in solution after equilibration was used to calculate the amount of dye bound to the exchanger.

Preparation of samples

Epichlorohydrin (10 g, 0.108 mol) was mixed with NH_4OH (0.074 mol) and 17.5% NaOH solution (10 g, 0.044 mol; in 0.781 mol of water), in a round-bottom flask connected to a condenser, was stirred (about 1 h) until the mixture became so viscous that it could not be stirred with a magnetic stirrer. After a total reaction time of 24 h at room temperature, the poly(hydroxypropylamine) (PHPA) product was washed with water until a neutral reaction of the eluate was obtained on pH paper. The product was then lyophilized (yield, 4.4134 g; N content, 10.72%).

WSS (1.62 g, 0.01 mol) was mixed with 16.2 g of 17.5% NaOH (0.071 mol) and activated at 28°C for 1 h. E (0.175 mol) and 28% NH_4OH (0.13 mol in the presence of 0.72 mol of water) were added and stirred at 28°C until the whole mixture become solid (within about 20 min), after which the reaction was continued, unstrirred, for 24 h total. The product was homogenized, washed until neutral washings were obtained and lyophilized (Table 1, sample 1). The yields were calculated on the dry basis to the weight of starch used. Alternatively, WS or WSS (1.62 g) were mixed with NH_4OH, E, and solid NaOH in ratios listed in Table 1. The mixture become solid after 2 h; after 24 h the reaction was stopped by diluting with water, adjusting to pH < 2, and washing with water until the eluate was neutral on pH paper. The product was washed with acetone, homogenized, and dried at 22°C in vacuum to constant weight. For some experiments, a sealed plastic reactor (120 ml volume) with a pressure releasing adapter (up to 839 kPa) was employed. WS or WSS (1.62 g) was mixed with NH_4OH, E, and solid NaOH, enclosed in the reactor, and maintained at 22°C for 24 h, then treated as described above. Molar ratios of reactants used are listed in Table 1.

RESULTS AND DISCUSSION

Poly(hydroxypropylamine) polymer (PHPA), which contained no starch, was examined by ^{13}C solid-state NMR to identify characteristic signals. With dry PHPA, signals were so broad that individual peaks could not be recognized. With the addition of water, as demonstrated previously (Morgan *et al.*, 1994), individual carbon signals could be distinguished. When CP was applied (45% water), the predominant signals were at 74.0, 72.2, 69.0, 64.5, 62.7, 58.6, 53.8 and 51.4 ppm. These chemical shifts are in the region expected for O—CH_2—, N—CH_2— and O—CH(OH)— groups found in polyamine-type resins (Vorchheimer, 1988). When the Bloch decay experiment was applied to PHPA, the signals observed with CP were split into many signals, but three groups of signals could still be distinguished (77.0 to 66.3 ppm, 64.6 to 60.0 ppm and 58.3 to 45.3 ppm; Fig. 1A).

The spectra of CP and Bloch decay experiments with unmodified WS in the absence of water were identical, with signals at 101.0 (C-1), 81.1 (C-4), 72.6 (C-2,3,5) and 62.4 (C-6) ppm. After addition of water (40%), the signals did not sharpen or shift, and no additional peaks were detected by CP or Bloch decay. Analysis in the presence of 50% of water by CP of WSS, crosslinked with E, but without NH_4OH present (Table 1, sample 12), produced sharp and intense signals at 72.9, 71.4, 70.0, 63.6, 61.7, 52.3 and 45.6 ppm. In addition, a lower intensity signal at 100.5 ppm was observed. The Bloch decay spectrum under the same conditions (sample 12, 50% of water) had signals at 100.3, 72.9, 71.3, 69.8, 63.5 and 61.3 ppm (Fig. 1B). We assume that these signals are related to O–CH_2 and CH(OH) groups of hydroxypropyl side-chains as well as to C-1 (100.3 ppm) and C-6 (63.5 and 61.3 ppm). These last two signals may arise from low molecular weight starch components that were able to change their mobility in the presence of water.

Analysis of E/NH_4OH crosslinked starch (Table 1, sample 17, 50% water-content) by the Bloch decay technique gave signals at 99.4, 76.6, 73.0, 71.1, 65.2, 62.4, 60.3, 57.3, 50.1 and 46.4 ppm (Fig. 1C). The signals at 99.4 ppm (C-1) and 62.4 ppm (C-6) are again

Table 1. Quantities of reactants and the properties of products obtained from crosslinking starch.

Sample No.	Moles of reactants[a]				Substrate	Yield (%)	PV (ml/g)	N (%)
	E	NH_4OH	NaOH	H_2O				
1	0.175	0.130	0.071	1.71	WSS	616[b]	6.6	10.19
2	0.120	0.120	0.120	2.08	WSS	333[b]	7.2	5.80
3	0.180	0.060	0.060	0.35	WSS	358[b]	–	6.09
4	0.180	0.060	0.100	0.35	WSS	344[b]	–	4.21
5	0.060	0.060	0.060	0.35	WSS	27[b]	–	0
6	0.060	0.060	0.060	2.78	WS	109[c]	–	2.09
7	0.090	0.090	0.090	2.78	WS	116[c]	–	2.65
8	0.120	0.120	0.120	2.78	WS	101[c]	–	3.10
9	0.030	0.120	0.030	2.78	WS	96[c]	–	0.43
10	0.120	0.120	0.120	2.78	WSS	45[c]	–	3.70
11	0.120	0	0.120	2.78	WSS	160[d]	–	0
12	0.060	0	0.060	0.35	WSS	134[b]	–	0
13	0.060	0.060	0.060	0.30	WS	237[b]	3.7	4.63
14	0.120	0.120	0.120	0.60	WS	277[b]	6.5	4.84
15	0.200	0.200	0.200	1.00	WS	464[b]	2.7	3.36
16	0.060	0.060	0.060	0.30	WS	215[b,e]	4.6	5.55
17	0.030	0.010	0.050	0.05	WS	138[b,e]	6.7	2.12

[a] Amount of starch used was 0.01 mol, anhydroglucose-content basis.
[b] water-insoluble ion exchanger.
[c] mixture of water-soluble and insoluble product.
[d] water-soluble product.
[e] run in a plastic reactor; for more details see experimental section.

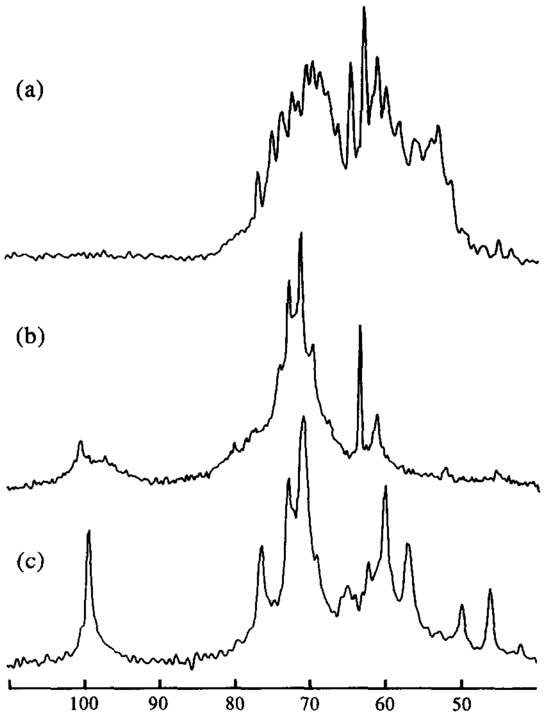


Fig. 1. Solid-state NMR spectra (Bloch decay decoupled experiments): (a) of PHPA copolymer (45% water-content); (b) starch crosslinked with epichlorohydrin; (c) starch crosslinked with epichlorohydrin in the presence of NH_4OH.

related to low molecular weight starch fragments or degradation products able to tumble in the presence of water. With nondecoupled Bloch decay, the signals at 99.4, 76.6, 73.0 and 71.1 ppm split into doublets (GH groups); the rest were overlapping triplets (CH_2 groups). By comparison with the spectra of PHPA and E-crosslinked starch, we conclude that the signal at 76.6 ppm belongs to the N–CH_2-group of polyamine side-chains because it was absent in E-crosslinked starch and shifted to 74.0 ppm in the spectra of PHPA. For a sample with higher polyamine content (Table 1, sample 14, 40% water-content), the Bloch decay spectrum contained many signals between 72.7 and 50.8 ppm, which indicate again the presence of CH_2-groups. Again, three separated groups of multiplets were present: the first at 72.7–69.3 ppm, second at 67.2–59.2 ppm and the third at 53.7–50.8 ppm. The signals of CH_2 groups of polyamine side-chains etherifically linked to polysaccharide could not be identified. Signals arising specifically from etherifically-linked CH_2 groups might be weak relative to all other hydroxypropyl CH_2 groups present because of their low abundance and mobility. The motion of the polyamine side-chains is probably much greater than that of the starch polymer main-chain, which remains relatively motionless despite the presence of water. Thus, the linkage of polyamine to WS could not be confirmed by solid-state NMR.

When a large excess of reactants was applied to WSS, a water-insoluble starch-polyamine copolymer was produced with a yield of 616% based on the weight of starch used in the reaction (Table 1, sample 1). The nitrogen content of this material (N = 10.19%) was close to the nitrogen percentage of the PHPA sample (N = 10.72%), indicating that the sample contained only a small amount of starch. With lower quantities of E and NH_4OH and slightly higher amounts of NaOH and water, the yield decreased but the polyamine component was still predominant (sample 2). At an E: NH_4OH ratio 3:1 and at a lower NaOH concentration than sample 2, higher product yields were obtained (samples 3 and 4). On the other hand, the yield and nitrogen content of insoluble product was very low when 0.06 molar quantities of E, NH_4OH and NaOH and same amount of water were reacted (sample 5). When equimolar quantities of NaOH, E and NH_4OH were used on WS (samples 6–8), the maximum nitrogen content of the product (a mixture of water-soluble and insoluble fractions) was 3.1% (sample 8) and the maximum yield was 116% (sample 7). The decrease in yield observed when using 0.12 mol of reactants (sample 8) indicates that there was degradation caused by NH_4OH. Degradation resulting from excess NH_4OH was also observed with sample 9, for which the ratio of E: NH_4OH was 1:4 and 0.03 mol of NaOH was applied. Yield decreased even more and the nitrogen content of the product was 0.43%. At the equimolar quantities of E, NH_4OH and NaOH on WSS, a water-soluble product was obtained (Table 1, sample 10) with a 45% yield and 3.70% nitrogen content.

The ^{13}C-NMR spectrum of WSS product (Table 1, sample 10, water-soluble fraction) in D_2O showed three anomeric signals (100.3, 100.2 and 98.7 ppm) and many signals between 73 and 47 ppm that were not present in the WSS spectrum. This indicates that all three hydroxyls of the glucopyranose unit were modified with hydroxypropyl groups. Treating WSS similarly, but without NH_4OH present (sample 11), produced a completely soluble material in high yield. The ^{13}C-NMR spectrum in D_2O of sample 11 gave dominant signals between 73 and 68 ppm, indicating hydroxypropyl polyether structures, while the polysaccharide signals were of minor intensity. This suggests that, at these starch concentrations (23–27 g/l) and a 7% NaOH concentration, most of the epichlorohydrin is consumed by side reactions resulting in water-soluble polysaccharide modified with poly(hydroxypropyl) side-chains. Such a product may be beneficial for subsequent modification of the formed exchanger material when the goal is the production of water-soluble products. By decreasing the amount of water in the mixture, even at 0.06 mol concentration of E and NaOH, water-insoluble product in good yield was obtained from water-soluble starting material (sample 12).

WS reacted under "dry" conditions, in which the water was introduced only as a part of the NH_4OH

solution (samples 13 and 14), produced water-insoluble material in greater yield and with higher nitrogen content than WS reacted in the presence of water (samples 6 and 8). Higher reactant amounts (0.2 mol; sample 15) further increased reaction yield (464%; 3.36% nitrogen content), yet the exchanger packed volume (PV) decreased, probably due to an increased quantity of hydroxypropyl bridges introduced in comparison to the experiment with 0.06 mol of added components (sample 13). These results indicate that reaction under low moisture (i.e. 'dry') conditions is preferable to highly hydrated reaction conditions.

WS modification in a sealed reactor was examined to determine whether a closed system (i.e. no loss of volatile reactants) would improve product yield. In a closed reactor, WS treated with 0.06 mol each of E, NaOH and NH_4OH (Table 1, sample 16) yielded slightly less product having a higher nitrogen and PV value, than a similarly constituted reaction run at atmospheric pressure (sample 13). (A higher PV indicates a lower density of crosslinking.) At lower concentrations of reactants, and a 3:1 E: NH_4OH reactants ratio (Table 1, sample 17), product yield and nitrogen content decreased in comparison to sample 16. The PV of 6.7 ml/g indicates that sample 17 was less well crosslinked than with the 1:1 E: NH_4OH ratio used to prepare sample 16. Thus, there is a marginal benefit to using a closed vessel for the crosslinking reaction.

Ion exchange capacity (Q) values determined by potentiometric titration using the residual-base method on samples in Cl^--form differed significantly from the values calculated from nitrogen content (Table 2). Titration of samples in acidic form with NaOH solution without pretreatment with an excess of NaOH gave lower Q values than those determined by elemental analysis, even when 1 h equilibrium intervals were used for the titration. In the presence of 1 M NaCl, no distinct endpoints were found. This observation is in agreement with results obtained on polyethylenimine (PEI) in the presence of salt (Jarvis & Wagner, 1995). There was only one titration endpoint for most of the samples when the starch content of the sample was greater than the content of the polyamine component. For sample 1 (Table 1), where the polyamine portion is predominant over starch, three titration endpoints were determined when the Cl^--form was titrated with NaOH solution. These endpoints may correspond to the ratio between quantities of tertiary, secondary and primary amine groups present in the material; for this sample the ratio was about 62:4:1. The titration of samples in free-base form (sample washed with 1% NaOH solution) with HCl gave even less distinct endpoints than samples in Cl^-- form titrated with NaOH. Also, the Q values determined in this way were lower than values calculated from nitrogen content. We conclude from these observations that simple potentiometric titration of the prepared starch-based exchangers is an unreliable measure of their total exchange capacity. This may result from some exchange sites having hindered access to ions.

Examination of dye binding by two samples (Table 1, samples 1 and 2) with different Q values, using F3B (a trivalent anionic dye) and ARS (a monovalent anionic dye), indicated increased dye-binding with lower solution pH (Fig. 2), behavior expected for weakly basic exchangers. The sample with higher Q value (sample 1, Table 1) bound both dyes more effectively than the exchanger with lower Q (sample 2). Dye binding by epichlorohydrin-alone crosslinked starch (Table 1, sample 11) was negligible over the whole pH range (data not shown). Similar to PEI (Horn, 1980), there seemed to be a point of zero charge on the samples close to pH 11 at which there was no dye binding (Fig. 2). The lack of dye binding at pH 11 confirms that there are no strong exchange sites (quaternary amonium groups) in

Table 2. Comparison of dye-binding and exchange capacities of selected samples.

Sample No.	Dye-binding capacity[a]		Exchange Capacity	
	ARS [mmol/g]	F3B [mmol/g]	$Q_{titr.}$[b] [mmol/g]	$Q_{calc.}$[c] [mmol/g]
1	4.2	1.4	6.0	6.2
2	3.8	1.3	3.8	4.1
13	2.9	0.8	3.4	3.3
14	2.9	1.0	2.5	3.5
15	3.2	1.1	2.6	2.4
17	1.5	0.3	2.0	1.5

[a] Determined at pH 3 after 20 h equilibration.
[b] $Q_{titr.}$ was determined by potentiometric titration.
[c] $Q_{calc.}$ was calculated from nitrogen content.

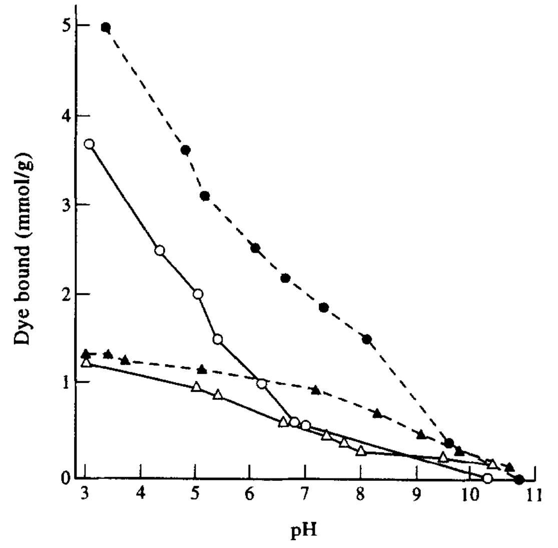


Fig. 2. Relation between dye-binding capacity and pH of weakly basic resins {Table 1, samples 1 (full symbols) and 2 (empty symbols)} with Alizarin Red S (circle) and hydrolysed Remazol Brilliant Red F3B (triangle).

the prepared materials. Dye-binding values (Table 2) of the samples examined were generally smaller than Q values determined by potentiometric titration or elemental analysis. This indicates that, like PEI (Horn, 1980), poly(hydroxypropylamine)starch is not fully protonated at low pH, which prevents full expression of its exchange capacity for anions.

Trivalent F3B bound less effectively, on a molar basis, than the smaller, monovalent dye (ARS) in the acidic pH region. However, when calculated on an equivalent basis, the amount of F3B bound to various samples (Table 2) was equal to, or slightly less than, the amount of ARS bound. This suggests that there is an approximate 1:1 stoichiometric relationship for the association of dye sulfonic acid groups and protonated nitrogen groups of poly(hydroxypropylamine)-starch. A similar relationship was observed with dyes, having one to three sulfonic acid groups, binding to chitosan (Maghami & Roberts, 1988). Therefore, dye binding to poly(hydroxypropylamine)-starch is not necessarily constrained by the size of the dye molecule; the amount of dye bound is determined by the number of positively charged exchange sites in the exchanger and number of anionic groups in the dye molecule.

CONCLUSIONS

Starch, preferably water-insoluble starch, crosslinked with E, NH_4OH, and NaOH in the presence of a limited amount of water, can be used to prepare weakly basic ion exchangers active in the acidic pH range. Such starch-based exchangers, because of their moderately high capacity and potentially low cost of manufacture, may be suitable for the treatment of various wastewaters. In particular, the decolorization of textile effluents containing anionic dyes using poly(hydroxypropylamine)-starch may be an economically-viable alternative to currently available technologies.

ACKNOWLEDGEMENTS

We thank to Mary M. Hallengren, Dr David Weisleder, Dr Jacob Lehrfeld, and Helena, Leštanská for their advice and assistance.

REFERENCES

Horn, D. (1980). Polyethylenimine–physico–chemical properties and applications, *Int. Symp. on Polymeric Amines and Ammonium Salts*, Pergamon Press, Oxford pp. 333–355.

Jarvis, N.V. & Wagner, J.M. (1995). Mechanistic study on ion binding to watersoluble polymers using potentiometry. *Talanta*, **422**, 219–226.

Kuniak, L. & Marchessault, R.H. (1972). Study of the crosslinking reaction between epichlorohydrin and starch. *Starch/Stärke*, **24**, 110–116.

Laszlo, J.A. (1994). Removing acid dyes from textile wastewater using biomass for decolorization. *Am. Dyestuff Reporter*, **83**, 17–21, 48.

Laszlo, J.A. (1995). Electrolyte effects on hydrolyzed reactive dye binding to quaternized cellulose. *Text. Chem. & Color.*, **274**, 25–27.

Maghami, G.G. & Roberts, G.A.F. (1988). Sudies on the adsorption of anionic dyes on chitosan. *Makromol. Chem.*, **189**, 2239–2243.

Morgan, K.R., Hemmingson, J.A., Furneaux & Stanley, R.A. (1994). A ^{13}C solid-state NMR study of ion-exchange resins derived from natural polysaccharides. *Carbohydr. Res.*, **262**, 185–194.

Vorchheimer, N. (1988). Polyamines and polyquaternary ammonium salts. *Encyclopedia of Polymer Science and Engineering. Wiley & Sons, New York, Vol.*, **11**, 489–507.