Pilot plant scale extraction of alginates from *Macrocystis pyrifera*
3. Precipitation, bleaching and conversion of calcium alginate to alginic acid

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Abstract

Three steps of the alginate production process were studied at pilot plant level. The effect of the amount of calcium chloride used during the precipitation was measured in terms of filtration time of the precipitated calcium alginate. Three different proportions of calcium chloride per gram of alginate were tested. The best proportion used was 2.2 parts of calcium chloride per one part of alginate, yielding a filtration rate of 97.9 L min^{-1} on a screen area of 1.32 m^2. The method of adding the solutions and the degree of mixing are discussed as other factors affecting the precipitation step. The effect of bleaching the calcium alginate with sodium hypochlorite (5%) was studied. Seven proportions, ranging from 0 to 0.77 mL of sodium hypochlorite per gram of sodium alginate were tested. The effect of hypochlorite was compared for alginates with three different viscosities. Using alginates with medium viscosity (300–500 mPa s), the best proportion was 0.4 mL hypochlorite per gram of alginate, yielding an alginate of light cream color with 20% less viscosity than the control. Alginates with lower viscosity showed a smaller loss of viscosity. The effect of pH during conversion of calcium alginate to alginic acid was determined using four combinations of pH, ranging from 2.2 to 1.6, in three acid washings. The extent of conversion was determined by measuring the percent reduction of the alginate viscosity (RV) in 1% solution before and after adding a sequestrant of calcium. When a pH 1.8 or 1.6 was used for each washing, only two washings were necessary to produce a RV lower than 40% (maximum recommended). The use of pH 2 required three acid washings to produce the same effect. The pH 2.2 did not remove enough calcium, even with three washings, the RV of the resulting sodium alginate being greater that 40%. The results of these experiments provide the information that producers need when deciding the best parameters to obtain a product with the desired characteristics.

Introduction

After the sodium alginate solution has been separated from the residual algae by filtration, the alginate can be recovered in solid form by precipitation, either as its calcium salt or as alginic acid. The precipitation with acid to produce alginic acid has two disadvantages; it is not possible to leave some calcium in the sodium alginate, to control the viscosity of the final product; precipitation with acid yields a gelatinous precipitate that is very difficult to dewater (Arvizu-Higuera et al., 1997). Calcium alginate can be precipitated as a fibrous material that can be readily separated on a metal screen, treated with chlorine to bleach the

product and then treated with dilute mineral acid to obtain alginic acid in a fibrous form. Alginic acid can be converted to sodium alginate by neutralization with an alkali (McHugh, 1987).

Green (1936) gave the amount of calcium chloride per volume of clarified extract to precipitate the calcium alginate, but did not relate the amount of calcium to the alginate content in the algae. Other authors mention the possibility of using 10% calcium chloride to precipitate the calcium alginate but do not give the amount of calcium to be used (Lukachyov & Pochkalov, 1965; McHugh, 1987). The first objective of this study was to determine the appropriate amount of calcium chloride to precipitate the calcium alginate at pilot plant level, yielding a fibrous form that allowed a fast filtration rate.

The next step of the alginate process is the bleaching of calcium alginate. Previous steps of formalin and acid treatment remove part of the products that impair the color of the alginate, however, the final color is still dark brown and for some food applications using higher grades alginates, the alginate must be bleached to improve the color. The best stage to bleach the product is as calcium alginate, because this stage is more resistant to degradation than alginic acid (Thornley & Walsh, 1931). Usually a sufficient quantity of sodium hypochlorite solution (12%) is added to a suspension of the calcium alginate in water. The amount of hypochlorite required varies with the seaweed used and the effectiveness of the previous stages. When a suitable color is obtained, the calcium alginate is separated on a metal screen (McHugh, 1987). No one has published the effect of the use of chlorine during the bleaching stage on the alginate viscosity. The second objective of this work was to analyze the effect of different amounts of chlorine used to bleach the calcium alginate at pilot plant level, on the color and viscosity loss (alginate degradation).

The bleached calcium alginate obtained must be converted to alginic acid. This is achieved by stirring the calcium alginate in a dilute mineral acid, using a three-step countercurrent system. The pH in the three tanks should be adjusted to less than 2 (McHugh, 1987), however no one has published the effect of different pH values in each of the three steps. The third objective of this work was to determine the effect of pH, during the conversion of calcium alginate to alginic acid at pilot plant level, on the residual calcium in the final product, the alginate viscosity and the percentage of viscosity reduction after sequestration of the residual calcium.

Materials and methods

Effect of calcium chloride on the filterability of calcium alginate

Ten kg of dried and milled alga *Macrocystis pyrifera* (L.) C. Agardh, collected from Bahía Tortugas, Baja California Sur, were placed in a tank to rehydrate overnight with 90 L of 0.1% formalin solution. The residual solution was drained off and the alga was washed with 100 L of hydrochloric acid solution at pH 4 in the same tank for 15 min with constant agitation (Hernández-Carmona et al., 1999a). The alga was transferred to an extraction kettle containing 166 L of water, the pH was adjusted to 10 with powdered sodium carbonate and heated at 80 °C for 2 hours, with constant stirring (Hernández-Carmona et al., 1999b). The paste was diluted to 45 mPa s and filtered in a rotary vacuum filter, using diatomaceous earth as filter aid (Hernández-Carmona et al., 1999b). During filtration, the filtrate (sodium alginate solution) was pumped to the precipitation tank and simultaneously a solution of 10% calcium chloride (anhydrous, commercial grade, 94% purity) was added to precipitate it as calcium alginate. The filtration and precipitation processes were continued until all the diluted solution passed through the filter. To carry through the residual solution in the filter pan, 50 L of water were added to the dilution tank. In order to have a constant volume for the next filtration, this filtration was stopped when the total volume in the precipitation tank reached 800 L (including the precipitate). To select the amount of calcium chloride to precipitate the alginate, the original alga was first analyzed at laboratory level to determine the alginate content, found to be 12.36%. The proportions of calcium chloride: alginate used in the experiment were 1.8, 2.0 and 2.2 parts of calcium chloride (anhydrous) per 1 part of alginate in the algal raw material. Therefore the quantities of calcium chloride used were 2.22 kg, 2.47 kg and 2.72 kg respectively, based on the use of 10 kg raw material.

After precipitating the calcium alginate, the suspension was filtered using a metal screen bed (US Sieve Series No.18 mesh) of 165 cm L × 80 cm W × 29 cm H, with a filtration area of 1.32 m^2. Filtration time was recorded for each experimental ratio and flow rate for the total liquid (800 L) was computed (L min^{-1}). One sample of the filtrate was taken and the residual calcium chloride in the water was measured by complexometric titration using the method described by Schwarzenbach & Flaschka (1969).

The calcium alginate was bleached using 700 mL sodium hypochlorite solution (5%), and converted to alginic acid with three acid washings at pH 2, 1.8, 1.8. To obtain the sodium alginate, enough ethanol was added to the fibers to give a proportion of 45% ethanol and 55% water and neutralized with powdered sodium carbonate until pH 7 was reached. The fibers were blended in a double planetary mixer for 40 min, drained on a screen and pressed in a hydraulic press. The sodium alginate fibers were separated and dried at 50 °C for sufficient time to yield a product with 12% moisture content. The percent alginate yield (based on the weight of original alga) was computed for each treatment. The experiment was run in triplicate for each ratio of calcium chloride.

Effect of bleaching on the alginate color and viscosity

(i) Bleaching at laboratory level
Calcium alginate was obtained from 10 kg alga, as described in the previous section, using a ratio of 2.2: 1 ($CaCl_2$: alginate). A portion of the filtered calcium alginate was carefully pressed to reduce the moisture content and divided into seven samples (each 20 g) that were suspended in water (each 200 mL) and treated with the following amounts of 5% sodium hypochlorite solution: 0.0, 1.0, 1.3, 1.6, 1.9, 2.2, and 2.5 mL. The suspensions were stirred for 15 minutes. The bleached calcium alginate was treated with hydrochloric acid to obtain alginic acid in a fibrous form, which was converted to sodium alginate by neutralization with sodium carbonate, then pressed and dried. Each of these steps was as described above, but reduced to laboratory scale. The yield, color and viscosity of sodium alginate were recorded for each experimental treatment. For color measurement, a 1% solution of sodium alginate was centrifuged to remove any turbidity and the color was measured as percent transmittance in a spectrophotometer at 510 nm. The same solution was used to measure viscosity, both before and after addition of sodium hexametaphosphate (0.5 g per 100 mL of alginate solution), using a Brookfield viscometer. Each experiment was run in triplicate; for each level of hypochlorite used, the three yields of sodium alginate were averaged and divided into the volume of hypochlorite to give the following measures of mL hypochlorite per gram of sodium alginate produced: 0, 0.31, 0.4, 0.48, 0.54, 0.74 and 0.77.

Three different types of alginate were treated, as described above, with the seven experimental amounts of hypochlorite: medium viscosity (~500 mPa s), low viscosity (~140 mPa s), and extra low viscosity (< 20 mPa s). The different types of alginate were obtained using raw materials collected at different dates and stored for different lengths of time. The three replicates for any one type of alginate were taken from three separate extractions at pilot plant level.

(ii) Bleaching at pilot plant level
Calcium alginate was obtained at pilot plant level as described above, in (i), using the alga containing medium viscosity alginate. After filtration, the calcium alginate was suspended in sufficient water (200L) to allow it to move freely in the suspension. Sodium hypochlorite solution (5%) was added using four different treatments: 0, 400, 600 and 700 mL (approximately equivalent to 0, 0.26, 0.38 and 0.49 mL g^{-1} of alginate produced). Stirring was maintained for 15 minutes. A sample (60 g) of the bleached product was removed and the isolation of sodium alginate was completed at laboratory level. Color of the sodium alginate was measured as described in (i).

Effect of pH during conversion of calcium alginate to alginic acid

In this experiment the same method was used as described above to obtain the calcium alginate. The fibers were suspended in water (150 L) and bleached with 5% sodium hypochlorite solution (700 mL). The calcium alginate obtained was divided in two parts and each half was used to test two different sets of pH. The fibers were suspended in water (125 L) and washed three times with constant agitation, adjusting the pH with hydrochloric acid to the following values: I (1.6, 1.6, 1.6), II (1.8, 1.8, 1.8), III (2.0, 2.0, 2.0) and IV (2.2, 2.2, 2.2). After each of the three washings, the precipitate was separated on a metal screen and a sample (60 g) was processed in the laboratory so that the extent of conversion to alginic acid could be estimated, using viscosity measurements. In the laboratory, the fibers were squeezed, placed in ethanol-water (50: 50, 400 mL) and neutralized with 10% sodium carbonate solution, with constant stirring. The sodium alginate obtained was drained with a cloth, pressed by hand, loosened and dried at 50 °C.

At pilot plant level the alginic acid, obtained after all three washings at a particular pH, was pressed in a hydraulic press to remove as much water as possible. The alginic acid cake was weighed, the fiber loosened and enough ethanol was added to reach a proportion of

Table 1. Effect of the calcium chloride: alginate ratio used to precipitate calcium alginate. Sample: 10 kg (dry wt) *Macrocystis pyrifera*, collected in Bahía Tortugas, B.C.S., October 1995. All results are shown as the average $\pm$ 1 SE

	Ratio $CaCl_2$: alginate		
	2.2: 1	2.0: 1	1.8: 1
Total calcium used (kg)	2.72	2.47	2.22
Filtration time (min)	8.5 $\pm$ 2.0	14.5 $\pm$ 2.8	56.3 $\pm$ 9.8
Filtration rate (L min^{-1})	97.9 $\pm$ 20.4	57.1 $\pm$ 12.5	14.6 $\pm$ 2.7
Residual $CaCl_2$ in filtrate (g)	1703 $\pm$ 91.7	1572 $\pm$ 6.0	604 $\pm$ 39.0
Alginate yield (%)	10.9 $\pm$ 1.0	9.9 $\pm$ 0.4	10.8 $\pm$ 1.0

ethanol-water (45: 55). The alginic acid was neutralized with powdered sodium carbonate and blended in a double planetary mixer for 40 minutes. The sodium alginate was pressed in a hydraulic press, loosened and dried at 50 °C.

For both laboratory and pilot plant samples, the viscosity of the sodium alginate obtained was measured in 1% solution at 22 °C with a Brookfield viscometer, using the method described in Hernández-Carmona et al. (1999a). The viscosity was measured again after adding sodium hexametaphosphate (0.5 g per 100 mL of alginate solution) to sequester the calcium ions present. The difference in viscosity was calculated and the percentage reduction in viscosity (RV) was used as the parameter of response for the experimental treatment; the less calcium alginate remaining after the acid treatment, the lower the RV. Experiments in the laboratory were run in triplicate and at least 5 replicates were run at pilot plant level.

The average, the standard error and the confidence interval ($\pm$1 SE) with a significance level of 95% were computed in each experiment. ANOVA was used to detect significant differences among treatments ($p < 0.05$) and the Tukey test was used to test the significance between means (Zar, 1984; StatSoft, 1995).

Results

Effect of calcium chloride on the filterability of calcium alginate

Filtration rates ranged from 14.6 L min^{-1} for 1.8: 1 ($CaCl_2$: alginate) to 97.9 L min^{-1} for the ratio 2.2: 1 (Table 1). The rate for the lowest ratio (1.8: 1) was significantly different from the other two ratios.

When using the lowest experimental amount of calcium chloride (1.8: 1), a small amount of calcium alginate formed as a gelatinous precipitate and during the filtration step this alginate formed a thin film on the screen, blocking the flow of residual liquid, increasing significantly the filtration time. This film formation was also observed for the ratio 2.0: 1 but not all the screen was covered. Although the difference was not statistically different from the ratio 2.2: 1, the filtration time tended to increase. One disadvantage of the use of ratio 2.0: 1, is that the gelatinous film must be removed before the screen can be used again, to avoid blocking in the next batch or when it is used to filter the calcium alginate after decoloration. The highest ratio, 2.2: 1, produced no gelatinous precipitate and gave an excellent flow rate.

The alginate yield range between 9.9% to 10.9% (Table 1) but the differences among treatments were not significantly different from the control, therefore the amount of calcium in the experimental range did not have a significant effect on the yield. This is because the thin layer of gelatinous alginate that was formed on the screen did not represent a significant amount of the total alginate obtained, although it was enough to block the holes in the screen and delay the process, with the consequent increase in cost of production. The average total amount of calcium in the residual water ranged from 604 g for the ratio 1.8: 1 to 1703 g for the ratio 2.2: 1 (Table 1). As expected, the amount of residual calcium chloride increased significantly as the calcium-alginate ratio was increased.

Effect of chlorine on the alginate color and viscosity

For samples bleached on the laboratory scale, the average percent transmittance (of all three viscosity grade alginates) in the treatment without hypochlorite was
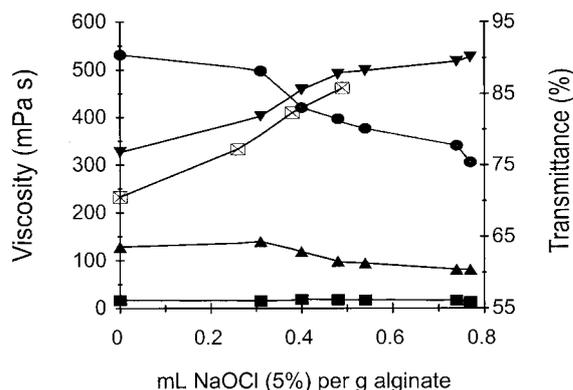
Figure 1. Effect of amount of sodium hypochlorite (5%) used (mL g^{-1} dry alga) on alginate color (% transmittance) and viscosity. Color obtained from samples of bleached calcium alginate taken at pilot plant level and processed at laboratory level (▼) and pilot plant level (⊠). Viscosity obtained, after addition of sodium hexametaphosphate, from alginates with medium viscosity (●), low viscosity (▲) and extra low viscosity (■). Mean ± 1 SE.

76.8, corresponding to a dark brown product; this increased to 90.1 for the treatment with the highest proportion of hypochlorite used, which produced an almost white alginate (Figure 1). Bleaching on the pilot plant scale was over a narrower range of hypochlorite concentrations and transmittance varied between 70.5 and 85.8, the last value corresponding to a cream color of the final product (Figure 1).

Viscosity of the medium viscosity alginate was reduced from 531 mPa s, with no hypochlorite treatment, to 305 mPa s when using the highest proportion of bleach (0.77 mL g^{-1}) (Figure 1), corresponding to a 42.6% viscosity loss; significant loss of viscosity (20.9%) resulted from 0.4 mL g^{-1}, falling to 420 mPa s. For alginates with low viscosity, the values changed from 140 mPa s (control) to 81 mPa s (0.77 mL g^{-1}) corresponding to 42.1% reduction; a significantly lighter color was obtained from 0.48 mL g^{-1} but with a viscosity loss of 29.4% (98.8 mPa s). Using alginates with extra low viscosity, the reduction in viscosity was not significantly different among treatments (average 16.7 mPa s).

Effect of pH during conversion of calcium alginate to alginic acid

For the samples that were taken after each of the three acid washings in the pilot plant and neutralised in the laboratory, the percent reduction in viscosity (RV) is shown in Figure 2a. The first washing, at any of the
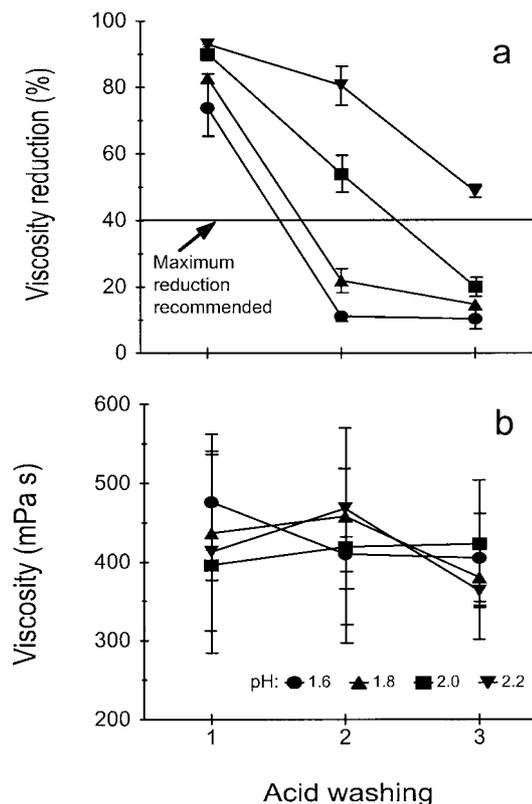

Figure 2. Effect of pH, after *each* of three acid washings to convert calcium alginate to alginic acid, on (a)% reduction of viscosity and (b) viscosity after addition of calcium sequestrant. Viscosity was measured using 1% solution of sodium alginate; viscosity reduction (%) was calculated after adding a calcium sequestrant to the same solution. Data from samples of alginic acid taken at pilot plant level and neutralised at laboratory level. Mean ± 1 SE.

four pH values, did not remove sufficient calcium with the RV varying from 73.8% at pH 1.6 to 93% for pH 2.2. A second washing did produce useful products for pH 1.6 (RV, 11.1%) and 1.8 (RV, 21.9%); however too much calcium remained at the two less acidic pH values, pH 2.0 (RV, 54%), pH 2.2 (RV 80.6%). A third washing at the lowest pH (1.6) gave only a small further reduction in RV (from 11.1% to 10.3%); at pH 1.8 the change was larger (from 21.9% to 14.7%) while pH 2.0 decreased the RV to 20%, an acceptable level. After the third washing at pH 2.2, the product still contained too much calcium (RV, 48.7%).

The average viscosity of the sodium alginate (as measured after sequestration of the calcium), from the twelve experimental treatments and neutralization at laboratory level, ranged from 363 mPa s (third wash-
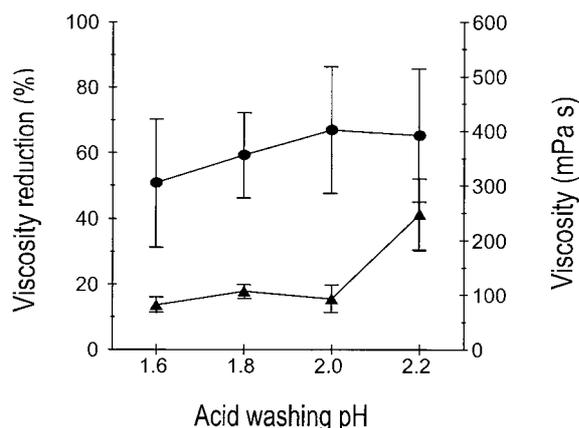
Figure 3. Effect of pH, after three acid washings to convert calcium alginate to alginic acid, on % reduction of viscosity (▲) and viscosity after addition of calcium sequestrant (●). Viscosity was measured using 1% solution of sodium alginate, viscosity reduction (%) was calculated after adding a calcium sequestrant to the same solution. Data obtained after alginic acid was neutralised at pilot plant level. Mean ± 1 SE.

ing, pH 2.2) to 476 mPa s (first washing, pH 1.6), (Figure 2b). No significant differences were found for any of the experimental values of pH or number of acid washings. At pilot plant level, the average RV (Figure 3) was similar to those obtained from acid-treated samples taken from the pilot plant and neutralised at laboratory level. Three acid washings at pH 2.2 produced a RV of 41.4% while pH values of 1.6, 1.8, and 2.0 yielded products with RVs ranging from 13.6% to 17.8%.

At pilot plant level, average viscosity of the sodium alginate (as measured after sequestration of the calcium) obtained after three washings ranged from 305 mPa s to 402 mPa s (Figure 3), and again no significant differences were found among the experimental pH values.

Discussion

The results show that the minimal ratio that can be used to precipitate the fibrous calcium alginate is 2.0: 1. However if the gelatinous film formation is to be avoided, to yield a maximum filtration rate, the ratio must be increased to 2.2: 1. This extra 10% calcium chloride is worthwhile, considering the following advantages: maximum filtration rate, harder fibers, no need to rinse the screen for the next batch, minimal increase in reagent, and low cost of the calcium chloride.

Green (1936) carried out the precipitation by adding the clear sodium alginate solution to calcium chloride solution in a proportion of about 45 kg calcium chloride and 362 kg water per 8 t alginate solution. Although the author did not mention the proportion, it is possible estimate it. Assuming the alginate solution, after dilution, contained between 0.25 to 0.5% alginate (Hernández-Carmona et al., 1999b) the proportion used was from 1.25: 1 to 2.5: 1, which is close to the experimental range used in our study. Green (1936) used 12.4% calcium chloride solution, Lukachyov & Pochkalov (1965) proposed using a 30% solution, but we found that a 10% solution is a better concentration to ensure complete solution of commercial grade calcium chloride. At higher concentrations an insoluble impurity appears to precipitate which requires vigorous stirring to keep it uniformly suspended.

An alternative method of recovering alginate from the sodium alginate solution is to add mineral acid to produce a precipitate of alginic acid. In this case, the pH is adjusted to give 2.8–3.2 (Le Gloahec & Herter, 1938), using for example 10% sulfuric acid solution. Others reduced the pH to 2 (Baranov et al., 1967) and in some cases suggest adding just enough acid to obtain alginic acid (Zvered et al., 1969). The precipitate, which is of a gelatinous consistency and holds large quantities of water, may be removed to baskets and allowed to drip and then pressed or centrifuged (Le Gloahec & Herter, 1938) but this is a time consuming and difficult process. On the other hand precipitation with calcium yields long threads of calcium alginate which can be washed, leached and bleached with great facility. When this calcium alginate is treated with mineral acid the resulting alginic acid has similar characteristics to the calcium alginate. These characteristic are very important for the economical production of the alginate on a large commercial scale.

One important feature of precipitation with calcium is the method of adding the solutions. Some authors recommend adding the dilute extract to the calcium chloride solution, because if the reverse is done, a gel will be obtained instead of fibers (McHugh, 1987). However it was found that better results were obtained when both solutions where added at the same time, calculating the precipitation time (which depends on the filtration rate) and adjusting the flow rate of the calcium chloride solution to finish at the same time as the filtrate. The degree of mixing is also important; too little will give a gel-type precipitate while too much may cause excessive breaking up of the

fibers, making it difficult to separate (McHugh, 1987). It was found that, stirring with a propeller, the speed should start at 300 rpm and then gradually increase to 500 rpm by the end of the step, but this condition may vary depending on the volume of the tank and the volume of the solution to be precipitated. Operator skill and experience are necessary to obtain consistent results (McHugh, 1987). During the filtration of the calcium alginate a large amount of water is discharged, containing unleached salts, soluble inorganic matter, excess soda ash, calcium chloride and some calcium carbonate (Green, 1936). In our pilot plant, the effluent contained a large amount of residual calcium (Table 1) but this excess calcium was needed to obtain a suitable form of calcium alginate. The total volume discharged ranged from 400 to 500 L kg^{-1}alginate produced (depending on the alginate content and viscosity in the raw material). Methods to recover these large quantities of water should be investigated.

Bleaching on the laboratory scale showed that the minimum amount of hypochlorite to give a significantly lighter color was 0.48 mL g^{-1} alginate which corresponds to the use of 700 mL at the pilot plant level. The color of the sodium alginate was light cream and was sufficiently bleached for use in any food applications. Bleaching trials were run in the pilot plant at 700 mL and lower levels of hypochlorite; it was found that, while 600 mL gave a cream product, 700 mL gave a lighter cream that would be more acceptable as a food additive. Use of 400 mL gave a light brown product that would be suitable only for a technical grade alginate.

In the production of sodium alginate, one of the serious factors causing breakdown of its molecular size is the amount of bleach used (Green, 1936). Results for the medium viscosity alginate showed there are two options. The first is to use 0.3 mL g^{-1} alginate proportion of hypochlorite with no significant reduction of viscosity and obtain a product that is still dark for some applications. The second option is to increase the hypochlorite proportion to 0.4 mL g^{-1} and obtain a light cream product for food applications, but losing 20.7% of its viscosity. Overall the results indicate that viscosity can be reduced by the use of hypochlorite and while low proportions produce an alginate with a color light enough for many uses, higher amounts may be needed to produce colors suitable for food applications. The reduction of viscosity by hypochlorite depends on the degree of polymerization (DP). Because of the low DP of the extra low viscosity alginate, the hypochlorite did not produce any viscos-

ity reduction in this product. Other methods have been used to reduce color. Le Gloahec and Herter (1938) used a jelly of hydrated alumina that was absorbed on the material and afterwards was separated by centrifugation, regenerated with solvents (eg. ethanol) and then recovered. Other authors recommend bleaching after acid treatment, washing the algae with an organic solvent (such as ethanol, acetone or other) to dissolve pigments, resins and other products soluble in organic solvents (Société Tech. de Rech. et d'Explot., 1949; Mizuno et al., 1982). Hasebe (1976) described a method for preparing seaweed powder which comprised adding hydrogen peroxide, or a compound that decomposes to produce hydrogen peroxide, to strip off the outer skin of the seaweed at 30–80 °C from 2–4 hours (depending on the species), and then drying the seaweed. However those methods required the use of expensive chemicals, increasing the production cost. Decolorization can also be achieved by the use of formaldehyde, tannic acid or other protein coagulants. Le Gloahec and Herter (1938) added formalin before digestion with soda ash solution. After the mixture has stood for one hour, the kelp was taken out and stored for 15–21 days. When the soda ash solution was added, only the alginous matter dissolved, and the pigments remained behind, 'fixed' by the protein-cellulose mixture; however this method required too much time.

The color of the alginate will depend on the seaweed used as raw material and the age of the plant harvested. However the results obtained here give the approximate amount of chlorine (as sodium hypochlorite) that can be used. If the product has still not the desired color, it can be adjusted by adding more sodium hypochlorite to reach the appropriate level by visual comparison.

In the conversion of calcium alginate to alginic acid, the method of sampling the alginic acid after each experimental acid washing, neutralizing it in the laboratory and measuring the reduction in viscosity (RV), provided an indirect technique to evaluate how much calcium was removed in each acid washing. The less the calcium remaining after acid treatment, the less will be the difference in viscosities before and after calcium sequestration and so the lower will be the percent reduction in viscosity (RV). The industry aims for an RV lower than 40%; above this figure the amount of residual calcium is judged to be excessive.

After the first washing, the average RV was significantly lower for pH 1.6 (73.8%) than for pH 2.0 (90.1%) or 2.2 (93.0%). In all cases the RV was higher

478

that the maximum recommended (40%), indicating that more acid treatment was necessary (Figure 2a). During the second washing all values were significantly different but the RVs obtained with pH 2.2 and 2.0 were still higher than the maximum reduction recommended, and another acid washing was needed. However, for pH 1.8 and 1.6 the RV was lower than the maximum recommended, indicating that two acid washings at either pH are enough to convert the calcium alginate to alginic acid, leaving some calcium to increase the viscosity. After the third washing, the RV was significantly higher for pH 2.2 than for all other treatments. Using pH 1.6 or 1.8, the third washing was shown to be unnecessary, because the calcium content was very low after two and the third treatment removed only a small fraction of calcium. The third treatment at pH 2.0 yielded an alginate with a RV of 20% and this can be used for many applications. However pH 2.2 still gave a product with a RV of 48.7% and so required more acid treatment to reduce calcium to an acceptable level. Therefore pH 2.2 is not recommended.

The graphs obtained provide information on how the calcium is removed by the treatment and the user can select the appropriate combination of pH values to obtain the desired reduction in viscosity. However the best compromise could be three acid washings at pH 2.0, 1.8 and 1.8, because the first washing exchanges only a small fraction of calcium, and it was not significantly higher when using pH 1.8. If pH 2.0 is selected for the first washing, less acid is used, reducing the costs. The second washing at pH 1.8 was selected, because it removed a significantly higher amount of calcium than pH 2.0, but it is not significantly higher than using pH 1.6. The third acid washing at pH 1.8 was selected because it was not significantly different from pH 1.6. If water supply is short, two acid washings at pH 1.8 or pH 1.6 (or a combination of these) can be enough, depending on the viscosity reduction allowed by the quality control adopted.

Green (1936), proposed three acid washings at pH 1.3, 1.5 and 1.8, but the results obtained in the present experiment demonstrate that when using a pH of 1.8 or lower, only two washings are necessary. In an industrial plant, this treatment can be carried out using three tanks in countercurrent, as described by McHugh (1987), but using 15 minutes for each washing, as proposed in this paper. The pH in the three tanks should be adjusted to the pH values according to the desired reduction in viscosity for the final product.

The viscosity of the sodium alginate produced in either the pilot plant or the laboratory did not depend on the pH of the washings or the number of washings. This was surprising, because it had been expected that higher depolymerization of alginate would occur during this step, especially at pH 1.6. However our results suggest that alginic acid is more resistant than expected, because this step was considered as one of the steps where more degradation may take place (Haug, 1964). This finding is also important, because it means that it is possible to carry out the acid washings at a pH as low as 1.6, removing almost all the calcium and reducing the number of acid washing to two, without significant degradation of the alginate. The results of these experiments on the precipitation, bleaching and acid washing of calcium alginate provide information that allows any producer to decide the best parameters to obtain an alginate with the desired characteristics.

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