



## Effect of G/M ratio on the radiation-induced degradation of sodium alginate

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### A B S T R A C T

Radiation-induced degradation of sodium alginate (NaAlg) having different G/M ratios was investigated. NaAlg samples were irradiated with gamma rays in air at ambient temperature in the solid state at low dose rate. Change in their molecular weights was followed by size exclusion chromatography (SEC). Changes in their rheological properties and viscosity values as a function of temperature, shear rate and irradiation dose were also determined. Chain scission yields,  $G(S)$ , and degradation rates were calculated. It was observed that G/M ratio was an important factor controlling the  $G(S)$  and degradation rate of sodium alginate.

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### 1. Introduction

Alginate is one of the natural polysaccharides that occur in substantial amounts in the brown seaweeds. Alginates are unbranched binary co-polymers of (1–4)-linked residues of  $\beta$ -D-mannuronic acid (M) and  $\alpha$ -L-guluronic acids (G). Alginic acid and its water-soluble sodium salt have been manufactured for decades for their great ability to give highly viscous solutions even at moderate concentrations and mainly used in food, pharmaceutical, and chemical industries as thickening agents, drug carrier, stabilisers, and plant growth stimulator (Østberg et al., 1994; Garcia and Ghaly, 1996; Hien et al., 2000). Aqueous solution of sodium alginate forms stable gels in the presence of multivalent cations such as  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ . Gel formation occurs due to the ionic interaction between guluronic acid residues from two or more alginate chains and cations, yielding a three-dimensional network of alginate molecules well described by the “egg-box model” (Grant et al., 1973). Functional and physical properties, mechanical strength, porosity, gel uniformity, biocompatibility, and influence on encapsulated cells properties of alginate gels vary widely depending on ratio of mannuronic to guluronic acids, the frequency and size of guluronic acid blocks and the molecular weight of the polymer (Smidsrod, 1974; Klock et al., 1994). Experimental studies have shown that selective binding of certain alkaline earth metal ions increase markedly with increasing content of the  $\alpha$ -L-guluronic acids (G) residues in the chain. On the other hand, poly-mannuronate blocks and alternating blocks are almost non-selective.

In recent years much more attention has been directed to radiation modification and degradation of natural polymers such as kappa carrageenan, sodium alginate and chitosan, and using of these low molecular weight polysaccharides or oligosaccharides in the plant growth promoter and plant protectors (Relleve et al., 2005; Thama et al., 2001; Nagasawa et al., 2000).

Recently, Nagasawa et al. (2000) investigated the effect of radiation on alginates in solid state and in aqueous solution. It was found that NaAlg degraded after irradiation in both solid state and aqueous solution; and the degradation in solution was remarkably greater than that in the solid state. Degradation yields,  $G(S)$ , of NaAlg were found in this study as 1.9 and  $55 \text{ g mol}^{-1} \text{ kGy}^{-1}$  for solid state and aqueous solution, respectively.

In another study performed by Wasikiewicz et al. (2005), ultrasonic, ultraviolet and gamma degradations of NaAlg and Chitosan were investigated. It was found for both polymers that the most effective method, from the energetic point of view, was gamma radiation with a yield of scission,  $G(S)$ , of  $0.55 \times 10^{-7} \text{ mol/J}$  for 1% alginate, and  $3.53 \times 10^{-7} \text{ mol/J}$  for 1% chitosan. However, considering the reaction time, the ultraviolet method was the most effective, with the reaction rate constant,  $k$ , of  $0.52 \text{ h}^{-1}$  for alginate, and  $1.6 \text{ h}^{-1}$  for chitosan.

Neither in these radiation-induced degradation studies of NaAlg, nor in other studies on the preparation of oligosaccharides of NaAlgs as the plant growth promoter and plant protectors (Nagasawa et al., 2000; Kume et al., 2002) the effect of configuration of NaAlg was considered by the authors.

In the present study, radiation-induced degradation of sodium alginate (NaAlg) having different G/M ratios was investigated for the first time. NaAlg samples were irradiated with gamma rays in air at ambient temperature in the solid state at low dose rates. Change in their molecular weights was followed by size exclusion chromatography (SEC). Changes in their rheological properties

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**Table 1**  
Initial molecular weights and G/M ratios of NaAlgs used.

Polymer	$M_n$	$M_w$	G/M
LF120	$340.0 \times 10^3$	$670.0 \times 10^3$	70/30
LF200	$310.0 \times 10^3$	$690.0 \times 10^3$	50/50
LF240	$275.0 \times 10^3$	$500.0 \times 10^3$	45/55

and viscosity values as a function of temperature, shear rate and irradiation dose were also determined. Chain scission yields,  $G(S)$ , and degradation rates were calculated. It was observed that G/M ratio was an important factor controlling the  $G(S)$  and degradation rate of sodium alginate.

## 2. Experimental

Sodium alginate samples were obtained from FCM Biopolymers Company, Norway and used as received. The initial molecular weights of NaAlgs, guluronic acids and mannuronic acid, G/M ratios and abbreviations used for these polymers are given in Table 1. The polymer samples were placed in tightly closed containers, and irradiated at the required doses (2.5, 5, 10, 15, 20, and 25 kGy) in a Gammacell 220 type  $^{60}\text{Co}$ -gamma irradiator at room temperature in air.

Irradiated and unirradiated samples were analysed using a Waters Breeze model Gel Permeation Chromatograph.  $\text{NaNO}_3$  (0.1 M) was used as the eluting solvent. Water 2000–1000–500 hydrogel columns were used for molecular weight analyses, and universal calibration was constructed by using narrow molecular weight poly(ethylene oxide) standards.  $K$  and  $a$  constants for NaAlg were taken as  $7.3 \times 10^{-5}$  and 0.92, respectively (DeRamos et al., 1997).

Change in their viscosity with concentration and irradiation dose, and all rheological properties were measured by using a Thermo MARS rheometer equipped with a controlled test chamber (CTC) for temperature control and using a cone–plate geometry (cone diameter 40 mm and angle  $4^\circ$ ). Viscosity measurements were performed in the shear rate range  $0.1$ – $500 \text{ s}^{-1}$ , and at  $30^\circ\text{C}$ .

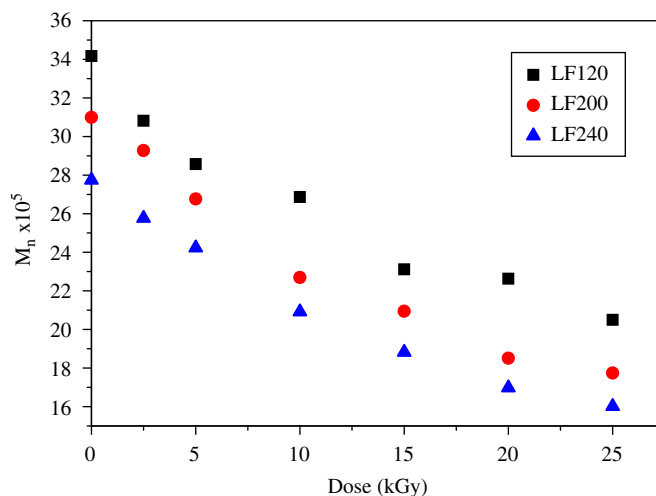
## 3. Results and discussion

### 3.1. Controlling of molecular weight of sodium alginates by G/M ratio

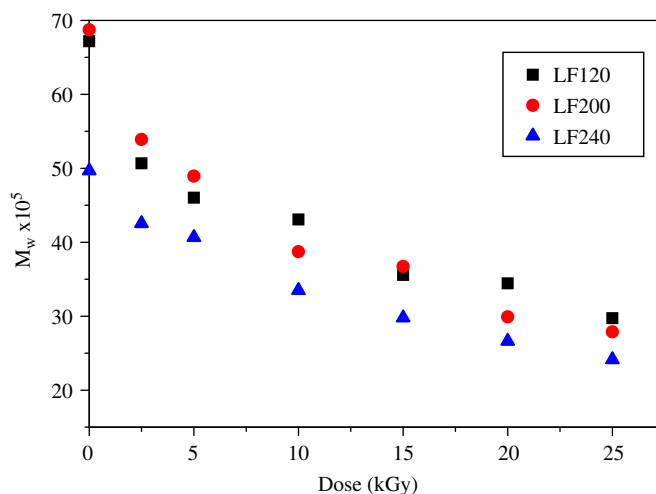
It is very well known that polysaccharides in dry form or in solution degrade when exposed to ionizing radiation. For the investigation of the effect of gamma rays on the molecular weight of NaAlgs, their  $M_w$  and  $M_n$  values were evaluated using SEC. Unimodal chromatograms were obtained for all NaAlg samples. As the irradiation dose increased, the SEC chromatogram of the NaAlg sample shifted to higher retention volumes indicating the molecular weight of the sample was decreased with irradiation. By using universal calibration curve, average molecular weight of NaAlg samples was evaluated. Changes in the weight and number average molecular weights ( $M_w$  and  $M_n$ , respectively) with irradiation dose are given in Figs. 1 and 2, respectively. As can be seen from these figures, both average molecular weights decreased rapidly up to 20 kGy.

### 3.2. Determination of chain scission yield and degradation rate constant

The efficiency of radiation-induced events is expressed by the so-called  $G$ -value. The  $G$ -value, equal to the number of events per



**Fig. 1.** Change in the number average molecular weight ( $\bar{M}_n$ ) of NaAlgs with dose.



**Fig. 2.** Change in the weight average molecular weight ( $\bar{M}_w$ ) of NaAlgs with dose.

100 eV of energy absorbed, has been customarily used to measure radiation chemical yield. Molecular weight values of NaAlgs are used for the determination of chain scission yield, ( $G(S)$ ), crosslink yield ( $G(X)$ ) and degradation rate. By using the related equations given in the literature zero  $G(X)$  values were found for each NaAlg. (Güven et al., 1992). If scission is the only mode of action of radiation then the radiation–chemical yield of degradation (scission)  $G(S)$  is determined from the Alexander–Charlesby–Ross equation (Charlesby, 1960):

$$1/\bar{M}_{nD} = 1/\bar{M}_{n0} + 1.04 \times 10^{-7} G(S)D \quad (1)$$

where the absorbed dose,  $D$ , is in kGy, and  $\bar{M}_{nD}$  and  $\bar{M}_{n0}$  are the number average molecular weights of the polymer before and after irradiation, respectively. Rapid decrease in  $\bar{M}_n$  and  $\bar{M}_w$  clearly indicated that the only mode of action of radiation on sodium alginate was degradation. For the determination of the  $G(S)$  values,  $1/\bar{M}_n$  was plotted against dose for all samples (Fig. 3). Then,  $G(S)$  values were calculated by using the intercepts. The calculated  $G(S)$  values were  $0.71 \pm 0.083$ ,  $0.97 \pm 0.048$ ,  $1.05 \pm 0.031$  for LF120, LF200, and LF240 NaAlg, respectively. It was concluded that  $G(S)$  values were dependent on the structure (guluronic acids(G) and mannuronic acid(M) ratio) of the sample and followed the order  $\text{LF240} > \text{LF200} > \text{LF120}$  or  $70/30 < 50/50 < 45/55$ , G/M ratio.

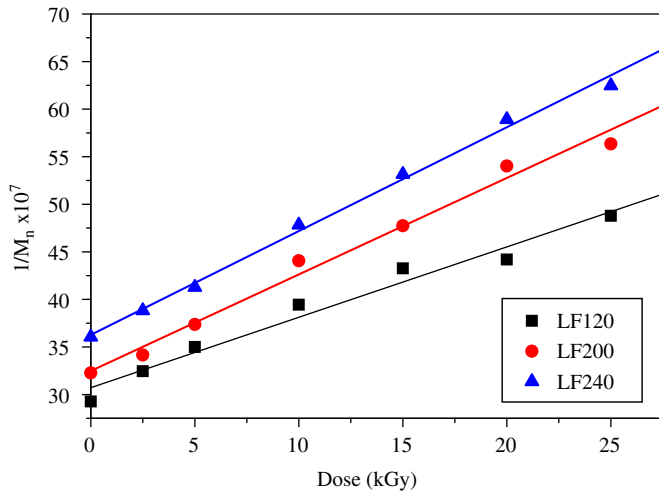


Fig. 3. Plot of  $1/M_n$  vs. dose for the determination of the  $G(S)$  values of NaAlgs.

When the chemical structures of NaAlgs were examined it was seen that the properties of NaAlg varied widely depending on the composition of the alginate molecule (i.e., the ratio of mannuronic to guluronic acids, the frequency and size of guluronic acid blocks, and the molecular weight of the polymer), and the concentrations of alginate and cation at the time of gelation. It could be concluded that the  $G(S)$  value increased with a decrease in the guluronic acids to mannuronic acid ratio. This increase was attributed to the decrease of radical-radical recombination reactions due to the decrease of stiff and extended nature of alginate chains. It is well known that the diaxial linkage in G-blocks results in a large, hindered rotation around the glycosidic linkage, which may account for the stiff and extended nature of alginate chains (Smidsrod et al., 1973).

The equation given by Jellinek (1955) was modified and used in the determination of the degradation rate. Where  $N$  is the average number of bond cleavages per original polymer molecule, this value can be calculated by:

$$N = \frac{\bar{M}_{n0}}{\bar{M}_{nD}} - 1 \quad (2)$$

where  $\bar{M}_{n0}$  and  $\bar{M}_{nD}$  are the  $\bar{M}_n$  before and after irradiation to a certain dose ( $D$ ), respectively.  $N$  is also named as the degree of scission. Assuming the rate of chain scission is independent of chain length and position of the link in the chain,  $N$  is anticipated to be a linear function of irradiation dose (first-order reaction)

$$N = \frac{\bar{M}_{n0}}{\bar{M}_{nD}} - 1 = k \left[ \frac{\bar{M}_{n0}}{m_o} \right] D \quad (3)$$

where  $k$  is the rate constant and  $m_o$  is the molecular weight of a monomer unit. Eq. (3) can be rewritten as:

$$\frac{1}{\bar{M}_{nD}} - \frac{1}{\bar{M}_{n0}} = \left( \frac{k}{m_o} \right) D \quad (4)$$

Degradation rate constants for NaAlgs were determined by using the  $(1/\bar{M}_n) - (1/\bar{M}_{n0})$  vs. dose curves. The determined degradation rate constants for NaAlgs are given in Table 2. They followed the order  $LF240 > LF200 > LF120$ . These results clearly indicated that degradation rate was also dependent on the G/M ratio for these initial molecular weights.

### 3.3. Examination of rheological behaviour of the NaAlg samples

Change in viscosity with irradiation at different polymer concentrations and shear rates was investigated for all NaAlg

Table 2  
Degradation rate constants of NaAlgs.

Polymer	G/M	$k$ (kGy <sup>-1</sup> )
LF120	70/30	$4.27 \times 10^{-10} \pm 3.2 \times 10^{-11}$
LF200	50/50	$5.86 \times 10^{-10} \pm 3.0 \times 10^{-11}$
LF240	45/55	$6.30 \times 10^{-10} \pm 2.0 \times 10^{-11}$

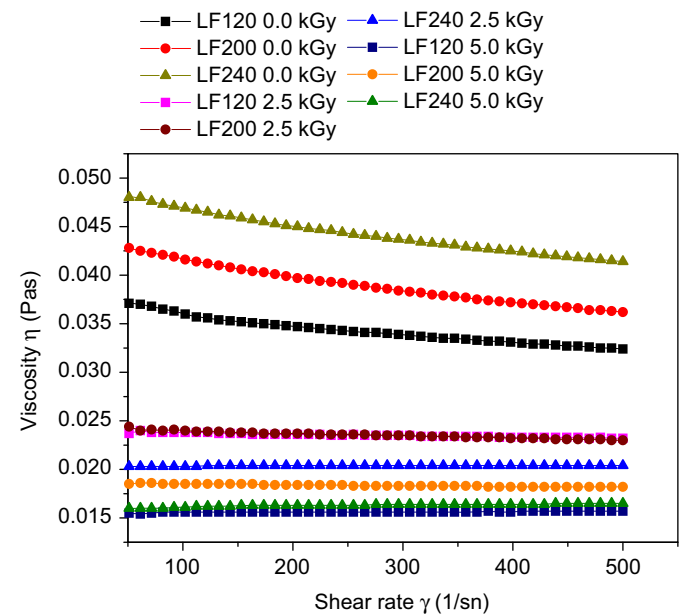


Fig. 4. Change in the viscosity with shear rate for LF120 NaAlg irradiated at the indicated doses.

samples. A representative figure for the change of viscosity with shear rate for the unirradiated, and the 2.5 and 5.0 kGy-irradiated LF120 NaAlg is given in Fig. 4.

Shear rate vs. shear stress, and shear rate vs. viscosity curves became linear at an irradiation dose of approximately 2.5 kGy for LF120 NaAlg. Same behaviour was also observed for the other NaAlgs. Linearity of these plots indicated that the samples showed the Newtonian fluid properties. We could conclude that NaAlg lost the pseudoplastic fluid behaviour, and that the flow behaviour change from non-Newtonian to Newtonian even upon irradiation at very small doses.

Viscosimetric studies performed in different polymers concentrations and at different shear rates clearly indicated that the change in specific viscosity of NaAlg solutions depended on the shear rate and concentration of the solution. For the determination of limiting viscosity number of NaAlgs for zero shear rate and concentration, the rheological data were reanalysed, and the  $\eta_{sp}/c$  values were plotted vs.  $(c + 0.001 \times \dot{\gamma})$ . The representative curves for the determination of  $(\eta)$  are given in Fig. 5. Similar curves were obtained for the other NaAlgs. Decrease percentage in  $(\eta)$  with irradiation is given in Fig. 6. As can be seen in this figure, the decrease in viscosity followed the order  $LF200 > LF120 > LF240$ . These results clearly indicated not only that the rheological properties of the irradiated and molecular weight-reduced alginates were only controlled by the G/M ratio, but also that the frequency and size of the guluronic acid blocks were important parameters. Despite the lower molecular weight and lower G/M ratio, the slower decrease in  $(\eta)$  value for LF240 with irradiation was probably due to the presence of more GG blocks in the main chain. Experimental viscosimetric data of alginate

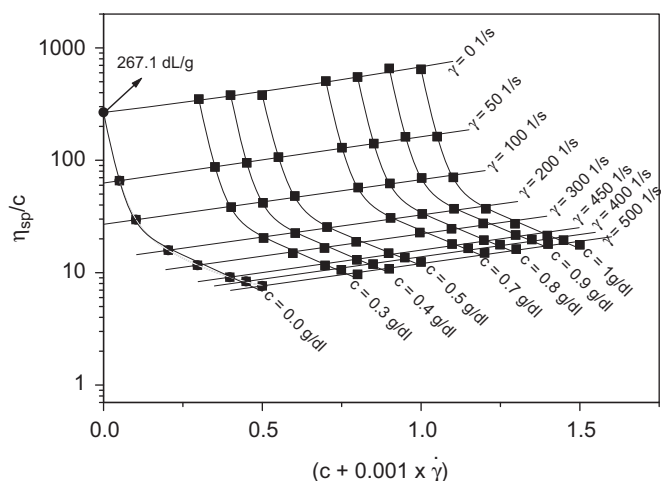


Fig. 5. A plot for the determination of the limiting viscosity number of LF120 NaAlg for zero shear rate and zero concentration.

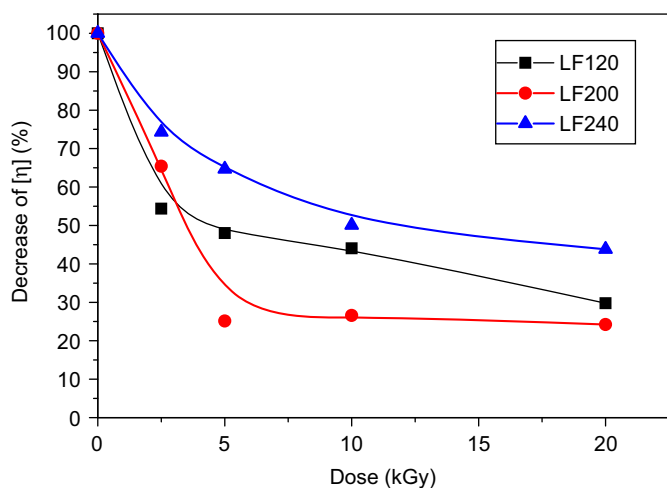


Fig. 6. Decrease percentage in the limiting viscosity number for LF120 NaAlg with irradiation dose.

solutions indicated that the stiffness of the chain blocks increased in the order  $MC < MM < GG$  (Smidsrod, et al., 1973).

#### 4. Conclusions

The effect of irradiation on the NaAlg samples was determined to be dependent on the chemical structure of the sample. As the

gluronic acid to mannuronic acid ratio decreased, the effect of irradiation on the molecular weight and rheological properties of the sample became more pronounced. It could be concluded that the G/M ratio was one of the factors affecting chain scission yield of NaAlg. All NaAlg samples showed a pseudoplastic behaviour up to a certain dose, and a Newtonian flow behaviour above that dose.

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