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DEGRADATION OF SODIUM ALGINATE BY γ-IRRADIATION AND BY OXIDATIVE–REDUCTIVE DEPOLYMERIZATION

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ABSTRACT

Methods are described for preparing degraded sodium alginate by oxidative–reductive depolymerization (ORD) and by γ-irradiation. The rate of degradation by ORD was shown to be governed by more than one rate constant, and the degraded products prepared by either method were shown to have gross chemical and biological properties similar to those of a derivative of sodium alginate prepared by hydrolytic degradation. The rate of degradation is discussed in the light of the known structure of alginic acid, and the value of the products as food additives to suppress strontium absorption is assessed.

INTRODUCTION

It has been shown in man[1] and in animals[2] that the addition of sodium alginate to the diet reduces the absorption of strontium without materially affecting the absorption of calcium. It has also been shown[3,4] that the greatest selectivity against strontium is obtained from alginate containing the highest proportion of guluronic acid.

Alginate (OG1) having a high content of guluronic acid, however, is prepared by a time-consuming method[5] which does not lend itself easily to larger scale preparation, and the main purpose of the present work was to explore alternative methods.

The method used for enriching commercial sodium alginate in guluronic acid causes degradation, but the reduced viscosity of the product increases its value as an additive by improving its palatability and water solubility. By sacrificing some of the qualities of high content of guluronic acid, an effective food additive may still be prepared by simple degradation of commerical alginate. The two methods investigated were oxidative–reductive depolymerization (ORD)[6] and γ-irradiation.

MATERIALS AND METHODS

The low-viscosity, commercial alginate Manucol SS/LD/2 (Alginate Industries Ltd., Henrietta Street, London, W.C.2) was used as the starting material in all investigations. Other chemicals were analytical grade, wherever possible.

Oxidative–reductive depolymerization. — Solutions (2%) of Manucol SS/LD/2 containing 1.5 mM ferrous ammonium sulphate and 500 mM hydrogen peroxide were stirred for time intervals ranging from 0.5 to 160 min. At selected times, the degraded product was precipitated at pH 1.5 with hydrochloric acid, collected by centrifugation, and washed with dilute acid. After neutralization with sodium hydroxide, water-insoluble material was removed by centrifugation, and the sodium salt was precipitated from the supernatant with ethanol.

The reaction was followed by measuring, in a capillary viscometer, the specific viscosities of 1% solutions of the product in 0.1M sodium chloride.

γ-Irradiation. — Samples of Manucol SS/LD/2 were irradiated at the Spent Fuel Assembly, Atomic Energy Research Establishment, Harwell, with ~1 MeV γ-radiation. The dose-rates, measured by ionization chamber (calibrated against a Fricke dosimeter), were maintained between 1.42 and 3.15 Mrad/h to give overall doses of between 2 Mrad and 200 Mrad. The products were dissolved in 0.1M sodium chloride to give 1% solutions, and their specific viscosities were determined after removing traces of insoluble material.

Measurement of stability constants. — Schubert's resin-exchange method was followed[4,7] by using barbitone buffer at an ionic strength of 0.16M and ^{47}Ca and ^{85}Sr at carrier levels of 10 μequiv./litre. A purified and graded Dowex resin was used (AG-50WX12, 100–200 mesh, Bio-Rad Laboratories Richmond, California).

The effects of the products on the uptake of calcium and strontium by surviving rat intestinal slices was measured by the literature technique[2]. The guluronic acid content was measured by the method of Haug and Larsen[5,8]. Sodium analyses were made by emission spectrophotometry after wet-ashing. Iron was measured by the thiocyanate method after wet-ashing.

RESULTS

It can be shown, for a first-order random depolymerization[9], that:

$$\frac{1}{[\eta]_t} - \frac{1}{[\eta]_0} = k't = \frac{\Delta 1}{[\eta]} \qquad (1)$$

where $[\eta]_0$ and $[\eta]_t$ are limiting-viscosity numbers at zero time and time t, respectively.

In the present series of experiments, the starting material and products give viscosity numbers (specific viscosity/concentration) which do not vary greatly with concentration, so that the specific viscosity (η_{sp}) of a 1% solution is approximately equal to the limiting-viscosity number. Therefore, substituting in Equation (1),

$$\Delta 1/\eta_{sp} = k't,$$

Fig. 1 shows the relation between $\Delta 1/\eta_{sp}$ and t for ORD.

Similarly $\Delta 1/\eta_{sp} = k'd$ (where d = radiation dose in Mrad), and Fig. 2 shows the relation between $\Delta 1/\eta_{sp}$ and d for γ-irradiation.

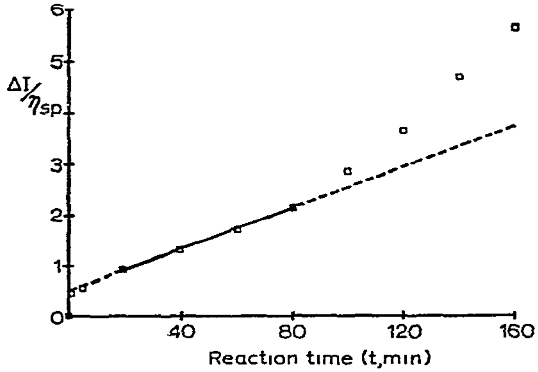

Fig. 1. Variation of extent of degradation ($\Delta 1/\eta_{sp}$) of sodium alginate with duration (t, min) of oxidative–reductive depolymerization [Regression equation: $(\Delta 1/\eta_{sp})_t = 0.516+0.02t$].

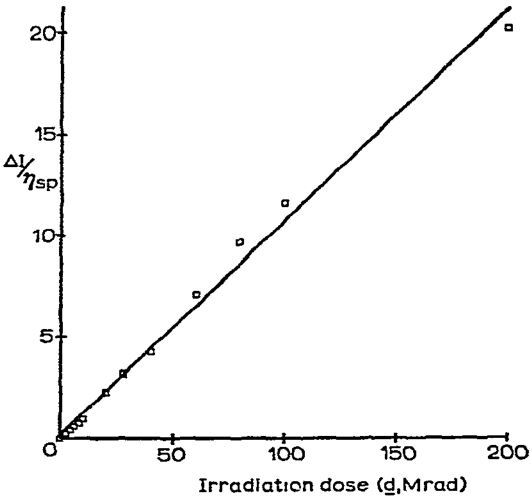

Fig. 2. Variation of extent of degradation ($\Delta 1/\eta_{sp}$) of sodium alginate with dose (d, Mrad) of γ-irradiation [Regression equation: $(\Delta 1/\eta_{sp})_d = 0.226+0.105d$].

Table I contains analytical data for (*a*) the product (**1**) of selective precipitation and hydrolysis (OG1)[5]; (*b*) the product (**2**) of ORD after 160-min reaction; and (*c*) the acid-insoluble fraction (**3**) obtained after 40-Mrad irradiation.

It was found that the graph relating $\Delta 1/\eta_{sp}$ with d for the acid-insoluble fractions of the products of γ-irradition showed a similar linearity to Fig. 2, but with a wider scatter.

DISCUSSION

Fig. 1 shows that, for a reaction time between 20 and 80 min, the relation between $\Delta 1/\eta_{sp}$ and t is linear ($k = 0.02\ \text{min}^{-1} \pm 0.0003$) and similar in value to that estimated from the data of Smidsrød *et al.*[10]. Above 80 min, the rate of increase of

TABLE I

PROPERTIES OF SODIUM ALGINATE DEGRADED BY ACID HYDROLYSIS (PRODUCT **1**), OXIDATIVE–REDUCTIVE DEPOLYMERIZATION (PRODUCT **2**), AND γ-IRRADIATION (PRODUCT **3**)

Product	**1**	**2**	**3**
Carbon content (%)	33.79	34.03	33.23
Calc. equiv. wt.	213.3	211.8	216.9
Sodium content (mg/g)	109.7	109.7	105.3
Iron content (mg/g)	<0.1	1.3	<0.1
Guluronic acid (%)	>95	88	92
$[\alpha]_D^{20}$ (c 0.1, water) (degrees)	−136	−121	−134
Specific viscosity (1%)	0.17	0.17	0.24
Stability constant (l/equiv.) Sr	388	418	337
Ca	74.3	112	88.4
Ratio Sr/Ca	5.22	3.73	3.81
Inhibition of uptake by intestinal slices (%) Sr	43.6	49.9	38.6
Ca	3.4	17.2	0.5

$\Delta 1/\eta_{sp}$ with time increases, as a result, perhaps, of an increasing proportion of acid-soluble material in the product.

Extrapolation of the linear section of the graph to zero time does not give a value of $\Delta 1/\eta_{sp} = 0$, showing that degradation is initially more rapid. Measurements made at 0.5 and 5 min did, indeed, give values of $\Delta 1/\eta_{sp}$ falling below the extrapolated line, but, at these short intervals (particularly 0.5 min), errors in the measurement of the reaction time were considerable.

It has been shown[11,12] that alginate molecules are made up of sections containing ~90% of guluronic acid residues (G-blocks), ~90% of mannuronic acid residues (M-blocks), and alternating guluronic and mannuronic acid residues (intermediate fraction); the G- and M-blocks were shown to be considerably more resistant to hydrolytic attack than the intermediate fraction. The increase in guluronic acid content, as a result of ORD, from a value of ~72% for the starting material to 88% for product **2**, suggests that the intermediate fraction is also more susceptible to ORD than the remainder of the molecule.

The relation between the degradation caused by γ-irradiation and dose (Fig. 2) is linear, but the intercept, although not zero, is smaller than that recorded for ORD (Fig. 1). The possibility of two rates of degradation is therefore less clearly seen, but the product of γ-irradiation shows a higher content of guluronic acid than the starting material, indicating that degradation caused by γ-irradiation also is selective.

The analytical values presented in Table I show that all three products have similar properties. Although the stability constant for the strontium salt of product **2** appears to suggest that this would be the most effective agent for suppressing strontium absorption[4], the ratio of strontium to calcium stability constants (the more important parameter[13]) points to product **1**, already known to be the best agent. Product **3**, however, which shows a stability constant ratio that is approximately equal to that of product 2 and does not cause such a high inhibition of slice-uptake of calcium, is technically very much easier to prepare.

Irradiation for 40–50 Mrad, followed by separation of the acid-insoluble fraction of the product, is therefore probably the optimal procedure for preparing a derivative of sodium alginate for use as a food additive to suppress strontium absorption, although prior testing for toxicity would, of course, be necessary.

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