

## Determination of Intrinsic Viscosity of Alginates

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A method for determination of intrinsic viscosity of alginates by extrapolation to zero rate of shear and zero concentration is described, and the importance of the extrapolation to zero rate of shear is discussed. The influence of salts and pH on the viscosity of alginate solutions has been investigated, and the viscosity was found to be independent of pH in the range from 4 to 12. Curves correlating the intrinsic viscosity with the relative viscosity of alginate solution at various concentrations determined at standardized conditions are given. A method for routine determination of intrinsic viscosity without isolation of the alginate by determining the apparent viscosity of the same solution at two different values of rate of shear is described.

From a commercial point of view, the most important property of alginates is their ability to form viscous solutions in water. It is therefore of considerable practical importance to be able to characterize an alginate sample with regard to its ability to form viscous solutions. Most producers describe their products by quoting the viscosity of an aqueous solution containing a certain amount of alginate, usually 0.3 or 1.0 g in 100 ml. In several of the works published about the properties of alginates, the viscosity of an alginate solution containing 0.25 g in 100 ml 0.1 N sodium chloride solution is used for characterizing the samples<sup>1-4</sup>.

For scientific purposes it is usual to characterize linear macromolecules by means of the intrinsic viscosity

$$[\eta] = \lim_{c \rightarrow 0} \left( \frac{\eta_r - 1}{c} \right)$$

where  $\eta_r$  is the relative viscosity of the polymer solution and  $c$  is the concentration, usually given as g per 100 ml. In a number of works the intrinsic viscosity of alginates has been determined. Donnan and Rose<sup>5</sup> investigated a series of alginates varying in intrinsic viscosity from 4.2 to 14.4 (100 ml/g). They reported that dependence of shear stress was only important at viscosities higher than 40 cp. Harkness and Wassermann<sup>6</sup>, investigating alginates with intrinsic viscosity in the order of 7 also reported that non-Newtonian

behaviour was only significant at high viscosity and therefore did not affect the intrinsic viscosity. Cook and Smith<sup>7</sup> investigated samples with intrinsic viscosities varying from 3.1 to 17.5. Only the sample with the highest intrinsic viscosity was found to give solutions with non-Newtonian behaviour. Vincent, Goring and Gordon Young<sup>4</sup> prepared alginates from some Canadian brown algae, and found values of intrinsic viscosities varying from 3.6 to 19.2. The influence of rate of shear was not discussed.

Observations in this laboratory<sup>8,9</sup> indicated that non-Newtonian behaviour could be observed at very low concentrations and that intrinsic viscosities, at least above 8–9, determined by extrapolation to zero concentration from measurements carried out at constant shear stress, would be influenced by the shear stress in the measurements.

The purpose of the present paper is to describe a method for the determination of a true intrinsic viscosity of alginate samples by extrapolation to zero rate of shear and zero concentration. The correlation between the intrinsic viscosity and quantities suited for routine determination is given.

As some confusion seems to exist in the literature concerning the effect of pH on the viscosity of alginates, an investigation of this effect and the influence of salts was necessary in order to find suitable conditions for the intrinsic viscosity measurements, and in order to compare the results given in the literature.

## EXPERIMENTAL

The sampling of seaweeds and the preparation of alginate have been described earlier<sup>10</sup>. When alginic acid of low molecular weight was desired, the weed, after the usual extraction with dilute acid, was suspended in 1 N H<sub>2</sub>SO<sub>4</sub> and maintained at 60°C for a certain time before the usual alkali extraction. The water content of the alginate samples was determined by heating in an oven at 105°C for 6 h, and the intrinsic viscosities are calculated on the basis of dry sodium alginate. All other results are calculated on the basis of sodium alginate containing 90 ± 1 % dry matter.

Solutions were prepared by adding alginate to water and shaking the solution in a slow moving shaker for 15–20 h. After the alginate was completely dissolved, the desired amount of inorganic salt was added and the solution stirred one hour before the measurements.

Five different viscometers were used: Ubbelohde Viscometer No. 1 ( $k = 0.00999$ ) and No. 2 ( $k = 0.1001$ ) (Jena Glas, Mainz), a Wagner-Russell Viscometer<sup>11</sup>, Höppler Visco-Waage<sup>8</sup> (glass ball, diameter 99.15 % of tube diameter) and a Höppler Viscometer (glass ball No. 2, diameter 98.00 % of tube diameter). The Ubbelohde viscometers were calibrated at the factory, and the other viscometers were calibrated against the Ubbelohde instruments by means of glycerol-water solutions. All results obtained in the capillary viscometers were corrected for kinetic energy according to the Hagenbach-Couette equation. All viscosity determinations were carried out at 20°C.

A Beckman Zeromatic pH meter, furnished with a glass electrode, was used for pH determinations.

The uronic acid composition of alginates was determined by chromatography of the hydrolysates of alginate samples on an anion exchange column<sup>12</sup> and determination of the amounts of the separated uronic acids by a colour reaction. The detailed procedure will be published later.

## RESULTS

The effect of addition of sodium chloride, sodium sulphate and sodium carbonate to a solution of sodium alginate in water is shown in Fig. 1.

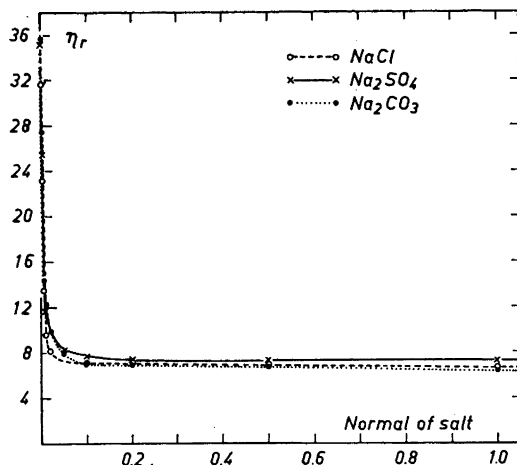


Fig. 1. The relative viscosity of a 0.125 % solution of sodium alginate (*L. digitata*, Munkaunet, 14/2-61,  $[\eta] = 27$ ) containing varying amounts of NaCl, Na<sub>2</sub>CO<sub>3</sub> and Na<sub>2</sub>SO<sub>4</sub> (Ubbelohde No. 2).

Table 1. The relative viscosity of a 0.125 % solution of sodium alginate (*Laminaria digitata*, Munkaunet, 14/2-61,  $[\eta] = 27$ ) containing potassium and sodium salts in a concentration of 0.25 N (Ubbelohde No. 2).

Salt	pH	$\eta_r$
NaCl	7.0	6.96
Na <sub>2</sub> CO <sub>3</sub>	11.2	6.87
KCl	6.4	6.88
KI	6.3	6.85
KBr	6.3	6.70
KNO <sub>3</sub>	6.3	7.17
K <sub>2</sub> CO <sub>3</sub>	11.1	6.89
K <sub>2</sub> SO <sub>4</sub>	7.5	7.01
K <sub>3</sub> PO <sub>4</sub>	11.9	6.46
Na-acetate	8.6	6.70
KH-phthalate	4.0	6.70
Na <sub>3</sub> -citrate	7.3	7.25
Dist. water	6.9	31.99

Table 2. The relative viscosity of 0.1 % solution of sodium alginate (*Laminaria digitata*, Munkaunet, 14/2-61,  $[\eta] = 27$ ) at various pH values. (Ubbelohde No. 2).

pH	Ionic strength	$\eta_r$	
4.95	0.04	5.68	
6.05	0.06	5.69	Phosphates
7.00	0.11	5.56	
8.21	0.15	5.48	
8.90	0.15	5.37	
9.90	0.15	5.36	
11.00	0.15	5.47	
12.00	0.15	5.46	

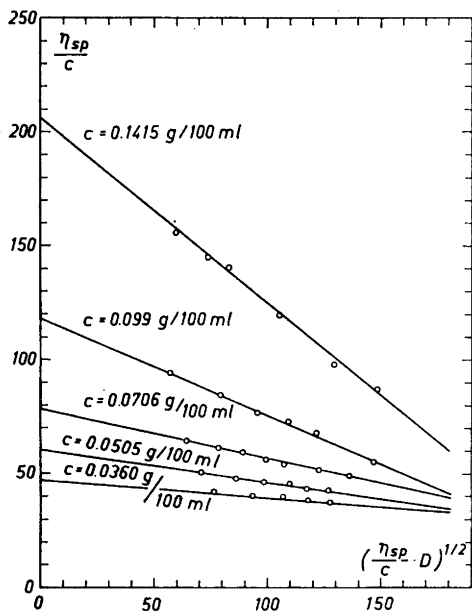


Fig. 2. Extrapolation to zero rate of shear, according to Golub, for sodium alginate from *L. digitata*, Tarva, 3/7-1957 ( $[\eta] = 30.0$ ).

The relative viscosity of solutions of sodium alginate containing various sodium and potassium salts in a concentration of 0.25 N is given in Table 1.

In Table 2 the relative viscosity of sodium alginate solutions at pH values varying from 5 to 12 is shown. The pH of the solutions was adjusted with phosphate or glycine buffers. Potassium chloride was added to the glycine buffer to obtain an ionic strength of approximately 0.15.

For the determination of intrinsic viscosity, 3% (0.565 N) sodium carbonate was chosen as a solvent, and the measurements were carried out in the Wagner-Russell viscometer. As rate of shear ( $D$ ) the maximum value, at the capillary wall, was used, and calculated according to the formula<sup>13</sup>

$$D = \frac{4V}{\pi R^3 t}$$

where  $V$  is the volume of the solution passed through the capillary with radius  $R$  in the time  $t$ . In order to extrapolate to zero rate of shear, the values of the reduced viscosity,  $\eta_{sp}/c$ , were plotted against

$$\left[ \left( \frac{\eta_{sp}}{c} \right) \cdot D \right]^{\frac{1}{2}}$$

according to Golub<sup>14</sup>. In Fig. 2 this is shown for five concentrations of a sodium alginate sample with intrinsic viscosity 30 prepared from *Laminaria digitata*. Assuming the curves to be linear over the whole range to  $D = 0$ , the values of

$$\left( \frac{\eta_{sp}}{c} \right)_{D=0}$$

were determined by extrapolation.

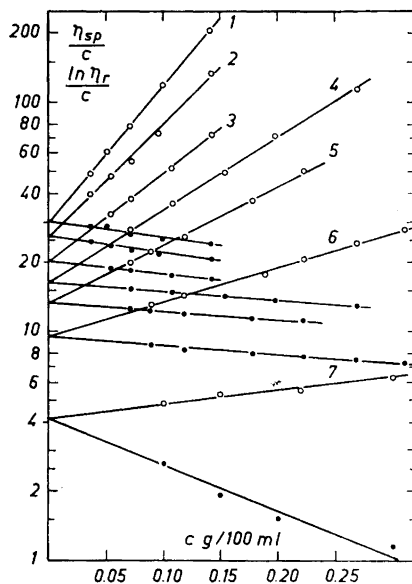


Fig. 3. Extrapolation to zero concentration of  $\eta_{sp}/c$  and  $(\ln \eta_r)/c$  for 7 different alginate samples.

1) <i>L. digitata</i> , Tarva, 3/7-57	$[\eta]$ : 30.0
2) » » Kråkvågøy, 3/7-57	$[\eta]$ : 26.0
3) » » Espevær, 8/1-53	$[\eta]$ : 20.1
4) <i>L. hyperborea stipes</i> , Hustad, 12/2-59	$[\eta]$ : 16.35
5) <i>L. digitata</i> , Munkaunet, 16/3-61 *	$[\eta]$ : 13.35
6) » » » 14/2-61 *	$[\eta]$ : 9.6
7) » » » 14/2-61 *	$[\eta]$ : 4.2

\* Treated with 1 N  $H_2SO_4$  at 60°C.

The intrinsic viscosity of alginate samples was determined by extrapolation to zero concentration of the curves of

$$\left(\frac{\eta_{sp}}{c}\right)_{D=0}$$

plotted against concentration in a semilogarithmic scale. To facilitate the extrapolation the values of  $[(\ln \eta_r)/c]_{D=0}$  were plotted in the same graph. The two expressions extrapolate to the same point at zero concentration. In Fig. 3 the extrapolation to zero concentration is shown for seven different alginate samples.

The importance of extrapolating to zero rate of shear is illustrated in Figs. 4 and 5, which show the extrapolation of  $\eta_{sp}/c$  and  $(\ln \eta_r)/c$  for different constant values of rate of shear and shear stress ( $\tau$ ) respectively, for the same alginate sample used for the determinations shown in Fig. 2. The curves for constant rate of shear were determined by interpolation of curves where

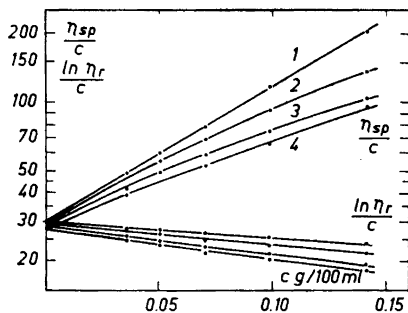


Fig. 4. Extrapolation to zero concentration of  $\eta_{sp}/c$  and  $(\ln \eta_r)/c$  for sodium alginate from *L. digitata*, Tarva, 3/7-1957 ( $[\eta] = 30.0$ ) for different constant values of rate of shear.

- |    |                            |                               |                     |
|----|----------------------------|-------------------------------|---------------------|
| 1: | $D = 0 \text{ sec}^{-1}$ , | $\tau = 0$                    | dyn/cm <sup>2</sup> |
| 2: | $D = 40 \text{ »}$         | $850 > \tau > 120 \text{ »}$  |                     |
| 3: | $D = 140 \text{ »}$        | $2200 > \tau > 220 \text{ »}$ |                     |
| 4: | $D = 260 \text{ »}$        | $3400 > \tau > 400 \text{ »}$ |                     |

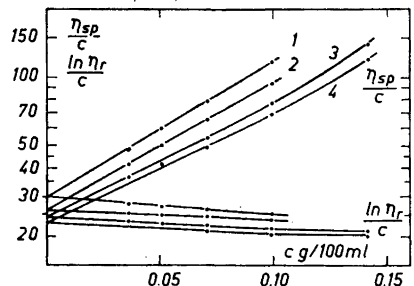


Fig. 5. Extrapolation to zero concentration of  $\eta_{sp}/c$  and  $(\ln \eta_r)/c$  for sodium alginate from *L. digitata*, Tarva, 3/7-1957 ( $[\eta] = 30.0$ ) for different constant values of shear stress.

- |    |                               |                           |                   |
|----|-------------------------------|---------------------------|-------------------|
| 1: | $\tau = 0 \text{ dyn/cm}^2$ , | $D = 0$                   | sec <sup>-1</sup> |
| 2: | $\tau = 355 \text{ »}$        | $140 > D > 35 \text{ »}$  |                   |
| 3: | $\tau = 1024 \text{ »}$       | $440 > D > 50 \text{ »}$  |                   |
| 4: | $\tau = 1682 \text{ »}$       | $380 > D > 100 \text{ »}$ |                   |

$\eta_{sp}/c$  was plotted as a function of  $D$  (see Ref. 9, p. 21). The same solutions as used for the determination of Fig. 3 were measured in a Höppler viscometer, and the "apparent intrinsic viscosities",  $[\eta]_H$ , determined by extrapolation.

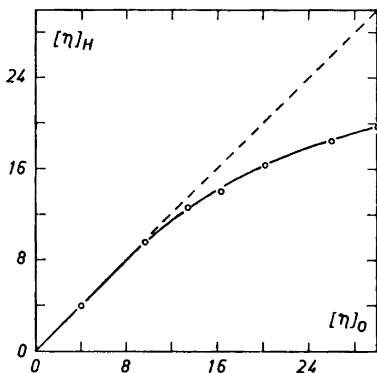


Fig. 6. The relation between the "apparent intrinsic viscosity"  $[\eta]_H$  determined from measurements in a Höppler viscometer and  $[\eta]_{D=0}$  determined by extrapolation to zero rate of shear.

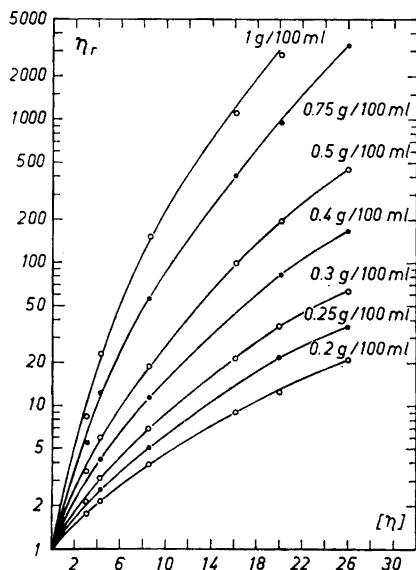


Fig. 7. Viscosity of alginate solutions of varying concentrations in 0.1 N NaCl for samples with different values of intrinsic viscosity (Ubbelohde No. 2).

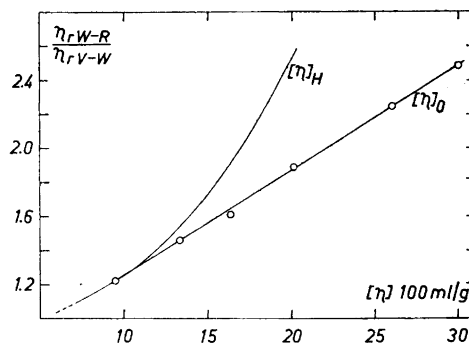


Fig. 8. The quotient between the apparent viscosities determined in the Wagner-Russell viscometer at the lowest rate of shear and in Höppler Visco-Waage with 10 g load as a function of  $[\eta]_{D=0}$  and  $[\eta]_H$ .

Fig. 6 gives the relation between  $[\eta]_H$  and intrinsic viscosity determined by extrapolation to zero rate of shear,  $[\eta]_{D=0}$ .

The correlation between the intrinsic viscosity and the relative viscosity of solutions of various concentrations in 0.1 N sodium chloride measured with Ubbelohde viscometer No. 2 is given in Fig. 7.

The non-Newtonian behaviour of alginate solutions is most pronounced at high values of intrinsic viscosity<sup>8</sup>. The quotient between the apparent viscosities of an alginate solution determined in two viscometers with different rate of shear may, therefore, empirically be correlated to the intrinsic viscosity of the alginate sample. Alginate solutions in 3% sodium carbonate were adjusted by dilution to an apparent relative viscosity between 4.8 and 5.2, measured in Höppler Visco-Waage with 10 g load. The apparent viscosity of the same solution was then determined in the Wagner-Russell viscometer at the lowest shear stress. The quotient between the two apparent viscosities  $\eta_{rW-R}/\eta_{rV-W}$  as a function of the intrinsic viscosity is shown in Fig. 8. The same quotient as a function of the "apparent intrinsic viscosity",  $[\eta]_H$ , determined from measurements in the Höppler viscometer is given in the same figure.

Table 3 gives the intrinsic viscosities of some commercial alginates, and some laboratory preparations.

Table 3.

	$[\eta]$
<i>Ascophyllum nodosum</i> , 17/3—57, Leangen	12.5
» » 3/7—57, Tarva	10.5
<i>Laminaria hyperborea</i> , fronds, 26/8, Munkholmen	19.0
» » stipes, 26/8, »	18.4
» » fronds, 24/8, »	13.2
» » stipes, 24/8, »	16.8
<i>Laminaria digitata</i> f. <i>flexicaulis</i> , 12/4, Flak	27.7
» » broad fronds, 12/4, »	20.0
Protanal /LF ( <i>Laminaria digitata</i> )	8.8
» /HF » »	14.0
Manucol SS/LM ( <i>Laminaria hyperborea</i> stipes)	10.5
» SH/LM ( <i>Ascophyllum nodosum</i> )	9.6
Noraltex MV ( <i>Laminaria digitata</i> )	7.9
Kelco SS ( <i>Macrocystis pyrifera</i> )	10.0

## DISCUSSION

Polyelectrolytes dissolved in water give solutions which show a marked drop in viscosity by addition of salts, and at the same time the curves of  $\eta_{sp}/c$  versus concentration changes to the shape normal for solutions of uncharged macromolecules.

In pure water at low concentrations the values of  $\eta_{sp}/c$  increase with decreasing concentration<sup>15</sup>. Alginate solutions have previously been shown to behave as typical polyelectrolyte solutions in this respect<sup>8</sup>. The very pronounced effect of addition of small amounts of inorganic salt shown in Fig. 1 should be taken into account when one wants to compare the viscosity of alginate samples. A difference between the viscosity of solutions of two different alginates in water may thus be caused by different amounts of salts as impurities in the two samples, and not by a difference in molecular weight. Determination of viscosity of solutions of alginate in water is, therefore, not suited for characterizing alginate samples. It may be mentioned, however, that the effect of salts is most pronounced at low concentrations of alginate. When the concentration of alginate is increased, the ionic strength increases, and at a sufficiently high alginate concentration (approximately 3%) an additional increase in ionic strength due to addition of inorganic salt to the solution will have little or no effect on the viscosity.

Harkness and Wassermann<sup>6</sup> and Thiele and Hallich<sup>16</sup> discussed the effect of the addition of different sodium salts on the viscosity of alginate solutions, and claimed that the decrease of viscosity depends on the anion of the sodium salt. As shown in Table 1 some difference between the different salts has been observed. The effect, however, is not very marked. It should be noted that all values of intrinsic viscosity given in this paper refer to 3% (0.565 N) sodium carbonate solutions. Referring to Fig. 1 and Table 1, it seems that the differences between the intrinsic viscosities in different salt solutions may be disregarded for most practical purposes, provided the amounts of salts are sufficient to reach the horizontal part of the curves in Fig. 1. For one of the samples used for the determination shown in Fig. 3 ( $[\eta] = 26.0$ ), the intrinsic viscosity was also determined in 0.1 N sodium chloride solution, in the way described above. No significant difference between the intrinsic viscosity in 0.1 N sodium chloride solution and 3% sodium carbonate solution could be observed.

Heen<sup>17</sup> and Bollinger and Münzel<sup>18</sup> claim that the viscosity of alginate solutions has a maximum at pH 7, while Donnan and Rose<sup>5</sup> found that the viscosity was independent of the pH of the solution between pH 6 and 8. Our results, given in Tables 1 and 2, show that the viscosity is independent of pH between pH 4 and 12, and the drop in viscosity observed by Heen and Bollinger and Münzel is probably an effect of increased ionic strength.

The extrapolation method of Golub<sup>14</sup> is derived from a formula of Bueche<sup>19</sup>:

$$\frac{\eta_{sp}}{c} = \left( \frac{\eta_{sp}}{c} \right)_{D=0} \cdot \left[ 1 - k \left( \frac{\eta_{sp}}{c} \cdot D \right)^{\frac{1}{2}} \right]$$

Golub investigated solutions of rubber in acetone at values of shear rate down to 1 sec<sup>-1</sup> and found that this formula was in good agreement with the experi-



mental results. As shown in Fig. 2, our results for alginate solutions, plotted according to Golub, gave straight lines in the range of shear rates investigated (20—550 sec<sup>-1</sup>). Even if the assumption that the formula is valid to zero rate of shear should not be correct, the extrapolation is well-defined and reproducible, and leads to values of intrinsic viscosity that are independent of the viscometer used for the measurements. Of the seven samples described in Fig. 3, only the one with the lowest intrinsic viscosity,  $[\eta] = 4.2$ , gave solutions which were found to be independent of rate of shear in the range investigated in this work.

The two expressions  $\eta_{sp}/c$  and  $(\ln \eta_r)/c$ , plotted against concentration on a semilogarithmic scale, give straight lines at low concentrations, as shown in Fig. 3. The extrapolation shown in Fig. 5, at different constant values of shear stress, corresponds to determinations of intrinsic viscosity in the conventional way, using capillary viscometers. The range of shear stress shown in Fig. 5 is very narrow and not outside those employed in ordinary laboratory viscometers of Ostwald or Ubbelohde types. As may be seen from Fig. 6, the values of "apparent intrinsic viscosity", determined from measurements in the Höppler viscometer differ significantly from those determined by extrapolation to zero shear rates for values of intrinsic viscosity higher than 10. It may be noted that the Höppler viscometer has been supposed to give values of apparent viscosity corresponding to zero rate of shear<sup>20</sup>.

Extrapolations at constant rate of shear, shown in Fig. 4, gave considerably higher values of "apparent intrinsic viscosity" than extrapolations at constant shear stress. This has earlier been shown to be the case for measurements made in Höppler Visco-Waage<sup>8</sup>. Instruments where the rate of shear is kept constant when measuring a series of solutions with varying polymer concentration, such as rotating cylinder viscometers, will, other conditions being equal, give values more close to the intrinsic viscosity at zero shear rate, than instruments of the capillary or falling ball type where the shear stress is kept constant. This will be the case for all solutions where the viscosity decreases with increasing rate of shear, according to the formula  $\eta = \tau/D$ .

The determination of intrinsic viscosity by extrapolation to zero rate of shear and zero concentration is a time-consuming process, and not possible to use for routine determinations. It is, however, possible empirically to correlate the intrinsic viscosity, determined as described above, with viscosity measurements carried out at well defined conditions (*e.g.* 0.3 % sodium alginate in 0.1 N sodium chloride solution, measured in Ubbelohde Viscometer No. 2 at 20°C). Correlation curves of this type, for several concentrations of alginate, are given in Fig. 7.

Another method, well suited for routine work, is to measure the apparent viscosity at two different values of rate of shear, and correlate the quotient of the two results with the intrinsic viscosity, as shown in Fig. 8. The solutions must be adjusted by dilution to a certain viscosity in one of the viscometers. This method can only be used for alginates with intrinsic viscosity over a certain value (with the set of instruments used in our laboratory higher than 7), but has the advantage of being independent of the concentration of alginate in the solution. We have in our laboratory found this method very useful, particularly for comparing different raw materials. The intrinsic viscosity can

be determined directly in the sodium carbonate extract, without isolation of the alginate.

Recent investigations have shown that the uronic acid composition of alginates from *Laminaria digitata* and *Laminaria hyperborea* stipes is appreciably different<sup>21</sup>. One of the samples used for the determination of the curve in Fig. 8 was prepared from *Laminaria hyperborea* stipes and the proportion between mannuronic and guluronic acid residues was approximately 0.6. The other samples were all prepared from *Laminaria digitata*, and the proportion between the two different uronic acid residues was approximately 2.5. As the result for the sample with much guluronic acid ( $[\eta]$ : 16.35) does not deviate from the curve, it may be concluded that the non-Newtonian behaviour of alginate solutions does not depend on the uronic acid composition of the alginate.

#### REFERENCES

1. Black, W. A. P. and Woodward, F. N. *Advances in Chem. Ser.* 11 (1954) 83.
2. Black, W. A. P., Cornhill, W. J. and Dewar, E. T. *J. Sci. Food Agr.* 3 (1952) 542.
3. Vincent, D. L., Goring, D. A. L. and Gordon Young, E. *J. Appl. Chem. (London)* 5 (1955) 374.
4. Vincent, D. L. *Can. J. Technol.* 34 (1956) 220.
5. Donnan, F. G. and Rose, R. C. *Can. J. Research* 28 B (1950) 105.
6. Harkness, M. L. R. and Wassermann, A. *J. Chem. Soc.* 1952 497.
7. Cook, W. H. and Smith, D. B. *Can. J. Biochem. and Physiol.* 32 (1954) 227.
8. Haug, A. *Report No. 8*, Norwegian Institute of Seaweed Research, Oslo 1955.
9. Haug, A. *Viscosity of Alginate Solutions*. In *Report No. 20*, Norwegian Institute of Seaweed Research, Trondheim 1958.
10. Haug, A. *Acta Chem. Scand.* 13 (1959) 601.
11. Wagner, R. H. and Russell, J. *Anal. Chem.* 20 (1948) 151.
12. Larsen, B. and Haug, A. *Acta Chem. Scand.* 15 (1961) 1397.
13. Wilson, K. *Svensk Papperstidn.* 54 (1951) 195.
14. Golub, M. A. *Can. J. Chem.* 35 (1957) 381.
15. Fuoss, R. M. and Strauss, U. P. *J. Polymer. Sci.* 3 (1948) 246.
16. Thiele, H. and Hallich, K. *Z. Naturforsch.* 13 B (1958) 580.
17. Heen, E. *Kolloid-Z.* 83 (1938) 204.
18. Bollinger, R. and Münzel, K. *Pharm. Acta Helv.* 33 (1958) 225.
19. Bueche, F. *J. Chem. Phys.* 22 (1954) 1570.
20. Schurz, J., Streitig, H. and Wurz, E. *Monatsh.* 87 (1956) 520.
21. Haug, A. *Colloques Internationaux du C.N.R.S.*, No. 103, Dinard 1960, p. 163. Paris 1961.

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