

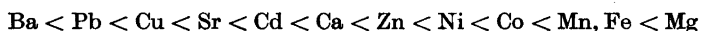
## The Effect of Divalent Metals on the Properties of Alginate Solutions

### II. Comparison of Different Metal Ions

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Addition of different divalent metal ions to solutions of sodium alginate, prepared from *Laminaria digitata* and the stipes of *Laminaria hyperborea*, revealed in each case that the concentration of the metallic ion required to bring about gel-formation and precipitation increased in the order:

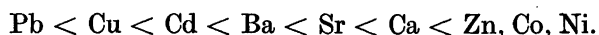


This order is not identical with that in which the ions are preferentially bound by the alginate. However, the efficacy of a divalent ion as a precipitant for alginate depends not only upon its affinity for the alginate, but also upon the amount of the ion which must be bound to the alginate for gel-formation to occur; it is shown that this last quantity is not independent of the nature of the ion.

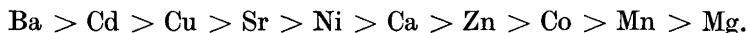
Added sodium ions markedly affect the concentration of divalent metal ion required for precipitation. When the divalent ion is strongly bound by the alginate, the effect of sodium ions is predominantly one of salting-out, whereas with weakly-bound cations the solubility of the alginate increases with increasing sodium-ion concentration, probably due to displacement of the ion-exchange equilibrium.

In the preceding paper the effect of added calcium ions on the properties of sodium alginate solutions was described. The ions of most divalent metals, like that of calcium, cause formation of gels and precipitates when added to alginate solutions, and the object of this paper is to compare the ion-exchange and precipitation properties of different divalent metals.

It is well known that different divalent metals show different affinities for alginate. Specker *et al.*<sup>1</sup> have studied alginate as an ion-exchange material. Thiele and Hallich<sup>2</sup> have arranged the divalent metals in an "ionotropic series" by measuring the capillary diameters in gels formed by allowing divalent metals to diffuse into alginate solutions. The diameter of the capillaries increased in the following order:



Thiele and co-workers also studied the ion exchange properties of alginates<sup>3</sup> and the precipitation of alginate by some divalent metals<sup>4</sup> (Pb, Cu, Cd, Ca, and Zn) and found that also for these properties the ionotropic series was followed. Recently Schweiger<sup>5</sup> studied the stability of different metal-alginate complexes by determining the viscosity of solutions obtained by adding EDTA to the insoluble alginates. He found that the stability decreased in the following order:



Magnesium alginate has been found to be soluble in water,<sup>6,4</sup> although light-scattering studies seem to indicate that magnesium alginate molecules are more compact than those of sodium alginate.<sup>7</sup>

In these investigations no attempts were made to distinguish between alginates from different sources or to determine whether there exists any correlation between the uronic acid composition and ion-exchange properties of the alginate.

Müller<sup>8</sup> investigated the ion-exchange between divalent metals and sodium for some alginates prepared from different raw materials and found the following series:

*L. hyperborea stipes*: Cu > Ba > Ca > Zn > Mn

*Macrocystis pyrifera*: Cu > Ca > Ba, Zn

*L. digitata*: Cu > Ba > Zn > Ca.

For Cu and Zn the degree of ion exchange was independent of the raw material, but for Ba and Ca the affinity for divalent metal decreased in the order, *L. hyperborea stipes*, *Macrocystis pyrifera*, *L. digitata*. Müller did not discuss the existence of residues of guluronic acid in the alginate and could not explain the observed difference.

McDowell,<sup>9</sup> studying alginates from *L. hyperborea stipes* and *Ascophyllum nodosum*, found that the minimum amount of divalent metal that started precipitation increased in the following order: Pb, Cu < Ca < Ni, Co, Zn < Mn. Smaller amounts of Pb and Ca were required to precipitate alginate from *L. hyperborea stipes* than from *A. nodosum*. By addition of sodium nitrate less Pb and more Ca was needed to bring about precipitation.

Haug,<sup>10</sup> measuring the decrease in pH brought about by addition of divalent metals to half-neutralized alginic acid, found that for alginate prepared from *L. digitata* (rich in mannuronic acid) the affinity series was different from that for alginate prepared from *L. hyperborea stipes* (rich in guluronic acid). The following series were obtained:

*L. digitata*: Pb > Cu > Cd > Ba > Sr > Ca > Co, Ni, Zn, Mn > Mg.

*L. hyperborea stipes*: Pb > Cu > Ba > Sr > Cd > Ca > Co, Ni, Zn, Mn > Mg.

The affinity follows the ionotropic series of Thiele only for *L. digitata*. The selectivity coefficient for the ion exchange reaction,  $\text{SrAlg}_2 + \text{Cd}^{2+} \rightleftharpoons \text{CdAlg}_2 + \text{Sr}^{2+}$ , was found to be higher than unity for alginates from *L. digitata* and lower than unity for alginate from *L. hyperborea stipes*, in agreement with the two series given above.

In the present paper the effects of different divalent metals on solutions of sodium alginate are compared by viscosity measurements, precipitation experiments, and by determining selectivity coefficients.

### EXPERIMENTAL

The preparation of the alginate solutions and the viscosity and precipitation experiments were described in the preceding paper.

The following divalent metals were investigated: Ba, Pb, Cu, Sr, Cd, Ca, Zn, Ni, Co, Mn, Fe, and Mg. In the viscosity experiments the anions of the salts were sulphates (for Zn, Mn, Fe, and Mg) and nitrates (for the other metals). The alginate solutions contained sodium nitrate before the addition of divalent metals.

In some centrifugation experiments with Cu, Ca, and Mn, the concentration of alginate in the soluble phase was determined by the phenol-sulphuric acid colour reaction.<sup>11</sup> The reaction was affected by nitrate in the solutions. Because of this, NaCl, CuCl<sub>2</sub>, CaCl<sub>2</sub>, and MnSO<sub>4</sub> were used in these experiments.

In order to determine the selectivity coefficients for the ion exchange systems Na—Cu, Na—Ba, Na—Co, and Na—Ca the method described earlier for calcium was used.<sup>12</sup> All the divalent metals were titrated with EDTA according to Schwarzenbach.<sup>13</sup>

The fractionation experiment with calcium chloride was carried out in the following way: Portions of 15 ml 0.2% alginate solutions containing 1 N NaCl were mixed with equal amounts of 0.012 M CaCl<sub>2</sub> and stirred with a mechanical stirrer for 30 sec. After centrifugation and decantation, the gel and solutions were precipitated with ethanol, washed with ethanol and ether and dried overnight at 40°C. The ratio of mannuronic to guluronic acid, M/G, was determined as described earlier,<sup>14</sup> and the results are corrected for the different rate of breakdown during hydrolysis.

Alginate from two different raw materials was used throughout this work: *L. digitata*, Tarva 3/7—57 and *L. hyperborea* stipes, Hustad 12/2—59. The M/G-ratios are given in Table 1.

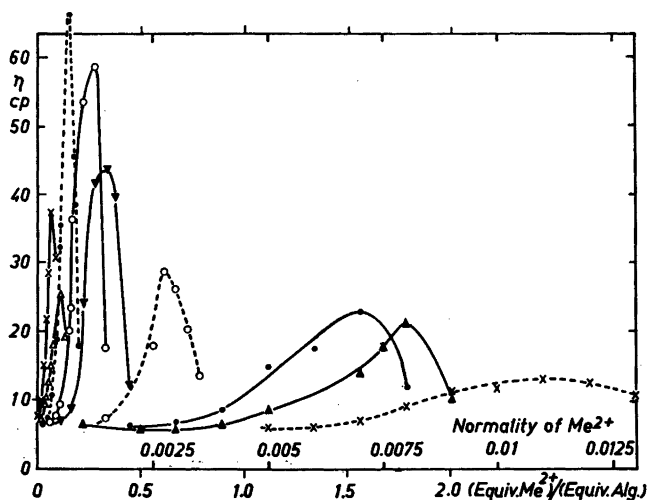


Fig. 1. Viscosity of 0.1% (0.0045 N) alginate solutions containing divalent metals. Alginate from *L. digitata*. Sodium nitrate concentration 0.05 N.

× — × = Ba; Δ — Δ = Pb; ● — — ● = Cu; ○ — — ○ = Sr; ▼ — — ▼ = Cd;  
○ — — ○ = Ca; ● — — ● = Zn; ▲ — — ▲ = Ni; × — — × = Co.

## RESULTS

Alginate solutions were mixed with increasing amounts of different divalent metals and the viscosity was determined. In Fig. 1 the results are given for mixtures containing 0.1 % alginate and 0.05 N sodium nitrate. The alginate was prepared from *L. digitata*. Results from similar experiments with alginate prepared from *L. hyperborea* are given in Fig. 2. At a certain concentration of divalent metal the viscosity reached a maximum value. This effect was de-

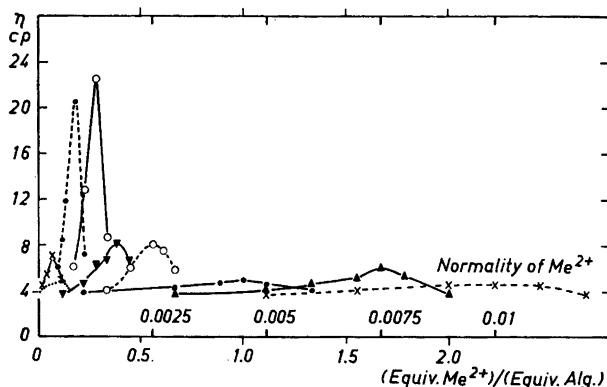


Fig. 2. Viscosity of a 0.1 % alginate solution containing divalent metals. Alginate from *L. hyperborea* stipes. Sodium nitrate concentration 0.05 N.

..... = Pb. The other symbols are the same as in Fig. 1.

scribed for calcium in the preceding paper. The concentration giving the maximum viscosity for both alginates increased in the following order:



The effects of manganous, ferrous and magnesium salts were also investigated. With these three metals, no increase in viscosity was observed. At concentrations above 0.05 N  $\text{MnSO}_4$  the viscosity was slightly reduced and a flocculent precipitate observed. The experiment with ferrous salt was not always reproducible because the autoxidation of the ferrous ions caused both degradation of the alginate<sup>15</sup> and some precipitation by the ferric ions formed. The concentration range at which precipitation occurred was in the same order as that for  $\text{MnSO}_4$  (0.05–0.1 N). Magnesium sulphate gave no decrease of viscosity and no precipitation could be observed.

It is well known that divalent metals have different affinities for alginate. If this difference alone determines the metal ion concentration giving maximum viscosity, the corresponding amount of divalent metal bound to the alginate should be equal in each case. A calculation of the amount of calcium bound to the alginate at maximum viscosity showed that this quantity was markedly higher than the total amount of barium added at maximum viscosity. Thus, both the different ion exchange properties and the different gel forming ability

Table 1. Selectivity coefficients,  $k$ , for different ion exchange reactions.

$$k = \frac{Me_g \cdot [Na_1]^2}{[Me_1] \cdot Na_g^2}$$

Metal ions	<i>L. digitata</i> , Tarva 3/7-57. M/G = 1.60	<i>L. hyperborea stipes</i> , Hustad 12/2-59. M/G = 0.45
Cu <sup>2+</sup> -Na <sup>+</sup>	230	340
Ba <sup>2+</sup> -Na <sup>+</sup>	21	52
Ca <sup>2+</sup> -Na <sup>+</sup>	7.5	20
Co <sup>2+</sup> -Na <sup>+</sup>	3.5	4

of the metal ions bound to the alginate is responsible for the different precipitation effects of the divalent metals.

In order to distinguish between these two effects, the selectivity coefficient for the ion exchange reaction between sodium and divalent metal was determined for the two alginates. The values given in Table 1 show that the metals differ markedly in ion exchange properties, and that alginate from *L. hyperborea stipes* has the higher affinity for divalent metal.

The total concentration of the divalent metal at maximum viscosity,  $Me_{max}$ , can be taken as a measure of the precipitation property of the metal. In Table 2 the values of  $Me_{max}$ , given as the equivalent ratios between the total amount of divalent metal and alginate, are given for the experiments shown in Figs. 1 and 2. The equivalent fraction of divalent metal bound to the alginate at maximum viscosity,  $Me_{g max}$ , was calculated for some metals using the selectivity coefficients in Table 1. The results given in Table 2 show that the  $Me_{g max}$  values increase in the sequence Ba < Cu < Ca < Co for both alginates.

Table 2. Viscosity maximum for different divalent metals. Concentration of alginate: 0.1 %, 0.0045 equiv./l.

Metal	<i>L. hyperborea stipes</i> , Hustad 12/2-59. [ $\eta$ ] = 16.4, M/G = 0.45 0.05 N sodium nitrate.		<i>L. digitata</i> , Tarva 3/7-57. [ $\eta$ ] = 26, M/G = 1.60			
	$Me_{max}$	$Me_{g max}$	$Me_{max}$	$Me_{g max}$	$Me_{max}$	$Me_{g max}$
Ba	0.067	0.066	0.067	0.065	0.033	0.025
Pb	0.111		0.111		0.045	
Cu	0.178	0.178	0.155	0.155	0.13	0.12
Sr	0.278		0.278		0.33	
Cd	0.378		0.333		0.67	
Ca	0.555	0.49	0.61	0.47	1.65	0.39
Zn	1.00		1.56		2.45	
Ni	1.66		1.78		3.33	
Co	2.22	0.72	2.45	0.71	5.55	0.50

The alginates have so far been tested at a sodium nitrate concentration of 0.05 N. The alginate from *L. digitata* was also investigated with 0.2 N sodium nitrate in the mixture. The results are given in Table 2. If the  $Me_{g \max}$ -values at 0.2 and 0.05 N sodium nitrate are compared, it is seen that the amount of divalent metals bound to the alginate at maximum viscosity ( $Me_{g \max}$ ) is lower at the highest concentration of sodium salt. This is due to the "salting-out" effect of the sodium salt, as described for calcium in the preceding paper. If the total amounts of divalent metals present,  $Me_{\max}$ , are compared, it is seen that this quantity decreases with increasing sodium nitrate concentration for Ba, Pb, and Cu but increases for the other metals which have lower affinity for alginate.

The effect of sodium salt was further investigated for copper and cobalt ions. In Fig. 3 the total amount of added divalent metal at maximum viscosity,  $Me_{\max}$ , and the calculated amount bound to the alginate,  $Me_{g \max}$ , are given as a function of the sodium nitrate concentration. The  $Me_{g \max}$ -values decrease

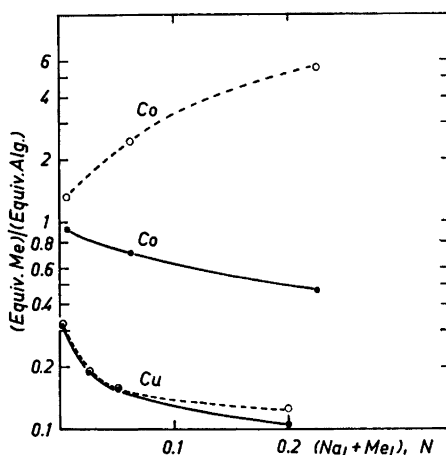


Fig. 3. Amounts of cobalt and copper giving viscosity maximum at different sodium nitrate concentrations. Alginate from *L. digitata*, 0.1 %.

--- = equiv. fraction added,  $Me_{\max}$   
 — = » » bound to  
 alginate,  $Me_{g \max}$

with increasing ionic strength for both metals, while the  $Me_{\max}$ -values decrease for Cu and increase for Co in agreement with the results given in Table 2.

The magnitude of the maximum viscosity decreases markedly as the ionic strength increases and makes determinations less accurate. The effect of sodium ions at higher concentrations was studied by means of precipitation experiments as described for calcium in the preceding paper. The effects of different concentrations of copper and cobalt ions in mixtures containing 0, 0.05, 0.2, and 1.0 N sodium chloride were determined. In Fig. 4 the volume-fraction of the gel after centrifugation is given. For both metals, a loose gel occupying the total volume of the mixture is obtained at a low sodium ion concentration. When the sodium ion concentration is increased the maximum volume fraction of the gel phase decreases. Increasing ionic strength has quite a different effect for copper and cobalt. Less copper and more cobalt is necessary to obtain

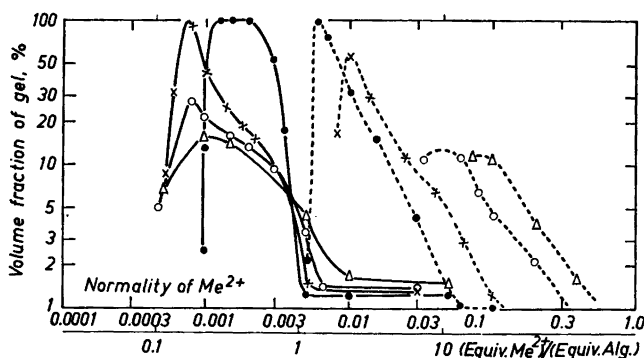


Fig. 4. Centrifugation experiment with copper and cobalt nitrate. Alginate from *L. digitata*, 0.1 %.

● = 0 N NaCl; × = 0.05 N NaCl; ○ = 0.2 N NaCl; △ = 1.0 N NaCl.  
 — = Cu; - - - = Co.

the maximum gel volume, as was the case when the viscosity maximum was considered.

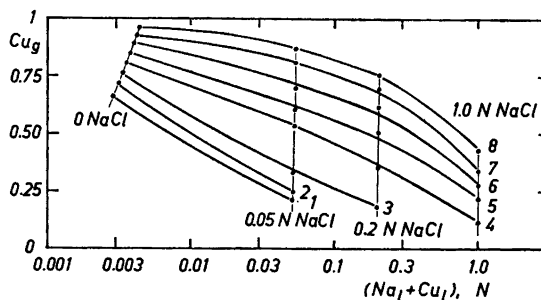


Fig. 5. The correlation between  $Cu_g$ , gel volume and concentration of sodium chloride in the solution (calculated from Fig. 4). Gel volume: 1 = 60 %; 2 = 40 %; 3 = 25 %; 4 = 15 %; 5 = 10 %; 6 = 7 %; 7 = 4 %; 8 = 2 %.

$Me_g$ -values corresponding to a number of different volume fractions were calculated. The results, given for Cu in Fig. 5 and for Co in Fig. 6, show that the  $Me_g$ -values necessary for obtaining a certain volume fraction of the gel decrease for both metals as the ionic strength increases.

It can be seen from Fig. 4 that increasing ionic strength tends to make the difference between the precipitation effect of Cu and Co larger. This is due to the large difference in the selectivity coefficient of the two metals. Using calculated  $Me_g$ -values one can compare the effect of the bound divalent metals on the gel volume, and thus eliminate the difference in selectivity coefficient of the metals. In Fig. 7 the gel volume is given as a function of  $Co_g$  and  $Cu_g$

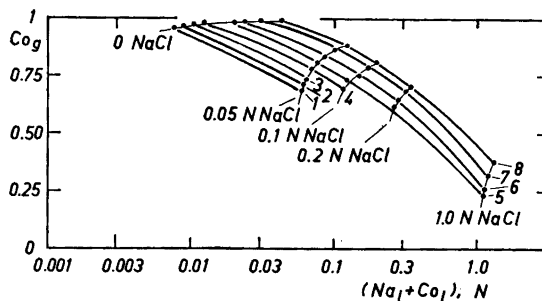


Fig. 6. The correlation between  $Co_g$ , gel volume and concentration of sodium chloride in the solution (calculated from Fig. 4). Volume fraction of gel as in Fig. 5.

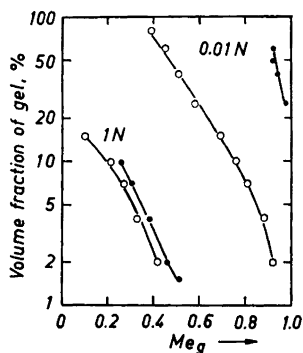


Fig. 7. Comparison at two different ionic strengths of the effect of bound cobalt and copper on the gel volume.

○ = copper; ● = cobalt.

at high (1 N) and low (0.01 N) concentrations of free cations. The difference between the effects of the two metals is markedly lower at the highest ionic strength, showing that the "salting-out" effect of the sodium salt is a major part of the total precipitation effect of the system at this concentration.

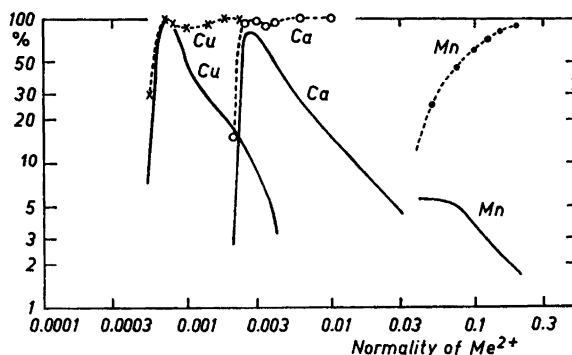


Fig. 8. Precipitation of alginate with divalent metals. Alginate from *L. digitata*, 0.1%. Sodium chloride concentration 0.05 N. — = volume fraction of gel; - - - = percentage of alginate precipitated.

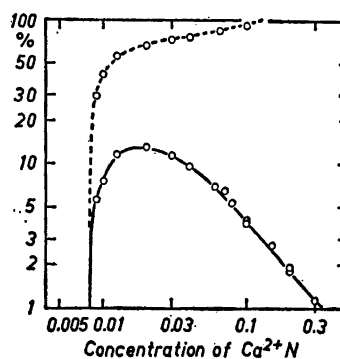


Manganous salt is used to fractionate alginate into a guluronic-rich and a mannuronic-rich fraction.<sup>16</sup> Precipitation experiments were also carried out with this metal. In Fig. 8 the volume fraction of gel and the fraction of alginate precipitated are given for Cu, Ca, and Mn in 0.05 N sodium chloride. It is seen from the figures that Mn in all cases gives a low volume fraction of gel and that a separation into a soluble and insoluble fraction is easily obtained by choosing a suitable concentration of manganous salts. Cu and Ca is clearly unsuitable for fractionation.

However, at higher ionic strength the decreased maximum volume fraction of gel result in a better fractionation curve for both Cu and Ca. This is demonstrated in Fig. 9 where calcium is used with 0.5 N sodium chloride in

Fig. 9. Precipitation of alginate with calcium at high ionic strength (0.5 N NaCl) Alginate from *L. digitata*, 0.1 %.

— = volume fraction of gel; - - -  
= percentage of alginate precipitated.



the mixture. The M/G-ratios of the two fractions, after precipitation with 0.012 N calcium chloride, were determined. The results given in Table 3 show that the precipitated part of the alginate is richer in guluronic acid than the soluble part.

#### DISCUSSION

In the preceding paper the effect of calcium on the viscosity of sodium alginate solutions was described. When calcium is added to the solution, the viscosity increases and passes through a maximum. Precipitation experiments have shown that the viscosity maximum corresponds to a loose gel structure

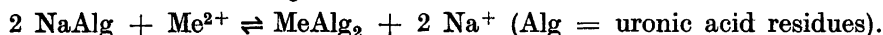
Table 3. Fractionation of alginate from *L. digitata*, Tarva 3/7-57. Concentrations in the mixture: 0.012 N CaCl<sub>2</sub>, 0.5 N NaCl.

	Alginate fractions		M/G
	mg	%	
Before fractionation	390		1.60
Precipitated phase	170	43.6	1.44
Soluble phase	168	43.2	2.0

occupying the total volume of the mixture. Further addition of calcium leads to a decrease in the volume of the gel and viscosity measurements become irreproducible as the mixture becomes heterogeneous.

In the present paper similar experiments with a number of different divalent metals are reported. With the exception of magnesium, manganous and ferrous ions, all divalent metals tested gave a viscosity maximum at a certain concentration of metal ions. This concentration ( $Me_{\max}$ ) is very different for the different metals (Figs. 1 and 2), and may be regarded as characteristic of the precipitation effect of the metal. By comparing the results given in Figs. 1 and 2 with the wellknown affinity series of alginates,<sup>10</sup> it is seen that the two series are not identical.

The total amount of divalent metal in the mixture at maximum viscosity ( $Me_{\max}$ ) is the sum of the divalent metal ions in the solution ( $Me_{\text{max}}$ ) and those bound to the alginate ( $Me_{g \max}$ ) according to the ion-exchange reaction:



When the selectivity coefficient of the ion-exchange reaction is known, it is possible to calculate the amount of metal bound to the alginate ( $Me_{g \max}$ ). The results in Table 2 show that the amount of metal which must be bound to the alginate to obtain gel formation,  $Me_{g \max}$ , differs from metal to metal. Thus, the precipitation effect of a certain divalent metal is determined by two different factors: 1) the affinity of the metal for alginate, which determines the amount of metal bound to the alginate ( $Me_g$ ) in a solution containing a certain concentration of metal ions and 2) the gel-forming ability of the metal, which determines the amount of metal which must be bound to the alginate to obtain a certain stage of insolubility.

In the preceding paper the effect of sodium ions on the gel formation of alginate with calcium ions was discussed. The sodium ions were found to have two oppositely directed effects: 1) that of displacing the ion-exchange equilibrium in the direction of lower values of  $Ca_g$  and 2) a "salting-out" effect due to the increased ionic strength. Table 2 and Fig. 3 illustrate the same effects for some other divalent metals. For metals with high affinity for alginate, *e.g.* copper, the "salting-out" effect predominates and less copper is needed for obtaining gel formation when the sodium chloride concentration is increased. For metals with low affinity, *e.g.* cobalt, the ion exchange effect predominates, and more cobalt ions are necessary for gel formation at higher sodium ion concentrations. Also, the amount of ions bound to the alginate at the viscosity maximum decreases with increasing sodium chloride concentration.

The same effects of increasing sodium ion concentration are illustrated in the precipitation experiments in Fig. 4, where the gel volume after centrifugation was determined as a function of the total amount of divalent ions added to the mixture. In Figs. 5 and 6 the same results are given as a correlation between free cations and  $Me_g$  for certain volume fractions of gel.

The selectivity coefficients given in Table 1 show that the guluronic acid-rich alginate has a higher affinity for divalent metals than the mannuronic acid-rich sample. This difference has been utilized in the fractionation of alginate with manganous ions.<sup>16</sup> Manganous ions have been chosen on account

of the flocculent precipitate formed. The formation of a voluminous gel occupying the total volume of the mixture, as is the case with most other divalent metals, makes fractionation impossible (see Fig. 8). However, the formation of voluminous gels with these divalent metals is avoided at higher ionic strength. Fractionation is possible under such conditions using calcium as the divalent metal (see Table 3).

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