g. of tartronic acid and made up to 50 ml. with water. In about thirty minutes iodine appeared in the solution. A 2-ml. aliquot was withdrawn periodically for an iodate determination. Sodium bicarbonate was added to an aliquot and the iodine present was reduced by titrating with 0.1 N arsenite in the presence of starch indicator. Acid and potassium iodide were added and the liberated iodine which is a measure of the remaining iodate was titrated with thiosulfate. The consumption of 0.115 ME of iodate was observed in six hours and 0.432 ME in five days.

Isolation and Characterization of Formic, Oxalic and Propionic Acids.—Five grams of malonic acid and 32 g. of sodium metaperiodate (3.1 ME) were permitted to react in a total volume of 500 ml. aqueous solution for six hours. The mixture was continuously extracted with ether for four days after which an aqueous layer had separated from the ether layer. Evaporation of the ether layer to dryness yielded 270 mg. of oxalic acid dihydrate, m. p. 101°. A mixed melting point with an authentic sample of oxalic acid showed no depression. Calcium chloride was added to 10% of the aqueous fraction described above and 45 mg. of calcium oxalate was obtained which gave 710 mg. as the total yield of oxalic acid dihydrate or 12%. The remainder of the aqueous phase was acidified with phosphoric acid and distilled to a small volume. An aliquot of the distillate was diluted to 0.01 N and the Duclaux constants[24] were 0.110, 0.141 and 0.220. An authentic sample of formic acid give 0.116, 0.149 and 0.227. The remaining aqueous fraction was neutralized with sodium hydroxide and evaporated to dryness. The resultant sodium salt was converted to the *p*-toluidide, m. p. 55°.

In a similar experiment 2 g. of malonic acid was oxidized by 10 ME of periodate and 2.6 mg. of calcium oxalate was obtained.

Five grams of α-ethyl malonic acid was oxidized as described above with 35 g. of sodium metaperiodate for three days. After ether extraction the ether layer was extracted with sodium hydroxide which in turn was acidified with phosphoric acid and distilled to a small volume. The total amount of volatile acid calculated as propionic acid was 2 g., a 71% yield. Duclaux constants determined on part of this solution were 0.278, 0.257 and 0.228. An authentic sample of propionic acid gave 0.282, 0.260 and 0.230. The sodium salt of this acid was converted to the *p*-toluidide, m. p. 122°.

Action of Periodate on Diethyl Malonate.—Five ml. of diethyl malonate was dissolved in a liter of water containing 3.5 ME of sodium metaperiodate. After four days at room temperature the solution was continuously extracted with ether. The ether was concentrated and the residue distilled. Three grams was collected: b. r. 185–190°; N, 1.4139; calcd. 1.4143.

Anal. Calcd. for $C_7H_{12}O_4$: C, 52.5; H, 7.5. Found: C, 52.3; H, 7.4.

Acknowledgment.—We wish to thank Professor K. P. Link and Professor C. A. Elvehjem for their helpful criticism and advice and Professor A. L. Wilds who reviewed a preliminary draft of this manuscript.

Summary

A new mode of oxidation by periodate in aqueous solution is described which consists in the conversion of an activated α-hydrogen to an α-hydroxyl group. The configuration necessary is a three-carbon system consisting of a free carboxyl or aldehyde, and α-carbon bearing at least one hydrogen, and a β-carbonyl group which may be part of an aldehyde, ketone, carboxyl, carbalkoxyl or a similar activating structure. This first reaction is then followed by the well-known oxidative cleavage of α-hydroxy acids and α-hydroxy aldehydes. Using malonic acid as a type compound, intermediates were secured and the mechanism indicated. The influence of structural modification on the oxidation is presented from which generalizations are made as to the specificity of periodate action.

MADISON, WISCONSIN RECEIVED APRIL 1, 1946

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[24] Duclaux constants were determined by a modification of the method of A. I. Virtanen and L. Pulkki, THIS JOURNAL, **50**, 3138 (1928).

[CONTRIBUTION FROM THE WESTERN REGIONAL RESEARCH LABORATORY[1]]

Shape and Size of Pectinic Acid Molecules Deduced from Viscometric Measurements

BY H. S. OWENS, H. LOTZKAR, T. H. SCHULTZ AND W. D. MACLAY

Until the 1930's chemical evidence had generally been interpreted to indicate that pectin has a small cyclic structure.[2] In 1923 Smolenski, however, offered the opinion that pectin is a high polymer comparable in structure to starch. X-Ray results agreed with that hypothesis, but indicated that comparison with cellulose is more logical. Schneider and co-workers showed that pectin nitrate in acetone solutions has a Staudinger constant between that for cellulose and starch nitrates. Later, Säverborn[3] and Snellman and Säverborn,[4] on the basis of measurements of flow birefringence, sedimentation and diffusion constants of various pectins and of pectin nitrates, also concluded that pectin has a high molecular weight and is a long chain compound like cellulose.

Shape and size of molecules of high polymers deduced from viscometric measurements of their solutions appear to agree with those calculated from measurements of other types.[5,6] Recent studies[7,8] of the viscosity behavior of pectic materials in electrolytic solutions have made it possible to determine their intrinsic viscosities. It appeared desirable to use these values for cal-

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[1] Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture. Article not copyrighted.

[2] The history of the development of pectin structure is reviewed in detail by K. H. Meyer, "High Polymers," Series 4, Interscience Publishers, Inc., New York, N. Y., 1942, pp. 364–370.

[3] (a) S. Säverborn, *Kolloid Z.*, **90**, 41 (1940). (b) Also "A Contribution to the Knowledge of the Acid Polyuronides," (thesis), Uppsala (1945). The latter publication appeared after the present manuscript was written.

[4] O. Snellman and S. Säverborn, *Kolloidchem. Beihefte*, **52**, 467 (1941).

[5] M. A. Lauffer, *Chem. Rev.*, **31**, 561 (1942).

[6] R. Simha, *J. Chem. Phys.*, **13**, 188 (1945).

[7] L. Malsch, *Biochem. Z.*, **309**, 283 (1941); *C. A.*, **37**, 4280 (1943).

[8] H. S. Owens, H. Lotzkar, R. C. Merrill and M. Peterson, THIS JOURNAL, **66**, 1178 (1944).

culations, by means of available equations, of the molecular size and shape of pectinic acids. For comparative purposes, number average molecular weights determined from osmotic pressure measurements are also presented.

Experimental

The methods of preparation and purification of the pectinic acids used in this investigation were the same as used before.[8,9] Some of the pertinent information on them is given in Table I.

TABLE I

ANALYTICAL DATA FOR THE PECTINIC ACID SAMPLES USED IN THIS INVESTIGATION

Pectinic acid[a]	Source	Uronic anhydride, %
10.7 No. 1	Citrus	85
10.7 No. 2	Citrus	80
10.7 No. 3	Citrus	87
10.5	Citrus	80
10.9	Apple	84
6.6A	Apple	86
7.9E	Citrus	81
5.8E	Citrus	82
5.2E (*in vitro*)	Citrus	87
4.4E	Citrus	83
3.6E	Citrus	83
0.7E	Citrus	82
7.8B	Citrus	86
6.6B	Citrus	87
6.2B	Citrus	87

[a] The sample numbers refer to the methoxyl content. The letter following the number stands for the method of deësterification: A = acid (Olsen, Stuewer, Fehlberg, and Beach; see ref. 13); B = base (R. M. McCready, H. S. Owens and W. D. Maclay, *Food Ind.*, **16**, 794 (1944)); E = enzyme (H. S. Owens, R. M. McCready and W. D. Maclay, *Ind. Eng. Chem.*, **36**, 936 (1944)).

The viscosity technique has been described in detail elsewhere.[8] Viscosity measurements were made on pectinic acids dissolved in 0.155 *M* sodium chloride solutions by means of Ostwald–Cannon–Fenske pipets[10] which were calibrated with various standards. All measurements were made in water baths at temperatures regulated to ±0.03°. The so-called natural *p*H is the *p*H of the pectinic acid solution without added base or acid. It is near 3 for the samples and concentrations used. Other *p*H values were obtained by partial neutralization of the most concentrated solution of the series after addition of sodium chloride. Dilutions made from that were not adjusted further.

Osmotic pressure measurements were made at 25°, using Lucite or stainless steel cells. The cells differ from those designed by Flory[11a] by having a series of concentric ridges[11b] instead of island supports for the membrane. The solvent was 0.155 *M* sodium chloride, a concentration that reduces the electroviscous effect to a minimum.[12] Membranes were prepared from Cellophane 300 and 450 PT, by swelling them in 7 *N* ammonium hydroxide for one hour, followed by washing with distilled water. Equilibrium was usually reached in a few hours, but the measurements were continued for a period of forty-eight hours.

Results

Figure 1 shows a semi-log plot of (η_{sp}/c) *vs.* c for some of the samples, where η_{sp} is the specific viscosity and c is in g./100 ml. In most cases the measurements were made at *p*H 6 because of the tendency of low-methoxyl pectinic acids to aggregate at low *p*H values.[9,13,14,15] The slopes of the lines show a tendency to increase with decreasing *p*H, becoming significantly greater at the natural *p*H with pectinic acids which have high intrinsic viscosities. The intercepts of the lines in the figure are the intrinsic viscosities which are believed to be a function of molecular chain length.

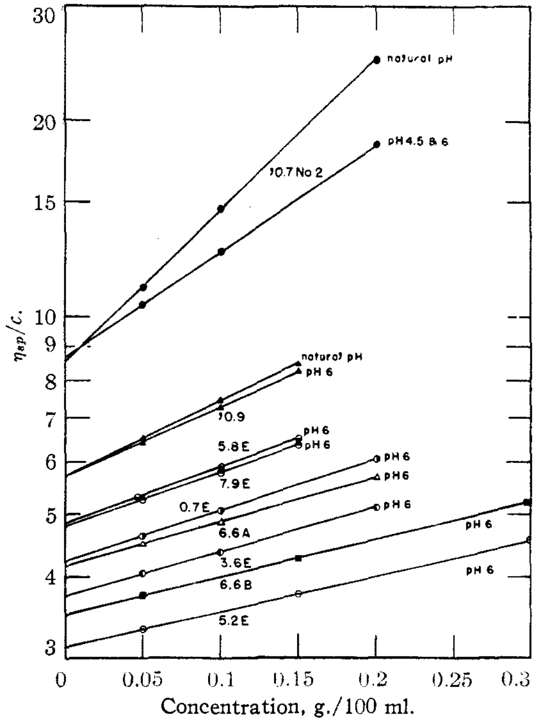

Fig. 1.—Plot of log of η_{sp}/c against concentration of various pectinic acids in the presence of 0.155 *M* sodium chloride.

Values of k', which are dependent upon the size and shape of solute submolecules and the co-

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(9) T. H. Schultz, H. Lotzkar, H. S. Owens and W. D. Maclay, *J. Phys. Chem.*, **49**, 554 (1945).

(10) M. R. Cannon and M. R. Fenske, *Ind. Eng. Chem., Anal. Ed.*, **10**, 297 (1938).

(11) (a) P. J. Flory, THIS JOURNAL, **65**, 372 (1943); (b) R. E. Montonna and L. T. Jilk, *J. Phys. Chem.*, **45**, 1374 (1941).

(12) H. Lotzkar, T. H. Schultz, H. S. Owens and W. D. Maclay, *J. Phys. Chem.*, **50**, 200 (1946).

(13) A. G. Olsen, R. F. Stuewer, E. R. Fehlberg and N. M. Beach, *Ind. Eng. Chem.*, **31**, 1015 (1939).

(14) G. L. Baker and M. W. Goodwin, *Agr. Expt. Sta. Del., Bull.*, 234 (1941).

(15) R. Speiser, C. H. Hills and C. R. Eddy, *J. Phys. Chem.*, **49**, 328 (1945).

hesional forces of solvent molecules and solute submolecules,[16] were obtained from the exponential equation $\eta_{sp}/c = [\eta]e^{k'[\eta]c}$, where $[\eta]$ is the intrinsic viscosity. They are shown in Tables II and III and were calculated by the method of least mean squares.

The effect of temperature on $[\eta]$ and k' of three pectinic acids at *p*H 4.5 is shown in Table II. A decrease in $[\eta]$ and k' is obtained on increasing the temperature. Changes in the same direction have been noted with polyvinyl chloride in cyclohexanone[17] and with a number of other colloidal materials.[18]

TABLE II

EFFECT OF TEMPERATURE ON $[\eta]$ AND k' OF PECTINIC ACIDS

T, °C.	k'	$[\eta]$
	Pectinic acid 10.7 No. 3	
10	0.41	1.27
50	0.35	1.17
	Pectinic acid 4.4E	
10	0.39	5.54
50	0.33	4.83
	Pectinic acid 10.5	
10	0.42	7.25
50	0.38	6.22

Figure 2 is a plot of the ratio of osmotic pressure in cm. of water to concentration in g./100 ml. against concentration. From these results it appears that single determinations of osmotic pressures of dilute pectin solutions are sufficient for the calculation of number average molecular weights ($\overline{M}_n$). The values for such molecular weights are given in Table III for some of the samples.

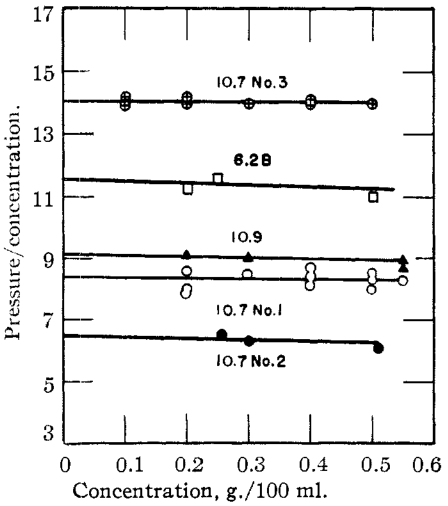

Fig. 2.—Relationship between the ratio of the osmotic pressure in cm. of water to concentration of pectinic acid and the concentration.

TABLE III

VALUES FOR k', INTRINSIC VISCOSITY AND AVERAGE MOLECULAR WEIGHTS FOR THE SAMPLES USED. MEASUREMENTS WERE MADE AT *p*H 6 AND AT 25°

Pectinic Acid	$[\eta]$	k'	$\overline{M}_n \times 10^{-4}$	$\overline{M}_w \times 10^{-4}$	From Simha's equation $\overline{M}$ 10^{-4}	From Huggins' equation $\overline{M} \times 10^{-4}$
10.7 No. 1[a,b]	4.2	.43	3.1	6.7	4.7	2.1
10.7 No. 2	8.7	.43	3.9	11.5	7.1	3.1
10.7 No. 3[a,b]	1.2	.41	1.8	2.7	2.3	1.2
10.5	7.9	.42	...	11.0	6.7	2.9
10.9[a]	5.7	.44	2.7	8.6	5.7	2.5
6.6A	4.2	.38	...	6.7	4.7	2.1
7.9E	4.8	.40	...	7.4	5.1	2.3
5.8E	4.8	.42	...	7.4	5.1	2.3
5.2E (*in vitro*)	3.1	.41	...	5.4	4.0	1.8
4.4E	6.0	.41	...	8.8	5.8	2.5
3.6E	3.7	.42	...	6.0	4.4	2.0
0.7E	4.2	.43	...	6.7	4.7	2.1
7.8B	3.6	.43	...	6.0	4.3	2.0
6.6B	3.5	.39	...	5.9	4.2	1.9
6.2B	3.3	.38	2.3	5.6	4.1	1.9

[a] These samples are the same as those used in a previous investigation (ref. 8). The lower intrinsic viscosity for sample No. 10.9 is the result of changes during storage and redrying. [b] Measured at natural *p*H.

Discussion

Calculations of molecular size and shape made from viscosity data are valid only if the measurements are not influenced by charge and aggregation effects. With a heteropolar molecule like pectinic acid, charge effects are appreciable. However, sodium chloride and other salts[12] reduce them so that reliable measurements of intrinsic viscosity may be made.[19] In solutions as dilute as 0.1%, aggregation is apparently not important with pectin, as was shown previously[8] and is also shown by the osmotic pressure results presented in Fig. 2 and by the work of Säverborn.[3b]

The fact that k' values in Table III do not change significantly when methoxyl groups are removed from pectin indicates that the change in surface characteristics, in the presence of electrolytes, is slight. The relative changes in k' values with temperature for pectinic acid 4.4E were practically the same as with pectinic acid 10.5 and 10.7 No. 3.[20] The decrease noted can be correlated with the expected decrease in hydration of pectinic acids as the temperature is increased in

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[16] M. L. Huggins, THIS JOURNAL, **64**, 2716 (1942).

[17] E. M. Frith, *Trans. Faraday Soc.*, **41**, 90 (1945).

[18] W. Philippoff, "Viscosität der Kolloide," Theodor Steinkopff, Dresden, 1942, pp. 203–211.

[19] H. Staudinger, "Die Hochmolekularen organischen Verbindungen," Julius Springer, Berlin, 1932.

[20] Another interpretation that may be placed on these data is obtained from W. J. Lyons, *J. Chem. Phys.*, **13**, 43 (1945), who has developed the equation $\eta_r = 1 + Bc/kT + (Bc)^2/2(kT)^2 + \ldots$) in which k is Boltzmann's constant and B is assumed to be a function of the flow gradient. This factor was essentially constant for a given pectinic acid for the temperature changes reported in Table II, although the maximum flow gradients varied as much as 1900 sec.$^{-1}$. This calculation may offer further confirmation that flow orientation is not a factor in these experiments.[8]

agreement with the equation of Frith.[21] The data in Tables II and III imply that the flexibility of the galacturonide chain is not affected by removal of methoxyl groups and that the viscosity equations can be applied with equal reliability to pectinic acids of various methoxyl contents.

Because the flexibility of the galacturonide chain is apparently unchanged by deësterification and the viscosities of pectin solutions decrease when the molecular weight of the pectin is decreased by heat,[22] acid[23] and alkali,[24] it is believed that intrinsic viscosities can be taken as measures of the molecular weight provided the shape of the distribution curve of molecular sizes is not markedly different among the various samples. Attempts to fractionate the samples without degradation have been unsuccessful. Despite the heterogeneity[23b] it is possible to make some calculations that are enlightening.

Application of Simha's equation[25a] yields values for the ratios of length to breadth of the pectinic acid molecules. Assuming an elongated ellipsoidal shape, a value for this ratio of 165 is obtained with sample 10.7 no. 2, 110 with sample 10.7 no. 1, and 53 with 10.7 no. 3.[25b] The high values of the ratios indicates a highly asymmetric molecule.[3]

X-Ray results obtained in this Laboratory by K. J. Palmer on pectinic acids used in this investigation show that the average length of the pyranose units along the chain is 4.3Å. and not 5.15 as in cellulose. Models were constructed which are compatible both with this residue length and the usually assumed bond angles and interatomic distances. These models clearly show that rotation around the glycosidic bond is much more restricted than in cellulose. Lack of rotation tends to stiffen the chain and accounts for the viscous nature of the pectinic acid solutions. X-Ray results obtained with sodium salts of pectinic and alginic acids, soda cellulose, and the free acids and free cellulose also show that pectin chains are less flexible.[26]

The asymmetry ratios that have been calculated may be applied to approximate minimal average molecular weights, provided it is assumed that the molecules are cylindrical, in order to obtain the dimensions from the known structure of the pyranose ring. The *α-trans* form of the methyl ester of anhydrogalacturonic acid has a calculated width averaging about 10 Å. Neglecting hydration, sample 10.7 No. 2 has an effective chain length of 1650 Å.; sample 10.7 No. 1, 1100 Å.; and 10.7 No. 3, 530 Å. Using a base weight of 185 which allows for partial deësterification and assuming that the effective and actual chain lengths are the same, the molecular weights are 7.1×10^4, 4.7×10^4 and 2.3×10^4, respectively. These represent neither number nor weight average molecular weights but are probably between them.

For comparative purposes the equation derived by Huggins[27] ($\eta_{sp}/c \approx \pi N a l^2 n^2/24000$, where N is Avogadro's number, a is the radius of the submolecule, l is its length, n the number of submolecules per molecule, and c is the concentration in submoles per liter), on the basis of a linear rod-like model, yields values of 3.1×10^4, 2.1×10^4 and 1.2×10^4, respectively. Molecular weights for the other samples calculated by means of Huggins' and Simha's equations are given in Table III. No corrections were made for nonuronide content or for changes in methoxyl content.

Molecular weights calculated from intrinsic viscosities are likely to be inaccurate for several reasons. Factors which influence the frictional ratio such as hydration, flexibility, or heterogeneity, will, in turn, influence the calculated molecular weight. Pectin is hydrated to about the same extent as proteins.[28] If the frictional ratios of pectins are calculated in the same way as for proteins,[29] the correction for hydration amounts to about 20%, which is to be added to the values given for Simha's equation. This adjustment for hydration may not be valid. In the range of values for the ratio of the length (L) to the diameter (d) found for pectin molecules (except sample 10.7 no. 3) Simha's equation can be placed in the approximate form $\eta_i = K(L/d)^2$ where η_i is the specific volume intrinsic viscosity. Since it is assumed that the molecules have a cylindrical shape, the volume concentration can be calculated from the equation $\phi = \pi d^2 l g N/(4 \times 185)$ where g is grams of pectin per ml. of solution. Combining these equations $[\eta_{sp}/g]_{g \rightarrow 0} = K'L^2$, so that the length of the molecule becomes independent of the diameter and, consequently, of the hydration. Hydration causes only a small decrease in the values for the molecular weight calculated from Huggins' equation. If the galacturonide chain is kinked, then the observed molecular weight would be somewhat higher than the one calculated from

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(21) E. M. Frith, *Trans. Faraday Soc.*, **41**, 17 (1945).

(22) E. F. Jansen and L. R. MacDonnell, *Arch. Biochem.*, **8**, 97 (1945).

(23) (a) Experiments run at *p*H 0.6 and 60° with a pectin similar to sample 10.7 No. 1 have indicated a decrease in $\overline{M}_n$ from 33,000 to 18,000 in three days, while the intrinsic viscosity dropped from 4.2 to 1.5. (b) See also R. Speiser and C. H. Eddy, THIS JOURNAL, **68**, 287 (1946).

(24) Sample 6.2B was prepared from sample 10.7 No. 1 and the number average molecular weights are indicative of the degradation. They are not quantitative measures of it, however, because the isolation and purification of the sample removed some of the small molecular weight fractions.

(25a) R. Simha, *J. Phys. Chem.*, **44**, 25 (1940).

(25b) A partial specific volume of 0.6 obtained from measurements of densities of pectin solutions was used to convert weight intrinsic to specific volume intrinsic viscosities. If reasonable dimensions for the submolecule and a cylindrical shape for the pectin molecule are assumed, the calculated specific volume is 1.1, which reduces the values for the ratios and the molecular weights derived from them by about 30%.

(26) K. J. Palmer and M. Hartzog, THIS JOURNAL, **67**, 1865 (1945).

(27) M. L. Huggins, *J. Phys. Chem.*, **42**, 911 (1938).

(28) R. Stuewer, *ibid.*, **42**, 305 (1938).

(29) J. L. Oncley, *Ann. N. Y. Acad. Sciences*, **41**, 121 (1941).

viscosity measurements and Simha's equation would not give precise values for the molecular weight, since, in this case, η_i would be proportional to (L/d) raised to a power less than two. Heterogeneity will cause the various types of average molecular weights to differ.

By reference to Table III, it is seen that there is little relationship between the intrinsic viscosity and number average molecular weights ($\overline{M}_n$) except that pectins from the same source yield values for these properties which follow the same trend. The reason for the lack of a more direct relationship between $[\eta]$ and $\overline{M}_n$, regardless of the source of the pectin, probably lies in the shapes of the molecular weight distribution curves.[30] If apple pectin has a flatter distribution curve than that for citrus pectin, a lower $\overline{M}_n$ at a given value for $[\eta]$ would be expected.

Speiser and Eddy[23b] have found that the intrinsic viscosities of pectin nitrates in acetone solutions follow the weight average molecular weight ($\overline{M}_w$). Although no values for $\overline{M}_w$ are available for the samples examined in this work, it is possible to correlate the viscosities of the pectin solutions with those of solutions of citrus pectins of known molecular weights examined by Säverborn.[3] An approximate equation interpolated from Säverborn's and the data reported previously,[8] which relates molecular weights to intrinsic viscosities, is $[\eta] = 1.4 \times 10^{-6} M^{1.34}$.[31a,b] Values for $\overline{M}_w$[31c] calculated from this equation are given in Table III. The proximity of the molecular weights from Simha's equations to these interpolated values strengthens the assumption that pectin is a fairly rigid, rod-like molecule.[3] Other arguments that favor the idea of a fairly rigid chain are based on the temperature effects. Increasing the temperature decreases the intrinsic viscosity, which may relegate segmental flow to a minor position,[32] although other factors such as change in flexibility or solvent conditions[21] complicate the interpretation. Since water may be classed as a good solvent for pectin, the zero slopes of the *p/c versus c* curves also indicate lack of flexibility of the galacturonide chains.[32]

The molecular weights noted in this and other recent work[33,34] are lower than those published by Schneider and co-workers.[35] The latter depend upon measurement of the viscosity of the nitrate in acetone. The preparation of the nitrate may involve some fractionation with loss of low molecular weight material; in addition, viscosity of the nitrate solutions may be strongly influenced by small quantities of polyvalent cations.

Acknowledgment.—The authors are grateful to R. M. McCready and H. A. Swenson for many analyses, to A. D. Shepherd for some of the samples, to J. R. Hoffman for constructing the osmometers, and to E. F. Jansen for the methyl glycoside of polygalacturonide methyl ester and the crystallized lactoglobin used to test the osmometers. They also appreciate the suggestions of K. J. Palmer on the structure of polygalacturonide chains.

Summary

The viscosity behavior of various pectins in 0.155 *M* sodium chloride solution has been studied under different conditions of *p*H and temperature. The intrinsic viscosity values were found to decrease with rise in temperature and to be essentially independent of *p*H and probably independent of the methoxyl content.

The intrinsic viscosity values for the samples used have been calculated and related to the physical features of the pectinic acid molecules. The length-to-diameter ratios calculated by means of Simha's equation varied from 53 to 165. Assuming a width of 10 Å., the length of the molecules varied from 530 to 1650 Å. The molecular weights ranged from 2.3×10^4 to 7.1×10^4.

The results indicate that pectin has a rigid, rodlike structure in aqueous salt solutions.

Osmotic pressure measurements were used to calculate the number average molecular weights, which varied from 1.8×10^4 to 3.9×10^4. The lack of agreement between these molecular weights and the viscosity and weight average molecular weights is believed to be due to the heterogeneity of the samples.

ALBANY, CALIF. RECEIVED FEBRUARY 11, 1946

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(30) E. O. Kraemer, "In the Ultracentrifuge," by Svedberg, Pedersen and others, Clarendon Press, Oxford, 1940, pp. 349–353.

(31a) In the work published by Säverborn[3a] relative viscosities of pectin in aqueous solutions were measured which do not yield intrinsic viscosity values. For this reason data obtained by us were used to obtain the interpolated values mentioned. (b) Of interest is a sample of methyl glycoside of polygalacturonide methyl ester prepared by the method of S. Morell, L. Baur, and K. P. Link (*J. Biol. Chem.*, **105**, 1 (1934)) which has an average molecular weight of 4400 and $[\eta]$ of 0.11. This point comes close to the curve for the equation given above. (c) Säverborn (ref. 3b) found that molecular weight values for pectin calculated from sedimentation and diffusion constants agreed fairly well with weight average molecular weights calculated from sedimentation equilibrium measurements.

(32) R. E. Powell and H. Eyring in *Advances in Colloid Science*, ed. by E. O. Kraemer, Vol. I, Interscience Publishing Inc., New York, N. Y., 1942, pp. 186–306.

(33) T. Svedberg and N. Gralen, *Nature*, **142**, 261 (1938).

(34) L. R. MacDonnell, E. F. Jansen and H. Lineweaver, *Arch. Biochem.*, **6**, 389 (1945).

(35) See G. G. Schneider and H. Bock, *Ber.*, **70**, 1617 (1937).