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SHORT COMMUNICATION

Specific refractive index increments of inulin

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The specific refractive index increments (d*n*/d*c*) of inulin in water, at 25 ºC, were measured at different wavelengths (436, 546 and 589 nm) using a BP-differential refractometer. The d*n*/d*c* at the operating wavelength (633 nm) of the laser light scattering photometer was calculated by an approximate method. This value can be used to determine the absolute molar mass and the second virial coefficient of inulin by light scattering photometry.

Keywords: inulin, specific refractive index increment.

INTRODUCTION

Inulin, a carbohydrate polymer $C_6H_{11}O_5(C_6H_{10}O_5)_nOH$, is a polyfructofuranoside with about 30 fructose units, bonded by -1,2-glucoside bonds, and with a saccharose at the end of the linear molecule. Investigations of medically important complexes with various ligands of the carbohydrate type (*e.g.,* inulin,[1] dextran[2]), show that the molar masses of the ligands and their distribution are of great importance to obtain stable compounds.

The determination of the absolute molar mass and the size of the polymer molecules by light scattering techniques requires the knowledge of the specific refractive index increment (d*n*/d*c*) of the polymer in solution. Since the square of d*n*/d*c* enters into the light scattering, Eq. (1), its value must be known with the greatest possible accuracy, in order to obtain precise results of the molecular parameters

$$ = \frac{32\ ^{3}\ n_0^2}{3\,N_A\ ^{4}}\ \frac{dn}{dc}^{\,2}\ c\,M \quad H\,c\,M \qquad (1)$$

H is an optical constant, is the comodated turbidity of the solution, *M* is the molar mass, *c* is the concentration of the polymer, N_A is Avogadro's constant, is wavelength of the light in vacuo, and n_0 the refractive index of the solvent. Most laser light

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scattering photometers utilize a helium–neon laser light source operating at a wavelength of 633 nm. Data for d*n*/d*c* of polysaccharides either at this wavelength or others, however, are very rare in the literature. There are some literature tables of d*n*/d*c* for different polymer–solvent systems,[3–7] but data pertaining to inulin was not found. Therefore, the quantities were determined using a differential refractometer in this work. A calibration constant (k) for the selected wavelength was calculated by Eq. (2):

$$n = k \quad d \qquad (2)$$

where d is the total slit image displacement in instrument units. n is an already known quantity for different reference solutions and different wavelengths.[8] The reading of the total displacement, d, corrected for the solvent zero reading was calculated as follows:

$$d = (d_2 - d_1)_{\text{solution}} - (d_2 - d_1)_{\text{solvent}} \qquad (3)$$

where d_1 is the zero reading, and d_2 is the reading at 180°.

The purpose of this communication is to provide precise values of the d*n*/d*c* of inulin at three different wavelengths (436, 546 and 589 nm) and an approximate value at 633 nm.

EXPERIMENTAL

The native source inulin (*Dahlia variabilis*) was separated into fractions with narrow molar mass distributions by preparative gel permeation chromatography on Sephadex G-25/G-50 columns. The obtained inulin fractions were purified by precipitation with ethanol, redissolution in water and filtration of the solution through a 0.45 nm cellulose nitrate membrane filter. The fractions were dried in vacuum at 80 °C.

The d*n*/d*c* increment for aqueous solutions of inulin were measured at 25.00   0.02 °C, at the wavelengths of 436, 546 and 589 nm using a Brice-Phoenix differential refractometer BP-2000-V. The limiting sensitivity of the BP-refractometer is about 3 units in the sixth decimal place of the refractive index difference, while the range is 0.01 units. Mercury vapor and sodium lamps were used as light sources, from which the monochromatic light beams of 436, 546 and 589 nm were isolated using corresponding filters. The BP-differential refractometer was calibrated with NaCl solutions using the data of Kruis[8] (0.1034, 0.5602, 1.1240, 2.0327 and 3.7307 g/100 cm^3). The measurements were performed at least 10 times for the inulin solutions (0.530, 1.020, 1.520, 2.010 and 3.010 % w/v).

RESULTS AND DISCUSSION

The dependence between the slit image displacement (d) and the concentration of the solution (c) was observed using a Brice-Phoenix differential refractometer. The calibration constants of the refractometer at the chosen wavelengths were determinated using the method of least squares from Eq. (2). Once the value of k had been determined, any reading of light of the selected wavelength may be directly converted to n. The differential method of determination of the refractive index difference between a solution and the solvent ($n = n_1 - n_0$) is suitable for the determination of the d*n*/d*c* of polymer solutions. The measurements were performed using inulin solutions of various concentrations (0.530, 1.020, 1.520, 2.010 and 3.010 % w/v). Using the already determined value of *k* under the specific conditions, the values for *n* were calculated for the solutions of inulin, compared to the water as the solvent. Repeating this for different concentrations of those solutions, the d*n*/d*c* can be determined. The resulting values are shown in Table I.

TABLE I. Refractive index difference (*n*) between inulin solutions and distilled water and specific refractive index increment (d*n*/d*c*) of inulin in water at 25 °C

Concentration/(g/100 cm^3)	*n* ^{6} at 25 °C		
	= 436 nm	= 546 nm	= 589 nm
0.530	766	758	756
1.020	1474	1458	1454
1.520	2196	2173	2168
2.010	2905	2875	2866
3.010	4350	4304	4292
	d*n*/d*c* (cm^3 g^{-1})		
Inulin	0.1445	0.1430	0.1426

Since d*n*/d*c* is inversely proportional to the wavelength,[6,7] the value of d*n*/d*c* at KMX-6 laser's operating wavelength of 633 nm was calculated by approximate method *via* Fig. 1, using the d*n*/d*c* values at 436, 546 and 589 nm. A good linear relationship is evident from Fig. 1. This linearity is a necessity for a reliable d*n*/d*c* calculation. The increment value of 0.1423 cm^3 g^{-1}, obtained by the approximate method for wavelength of 633 nm, can be used to determine the absolute values M_w for inulin.

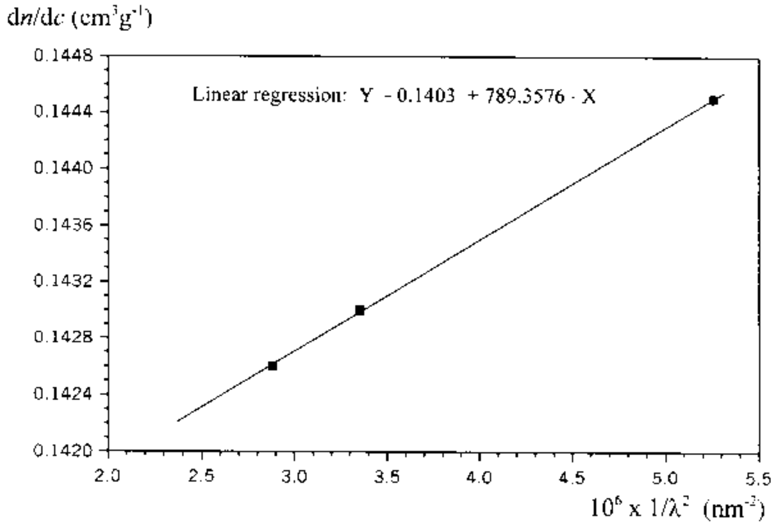

Fig. 1. Specific refractive index increment (d*n*/d*c*) of inulin as a function of wavelength ( ).

Results of d*n*/d*c* determination of another carbohydrates,[4,6,9] in comparison with the values for inulin, are given in Table II. The corresponding values of the inulin monomers (glucose and fructose) are also included in Table II. As is well known, the d*n*/d*c* value is a function of temperature, the laser light wavelength and the solvent in which the polymer is dissolved. As can be seen from the d*n*/d*c* values compiled from the literature, the ratio of d*n*/d*c* values at the different wavelengths is almost constant for most of the different polymer/solvent systems.[10] So, for example, the data in the Table II show that the d*n*/d*c* of dextran in water is not independent of the molar mass, but increases steadily with increasing mass. In the range of wavelengths from 436 to 633 nm, the specific refractive index increment of inulin in water increases with decreasing wavelength (Fig. 1), according to the literature data.[10] It is gratifying to see from Table II that the agreements among the results of different carbohydrates, obtained by independent authors, are remarkably good.

TABLE II. Comparison of the specific refractive index increments of some carbohydrates in water at 25 ºC

Name	Molar mass g mol^{-1}	(d*n*/d*c*)/cm^3 g^{-1}				Ref.
		436 nm	546 nm	589 nm	633 nm	
Inulin	5635	0.1445	0.1430	0.1426	0.1423	–
Dextran	7300	0.1449	0.1411	0.1401	0.1395	6
Dextran	2040	0.1432	0.1396	0.1387	0.1379	6
Dextran	1150	0.1427	0.1392	0.1383	0.1375	6
Dextrose	–	0.1470	–	–	–	4
Sucrose	342	0.1448	0.1429	–	–	4
Fructose	180	0.1392	0.1373	–	–	9
Glucose	180	0.1265	0.1243	0.1238	0.1231	6
Hydroxyethyl cellulose	–	0.1410	0.1390	–	–	4

The details of the determination of the absolute molar mass and second virial coefficient of purified inulin (5635 g mol^{-1} and 7.28 10^{-4} mol cm^3 g^{-2}, respectively) by laser light scattering will be published elsewhere.

CONCLUSION

On the basis of the obtained results, it may be concluded that the d*n*/d*c* value of aqueous solutions of inulin is a function of the laser light wavelength, and showed a good linear relationship. The d*n*/d*c* values, measured using a BP-2000-V differential refractometer at 25 ºC at the wavelengths of 436, 546, 589 and 633 nm, increases with decreasing wavelength. These values can be used to determine the absolute molar mass and the second virial coefficient of inulin by light scattering photometry.

И З В О Д

СПЕЦИФИЧНИ ПРИРАШТАЈ ИНДЕКСА ПРЕЛАМАЊА ИНУЛИНА

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Мерење специфичног прираштаја индекса преламања (d*n*/d*c*) инулина у води, Brice-Phoenix диференцијалним рефрактометром, вршено је на 25 ºC за три различите таласне дужине (436, 546 и 589 nm). Вредност d*n*/d*c* прираштаја на радној таласној дужини light-scattering ласера (633 nm), израчуната је апроксимативном методом. Добијене вредности d*n*/d*c* могу се користити за одређивање апсолутне моларне масе и другог виријалног коефицијента инулина помоћу light-scattering фотометра.

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REFERENCES

1. G. Nikolić, M. Cakić, S. Žerajić, *4th Int. Conf. on Fundamental and Applied Aspects of Physical Chemistry,* Belgrade, Yugoslavia, 1998, Conference Proceedings, pp. 476–478
2. A. Müller, *Arznein. Forsch.* **8** (1967) 921
3. M. B. Huglin, *J. Appl. Polym. Sci.* **9** (1965) 3963
4. J. Brandrup, *Polym. Handbook*, Int. Publ. John Wiley and Sons, New York, 1966, IV-279
5. M. B. Huglin, *Light-scattering from Polymer Solution,* Academic Press, N. Y. 1972, p. 165
6. A. M. Basedow, K. H. Ebert, U. Ruland, *Makromol. Chem.,* **179** (1978) 1351
7. V. E. Eskin, *Raseyanie sveta rastvorami polimerov,* Mir, Moskva, 1973
8. A. Kruis, *Z. Phys. Chem.* **34B** (1936) 13
9. G. Nikolić, M. Cakić, Lj. Ilić, *J. Chem. Technol. Serb.* **42** (2000) 33
10. M. B. Huglin, *Polymer Handbook,* Wiley, New York, 1989, Chapt. VII.