



Dilute solution properties and degree of chain branching for dextran

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ABSTRACT

Hydrodynamic volume, radius of gyration, and viscometric constants, K and a , for dextran, with a wide molecular weight range were calculated using experimental reported average-molecular weights (M_n , M_w), and intrinsic viscosity, $[\eta]$, data in water and 0.05 M Na_2SO_4 . Degree of chain branching for dextran was also determined using different procedures. This study demonstrated that hydrodynamic volume and radius of gyration of a dextran sample with $M_w < 20$ kDa and its linear counterpart with equal M_w are almost identical, whereas the latter parameters for a dextran sample with $M_w > 20$ kDa was smaller than that of its linear counterpart. Values of 0.506 in water and 0.512, 0.425 and 0.273 in 0.05 M Na_2SO_4 for the exponent a were obtained. A smaller value for a was obtained for a larger M_w range. Molecular weights of desirable nano-particles for various branches of nanotechnology can be estimated from a derived radius of gyration–molecular weight relationship.

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1. Introduction

Dextran is a neutral and a branched naturally occurring polysaccharide. It is a product of microbial biosynthesis. The synthesized dextrans usually consist of α -D-(1-6)-linked glucose units in main chains and various amounts of side branches linked at positions 2, 3, or 4 (Khalikova, Susi, & Korpela 2005; Nordmeier, 1993). Branching at position 3 occurs in most dextrans, whereas occasionally at position 2 (Alger, 1997). A detailed description of molecular structure of dextrans synthesized by microorganisms has been given in Section 2.

Dextran is used in food industries as a thickener, an emulsifier and a stabilizer (Belitz, Grosch, & Schieberle, 2009; Cheng, 1996; Van Aken, 2006). It is used in the production of confectionary, baking products, beverages and ice cream. The principal potential uses of dextran in foods appear to be related to its capacity to prevent crystallization and retain moisture (BeMiller, 2003). The European Union has approved its uses as a food ingredient in bakery products (Scientific Committee on Food, 2000). It is widely used in many biomedical areas (Huguët, Prouchayret, Grandgeorge, & Dellacherie, 1993; Kim, Won, & Chu, 1999; Kokotilo et al., 2010; Xie, Lv, Yu, Lina, & Huang, 2010; Yalpani & Sandford, 1987). A complex of ferric hydroxide and dextran is used in the treatment of anemia. Dextran sulphate was employed as a stabilizer in the preparation of polyalkyl cyanoacrylate (PACA) nano-particles, which slowed down the release rates of drugs (Flexner et al., 1991;

Hosoya, Balzarini, Shigeta, & Clercq, 1991; Kumar, Sameti, Kneuer, Lamprecht, & Lehr, 2004).

Original dextrans are polydisperse and may contain a broad range of molecular weights from oligomers to macromolecules having $M_w \geq 1.0 \times 10^5$ kDa (Yalpani, 1988). The original dextrans mostly are not suitable for technological applications. However, hydrolyzed (by acids or enzymes) and fractionated dextrans possess a significant commercial interest in cosmetic, drug and food formulations (Moulis et al., 2008). Fractionated dextrans have been used as standard materials for M_w determination of water-soluble polymers as well as for the construction of a universal calibration curve for the evaluation of size exclusion chromatography (SEC) results (Wang & Cui, 2005a). Dextrans and their derivatives have been also used as a blood plasma extender, when partially hydrolyzed (Rodriguez, 1989; Yalpani & Sandford, 1987). A sterile solution of dextran with a specific M_w has been used to restore blood volume in patients suffering shock as a result of blood loss (Madigan & Martinko, 2006). A continuous fermentation processes has been developed for the preparation of clinical dextrans with M_w of 30–100 kDa (Yalpani, 1988). Dextran with a desired M_w can be safely injected into a blood stream. The molecular structure (with a particular attention on the position of branch linkages, the length of branch chains and the degree of chain branching, g) as well as M_w of dextrans varies with change in microbial strains, growth rates and reaction conditions (sucrose and enzyme concentrations, and reaction temperature: the temperature at which dextran is synthesized) (Yalpani, 1988).

Various properties and applications of the polymer are closely related to its M_w , g , hydrodynamic volume, V_h , radius of gyration, $\langle S^2 \rangle^{1/2}$, and polymer conformation. With knowledge on the latter parameters, its properties (such as rheological properties) and its

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efficiencies in various food and medical applications can be predicted. Background and theoretical basis on the above-mentioned parameters are described in Section 3.

The objectives of this work were: to determine the values of K and a , for dextran as a function of molecular weight range with taking into account the polydispersity, q_{MHS} , of the polymer; to determine hydrodynamic volume, and radius of gyration for each dextran sample and to compare them with corresponding data for its linear counterpart, pullulan; and to estimate the degree of branching for dextrans relative to pullulans.

2. Molecular structure of dextrans synthesized by microorganisms

Dextran is an extracellular polysaccharide synthesized from sucrose and dextransucrase. The latter compound is produced by a number of bacteria from the family of *Lactobacillaceae*: *Leuconostoc*, *Lactobacillus*, and *Streptococcus* (Kenne & Lindberg, 1983; Khalikova et al., 2005; MacGregor, 2002; Madigan & Martinko, 2006; Robyt, 1985; Ul-Qader et al., 2001; Yalpani & Desrochers, 1987). Other bacteria, *Acetobacter capsulatus* (renamed *Gluconobacter oxydans*) and *Acetobacter viscous*, produce dextransucrase (EC2.4.1.2) and convert dextrans to dextrans (Sims, Thomson, Hubl, Larasen, & Furneaux, 2001; Tirtaatmadja, Dunstan, & Boger, 2001). Dextrans are also produced from a number of molds such as *Rhizopus* spp. or a number of yeasts (Cerutti de, Diez, Cardenas, & Oliver, 2000; Irvine, 1981; Jiménez, 2009; Sankpal, Joshi, Sainkar, and Kulkarni, 2001; Wang, Suzuki, Tanaka, Kumura, & Shimazaki, 2004). *Streptococcus* mutant produces dextran only when sucrose and dextransucrase are present. It is found predominantly in dental crevices and small fissures (Madigan & Martinko, 2006). Two principal bacteria those produce enzymes are *Leuconostoc* and *Streptococcus*. Most of commercial production of dextran is derived from *Leuconostoc mesenteroides* strain B512-F (Khalikova et al., 2005). Dextrans are also produced in harvested sugar canes and beets (Jiménez, 2009). In sugar production, dextran is synthesized by contaminant microorganisms (some bacterial strains, filamentous fungi and a small number of yeasts). Undesirable formation of dextran reduces in the sugar recovery and results in a significant loss (Jiménez, 2009). Dextrans may be synthesized in the production of milk or cheese due to the presence of some molds or yeasts in their production processes (Wang et al., 2004). Some strains of *Leuconostoc* produce large amounts of dextran when cultured on sucrose (Madigan & Martinko, 2006). Sucrose is mainly required for dextran formation. No dextran is generally formed when one of the above-mentioned microorganisms is cultured on a medium with glucose or fructose (Madigan & Martinko, 2006).

Dextrans can be divided into three classes based on their molecular structures: class-1 dextrans contain a main chain of consecutive α -D-(1-6)-linked glucosyl residues, with branching at positions 2, 3, or 4; class-2 dextrans contain non-consecutive α -D-(1-3)- and α -D-(1-6)-linkages and α -D-(1-3)-branch linkages; and class-3 dextrans contain consecutive α -D-(1-6)-linkages and α -D-(1-6)-branch linkages. Class 1 comprises most of commercially synthesized dextrans. *L. mesenteroides* B-512 F dextran is the classical dextran (class 1) containing a high percentage (95%) of consecutive α -D-(1-6)-linkages and a low percentage (5%) of α -D-(1-3)-branch linkages (Nordmeier, 1993).

3. Theoretical considerations

3.1. Calculation of viscometric constants and polydispersity correction factor, q_{MHS}

The value of $[\eta]$ varies with viscosity-average molecular weight, M_v , for a homologous series according to the

Mark–Houwink–Sakurada (MHS) equation (Flory, 1953; Tanford, 1961).

$$[\eta] = KM_v^a \quad (1)$$

Determination of constants, K and a , from $[\eta]$ data, requires a series of mono-disperse polymer samples with known M_w or a series of polydisperse polymer samples with known M_v . In general, M_v is not experimentally accessible, whereas other average molecular weights are experimentally achievable from different experimental methods. Eq. (1) can be rearranged and resulted in a modified MHS equation as follows:

$$[\eta] = KM_v^a = K \left(\frac{M_v}{M_w} \right)^a M_w^a = Kq_{\text{MHS}}M_w^a \quad (2)$$

The value of q_{MHS} is a statistical function of molecular weight distribution, MWD. The value of q_{MHS} varies from one sample to another one. It is a function of a and average-molecular weights (M_v , M_w). The value of q_{MHS} is determined from $(M_v/M_w)^a$. Alternatively the value of q_{MHS} can be calculated using a numerical method and other-average molecular weights (M_n , M_w , M_z) according to (Bareiss, 1999; Kasaai, 2006):

$$q_{\text{MHS}} = \left(\frac{M_w}{M_n} \right)^b \left(\frac{M_z}{M_w} \right)^c \quad (3)$$

On the whole, the correction factor, q_{MHS} , is a function of exponent a , and (M_n , M_v , M_w , M_z). The precision of q_{MHS} value depends on the precision of both a and average-molecular weights.

3.2. Hydrodynamic volume and radius of gyration

V_h may be expressed as the product of $([\eta])$ and one of the average-molecular weights ($[\eta]M_n$, $[\eta]M_w$ or $[\eta]M_v$) (Kasaai, 2006). Generally, SEC has been used to determine V_h of a polymer via construction of V_h versus elution volume, V_e . The products for a linear and a branched molecule having the same V_h at a constant V_e are the same as follows:

$$([\eta]M)_l = ([\eta]M)_{br} \quad (4)$$

This equation offers a method to determine molecular weights of branched polymers as follows: if the SEC universal calibration curve for a linear polymer is available, one can determine the V_e and $[\eta]$ of a branched sample. Then the product $[\eta]M$ corresponding to the V_e of the branched sample is found from the universal calibration curve. This value divided its $[\eta]$ gives molecular weight of the branched sample. One can also calculate the $[\eta]$ of the linear polymer with equal M_w by using the corresponding MHS equation (Van Krevelen, 1990; Yu & Rollings, 1987).

Properties of dilute polymer solutions depend on the root mean square (RMS) radius of gyration, $\langle S^2 \rangle^{1/2}$ rather than root mean square (RMS) end-to-end distance, $\langle R^2 \rangle^{1/2}$ (Chanda, 2000). Thus $\langle S^2 \rangle^{1/2}$ is used to characterize the dimension of branched macromolecules (Chanda, 2000). The $\langle S^2 \rangle^{1/2}$ is directly measured by light scattering, neutron scattering, and small angle scattering experiments (Chanda, 2000). Alternatively, the value of $\langle S^2 \rangle^{1/2}$ for a random coil as well as a flexible chain is determined from Flory–Fox equation (Tanaka, 1982):

$$\langle S^2 \rangle^{3/2} = \frac{[\eta]M_v}{6^{3/2}\phi} \quad (5)$$

where ϕ is shape factor.

3.3. Degree of chain branching

The value of g is usually determined by the ratio of mean square radius of gyration for a branched chain, $\langle S_{br}^2 \rangle^{1/2}$ to that of its

linear counterpart chain, $(S_l^2)^{1/2}$, with the same molecular weight as follows (Styring, Armonas, & Hamielec, 1987; Yu & Rollings, 1987):

$$g = (S_{br}^2)^{1/2} / (S_l^2)^{1/2} \leq 1 \quad (6)$$

Alternatively, the value of g is determined from the ratio of intrinsic viscosity for the branched chain, $[\eta]_{br}$, to that of its linear counterpart chain, $[\eta]_l$, with the same molecular weight as follows (Ioan, Aberle, & Burchard, 2000; Mirabella & Wild, 1988; Styring et al., 1987; Yu & Rollings, 1987):

$$g^e = \frac{[\eta]_{br}}{[\eta]_l} \quad (7)$$

where the value of exponent e varies from 0.5 to 1.5, depending upon the particular theoretical assumption in the model development and type of chain conformation (Styring et al., 1987; Yu & Rollings, 1987).

4. Materials and methods

4.1. Materials

Dextran samples with following range of M_w and M_w/M_n ($0.18 \leq M_w \leq 5900$ kDa; $1.00 \leq M_w/M_n \leq 2.62$) (American Polymer Standards Corporation, Mentor, Ohio) were used in this study.

4.2. Determination of intrinsic viscosity and average-molecular weights

The $[\eta]$ for dextran in water at 25 °C and in 0.05 mol L⁻¹ Na₂SO₄ at 30 °C and the values (M_n , M_w , M_z), of the polymer samples have been determined by American Polymer Standards Corporation. Light scattering and end group titration were used to determine M_w , and M_n , respectively. SEC was used to determine different M (M_n and M_w) values and MWD. Differential refractometer and differential viscometer were used as detectors. The values of M_n , M_w , and $[\eta]$ are listed in Table 1.

4.3. Determination of viscometric constants and polydispersity correction factor, q_{MHS}

The values of a , K , and q_{MHS} , were calculated using a modified MHS equation through a numerical method and Eqs. (2) and (3) (Kasaai, 2006, 2008).

4.4. Determination of hydrodynamic volume, radius of gyration and degree of branching

The values of $[\eta]M_v$ and $(S^2)^{1/2}$ were calculated using $[\eta]$, M_v , and Eq. (5). The value of g was estimated from several parameters for dextran in comparison with corresponding parameters for pullulan as follows: (1) $a_{dextran}/a_{pullulan}$; (2) $[\eta]_{dextran}/[\eta]_{pullulan}$; (3) $V_{h,dextran}/V_{h,pullulan}$; and (4) $(S_{dextran}^2)^{1/2}/(S_{pullulan}^2)^{1/2}$. In each case, the same M_w or the same molecular weight range has been used. The solvent-temperature system was the same for the determination of a , $[\eta]$, V_h and $(S^2)^{1/2}$ for both dextran and pullulan.

5. Results and discussion

5.1. MHS equations

Fig. 1 shows $(\log[\eta] - \log q_{MHS})$ versus $\log M_w$ for dextran with M_w range of 0.18–158 kDa in 0.05 M Na₂SO₄ at 30 °C. The values of 0.512 and 8.32×10^{-4} were obtained for a and K , respectively. The values of M_w/M_n for the polymer samples were ($1.0 < M_w/M_n < 2.28$). The experimental points have properly fitted

Table 1
The values of M_n , M_w , and $[\eta]$ for dextrans.

Sample	Low M_w			Medium M_w			High M_w			Ultra high M_w			Low to high M_w		
	M_n (kDa)	M_w (kDa)	$[\eta]^a$ (dL g ⁻¹)	M_n (kDa)	M_w (kDa)	$[\eta]^a$ (dL g ⁻¹)	M_n (kDa)	M_w (kDa)	$[\eta]^a$ (dL g ⁻¹)	M_n (kDa)	M_w (kDa)	$[\eta]^a$ (dL g ⁻¹)	M_n (kDa)	M_w (kDa)	$[\eta]^b$ (dL g ⁻¹)
1	0.18	0.18	0.012	34.1	44.1	0.205	160.7	226.7	0.396	820.3	1907	0.736	349.3	676	0.708
2	0.342	0.342	0.016	30.25	47.25	0.206	123.9	236.1	0.467	928.8	2025	0.713	239.9	403.0	0.554
3	0.505	0.505	0.020	41.2	55.5	0.239	204.6	275.9	0.463	1600	2400	0.803	167.5	262.0	0.494
4	0.51	1.01	0.028	42.4	59.9	0.208	204.2	291.6	0.433	1970	2800	0.745	98.55	143.0	0.394
5	0.9	1.2	0.028	40.8	62.6	0.232	226.7	310.6	0.472	1230	3000	0.767	55.6	79.8	0.315
6	2.75	3.65	0.048	50.6	68.9	0.235	205.0	326.6	0.350	2000	3450	0.828	36.65	50.8	0.230
7	3.25	4.3	0.055	50.7	72.7	0.238	186.8	344.8	0.490	1500	3800	1.685	17.95	22.7	0.169
8	2.16	4.3	0.058	53.0	84.6	0.260	229.6	431.8	0.552	2100	5500	0.889	8.0	11.7	0.112
9	3.16	7.2	0.085	63.25	91.1	0.263	204.1	443.2	0.538	1500	5500	0.889	3.325	5.7	0.078
10	6.25	9.2	0.085	64.7	97.0	0.273	318.4	515.9	0.513	3000	5900	1.045	1.032	1.27	0.028
11	6.70	9.90	0.089	64.0	102.0	0.293	371.0	534.0	0.633						
12	14.3	20.1	0.106	64.5	111.0	0.312	346.5	548.3	0.560						
13	7.87	16.23	0.118	96.9	123.4	0.305	432.1	606.2	0.659						
14	11.65	17.9	0.119	97.9	131.4	0.324	488.9	655.2	0.692						
15	17.9	25.5	0.130	81.3	144.1	0.338	982.1	714.5	0.692						
16	23.3	31.2	0.173	108.1	158.1	0.350	513.4	759.4	0.610						
17	22.8	33.5	0.158	35.35	60.3	0.223	705.0	1185	0.862						
18	25.3	36.3	0.181	27.7	46.8	0.190	877.8	1360	0.918						
19	28.7	42.75	0.178	26.2	39.9	0.186									
20	30.2	41.4	0.180												
21															

^a $[\eta]$ in 0.05 M Na₂SO₄ at 30 °C.

^b $[\eta]$ in water at 25 °C.

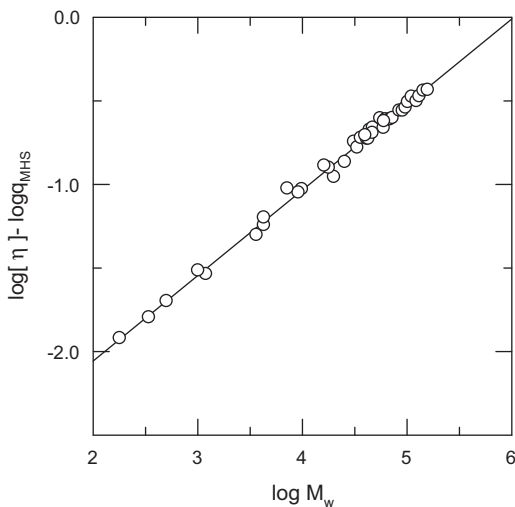


Fig. 1. A typical plot of $\log[\eta] - \log q_{MHS}$ versus $\log M_w$ for dextran with M_w range of 0.18–158 kDa in 0.05 M Na_2SO_4 at 30 °C ($R^2 = 0.995$).

on the linear plot and the regression value for the plot was found to be ($R^2 = 0.995$). The values of z -average molecular weights, M_z , for the polymer samples are not available. The values of q_{MHS} for the polymer samples were calculated by assuming the following equation:

$$\frac{M_z}{M_w} = \frac{M_w}{M_n} \quad (8)$$

This equation is valid for symmetrical curve, which is obtained from SEC. Such curve is usually obtained for Gaussian distribution (Kasaai, 2006; Netopilik, 2001). Calculation of the constants by taking into account the polydispersities of polymer samples results in higher precision compared to ignoring them. The values of q_{MHS} ($0.870 < q_{MHS} < 1.00$) for the polymer samples are given in Table 2. The MHS equation for dextran in 0.05 M Na_2SO_4 at 30 °C with M_w range of 0.18–158 kDa was obtained as follows:

$$[\eta] = 8.32 \times 10^{-4} M_v^{0.512} = 8.32 \times 10^{-4} q_{MHS} M_w^{0.512} \quad (9)$$

where $[\eta]$ is expressed in dL g^{-1} . The plots of $\log[\eta] - \log q_{MHS}$ versus $\log M_w$ for dextrans having different M_w ranges (226–1360 kDa and 1907–5900 kDa) in 0.05 M Na_2SO_4 at 30 °C and dextrans (1–676 kDa) in water at 25 °C were also linear. The experimental points identified by bold in Table 1 are as aberrant experimental points and have been removed from the linear plots. The values of q_{MHS} for the polymer samples are given in Tables 2 and 3. The information resulting from the plots (three plots) are given in Table 4.

Fig. 2 shows the plot of $\log K$ versus a . An inverse linear relationship between $\log K$ and a was obtained as follows:

$$\log K \times 10^{-4} = 3.54 - 5.1 \times \text{exponent } a \quad (R^2 = 0.999) \quad (10)$$

The MHS equation constants obtained in this study were compared with the literature values (Bahary, Hogan, Jilani, & Aronson, 1995; Bose, Rollings, Caruthers, Okos, & Tsao, 1982; Çatiker & Güner, 2000; Eremeeva & Bykova, 1998; Gekko, 1971; Granath, 1958; Ioan et al., 2000; Senti et al., 1955) (see Table 4 and Fig. 2). The following equation was obtained from both this study and literature reports (except the result reported by Gekko, 1971):

$$\log K \times 10^{-4} = 3.54 - 5.0 \times \text{exponent } a \quad (R^2 = 0.933) \quad (11)$$

The two equations ((10) and (11)) are nearly identical. The values reported by Gekko (1971) were highly deviated from other reported data and thus was removed from the linear plot. The result given in Fig. 2 is not surprising because similar results obtained for other polymers (Kasaai, 2006, 2007). The plots of Fig. 2 (Eq. (10) or (11))

Table 2
The values of q_{MHS} , M_v , $[\eta]$, $[\eta]M_v$, and $(S^2)^{1/2}$ for each dextran sample.

Sample	Low and medium M_w					Low and medium M_w				
	q_{MHS}	M_v (g mol ⁻¹)	$[\eta]^a$ (dL g ⁻¹)	$[\eta]M_v$ (dL mol ⁻¹)	$(S^2)^{1/2}$ (nm)	q_{MHS}	M_v (g mol ⁻¹)	$[\eta]^a$ (dL g ⁻¹)	$[\eta]M_v$ (dL mol ⁻¹)	$(S^2)^{1/2}$ (nm)
1	1.0	184	0.012	2.2	1.7	0.96	46,898	0.205	9614.1	28.5
2	1.0	322	0.016	5.2	2.3	0.94	47,346	0.206	9753.2	28.6
3	1.0	498	0.020	10.0	2.9	0.96	63,287	0.239	15125.7	33.1
4	0.894	960	0.028	26.9	4.0	0.95	48,248	0.208	10035.5	28.9
5	0.958	960	0.028	26.9	4.0	0.94	59,718	0.232	13854.5	32.1
6	0.959	2752	0.048	132.1	6.8	0.96	61,235	0.235	14390.3	32.5
7	0.949	3591	0.055	197.5	7.8	0.95	62,771	0.238	14939.6	33.0
8	0.936	3983	0.058	231.0	8.2	0.93	74,603	0.260	19396.7	36.0
9	0.949	8402	0.085	714.2	12.0	0.95	76,293	0.263	20065.1	36.4
10	0.956	8402	0.085	714.2	12.0	0.94	82,061	0.273	22402.8	37.7
11	0.933	9192	0.089	818.09	12.5	0.93	94,213	0.293	27604.4	40.4
12	0.947	12,933	0.106	1370.9	14.9	0.92	106,514	0.312	33232.2	43.0
13	0.962	15,946	0.118	1881.6	16.5	0.96	101,896	0.305	31078.3	42.1
14	0.940	16,211	0.119	1929.1	16.7	0.96	114,661	0.324	37150.3	44.6
15	0.957	19,267	0.130	2504.7	18.2	0.91	124,537	0.338	42093.6	46.5
16	0.943	33,666	0.173	5824.2	24.1	0.94	133,319	0.350	46661.6	48.2
17	0.946	28,201	0.158	4455.7	22.0	0.92	55,277	0.223	12326.7	30.9
18	0.953	36,774	0.181	6656.0	25.2	0.92	40,430	0.190	7681.6	26.4
19	0.940	35,593	0.178	6335.5	24.8	0.94	38,784	0.186	7213.8	25.9
20	0.953	36,378	0.180	6548.0	25.0					

^a $[\eta]$ in 0.05 M Na_2SO_4 at 30 °C.

Table 3The values of q_{MHS} , M_v , $[\eta]$, $[\eta]M_v$, and $(S^2)^{1/2}$ for dextrans.

Sample	High molecular weight					Ultra high molecular weight					Dextrans ($[\eta]^b$ in water at 25 °C)			
	q_{MHS}	M_v (g mol ⁻¹)	$[\eta]^a$ (dL g ⁻¹)	$[\eta]^a M_v$ (dL mol ⁻¹)	$(S^2)^{1/2}$ (nm)	q_{MHS}	M_v (g mol ⁻¹)	$[\eta]^a$ (dL g ⁻¹)	$[\eta]^a M_v$ (dL mol ⁻¹)	$(S^2)^{1/2}$ (nm)	M_v (g mol ⁻¹)	$[\eta]^b$ (dL g ⁻¹)	$[\eta]^b M_v$ (dL mol ⁻¹)	$(S^2)^{1/2}$ (nm)
1	0.952	182,993	0.396	72465.0	55.8	0.902	1,859,447	0.736	1368553.0	148.6				
2	0.904	269,746	0.467	125971.4	67.1	0.911	1,655,302	0.713	1180230.4	141.4				
3	0.958	264,341	0.463	122389.9	66.4	0.958	2,558,489	0.803	2054466.6	170.1				
4	0.950	225,793	0.433	97768.4	61.6	0.964	1,944,101	0.745	1448355.2	151.4				
5	0.956	276,591	0.472	130550.8	67.9	0.894	2,162,797	0.767	1658865.6	158.4				
6			0.350			0.941	2,862,567	0.828	2370205.6	178.4				
7	0.910	302,053	0.490	148005.8	70.8	–		1.685	–	–				
8	0.907	399,789	0.552	220683.5	80.9	0.886	3,713,965	0.889	3301714.9	199.2				
9	0.883	376,339	0.538	202470.5	78.6	0.924	6,714,813	1.045	7016979.1	256.2				
10	0.930	336,478	0.513	172613.0	74.5									
11	0.949	551,757	0.633	349261.9	94.2	0.898								
12	0.934	413,556	0.560	231591.3	82.2	0.921					461,638	0.708	326,840	92.2
13	0.953	606,570	0.659	399729.7	98.6	0.933					284,302	0.554	157,504	72.3
14	0.959	680,475	0.692	470888.5	104.1	0.945					226,671	0.494	111,975	64.5
15			0.692			0.946					144,965	0.394	57,116	51.5
16	0.945	505,739	0.610	308501.0	90.4	0.952					93,153	0.315	29,343	41.3
17	0.925	1,140,999	0.862	983541.3	133.1	0.966					50,035	0.230	11,508	30.2
18	0.937	1,323,134	0.918	1214637.4	142.8	0.943					27,212	0.169	4599	22.3
19						0.918					12,069	0.112	1352	14.8
20						0.970					5904	0.078	461	10.3
											780	0.028	21.8	3.7

^a $[\eta]$ in 0.05 M Na₂SO₄ at 30 °C.^b $[\eta]$ in water at 25 °C.**Table 4**

MHS equation constants for dextran and pullulan at different solvent–temperature systems.

Polymer	Solvent	T (°C)	M_w range (kDa)	M_w/M_n range	No. of samples	q_{MHS} range	a	K (dL g ⁻¹)	R^2	Reference
Dextran	H ₂ O	25	1.27–676	1.23–1.94	10	0.899–0.971	0.506	9.636×10^{-4}	0.987	This work
Dextran	0.05 M Na ₂ SO ₄	30	0.18–158	1.0–2.28	39	0.870–1.00	0.512	8.320×10^{-4}	0.995	This work
Dextran	0.05 M Na ₂ SO ₄	30	226–1360	1.34–2.17	16	0.883–0.959	0.425	2.297×10^{-3}	0.917	This work
Dextran	0.05 M Na ₂ SO ₄	30	1907–5900	1.42–2.62	8	0.887–0.942	0.273	1.430×10^{-2}	0.826	This work
Pullulan	0.05 M Na ₂ SO ₄	30	5.9–788	1.06–1.29	8	0.966–0.992	0.667	1.956×10^{-4}		Kasaai (2006)
Pullulan	0.05 M Na ₂ SO ₄	30	30–960	2.39–2.61	6	0.880–0.920	0.657	2.263×10^{-4}		Kasaai (2006)
Literature values for MHS equation constants for dextran at different solvent–temperature systems										
Dextran	0.5 M NaOH	20	500–2000				0.478	13.2×10^{-4}		Bose et al. (1982)
Dextran	Formamide	25	2–32				0.49	1.65×10^{-4}		Gekko (1971)
Dextran	H ₂ O	25	20–100				0.50	9.78×10^{-4}		Senti et al. (1955)
Dextran	H ₂ O	20	10–500				0.43	44.3×10^{-4}		Granath (1958)
Dextran	H ₂ O	20	66.2–522				0.62			Ioan et al. (2000)
Dextran	0.2 M NaNO ₃		40–590	1.6	5		0.39	48.5×10^{-4}		Bahary et al. (1995)
Dextran	Ethylene glycol	25	29.5–191.5				0.562	55.1×10^{-4}		Çatiker and Güner (2000)
Dextran	Ethylene glycol	45	29.5–191.5				0.562	48.5×10^{-4}		Çatiker and Güner (2000)
Dextran	0.4 M NaCH ₃ COOH/0.4 M CH ₃ COOH	25	10–2000				0.48	13.8×10^{-4}		Eremeeva and Bykova (1998)

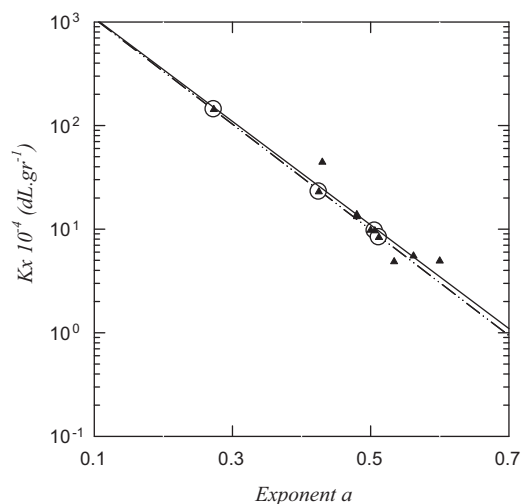


Fig. 2. The value of K versus exponent a . Open circle symbols refer to this work and filled triangle symbols represent literature results. Dash-dot line refers to this work and solid line represents entire results.

can be used to estimate the value of K in a solvent, if a in that solvent is known or vice versa.

5.1.1. The effect of solvent–temperature system on MHS equation constants

The values of 0.506 and 0.512 for dextrans with low and medium M_w and similar MWD ranges in two solvents (water at 25 °C and 0.05 M Na_2SO_4 at 30 °C) indicate that dextran behaves as a highly flexible chain (compact conformation) in both solvents. The presence of a small amount of salt in water may improve slightly the quality of the solvent. This data is consistent with the literature data for dextran in water (see Table 4 and Fig. 2).

5.1.2. The effect of molecular weight range on MHS equation constants

The values of 0.512, 0.425, and 0.273 were found for a in 0.05 M Na_2SO_4 at 30 °C. The value of a for dextran decreased with an increase in M_w . A value of 0.273 for ultra-high M_w indicates that the samples possess highly branched structures. The dispersity of the experimental points and deviation of the regression values from unity for the three plots increased with an increase in M_w (see Table 4). The higher the M_w , the greater the deviation from linearity. This data indicates that the number of branched points increased significantly with an increase in M_w . Thus, larger macromolecules with more branched points are more compact than those of the smaller ones.

5.2. Dilute solution properties of dextran and estimation of degree of chain branching for dextran relative to pullulan

(1–4)-Linkage provides a highly symmetrical structure and facilitates intermolecular association between units of different chains of carbohydrate polymers. (1–3)-Linkage imparts less symmetry and increases the solubility of carbohydrate polymers. (1–6)-Linkage dramatically improves water solubility of carbohydrate polymers. α -Configuration improves solubility of carbohydrate polymers in comparison with β -configuration (Izydorczyk, 2005). Dextran is more soluble in water than that of its linear counterpart, since the chain–chain interaction is less pronounced (Belitz et al., 2009; Izydorczyk, 2005; Van Aken, 2006). The degree of solubility in water decreases with an increase in g value. Dextrans with 43% branching through 1,3-linkages have been considered water insoluble (Mehvar, 2000). Dextran is stable in water, methyl sulphoxide,

formamide, glycerol, 4-methyl morpholine oxide, and hexamethyl phosphamide (De Belder, 2001). Among various reported solvents (see Table 4), water or an edible salt aqueous solution is the best solvent. This is due to: complete dissolution of the polymer; similar chain conformation of the polymer in comparison with other solvents; and an environmentally friendly material of water; and a safe solvent for food and medical applications. Dextran in comparison with its linear counterpart of equal M_w and equal concentration has a lower solution viscosity (Belitz et al., 2009; Cui, 2005). Dextran is a non-gelling polysaccharide and behaves almost Newtonian (Van Aken, 2006; Wang & Cui, 2005b).

Generally, the nature of a solvent and its interaction with a polymer would have an impact on that polymer conformation. In the presence of polar–polar interactions, $[\eta]$ as well as V_h polymer solution is greater and polymer conformation is larger than that of flexible polymers. If no major difference exists in polymer–solvent interactions of biopolymers in comparison with synthetic polymers, the conformation of biopolymers is flexible and the polymers behave like synthetic polymers. Dextran and pullulan are neutral and flexible polymers and no major differences in their interactions with different solvents in comparison with synthetic polymers were observed (see Table 4).

M_v , V_h , and $(S^2)^{1/2}$ for dextrans and pullulans were calculated and are given in Table 5. As described in Section 4.4, four different ratios have been used to estimate the value of g for dextran relative to pullulan. The difference between these two polymers is: change in their chemical structures, dextran is the branched counterpart of pullulan. The values of 0.512 and 0.425 for a were found for dextrans with low–medium (0.18–158 kDa) and high (226–1360 kDa) M_w , respectively. The value of 0.662 ± 0.005 for a has been already reported for pullulans with M_w range of 6–960 kDa (Kasaai, 2006). This data also indicates that: chain branching was observed in dextrans; and the number of chain branching points increased with an increase in M_w , as a decreased with an increase in M_w range.

The ratio $a_{\text{dextran}}/a_{\text{pullulan}}$ was found to be 0.771 ± 0.008 for M_w range of 1–158 kDa and 0.643 ± 0.006 for 226–767 kDa. Different ratios were also determined and given in Table 5. The values of different ratios are not the same, but there are correlation between the ratios and M_w . Each of the four ratios can be used as a measure of g for dextran relative to pullulan.

Fig. 3A shows $\log S$ (radius of gyration) versus $\log M_w$ for dextrans and pullulans. The slopes were found to be 0.49 and 0.56, respectively. Chain branching was not observed in a low- M_w sample $[(S^2_{\text{dextran}})^{1/2} \approx (S^2_{\text{pullulan}})^{1/2}]$ with equal M_w at $M_w < 20$ kDa, whereas a dextran sample with $M_w > 20$ kDa was smaller than that of pullulan. The larger M_w results in the greater divergence. Bahary et al. (1995) have found the values of 0.38 and 0.48 for the slopes ($\log S$ versus $\log M_w$). Nordmeier (1993) have also found the values of 0.42 and 0.56 for the slopes. Ioan et al. (2000) have found the value 0.43 for dextran. The lower values reported in the literature in comparison with the values obtained in this study (0.49 and 0.56), could be due to the narrower polydispersity and smaller g values for dextran samples those used in this study.

The plot $\log[\eta] - \log q_{\text{MHS}}$ versus $\log M_w$ for entire M_w range of dextran (0.18–5900 kDa) in 0.05 M Na_2SO_4 at 30 °C was illustrated in Fig. 3B. This plot is not linear and deviation from linearity increases with an increase in M_w . The dispersity of the experimental points also increased significantly in upper part of the plot. The macromolecule first consists of linear chains and latter the branch density increases slowly. The asymptotic region with a significant increase in chain branching is apparently attained when the M_w is enough large. Ioan et al. (2000) have reported that the plot of $\log[\eta]$ versus $\log M_w$ is non-linear for a wide range of M_w . They reported that for hyperbranched polymers the ratio, M_w/M_n does not remains constant for different M_w range.

Table 5
The values of M_w , $[\eta]$, $[\eta]M_w$, and $(S^2)^{1/2}$ for pullulans and dextrans.

Pullulan			Dextran			Ratio						
M_w (g mol ⁻¹)	$[\eta]^a$ (dL g ⁻¹)	$[\eta]^a M_w$ (dL mol ⁻¹)	(S^2) (nm ²)	$(S^2)^{1/2}$ (nm)	M_w (g mol ⁻¹)	$[\eta]^a$ (dL g ⁻¹)	$[\eta]^a M_w$ (dL mol ⁻¹)	(S^2) (nm ²)	$(S^2)^{1/2}$ (nm)	$([\eta]M_w)_{\text{dL}}/([\eta]M_w)_p$	$([\eta])_{\text{dL}}/([\eta])_p$	S_{dL}/S_p
6000	0.0659	395.4	96.5	9.8	6000	0.0715	429.2	101.9	10.1	1.085	1.085	1.03
10,000	0.0927	926.5	170.2	13.0	10,000	0.0929	929.2	170.5	13.1	1.003	1.003	1.008
20,000	0.1471	2942.2	367.6	19.2	20,000	0.1325	2650.2	342.9	18.5	0.901	0.901	0.964
40,000	0.2336	9343.0	794.3	28.2	40,000	0.1890	7558.5	689.6	26.3	0.809	0.809	0.932
50,000	0.2711	13553.0	1017.8	31.9	50,000	0.2118	10591.7	863.5	29.4	0.782	0.781	0.922
70,000	0.3393	23748.2	1479.3	38.5	70,000	0.2517	17616.2	1212.2	34.8	0.742	0.742	0.904
100,000	0.4304	43038.1	2198.9	46.9	100,000	0.3021	30208.1	1736.7	41.7	0.702	0.702	0.889
130,000	0.5127	66649.5	2943.3	54.3	130,000	0.3455	44916.5	2262.4	47.6	0.674	0.674	0.877
150,000	0.5640	84605.3	3450.7	58.7	150,000	0.3718	55766.5	2613.5	51.1	0.659	0.659	0.871
200,000	0.6833	136669.0	4750.6	68.9	200,000	0.4112	82248.6	3386.3	58.2	0.602	0.602	0.845
226,000	0.7414	167552.9	5441.7	73.8	226,000	0.4332	97896.1	3803.2	61.7	0.584	0.584	0.836
300,000	0.8956	268666.9	7455.0	86.3	300,000	0.4886	146574.6	4977.5	70.6	0.546	0.546	0.818
400,000	1.0850	433997.4	10263.5	101.3	400,000	0.5521	220849.5	6541.9	80.9	0.509	0.509	0.799
500,000	1.2591	629558.5	13152.0	114.7	500,000	0.6070	303524.1	8086.6	89.9	0.482	0.482	0.784
600,000	1.4219	853161.4	16106.1	126.9	600,000	0.6560	393574.1	9615.8	98.1	0.461	0.461	0.773
700,000	1.5759	1103142.2	19115.7	138.3	700,000	0.7004	490259.1	11132.3	105.5	0.444	0.444	0.763
760,000	1.6648	1265229.6	20945.1	144.7	760,000	0.7253	551214.2	12037.0	109.7	0.436	0.436	0.758
900,000	1.8477	1662893.3		158.5	900,000	0.7793	701385.2	14134.3	118.9	0.422	0.422	0.750

^a $[\eta]$ in 0.05 M Na₂SO₄ at 30 °C.

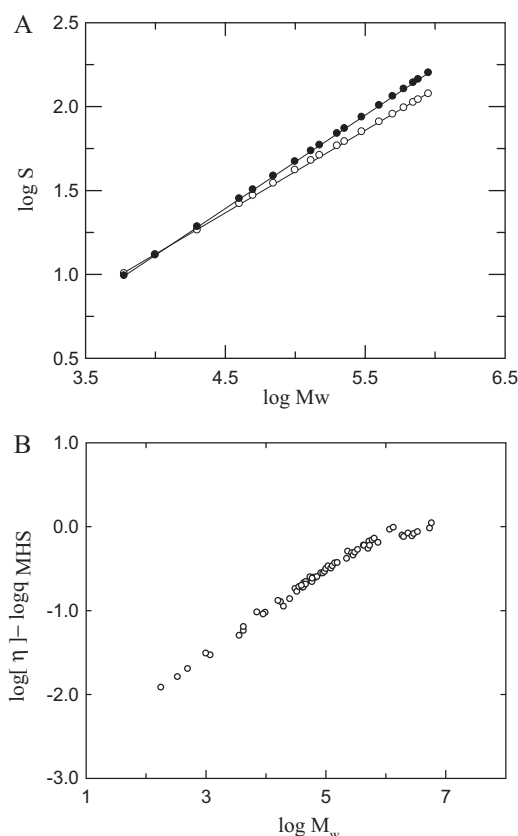


Fig. 3. (A) Log S versus $\log M_w$. Open symbols refer to dextran samples and filled symbols correspond to pullulan samples (); and (B) $\log [\eta] - \log q_{\text{MHS}}$ versus $\log M_w$ for dextrans in the entire M_w range (0.18–5900 kDa).

All of the results given in this study (a , k , g , $(S^2)^{1/2}$ versus M_w , and plot of $\log [\eta] - \log q_{\text{MHS}}$ versus $\log M_w$ for entire M_w range for dextrans strongly demonstrated that: (i) small molecules (less than 20 kDa) of dextrans and pullulans are almost identical; and (ii) very large macromolecules of dextrans are highly branched and have highly flexible structures. On the whole: (1) the results presented in Fig. 3A and B are in agreement with the results obtained for a , k and g values; and (2) all of the results given in this study are internally consistent.

The results given in this study can be used by researchers as a reliable source for comparison. This is because: experimental conditions (solvent–temperature) for dextran and its counterpart are identical; the polydispersity is taken into consideration; and experimental points on the plots are large in comparison with the literature reported data.

The results presented in this manuscript are useful for research groups who are interested in determining M_w , $(S^2)^{1/2}$, MWD, and g of the resulting polymers obtained from degradation; hydrolysis; fragmentation/depolymerization; and fractionation of the original dextrans and to compare them with the corresponding data of the original ones. The results can be also used to characterize fractionated dextran samples as polymer standards. Knowledge on the above-mentioned parameters enables one to improve properties and consequently enhance efficiency of the polymer. In addition, the latter information may be useful for nano-technology as follows.

5.3. The use of molecular weight and molecular size data in nanotechnology

The properties and applications of oligomers and polymers depend on their M_w as well as their molecular sizes. Hydrocolloids

such as dextrans are used in various branches of food and medicine because of their versatile properties. To optimize their efficiency in various branches of food and medicine, it is necessary to characterize their M_w as well as their $(S^2)^{1/2}$ accurately. The plot $(S^2)^{1/2}$ versus M_w for dextran can be used to estimate M_w of desirable nano-particles from their corresponding $(S^2)^{1/2}$. Thus, the information given in this manuscript may be used in various branches of nanotechnology.

6. Conclusions

This study resulted in the following MHS equations for dextrans:

$$[\eta] = 9.64 \times 10^{-4} M_v^{0.506} = 9.64 \times 10^{-4} q_{MHS} M_w^{0.506} \\ = 9.01 \times 10^{-4} M_w^{0.506}$$

for the polymer samples in water at 25 °C with M_w range of 1–676 kDa, M_w/M_n ($1.23 < M_w/M_n < 1.94$), q_{MHS} ($0.899 < q_{MHS} < 0.971$) and average value of 0.934 for q_{MHS} ;

$$[\eta] = 8.32 \times 10^{-4} M_v^{0.512} = 8.32 \times 10^{-4} q_{MHS} M_w^{0.512}$$

with M_w (0.18–158 kDa), M_w/M_n ($1.00 < M_w/M_n < 2.28$), and q_{MHS} ($0.870 < q_{MHS} < 1.00$) in 0.05 M Na_2SO_4 at 30 °C;

$$[\eta] = 2.297 \times 10^{-3} M_v^{0.425} = 2.297 \times 10^{-3} q_{MHS} M_w^{0.425}$$

with M_w (226–1360 kDa), M_w/M_n ($1.34 < M_w/M_n < 2.17$), and q_{MHS} ($0.883 < q_{MHS} < 0.959$) in 0.05 M Na_2SO_4 at 30 °C; and

$$[\eta] = 1.43 \times 10^{-2} M_v^{0.273} = 1.43 \times 10^{-2} q_{MHS} M_w^{0.273}$$

with M_w (1907–5900 kDa), M_w/M_n ($1.42 < M_w/M_n < 2.62$), and q_{MHS} ($0.887 < q_{MHS} < 0.942$) in 0.05 M Na_2SO_4 at 30 °C, where $[\eta]$ is expressed in dL g^{-1} .

The exponent values of 0.506 in water at 25 °C and 0.512, 0.425, and 0.273 in 0.05 M Na_2SO_4 at 30 °C indicate that dextran behaves as a compact conformation in both solvents. Smaller values for a were obtained for higher molecule weights. The values of a and their corresponding K values were highly inversely correlated. The g value decreased with an increase in M_w . Molecular size parameters (V_h , $[\eta]$, and $(S^2)^{1/2}$) for small macromolecules (less than 20 kDa) of dextrans and pullulans are almost identical, whereas the latter parameters for dextrans with $M_w > 20$ kDa were smaller than those of their linear counterparts with equal M_w . Chain branching was not observed in low M_w .

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