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Phase equilibrium in polyethylene glycol/maltodextrin aqueous two-phase systems

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Abstract

The polysaccharide maltodextrin (MD) can provide a low cost alternative to the fractionated dextran for the use with polyethylene glycol (PEG) in aqueous two-phase, two-polymer systems. The physical characteristics of these MD/PEG systems are similar in many respects to dextran/PEG systems. These systems' behaviour was studied at 25°C and local atmospheric pressure (727 mmHg) under several concentration ratios and molecular weights of MD/PEG. Molecular weight of PEG was 1450, 8000 and 10 000; and MD's molecular weight was 2000 and 4000. This paper establishes phase diagram data, subsequently adjusting the NRTL model for the calculation of activity coefficients. The results are satisfactory and show the good descriptive quality of the model. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Liquid–liquid equilibrium; Aqueous two-phase systems; Maltodextrin; Polyethylene glycol; NRTL

1. Introduction

Aqueous two-phase systems can be formed by combining either two "incompatible" polymers or a polymer and a salt in water, above a certain critical concentration (Szlag, Giuliano & Synder, 1990). Many authors have tested systems like that and determined their phase diagrams (Cheluget, Gelinas, Vera & Weber, 1994; Kula, 1979; Silva, Coimbra & Meirelles, 1997; Snyder, Cole & Szlag, 1992; Walter, Brooks & Fisher, 1985). Comprehensive reviews have been compiled by Albertsson (1986) and Zaslavsky (1995). PEG/salt systems have been used in large scale for protein separation. These systems are attractive because of low-cost, rapid phase disengagement and the availability of commercial separators, which allow faster continuous protein separation (Coimbra, Thömes & Kula 1994; Coimbra, Thömes, Meirelles & Kula, 1995; Coimbra, Mojola & Meirelles, 1998). Polymer/salt systems, on the other hand, are not very selective, can damage fragile proteins and in some cases, when high salt concentration is used, became a waste disposal problem. In contrast polymer/polymer systems can be more selective by incorporating appropriate ions or ligands in the system. The most common polymer/polymer system is composed by dextran and PEG, but this system is very expensive for scale up. This problem can be solved by the use of alternative polymers (Atkinson & Johns, 1994; Chistian, Manley-Harris & Richards, 1998; Szlag et al., 1990). Here the possibility of using low-cost starch derivatives (maltodextrin) as replacements for dextran was investigated.

Several attempts have been made in the literature to describe the liquid–liquid equilibrium in aqueous two-phase systems (Kang & Sandler, 1987; Wu, Zhu, Lin & Mei, 1996; Wu, Lin & Zhu, 1998; Wu, Zhu, Lin & Lian, 1999). The thermodynamic description of this phase equilibrium is important as it could provide the basis for extrapolating experimental data and predicting phase compositions when such data are not available. The thermodynamic representation of phase equilibrium could also aid the design and process optimisation of the aqueous two-phase extraction technique. In this paper the NRTL model for the activity coefficients was used to correlate the equilibrium data for PEG/MD systems.

2. Materials and methods

MD 2000 and 4000 (commercial name Loremalt 2030 and Loremalt 2001, respectively) were kindly supplied by Companhia Lorenz (Blumenau, SC, Brazil). Polyethylene glycols 1450, 8000 and 10 000 were purchased from SIGMA.

The polymers were analysed by Gel Permeation Chromatography (GPC) in a Waters chromatograph using

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Nomenclature

A	Parameter of NRTL model
C	Number of components
G	Parameter of NRTL model
M	Molecular weight
N	Total number of tie-lines in a given group of data
T	Absolute Temperature (K)
w	Weight fraction
Superscripts/subscripts	
calc	Calculated data
ex	Experimental data
i, j, k	Components
n	Tie-line
I, II	Phases
Greek letters	
α	NRTL parameter
γ	Activity coefficient
τ	NRTL parameter

the following experimental conditions: water as the mobile phase at a rate of 0.8 μl/min, injection temperature of 40°C, refraction index detector, sample injection 100 μl. This methodology was used in order to obtain the polymers' molecular distribution and polydispersity index (M_w/M_n). The results are shown in Table 1.

The water content of each polymer was determined through Karl Fisher titration using a Metrohm equipment (Swiss). The water content was taken into account for preparing the stock solutions.

2.1. Phase diagrams

Stock solutions, containing 55 and 50% of MD and PEG, respectively, were prepared by the addition of Milli Q water to a known quantity of polymer. The diagrams for PEG (1450, 8000 and 10 000) and MD (2000 and 4000) were determined in centrifuge tubes. Mixtures consisting of known weights of polymer stock solutions were prepared on an analytic balance (A200 S Sartorius, Germany), accurate to 0.0001 g. Typically 15 g of a system were prepared. Systems were mixed for 10 min and then centrifuged (BR4i model, Jouan, France) at 2900 g for 40 min at 25°C. The tubes were brought to equilibrium in a thermostatic bath (Viscotherm VT2, Physica, Germany) at 25 ± 0.1°C and local atmospheric pressure (727 mmHg) for 5 h. After this treatment, the two phases became clear and transparent, and the interface was well defined. Samples of the two phases were analysed.

Table 1
Polymer characterization by GPC

Product	Average molecular weight	Polydispersity index
Loremalt 2030	2017	1.31
Loremalt 2001	4004	1.98
PEG 1450	1468	1.03
PEG 8000	8768	1.09
PEG 10000	11 589	1.10

2.2. Analysis of phase compositions

The phase compositions were determined using a combination of polarimetry and freeze drying, according to the methodology suggested by Chistian et al. (1998) for a polysaccharide/PEG system. The MD concentration was determined by polarimetry (Carl Zeiss Jena, POLAMAT A model, equipped with a mercury lamp at 546 nm, Germany), since PEG is optically inactive. The water concentration was determined by freeze drying (EZ DRY model, FTS Systems, New York, USA) at −54°C and 100 m Torr for 48 h. The PEG concentration was determined by difference. The PEG concentration was further checked by refractometry (Polskie Zaklady Optyczne (PZO), Minska 25, 03-808 Warszawa-RL3, Germany). The average standard deviations of the phase concentrations (analysed in triplicate) were ±0.004% for water, ±0.006% for MD and ±0.007% for PEG.

3. Results

Several equilibrium diagrams were built for the system PEG/MD containing different polymer's molecular weight. The PEG 1450/MD 2000 mixture does not form aqueous two-phase system at the concentrations selected here. All the results are expressed as weight percentage. Four tie lines were determined for each polymer combination, except for the system PEG 1450/MD 4000, for which only three tie lines were determined. The experimental compositions for all systems are given in Tables 2 and 3.

The complete diagram for the PEG 1450/MD 4000

Table 2
Phase compositions for PEG/MD 2000 systems

System	Total system			Top phase			Bottom phase		
	MD	PEG	Water	MD	PEG	Water	MD	PEG	Water
PEG 8000/MD 2000	34.80	11.22	53.97	28.65	15.59	55.75	50.42	0.81	48.75
	36.81	12.97	50.21	27.19	18.57	54.23	54.99	0.60	44.40
	38.78	15.05	46.15	24.36	25.42	50.20	59.21	0.53	40.25
	40.50	16.69	42.80	23.27	29.08	47.64	63.87	0.89	35.23
PEG 10000/MD 2000	34.63	10.72	54.64	27.50	14.64	57.84	50.59	0.45	48.94
	35.77	12.38	51.84	25.61	18.04	56.34	54.99	0.73	44.27
	38.79	15.15	46.05	23.47	26.09	50.42	59.23	0.91	39.84
	40.26	16.81	42.91	22.47	29.60	47.92	60.91	0.90	38.17

system at 25°C is shown in Fig. 1. It can be seen that good linear fittings were obtained for the experimental data. The tie lines were determined by linear regression of each corresponding set of total, bottom phase, and top phase concentrations. The tie lines compositions were confirmed, within an experimental error of 3%, by performing mass balances on the top and bottom equilibrium compositions to determine the amounts of PEG and MD used to generate the total mixture. These mass balance deviations are in good agreement with results reported in the literature for other systems (Albertsson, 1986; Cheluget et al., 1994; Silva et al., 1997; Snyder et al., 1992).

For most systems the PEG concentration in bottom phase is very small, and in some cases PEG is almost excluded from this phase. Similar results were reported by Szlag et al. (1990). The only exception in this behaviour was observed for the PEG 1450/MD 4000 system. All PEG/MD systems were characterised by the presence of considerable quantities of MD in the top phase. This quantity increased markedly as the molecular weight of the polymers decreased. The same behaviour is observed by Atkinson and Johns (1994). In all systems studied in the present work the MD concentration in top phase is relatively high when compared to systems formed with PEG/dextran or PEG/salt (Albertsson, 1986; King, Blanch & Prausnitz, 1988; Silva et al., 1997; Zaslavsky, 1995).

The concentration of MD required to form two phases with PEG is much greater than that of dextran or salt. The systems with the lowest PEG molecular weight required particularly large concentrations of both polymers to exhibit phase splitting. For this reason more concentrated stock solutions (PEG 55%, MD 60%) were used in this case.

Retrogradation of MD has been reported to be a further disadvantage of its use as a phase-forming aqueous two-phase system polymer (Szlag et al., 1990). In this work, no retrogradation was observed due to the relatively low MD molecular weight. MD is produced by Companhia Lorenz using a combination of both acidic and enzymatic hydrolysis. Such processing strategy reduces the tendency to retrogradation (Atkinson & Johns, 1994). This gives to the low molecular weight MD a distinct cost advantage over other specifically modified starch derivatives, used to retard retrogradation.

Fig. 2 shows the binodal curves for the PEG/MD 4000 systems. These curves were determined by fitting a sigmoidal (Boltzmann) equation to the experimental data. The decrease in PEG molecular weight caused a decrease on the two-phase region, a behaviour already reported in the literature for similar systems (Albertsson, 1986; Szlag et al., 1990). The advantage of using less PEG of higher molecular weight is, however, offset by the higher viscosity of such solutions.

Table 3
Phase compositions for PEG/MD 4000 systems

System	Total system			Top phase			Bottom phase		
	MD	PEG	Water	MD	PEG	Water	MD	PEG	Water
PEG 1450/MD 4000	33.82	10.96	55.21	30.86	12.68	56.44	43.48	5.92	50.59
	36.18	12.54	51.26	27.18	17.76	55.04	50.46	4.31	45.21
	43.18	13.75	43.05	25.91	24.59	49.49	61.42	2.29	36.28
PEG 8000/MD 4000	34.98	11.02	53.99	18.55	18.84	62.60	53.54	0.00	46.45
	36.71	13.23	50.04	17.70	23.15	59.13	58.12	0.00	41.87
	38.93	15.05	46.00	15.43	30.19	54.37	60.15	0.00	39.84
	40.11	16.81	43.06	14.97	36.53	48.49	63.91	0.12	35.96
PEG 10000/MD 4000	34.02	10.61	55.36	18.15	18.75	63.08	53.13	0.00	46.86
	35.77	12.52	51.69	17.31	23.02	59.66	57.60	0.00	42.39
	37.84	14.55	47.60	15.17	30.14	54.68	58.11	0.04	41.84
	39.86	16.43	43.70	14.56	32.70	52.72	63.40	0.50	36.08

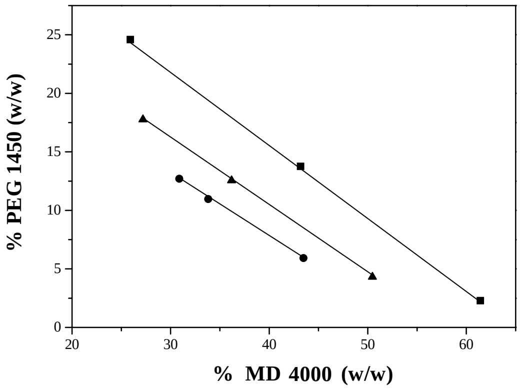

Fig. 1. Experimental tie lines for the system PEG 1450/MD 4000 at 25°C.

3.1. Modelling

The experimental tie line data were utilised in adjusting the parameters of the NRTL model for the activity coefficients. The weight fraction was used as the unit of concentration due the large difference in molecular weights between the components in the systems. The same procedure was suggested by Oishi and Prausnitz (1978) for modelling solvent activities using the UNIQUAC and UNIFAC models.

The Eq. (1) shows the NRTL model expressed in weight fraction:

$$\ln \gamma_i = \frac{\sum_{j}^{C} \frac{\tau_{ji} G_{ji} w_j}{M_j}}{\sum_{j}^{C} \frac{G_{ji} w_j}{M_j}} + \sum_{j=1}^{C} \left[\frac{w_j G_{ji}}{M_j \sum_{k}^{n} \frac{G_{kj} w_k}{M_k}} \left(\tau_{ij} - \frac{\sum_{k}^{C} \frac{\tau_{kj} G_{kj} w_k}{M_k}}{\sum_{k}^{C} \frac{G_{kj} w_k}{M_k}} \right) \right] \tag{1}$$

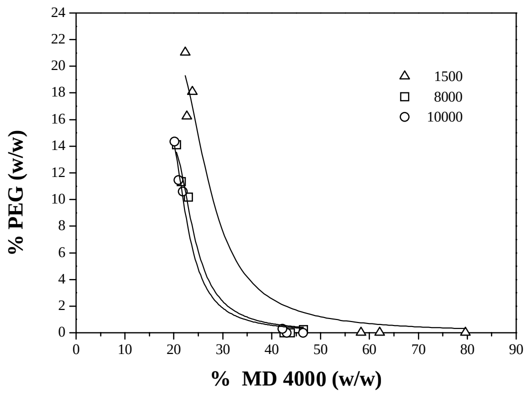

Fig. 2. Binodal curves for the systems PEG/ MD 4000 at 25°C.

Table 4
Adjusted parameters of the NRTL model ((1) MD 2000; (2) water; (3) PEG 8000; (4) PEG 10 000; (5) MD 4000; (6) PEG 1450)

Parameters	A_{ij} (K)	A_{ji} (K)	$\alpha_{ij} = \alpha_{ji}$
12	−1415.9	2876.7	0.27937
13	−4800.2	−548.03	0.42858
14	−4419.3	−188.24	0.46957
23	−1588.0	−3997.9	0.20001
24	695.67	−3095.5	0.29902
52	−1449.4	3059.30	0.27937
53	−4441.1	−148.26	0.42858
54	−2129.7	−656.49	0.47000
56	−3228.3	−150.42	0.4700
62	−5000.1	−2295.5	0.20125

where

$$G_{ij} = \exp(-\alpha_{ij}\tau_{ij}) \tag{2}$$

$$\tau_{ij} = A_{ij}/T \tag{3}$$

$$\alpha_{ij} = \alpha_{ji}. \tag{4}$$

Following the procedure developed by Stragevitch and d'Avila (1997), adjustments of the parameters were made by minimisation of the maximum likelihood objective function. Table 4 shows the parameters estimated from the experimental data. Figs. 3 and 4 show the experimental and calculated tie lines and the calculated binodal curves for the systems PEG 8000/MD 4000 and PEG 8000/MD 2000. For these aqueous two-phase systems the correlation was successful. The experimental data were compared to the calculated values by liquid–liquid flash using the adjusted parameters. The deviation between the experimental and calculated weight fractions for each system is given in Table 5, calculated according to Eq. (5):

$$\Delta w = 100\sqrt{\frac{\sum_{n=1}^{N}\sum_{i=1}^{C}\left[(w_{n,i}^{I,ex} - w_{n,i}^{I,calc})^2 + (w_{n,i}^{II,ex} - w_{n,i}^{II,calc})^2\right]}{2NC}}. \tag{5}$$

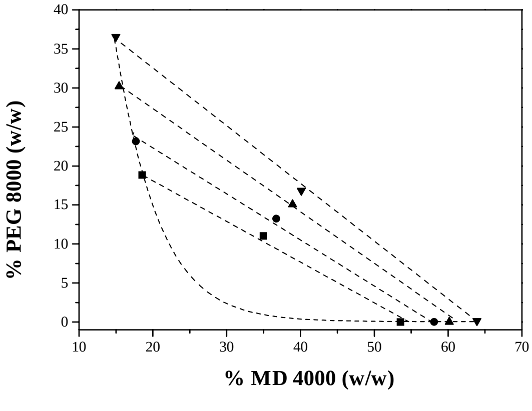

Fig. 3. Experimental and calculated tie-lines and the calculated binodal curve for the PEG 8000/MD 4000 System.

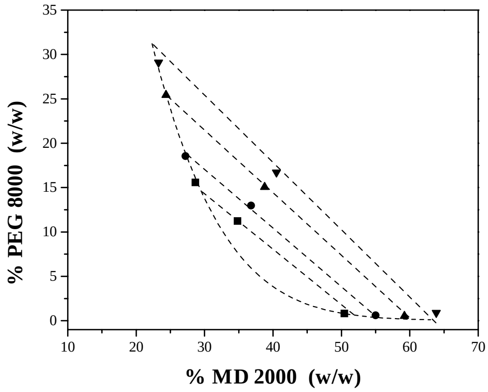

Fig. 4. Experimental and calculated tie-lines and the calculated binodal curve for the PEG 8000/MD 2000 System.

The best results were obtained for the system PEG 8000/MD 4000. For the system PEG 1450/MD 4000 the results shown are not satisfactory if compared with other systems. The low average deviation of the NRTL model for the MD/PEG/Water systems shows that it is possible to obtain a significant set of parameters, which describes well such systems.

4. Conclusions

The present work analysed the equilibrium phase behaviour of MD and PEG systems at 25°C and atmospheric pressure, under several conditions of concentrations and molecular weights of the polymers. The PEGs with molecular weights 1450, 8000 and 10 000 and MD with molecular weights 2000 and 4000 were combined to form the systems.

The PEG 1450/MD 2000 mixture does not form an aqueous two-phase system at the concentrations selected in this work. For the other systems the PEG concentration in bottom phase is very small, and in some cases PEG is almost excluded from this phase. The only exception in this behaviour was observed for the PEG 1450/MD 4000 system, where the molecular weight of MD is higher than PEG.

In all systems the MD concentration in top phase is relatively high when compared to systems formed with PEG/dextran or PEG/salt. The concentration of MD required to form two phases with PEG is much greater than that of dextran or salt. The decrease in PEG molecular weight caused a decrease in the two-phase region.

Table 5
Percent deviation of experimental to calculated weights fractions

PEG/MD system	$\Delta w\%$
2000/8000	0.7777
2000/10000	0.6992
4000/1450	2.6272
4000/8000	0.3430
4000/10000	0.6271
Average deviation	1.0148

The NRTL model for the MD/PEG/Water systems showed that it was possible to obtain a significant set of parameters that well describes such systems.

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