Liquid−Liquid Equilibrium for Ternary Systems Containing a Sugar + a Synthetic Polymer + Water

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This paper presents phase equilibrium data for ternary liquid mixtures containing glucose or maltose + poly(propylene glycol)s or block copolymers of ethylene oxide and propylene oxide units + water. The nonrandom two-liquid (NRTL) and the universal quasi-chemical activity coefficient (UNIQUAC) models were employed to model phase equilibrium, allowing the calculation of phase compositions. Good agreement between the experimental data and the model results was achieved.

Introduction

The use of aqueous two-phase systems (ATPS) for purification and recovery of biocompounds has gained attention since the early studies of Albertsson et al.[1] The ATPS are capable of promoting the separation of macromolecules, viral particles, cell fragments, cell organelles and; whole cells in a mild medium, in a way they can be partitioned and purified without loss of biological activity. The most common polymer + polymer system is composed of poly(ethylene glycol) (PEG) and dextran.[1,2] Poly(propylene glycol) (PPG) is a polymer that is structurally closely related to PEG. PPGs of low molecular weight are soluble in water, while high molecular mass ones are only partially soluble.[3] Some recent purification techniques employing ATPS suggest the use of thermoseparating polymers, such as copolymers of ethylene oxide (EO) and propylene oxide (PO) units, to reduce the cost of polymer recovery.[4,5]

Dextran is a high cost polymer which makes difficult the use of ATPS in large scale processes. Maltodextrin (MD) can be used as a lower cost substitute for dextran.[6,7] MD is a commercial polymer of D-glucose units linked primarily by $\alpha(1\rightarrow4)$ bonds. This polymer is obtained by acidic and enzymatic hydrolysis of starch. Low molecular mass saccharides, such as glucose, maltose, and sucrose, can also be used for dextran replacement, with the advantage that such compounds are of common occurrence in the food industry.

Thermodynamic models, namely the NRTL and UNIQUAC equations, are frequently applied to model phase equilibrium in many polymeric systems such as PEG + dextran,[8,9] PPG + maltodextrin,[6] and PEG + maltodextrin.[7] On the other hand, polymers of propylene oxide and/or ethylene oxide might form two-phase systems when mixed with some sugars in aqueous media.

In this work, two-phase liquid systems composed by a synthetic polymer (either poly(propylene glycol) of various molar masses or block copolymers of EO and PO units) + a sugar (glucose or maltose) + water were prepared and phase compositions measured. Binary interaction param-

eters for the NRTL and UNIQUAC thermodynamic models were obtained. The models showed good agreement with experimental data.

Materials and Methods

A total of seven two-phase systems containing water, a sugar, and a synthetic polymer were investigated. The following reagents were employed: poly(propylene glycol) (PPG) of molar masses 400, 725, and 1200; block copolymers of EO and PO units with molar masses 1925 (PE 61) (10%; EO, corresponding to 10% ethylene oxide units and 90% propylene oxide units in the polymer chain), 2100 (PE 62) (20%; EO), and 2450 (PE 64) (40% EO); poly(ethylene glycol) (PEG) of molar masses 8000, 10 000, and 20 000; maltose and glucose. PPGs 400 and 1200 were purchased from Fluka, and PPG 725 was purchased from Aldrich; block copolymers were kindly supplied by Oxiteno Brasil S/A (São Paulo, Brazil); PEGs 8000 and 10 000 were purchased from Sigma, and PEG 20 000 was purchased from Merck; maltose and glucose were purchased from Merck. All systems were prepared in test tubes by dissolving the desired pairs of the above reagents in distilled and deionized water (Millipore, USA).

The mixtures were centrifuged at $4000g$ for 20 min in a thermostated centrifuge (Jouan BR 4i, France). After centrifugation, the tubes were kept overnight (at least for 12 h) in a constant temperature water bath controlled to ±0.1 °C (Cole Parmer, USA). After this treatment the systems reached an equilibrium state characterized by a well-defined interface and clean, transparent bottom and top phases.

Samples of the phases were collected with syringe and needles. After proper dilution, the water content of the sample was obtained by freeze-drying (EZ-Dry model, FTS Systems, New York, USA). The sugar concentration was obtained by polarimetry (Carl Zeiss Polamat A, Germany), and the third component concentration was calculated by difference. This technique was previously used by Silva and Meirelles[6,7] with good results.

To have a better estimation of the experimental uncertainties, each tie line was measured at least twice. This means that solutions with practically the same total

Table 1. Phase Compositions (mass %) for the Systems Containing Glucose + Synthetic Polymer + Water at 298.15 K

system	overall composition			upper phase			lower phase		
	glucose	polymer	water	glucose	polymer	water	glucose	polymer	water
PPG 400	12.13	42.02	45.85	5.18	62.82	32.00	14.47	35.81	49.72
	19.98	35.03	44.99	2.08	77.34	20.58	29.06	15.88	55.06
	18.00	48.05	33.95	1.90	80.32	17.78	38.42	11.07	50.51
	24.95	39.92	35.13	1.53	83.89	14.58	45.38	8.33	46.29
	19.94	50.07	29.99	1.55	83.48	14.97	45.53	7.15	47.32
PPG 1200	13.36	40.00	46.64	0.12	94.05	5.83	23.35	2.09	74.56
	13.29	46.79	39.92	0.07	94.59	5.34	26.50	2.11	71.39
	17.33	43.38	39.29	0.09	94.93	4.98	32.63	1.43	65.94
	20.07	46.72	33.21	0.11	95.44	4.45	40.92	1.18	57.90
PE 61	7.62	30.25	62.13	0.84	83.03	16.13	10.92	3.55	85.53
	10.04	40.07	49.89	0.73	86.98	12.29	17.37	3.51	79.12
	13.30	33.17	53.53	0.67	88.63	10.70	20.45	3.01	76.54
	20.10	40.06	39.84	0.75	91.43	7.82	35.69	1.25	63.06

Table 2. Phase Compositions (mass %) for the Systems Containing Maltose + Synthetic Polymer + Water at 298.15 K[a]

system	overall composition			upper phase			lower phase		
	maltose	polymer	water	maltose	polymer	water	maltose	polymer	water
PPG 400	15.93	30.99	53.08	2.75	62.81	34.44	20.78	22.03	57.19
	19.96	35.34	44.70	2.17	71.46	26.37	29.04	17.70	53.26
	18.02	45.04	36.94	1.45	75.78	22.77	36.38	11.90	51.72
	17.97	48.15	33.88	1.01	80.12	18.87	41.00	9.36	49.64
PPG 725	11.40	28.69	59.91	0.17	88.90	10.93	15.50	7.12	77.38
	16.61	32.12	51.27	0.03	89.57	10.40	24.39	4.49	71.12
	19.42	33.46	47.12	0.04	89.98	9.98	30.57	2.42	67.01
PPG 1200	13.28	46.48	40.24	0.09	93.94	5.97	26.42	0.87	72.71
	16.61	43.38	40.01	0.03	94.87	5.10	31.24	1.33	67.43
	19.62	39.12	41.26	0.08	94.14	5.78	35.05	0.01	64.94
	20.00	46.74	33.26	0.03	94.95	5.02	41.03	0.51	58.46
PE 61	5.17	35.03	59.80	0.54	80.81	18.65	7.97	5.04	86.99
	9.96	35.04	55.00	0.72	83.73	15.55	15.9	2.95	81.15
	12.01	40.02	47.97	0.66	85.78	13.56	21.39	2.21	76.40
	16.90	49.68	33.42	0.41	90.45	9.14	37.44	0.39	62.17

[a] Except for the system maltose + PPG 725 + water, measured at 303.15 K.

composition were prepared at least in duplicate and the concentrations of the phases were measured for both experiments separately. The values for overall and phase compositions published in the present work are the averaged ones. The average standard deviations in the overall compositions were 0.16% for polymer, 0.22% for sugar, and 0.32% for water. These values can be considered as an estimation of the experimental uncertainties in the composition of the overall mixture. The standard deviations of the phase compositions were 0.29% in the top phase and 0.38% in the bottom phase for the synthetic polymer, 0.04% in the top phase and 0.49% in the bottom phase for sugar, and 0.28% in the top phase and 0.52% in the bottom phase for water. It should be noted that such uncertainties are relatively higher than those reported for similar systems in the literature. This occurs because the uncertainties in the present work reflect the errors accumulated in the whole experimental procedure, while for most experimental results reported in the literature the uncertainties reflect only the errors in the analytical procedures used for determining the phase compositions.

For the mathematical modeling, the NRTL[10] and the UNIQUAC[11] models were employed. Software was used for parameter adjustment, which employs a modified Simplex method for the minimization of the composition objective function expressed by eq 1:[12]

$$S = \sum_{m}^{D} \sum_{n}^{N} \sum_{i}^{K-1} \left[\left(\frac{w_{i,n,m}^{I,ex} - w_{i,n,m}^{I,calc}}{\sigma_{w_{i,n,m}^{I}}} \right)^2 + \left(\frac{w_{i,n,m}^{II,ex} - w_{i,n,m}^{II,calc}}{\sigma_{w_{i,n,m}^{II}}} \right)^2 \right] \quad (1)$$

in which $w_{i,n,m}$ is the mass fraction of component i in the

tie line n and in the data group m, the superscripts I and II stand for phases I and II, respectively, the superscripts ex and calc stand for experimental and calculated data points, respectively, D is the total number of data groups, N is the total number of tie lines in each group m, and K is the number of components in the mth data group. The subscripts i, n, and m are compound, tie line, and group number, respectively. $\sigma_{w_{i,n,m}^{I}}$ and $\sigma_{w_{i,n,m}^{II}}$ are the average standard deviations observed in the compositions of the two liquid phases.

Results and Discussion

The phase compositions for the systems studied are given in Tables 1 and 2. All concentrations are expressed in mass percent. The phase diagrams obtained are shown in Figures 1−7. In general it was observed that the higher the molar mass of the polymers, the larger the region of phase splitting, which is in agreement with literature data for other kinds of polymeric systems.[1,6,7]

It was observed that for the higher molar mass PPGs (725 and 1200) the systems showed a similar behavior, with a top phase with very low sugar concentration, varying in a narrow range for all the overall compositions. In the case of PPG 725 the bottom phase contains significant amounts of the synthetic polymer, but for PPG 1200 the corresponding concentrations are lower. These results are in accordance with the data reported by Silva and Meirelles[6] for PPG + maltodextrin systems.

The systems containing PE 61 showed a behavior comparable to that of the systems containing PPG 725. It is interesting to notice that the molar mass of PE 61 is more than two times greater than the molar mass of PPG
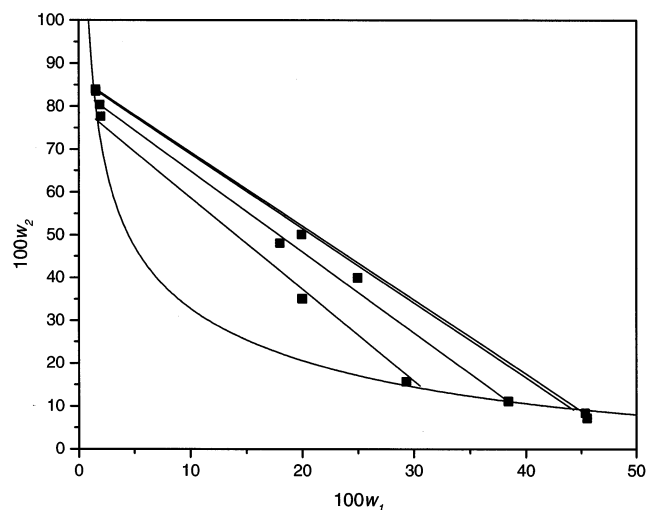
Figure 1. System containing glucose (1) + PPG 400 (2) + water (3): ■, experimental; —, NRTL.
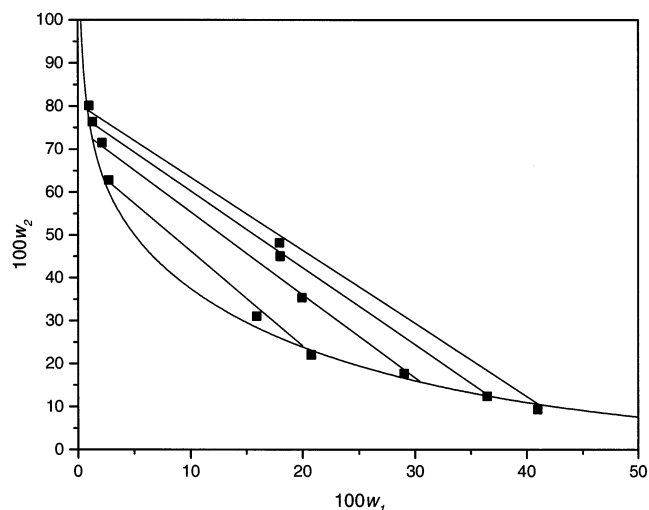

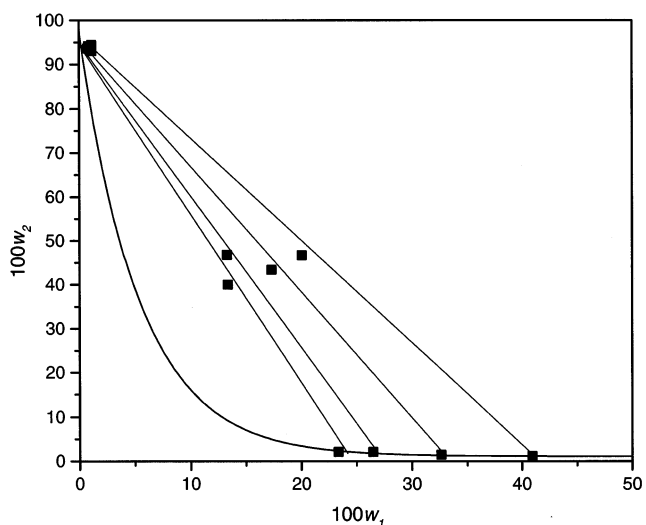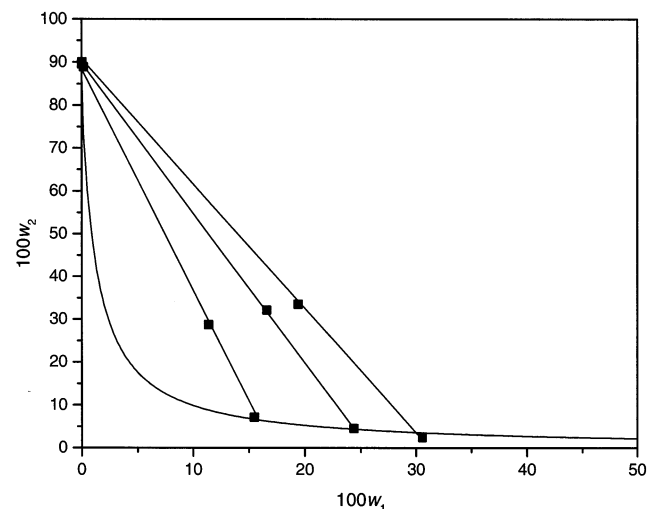**Figure 2.** System containing glucose (1) + PPG 1200 (2) + water (3): ■, experimental; —, NRTL.

Figure 3. System containing glucose (1) + PE 61 (2) + water (3): ■, experimental; —, NRTL.

Figure 4. System containing maltose (1) + PPG 400 (2) + water (3): ■, experimental; —, NRTL.

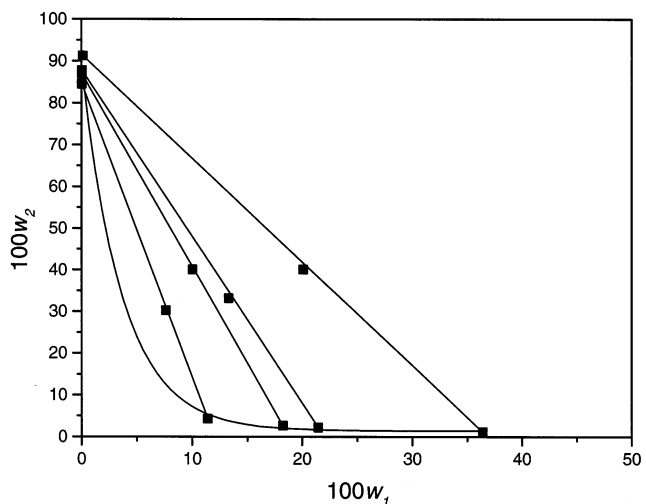**Figure 5.** System containing maltose (1) + PPG 725 (2) + water (3): ■, experimental; —, NRTL.
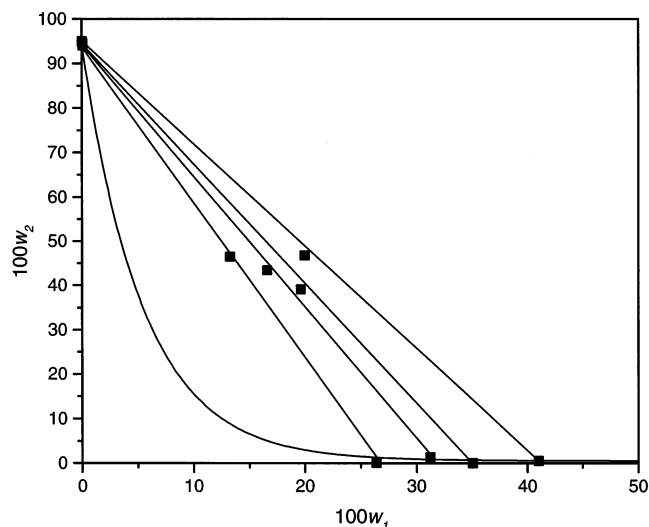

Figure 6. System containing maltose (1) + PPG 1200 (2) + water (3): ■, experimental; —, NRTL.

725, which means that the inclusion of only 10% ethylene oxide groups in the molecule is more important for reducing the hydrophobicity of the molecule than the higher molar

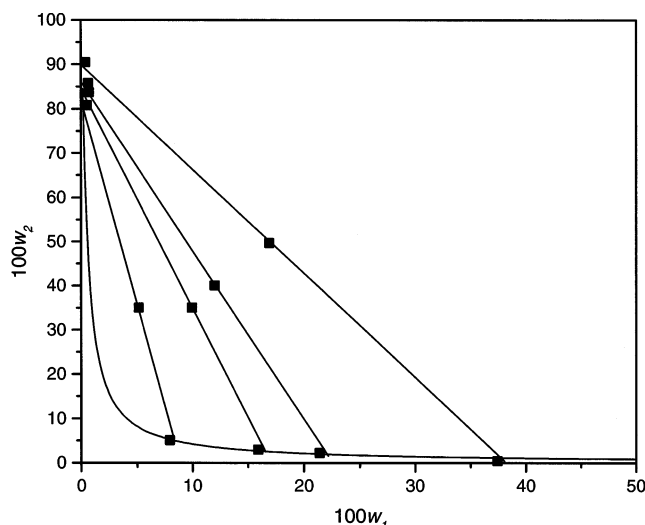mass is for increasing the concentration range of phase splitting.

Figure 7. System containing maltose (1) + PE 61 (2) + water (3): ■, experimental; —, NRTL.

Table 3. NRTL Interaction Parameters

i	j	substance i	substance j	A_{ij}/K	A_{ji}/K	α_{ij}
1	2	water	PPG 400	2382.5	−1370.7	0.208 50
1	3	water	maltose	−4997.5	−2073.1	0.2036 2
1	4	water	glucose	−1688.6	−1250.6	0.335 20
1	5	water	PPG 1200	2755.2	−855.16	0.253 55
1	6	water	PE 61	4158.0	−1889.4	0.201 07
1	7	water	PPG 725	2989.3	−1110.8	0.200 00
2	3	PPG 400	maltose	−0.060887	414.50	0.468 61
2	4	PPG 400	glucose	1928.8	890.37	0.277 45
3	5	maltose	PPG 1200	4997.3	369.78	0.464 22
3	6	maltose	PE 61	2678.9	715.35	0.200 00
3	7	maltose	PPG 725	1666.3	146.31	0.359 52
4	5	glucose	PPG 1200	3437.6	−2914.0	0.308 90
4	6	glucose	PE 61	1737.9	3.6394	0.200 08

In the case of the systems prepared with glucose and block copolymers or maltose and block copolymers in water, those block copolymers with higher EO content (20 and 40%) in the molecule did not show phase splitting at the temperatures chosen for analysis (298.15 and 303.15 K). Furthermore, mixtures of PEG and glucose or PEG and maltose in water were also tested and did not show phase splitting for all PEG molar masses until 20 000.

None of the systems discussed above showed the typical behavior of the PEG + maltodextrin systems,[7] which in general have almost no polymer in the bottom phase and large amounts of carbohydrate in the top phase. It was also observed that the composition of the polymer-rich phase did not vary significantly, except for the systems containing PPG 400. The phase composition varied almost only at the sugar-rich phase. Figures 2, 3, and 5 to 7 show this behavior.

For the systems containing PPG 400, a different behavior was observed, with some sugar in the polymer-rich phase and a reasonable amount of synthetic polymer in the sugar-rich phase. It is interesting to notice that PPG 400 is the only synthetic polymer completely miscible with pure water, but it showed phase splitting when dissolved in an aqueous sugar solution. Also, the tie lines are almost parallel to each other, meaning varying compositions for both phases alongside the overall compositions prepared (Figures 1 and 4). These systems also possess a small two-liquid-phase region.

Tables 3 and 4 show, respectively, the binary interaction parameters obtained for the NRTL and UNIQUAC models. Both models presented good adjustment to the experimen-

Table 4. UNIQUAC Interaction Parameters

i	j	substance i	substance j	A_{ij}/K	A_{ji}/K
1	2	water	PPG 400	−212.09	201.34
1	3	water	maltose	−504.87	−222.83
1	4	water	glucose	847.74	−531.35
1	5	water	PPG 1200	−57.640	292.01
1	6	water	PE 61	−118.95	278.09
1	7	water	PPG 725	−151.77	506.28
2	3	PPG 400	maltose	141.49	−2.8945
2	4	PPG 400	glucose	675.76	−80.276
3	5	maltose	PPG 1200	−10.437	−73.181
3	6	maltose	PE 61	4598.4	−595.78
3	7	maltose	PPG 725	−167.33	−15.816
4	5	glucose	PPG 1200	−15.179	205.65
4	6	glucose	PE 61	4455.4	184.39

Table 5. Mean Deviations in Phase Compositions (mass %)

	Δw	
system	UNIQUAC	NRTL
PPG 400 + glucose	0.96	0.75
PPG 400 + maltose	0.80	0.91
PPG 725 + maltose	2.01	0.39
PPG 1200 + glucose	0.62	0.47
PPG 1200 + maltose	0.55	0.33
PE 61 + glucose	0.71	0.80
PE 61 + maltose	1.66	0.61
global deviation	1.11	0.67

tal data, with results slightly better for the NRTL model (Table 5). The deviations given in Table 5 were calculated according to eq 2 below:

$$\Delta w = 100 \sqrt{\frac{\sum_{n}^{N}\sum_{i}^{K}[(w_{i,n}^{\text{I,ex}} - w_{i,n}^{\text{I,calc}})^2 + (w_{i,n}^{\text{II,ex}} - w_{i,n}^{\text{II,calc}})^2]}{2NK}} \tag{2}$$

Conclusion

In this work, phase diagrams for systems containing a sugar + a synthetic polymer + water were obtained. Binary interaction parameters of the NRTL and UNIQUAC models were adjusted for the various ternary mixtures studied. With these data, one may estimate the behavior of ternary or higher aqueous mixtures formed by the selected substances. Such results may contribute to the development of new low cost aqueous two-phase systems for downstream processing and also to a better understanding of aqueous solutions containing sugars and polymers, which represent mixtures of interest for the food industry.

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