Measurement and Correlation of Solubility of Uracil in Supercritical Carbon Dioxide

Ran Zhou, Shufen Li,* and Tongju Liu

Key Laboratory for Green Chemical Technologies of the Ministry of Education, School of Chemical Engineering and Technology, Tianjin University, C.P.300072, P. R. China

The solubility of uracil in supercritical carbon dioxide (SC-CO_2) was determined with dynamic method at temperatures (308, 318, 328, 338, and 348) K and various pressures in the range of (15 to 40) MPa. The mole fraction solubilities of uracil ranged from $(0.769 \text{ to } 14.98) \cdot 10^{-8}$. The solvent capacity of SC-CO_2 to dissolve uracil increases by increasing the pressure at a constant temperature. The crossover point was observed around a pressure of 15 MPa. Experimental data of the solubility of uracil were correlated by the Chrastil model and the modified Chrastil model. The results indicated that the two models were able to successfully correlate the experimental solid uracil + SC-CO_2 solubility data, and the values of average absolute relative deviation (AARD) were 12.8 % and 10.1 %, respectively.

Introduction

Supercritical fluid extraction (SCFE) is an increasing important technology for obtaining natural products,[1,2] which possesses several advantages over traditional liquid-solvent-based extraction methods including improved selectivity, expeditiousness, automation, and environmental safety. The solubility data of solids and liquids in supercritical fluids (SCFs) are very important in developing any supercritical extraction process.

Uracil (1*H*-pyrimidine-2,4-dione, [66-22-8], $C_4H_4N_2O_2$) is a nucleic acid base (nucleoside) present in an RNA molecule. The function of uracil in the body is to help synthesize many enzymes necessary for cell function through bonding with riboses and phosphates.[3] Uracil serves as an allosteric regulator and a coenzyme for reactions in the human body and in plants. Uracil is also involved in the biosynthesis of polysaccharides and the transportation of sugars containing aldehydes.[4] Furthermore, uracil is one of the most important components, which have an inhibitory effect on monoamine oxidase activity in antler velvet.[5,6] Studies have shown that uracil can significantly inhibit the activity of monoamine oxidase of brain in old mice and increase the content of 5-hydroxytryptamine and dopamine. Therefore, it may be a choice for the treatment of senile diseases such as melancholia and Parkinson's disease.

To extract uracil from antler velvet with SCFE effectively, the solubility of uracil in supercritical carbon dioxide (SC-CO_2) needs to be known. Only Burgos-Solórzano had previously reported the solubility of uracil in SC-CO_2 under the temperatures of (313.15 and 333.15) K and pressures up to 30 MPa.[7] Therefore, the present investigation was undertaken to determine the solubility of uracil in SC-CO_2, under the conditions of temperatures from (308.15 to 338.15) K and pressures from (15 to 40) MPa. The solubility data were correlated using density-based correlation models (Chrastil and modified Chrastil models).

Experimental Section

Materials and Chemicals. The carbon dioxide stored in a cylinder with a dip tube with a purity of 99.9 % was purchased from Liu Fang Gas Co. (Tianjin, China). Uracil (Ultra pure) was purchased from Amresco (USA). Distilled deionized water was made by our laboratory and was used as received. Methanol (HPLC grade) was supplied by Tianjin Chemical Reagent Factory (Tianjin, China). All chemicals were used without further purification.

Experimental Apparatus and Procedure. The solubility measurements were performed with a Spe-ed SFE instrument (Applied Separations Inc., Allenton, PA, USA), shown schematically in Figure 1. A portion of 5 g of uracil was suspended in a stainless steel extraction column (10 cm^3 capacity)[6] with layers of glass wool and 30−60 mesh glass beads. The packing helps minimize channeling of the supercritical fluid and prevents entrainment of the solute.[8,9] The extraction column was placed in the oven[7] of the system, and its temperature was indicated and controlled by the thermocouple[12] within ± 0.1 K during the solubility determination. Liquid CO_2 was pressurized with a gas booster pump[3] and then charged into the extraction column[6] to the desired pressure. The system pressure was regularly calibrated using a test gauge (OMEGA DP-41, uncertainty 0.1 MPa) with the accuracy of 1 % of the set pressure. The flow rate was controlled by the micrometer valve, varying from (0.10 to 0.15) standards $L \cdot min^{-1}$. This flow rate has been checked by preliminary experiments to be sufficiently low to obtain a saturated solvent at the outlet-valve. The supercritical CO_2 with dissolved solutes passed from the extraction column through a heated micrometer valve[9] and expanded to ambient pressure. The solid solute was absorbed by distilled deionized water in the vial. The gas flowed through the vial further to the wet-test meter[11] to measure its volume. Finally, the micrometer valve was washed with distilled deionized water and was collected in the collection vial. The final volume of the solution was adjusted to 5 mL. The procedure was repeated in triplicate at the identical operating conditions, and the average value was used as the solubility.

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* Corresponding author. Tel.: +86 22 27402720. Fax: +86 22 27402720. E-mail: shfli@tju.edu.cn.

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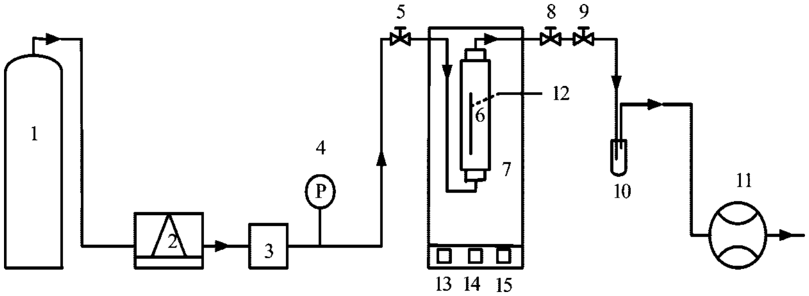

Figure 1. Schematic diagram of the experimental apparatus. 1, CO_2 cylinder; 2, liquid-cooled bath; 3, gas booster pump; 4, pressure gauge; 5, inlet valve; 6, extraction column; 7, constant-temperature oven; 8, outlet valve; 9, micrometer valve; 10, collecting valve; 11, wet-test meter; 12, thermocouple; 13, oven temperature; 14, column temperature; 15, micrometer valve temperature indicator.

Table 1. Uracil Solubility in SC-CO_2: Pressure *P*, Temperature *T*, SC-CO_2 Density *ρ*, Mass Fraction Solubility *S*, and Mole Fraction Solubility *y*

T/K	P/MPa	ρ/g·L^{-1}	10^8S	10^8y	P/MPa	ρ/g·L^{-1}	10^8S	10^8y
308.15	15	815.0	1.959	0.769	30	929.6	7.640	2.999
	20	866.1	4.165	1.635	35	952.8	8.933	3.506
	25	901.7	6.371	2.501	40	972.8	10.572	4.150
318.15	15	742.4	2.463	0.967	30	891.0	13.643	5.356
	20	813.3	6.202	2.435	35	917.8	15.193	5.964
	25	857.9	8.825	3.464	40	940.4	16.686	6.550
328.15	15	653.8	2.374	0.932	30	850.9	17.394	6.828
	20	755.0	7.136	2.801	35	881.9	27.418	10.763
	25	811.2	11.185	4.391	40	907.5	34.256	13.447
338.15	15	555.5	2.237	0.878	30	809.5	22.018	8.643
	20	692.3	11.295	4.434	35	845.2	29.755	11.680
	25	762.4	15.367	6.032	40	877.3	38.161	14.980

Analysis of the solute was carried out off-line by using a high-performance liquid chromatography system equipped with an isocratic pump (LabAlliance, model Series III, USA) and an ultraviolet−visible detector (LabAlliance, model 500, USA). The wavelength was set at 254 nm. Stock solutions of the solid sample were prepared by dissolving appropriate amounts of the solid sample in distilled deionized water. A set of eight standard solutions were then prepared by appropriate dilution of the stock solution. The calibration curves obtained (with regression coefficients better than 0.998) were used to establish the concentration of the uracil in the collection vial. The mole fraction compositions of the solutes were obtained according to a standard curve.

Correlation of Solubility Data. The experimental results from the present study were correlated by two different density-based correlation models (Chrastil and modified Chrastil models).

Chrastil Model Correlation. The Chrastil model gives a macroscopic description of the number of molecules surrounding the solute in the fluid phase.[10] It is based on the hypothesis that one molecule of a solute S (i.e., uracil) associates with *k* molecules of a solvent, B (i.e., CO_2), to form one molecule of a solvation complex, SB_k, which is in equilibrium with the fluid. The Chrastil equation is given below

$$\ln S = k \ln \rho + \frac{\alpha}{T} + \beta \qquad (1)$$

where *S* is the uracil solubility in SC-CO_2 (g·L^{-1}); *k* is the association number; *ρ* is SC-CO_2 density; α is dependent on the heat of solvation and vaporization of the solute; and *β* is a function of *k*. The parameters, *k*, α, and *β* are obtained performing a multiple linear regression on the experimental data.

Modified Chrastil Model Correlation. A model representing solubility behavior proposed by Wang is derived from the solvation concept,[11] the equilibrium constant of high-pressure reaction, and the rules of phase equilibrium. Therefore, an improved correlation, with four adjustable parameters b_0, b_1, b_2, and b_3, is obtained as follows

$$\ln y = b_0 + \frac{b_1}{T} + b_2\rho + b_3 \ln P \qquad (2)$$

with

$$b_0 = \Delta S^0/R + \ln P^{\mathrm{sat}}$$

$$b_1 = -(\Delta H^0 + V_S^S P^{\mathrm{sat}})/R$$

$$b_2 = -[2(B_{12} - kB_{22}) + (k-1)B] + V_S^S$$

$$b_3 = k - 1$$

In the above expressions, *y* is the solute mole fraction in the supercritical solution; *ρ* is SC-CO_2 density; and *T* and *P* are the operating temperature and pressure. The parameter b_0 is dependent on the heat of solvation and vaporization of the solute; b_1 is dependent on the heat of salvation, molar volume, and vaporization of the solute; b_2 is related to the second virial coefficient of the solute, mixed second virial coefficient, molar volume, and association number; and b_3 is a function of *k*. Finally, b_0, b_1, b_2, and b_3 can be obtained by regression of the experimental data.

The errors of the experimentally measured solubility and the solubility given by every model were estimated by calculating the average absolute relative deviation (AARD) between the experimental and the calculated solubility data using the following equation

$$\mathrm{AARD}\ (\%) = \frac{100}{N}\sum_n \frac{|y^{\mathrm{cal}} - y^{\mathrm{exp}}|}{y^{\mathrm{exp}}} \qquad (3)$$

where *N* is the number of solubility experimental data; y^{cal} are the calculated solubilities; and y^{exp} are the experimental solubility data.

Results and Discussion

Experimental Solubility Results. The reliability of the apparatus was preliminarily checked by measuring the solubilities (mole fraction, *y*) of benzoic acid in supercritical carbon dioxide at (308.15 and 318.15) K. The values are obtained from an arithmetic average of three replicate measurements with relative deviations less than 5 %. The solubilities obtained are in fair agreement with those reported in the literature.[12,13] The solubility data of uracil in SC-CO_2 at different temperatures of (308.15, 318.15, 328.15, and 338.15) K, in the pressure range of (15 to 40) MPa, were measured, as shown in Table 1 and Figure 2. The density of CO_2 at the given pressure and temperature, obtained from the IUPAC International Thermodynamic Tables,[14] is also included in Table 1. Each data point is an average of, at least, three experimental measurements, with

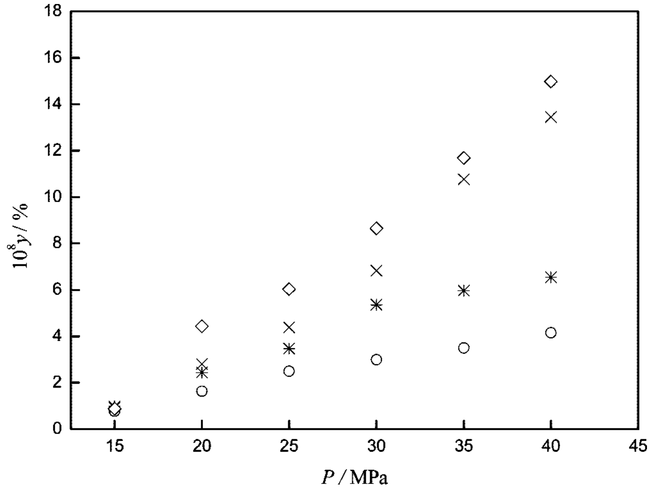

Figure 2. Solubility of uracil in the pressure range of (15 to 40) MPa at various temperatures. ○, 308.15 K; *, 318.15 K; ×, 328.15 K; ◇, 338.15 K.

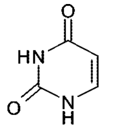

Figure 3. Structural formula of uracil.

reproducibility within ± 16 %. The relatively poor solid solubility in SC-CO_2 (10^{-7} to 10^{-8}) was expected, considering the quite high polarity of this compound with two carbonyl groups on the benzene ring and the well-known weak solvation power of SC-CO_2 for polar molecules. The structural formula of it is shown in Figure 3.

The solubilities of uracil above 15 MPa were studied, considering its poor solubility property in SC-CO_2 which may bring a larger error in lower pressure. In Figure 2, the solubility of uracil in SC-CO_2 increases with pressure while showing different trends at different temperatures. At (308.15 and 318.15) K, the solubility increases with pressure up to 30 MPa where it reaches a plateau. At (328.15 and 338.15) K the solubilities are increased almost linearly until 40 MPa and no plateau is observed. The solubility of uracil in SC-CO_2 is strongly influenced by the system pressure, which determines the density of the SC-CO_2 as a solvent.[15] The effect of temperature is more complex, and crossover points in Figure 2 are observed at about 15 MPa. The crossover pressure can be defined as the point where the slope of the plot of solubility versus temperature changes sign. It should not change with temperature. At the crossover pressure, the effects of solute vapor pressure and solvent density on solid solubility balance each other.[16,17] At pressures above the crossover pressure, the solubility of uracil increases with temperature.

Comparison of Uracil Data. The solubility data of uracil were compared between this work and existing data by Burgos-Solórzano.[5] Both data have obvious differences from each other. The main reasons may come from the tested uracil, which were purchased from different sources and different equipment applied for the solubility measurement. Additionally, operation and measurement errors may also exist. Although there is a great deviation of solubility data of uracil between this work and Burgos-Solórzano's work, crossover pressures of both data were about 15 MPa.

Correlation Results. Correlation and prediction of the solubility of uracil in supercritical CO_2 is very important for the application of supercritical fluid technology in processing this bioactive compound. In this work, the two density-based (Chrastil and modified Chrastil models) correlation results of uracil are represented in Figures 4 and 5. The linearity is excellent, and the correlation parameters are obtained. To both employed models, the quality of the correlation is expressed in terms of AARD.

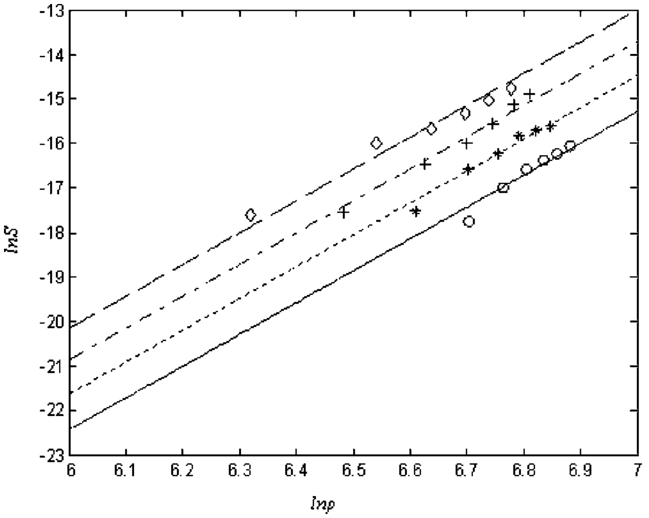

Figure 4. Plots of $\ln S$ vs $\ln \rho$ using the Chrastil model for uracil in the pressure range of (15 to 40) MPa at various temperatures. ○, 308.15 K; *, 318.15 K; ×, 328.15 K; ◇, 338.15 K. The lines represent the results of the Chrastil model correlation. —, 308.15 K; - - -, 318.15 K; — - —, 328.15 K; — —, 338.15 K.

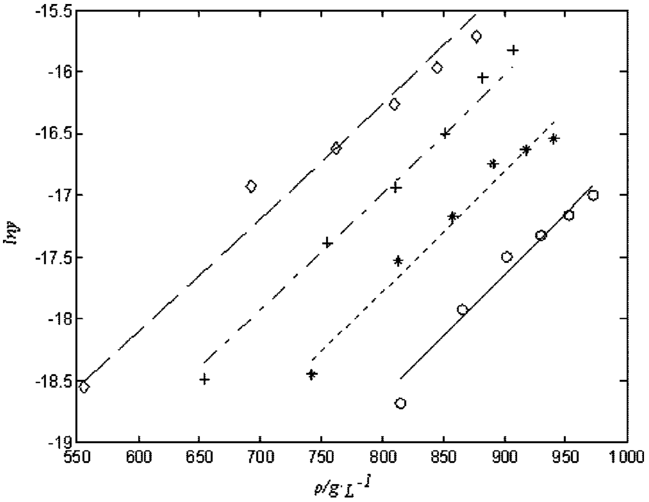

Figure 5. Plots of $\ln y$ vs ρ using the modified Chrastil model for uracil in the pressure range of (15 to 40) MPa at various temperatures. ○, 308.15 K; *, 318.15 K; ×, 328.15 K; ◇, 338.15 K. The lines represent the results of the modified Chrastil model correlation. —, 308.15 K; - - -, 318.15 K; — - —, 328.15 K; — —, 338.15 K.

In the first model, the Chrastil equation has the advantage of having only three parameters to fit all the experimental data, no matter at which temperature they were obtained. In the Chrastil equation, parameter k is related to the average number of molecules that forms the solvato-complex. With a higher value of k, it can be observed that the component extraction occurred at lower CO_2 density. Regarding parameter α, it takes into account the heat of solvation and vaporization of the solute, and therefore, it indicates the influence of the temperature changes inside the extraction vessel. Consequently, a higher absolute value of α is associated with higher temperature influence. Parameter β is associated with the solute molecular weight and melting point. They can possess molecular weights of the same order as well as similar melting points with quite similar value of β. By performing a multiple linear regression on ln S as a function of ln ρ and $1/T$, one obtains $k = 7.1$, $\alpha = -7918.4$ K, $\beta = -39.6$. The thermodynamic quantity, ΔH, can be calculated directly from a, resulting in a value of -65.83 $\text{kJ}\cdot\text{mol}^{-1}$. The average absolute relative deviation of the fitted Chrastil equation from experimental data was calculated to be 12.8 %. It can be also noted that there exists a relatively significant error at the lower pressures, especially near the critical point, which might contribute to the remarkable variation of the solvent properties under such operation conditions. In the second method, considering the effect of high pressure on the association balance migration, eq 2 was used. Four sets of parameters with clear physics significance were obtained for four different temperatures, as given in Figure 5. The results for the modified Chrastil models were obtained: $b_0 = -1.70$; $b_1 = -7692.0$; $b_2 = -0.00$; $b_3 = 0.20$; AARD = 10.1 %. As can be seen in Figures 4 and 5 and with obtained AARD values, both density-based models were able to successfully correlate the experimental solid uracil + SC-CO_2 solubility data. The correlates for the improved equation have better correlation accuracy than that of the Chrastil model.

Conclusions

The solubility of uracil in SC-CO_2 was measured in the pressure range of (15 to 40) MPa at different temperatures of (308.15, 318.15, 328.15, and 338.15) K. Equilibrium solubility data, expressed in terms of uracil's mole fraction, range from $(0.769 \text{ to } 14.98)\cdot 10^{-8}$. At a constant temperature, the solvent capacity to dissolve uracil increases by increasing the pressure. The crossover point was observed around a pressure of 15 MPa. The Chrastil equation and an improved version were used to correlate the solubility data. The improved version with an overall AARD of 10.1 % has better correlation accuracy than that of the Chrastil equation. The solubility results obtained in this work are very promising for the development of supercritical extraction processes for uracil from natural products.

Literature Cited

(1) Chang, C. J.; Wu, S. M.; Yang, P. W. High-pressure carbon dioxide and co-solvent extractions of crude oils from plant materials. *Innovative Food Sci. Emerging Technol.* **2001**, *1*, 187–191.

(2) Pasquini, D.; Pimenta, M. T. B.; Ferreira, L. H.; Curvelo, A. A d. S. Extraction of lignin from sugar cane bagasse and Pinus taeda wood chips using ethanol-water mixtures and carbon dioxide at high pressures. *J. Supercrit. Fluids* **2005**, *36*, 31–39.

(3) Garrett, R. H.; Grisham, C. M. *Principals of Biochemistry with a Human Focus*; Brooks/Cole Thomson Learning: United States, 1997.

(4) Brown, E. G. *Ring Nitrogen and Key Biomolecules: The Biochemistry of N-Heterocycles*; Lluwer Academic Publishers: Boston, 1998.

(5) Yang, X. W. HPLC analysis and inhibitory effect of base components in pilose antler of sika deer and red deer on monoamine oxidase activity. *Chin. Tradit. Herb. Drugs* **1995**, *26* (1), 17–19.

(6) Chen, X. G.; Wang, B. X.; Zhang, J.; Zhang, H. Inhibitory effect of uracil on monoamine oxidase activity. *Chin. Biochem. J.* **1992**, *8* (1), 81–85.

(7) Burgos-Solórzano, G. I.; Brennecke, J. F.; Stadtherr, M. A. Solubility measurements and modeling of molecules of biological and pharmaceutical with supercritical CO_2. *Fluid Phase Equilib.* **2004**, *220*, 57–69.

(8) Maxwell, R. J. Solubility Measurements of Lipid Constituents in Supercritical Fluids. *Supercritical Fluid Technology in Oil and Lipid Chemistry*; AOCS Press: Champaign, IL, 1996; pp 20−34.

(9) Lee, H.; Kim, C.; Kim, S.; Choi, C. Solid Solubilities of Methoxyphenylacetic Acid Isomer Compounds in Supercritical Carbon Dioxide. *J. Chem. Eng. Data* **1994**, *39*, 163–165.

(10) Chrastll, J. Solubility of Solids and Liquids in Supercritical Gases. *J. Phys. Chem.* **1982**, *86*, 3016–3021.

(11) Wang, L. *Studies on modeling phase equilibrium for supercritical fluid-solid System*; Guizhou university, 2005; pp 15−26.

(12) Dobbs, J. M.; Wong, J. M.; Lahiere, R. J.; Johnston, K. P. Modification of Supercritical Fluid Phase Behavior Using Polar Cosolvents. *Ind. Eng. Chem. Res.* **1987**, *26*, 56–65.

(13) Kurnik, R. T.; Holla, S. J.; Reid, R. C. Solubility of Solids in Supercritical Carbon Dioxide and Ethylene. *J. Chem. Eng. Data* **1981**, *26*, 47–51.

(14) Angus, S.; Armstrong, B.; de Reuck, K. M. *International Thermodynamic Tables of the Fluid State, Carbon Dioxide*; Pergamon Press: Oxford, U.K., 1976.

(15) Khimeche, K.; Alessi, P.; Kikic, I.; Dahmani, A. Solubility of diamines in supercritical carbon dioxide; experimental determination and correlation. *J. Supercrit. Fluids* **2007**, *41*, 10–19.

(16) Johnston, K. P.; Barry, S. E.; Read, N. K.; Holcomb, T. R. Separation of Isomers Using Retrograde Crystallization from Supercritical Fluids. *Ind. Eng. Chem. Res.* **1987**, *26*, 2372–2377.

(17) Foster, N. R.; Gurdial, G. S.; Yun, J. S. L.; Keat Liong, K.; Tilly, K. D.; Ting, S. S. T.; Lee, J. H. Significance of the Crossover Pressure in Solid-Supercritical Fluid Phase Equilibria. *Ind. Eng. Chem. Res.* **1991**, *30*, 1955–1964.

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