J. Chem. Thermodynamics 89 (2015) 142–147

Contents lists available at ScienceDirect

J. Chem. Thermodynamics

journal homepage: www.elsevier.com/locate/jct

Measurement and correlation of the solubility of genistin in eleven organic solvents from $T = (283.2 \text{ to } 323.2)$ K

Jie-Ping Fan [a],*, Xiao-Kang Xu [a], Gao-Lin Shen [a], Xue-Hong Zhang [b]

[a] *Department of Chemical Engineering, Nanchang University, Nanchang 330031, China*
[b] *School of Foreign Language, Nanchang University, Nanchang 330031, China*

ARTICLE INFO

Article history:
Received 31 January 2015
Received in revised form 13 May 2015
Accepted 17 May 2015
Available online 25 May 2015

Keywords:
Genistin
Solubilities
Simplified thermodynamic model
Modified Apelblat model
λh model
NRTL model

ABSTRACT

The solubilities of genistin in pure solvents including tetrahydrofuran, acetone, ethyl acetate, acetonitrile, isopropanol, n-butyl alcohol, methanol, cyclohexane, n-hexane, chloroform and ethanol were determined by the high performance liquid chromatography (HPLC) analysis method at $T = (283.2, 293.2, 303.2, 313.2$ and $323.2)$ K. The fusion enthalpy of genistin was estimated by the group contribution method. The solubility data of genistin were correlated by the simplified thermodynamic model, the modified Apelblat model, λh model and NRTL (Non-Random Two-Liquid) model. The calculated values by all models were in good agreement with the experimental values and however, the NRTL model could give better correlation results than other three models.

1. Introduction

Genistin ($C_{21}H_{20}O_{10}$, Molar mass: 432.37 g · mol^{-1}, CAS number: 529-59-9, figure 1) is one of the most effective isoflavone in soybean or *Radix Puerariae Lobatae*. Chemically, it is the 7-O-β-D-glucoside form of genistein. Genistin has attracted much attention due to its wide variety of pharmacological activities, such as weak oestrogen activity [1], antioxidant activity [2,3], hemolysis activity and antibacterial activity [4]. It is also considered as an effectual preventive for breast cancer [5], colon cancer [6], osteoporosis [7] and so on. Therefore, Genistin is often used as dietary supplements [8]. In soybean and *Radix Puerariae* some other similar isoflavones, *i.e.* daidein and genistein, coexist with genistin. In order to separate and purify them, it is necessary to understand their solubilities in various solvents. In the literatures, the solubilities of daidein [9,10] and genistein [11] have been reported. However, the solubility of genistin has not been systematically studied. So, in this research the solubility data of genistin and the thermodynamic parameter (Gibbs free energy, enthalpy and entropy) in various solvents were reported.

2. Materials and methods

2.1. Materials

Genistin (mass fraction purity ⩾ 0.98) was purchased from Xi'an Haoxuan Biotechnology Co. Ltd, Shanxi, China (table 1), and used without further treatment. Its structure was confirmed by UV and ^{1}HNMR spectra. The source and the purity of all the organic solvents were listed in table 1.

2.2. Solubility measurement

The solubility of genistin was measured by the HPLC analysis method according to the literatures [12,13] with minor modification. The experimental saturated solutions were prepared in a glass vial by adding excess genistin in the solvent. The vial was stoppered and sealed up with tape to prevent solvent from escaping, and then laid in a low temperature thermostatic reaction bath (type DFY5/40, China) with a temperature uncertainty of ±0.1 K. The solution was continuously stirred for at least 48 h by a magnetic stirrer to ensure equilibrium, and then stood for another 12 h to obtain a clear saturated solution before sampling. For each vial, three samples of approximately 0.5 mL were extracted from the saturated supernatant by a preheated disposable syringe equipped with a filter. The syringes with saturated solution were

---

* Corresponding author.
E-mail address: jasperfan@163.com (J.-P. Fan).

http://dx.doi.org/10.1016/j.jct.2015.05.019
0021-9614/

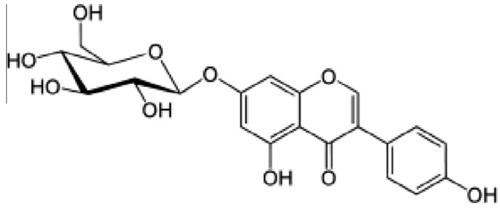

FIGURE 1. Chemical structure of genistin.

TABLE 1
The provenance and mass fraction purity of chemicals.

Chemicals	Mass fraction purity	Method of purification	Analysis method	Provenance
Genistin	⩾0.980	None[a]	HPLC[b]	Xi'an Haoxuan Bio-technique Co., Ltd, Shanxi, China
Ethyl acetate	⩾0.995	None	GC[c]	Tianjin Hengxin Chemical Preparation Co., Ltd.
Acetonitrile	⩾0.999	None	GC	Guangdong Xilong Chemical Co., Ltd.
Cyclohexane	⩾0.995	None	GC	Shanghai Sinopharm Chemical Reagent Co., Ltd.
n-Hexane	⩾0.970	None	GC	Shanghai Sinopharm Chemical Reagent Co., Ltd.
Chloroform	⩾0.990	None	GC	Shanghai Rich Joint Chemical Reagents Co., Ltd.
Isopropanol	⩾0.995	None	GC	Guangdong Xilong Chemical Co., Ltd.
n-Butyl alcohol	⩾0.995	None	GC	Tianjin Hengxin Chemical Preparation Co. Ltd.
Methanol	⩾0.998	None	GC	Shanghai Sinopharm Chemical Reagent Co., Ltd.
Ethanol	⩾0.997	None	GC	Tianjin Hengxin Chemical Preparation Co. Ltd.
Tetrahydrofuran	⩾0.990	None	GC	Tianjin Hengxin Chemical Preparation Co. Ltd.
Acetone	⩾0.995	None	GC	Shanghai Rich Joint Chemical Reagents Co., Ltd.

[a] No further purification.
[b] The purity was determined by high-performance liquid chromatography (HPLC).
[c] The purity was determined by gas chromatography (GC).

weighed on an analytical balance (type FA1104N, Shanghai, China) with an uncertainty of ±0.1 mg. In order to prevent solvent from evaporating in the weighing process, the syringe needle was closed with a silicon rubber. The saturated solution was injected into a volumetric flask (10 ml) quickly, the syringe was thoroughly eluted by methyl alcohol and all elutions were also transferred into the volumetric flask. Then the sample solution was diluted to the mark prior to the HPLC determination. The concentration of genistin in sample solution was determined by the HPLC method according to the literatures [14,15]. Each experiment was repeated at least twice to check the repeatability and three samples were taken for each solution at each temperature. In this study, the relative uncertainties of the experimental solubility data were within ±0.10. The saturated mole fraction solubility of genistin (x) can be obtained from the equation (1).

$$x = \frac{m_1/M_1}{m_1/M_1 + m_2/M_2}, \tag{1}$$

where m_1 and m_2 represent the mass of genistin and solvent, respectively; M_1 and M_2 are the molar mass of genistin and solvent, respectively.

2.3. HPLC conditions

The chromatographic analysis was carried out on an Agilent 1100 HPLC system (Agilent Technologies, USA) equipped with a vacuum degasser (type G1379A), a quaternary pump (type G1311A), an auto-sampler (type G1313A) and a diode-array detector (type G1315A). The detection wavelength of genistin was 260 nm. The separation column was a Hypersil ODS-C18 column (4.6 · 200 mm, 5 μm) at T = 303.2 K. The mobile phase was a mixture of acetonitrile–water (57:43, v/v) and the flow rate was 1.0 mL min^{-1}.

2.4. Thermal analysis

The melting point of genistin was determined by a thermogravimetry–differential scanning calorimetry (TG/DSC, type SDT Q600, American TA Co.). The sample was scanned from (50 to 500) °C at a heating rate of 5 °C · min^{-1} with high purity nitrogen at constant flow rate of 100 mL · min^{-1}. In this study, the melting temperature was used as the onset point of DSC curve which was the intersection of the extension of the baseline with the tangent at the point of greatest slope (inflexion point) of the DSC curve.

3. Results and discussion

3.1. Evaluation of thermochemical properties

The thermal analysis (TGA/DSC) of genistin was shown in figure 2. According to the DSC analysis, the determined melting point (T_m) of genistin was T = 547.47 K, and the determined molar fusion enthalpy ($\Delta_{fus}H_m$) of genistin was 65.6 kJ · mol^{-1}. The standard uncertainty of the melting point value was T = 0.01 K, and the relative standard uncertainty of the calorimetric measurement was 0.02. In this study, the initial melting temperature of genistin was 537.15 K according to the DSC curve, however the melting point was used as the onset point of the DSC curve which was the intersection of the extension of the baseline with the tangent at the point of greatest slope (inflexion point) of the DSC curve. Jeong *et al.* [16] reported that the melting point of genistin was about T = 537.55 K determined on a melting point apparatus. There were some deviations in the values determined for the melting point, which was probably due mainly to the different determination methods. Moreover, as shown in the TGA of genistin in figure 2, decomposition of genistin began at almost the same time as melting. It revealed that the determined fusion enthalpy of genistin (65.6 kJ mol^{-1}) in figure 2 contained decomposition heat, and the conventional calorimetric test could not be used to measure genistin's fusion enthalpy due to the thermal unstability of genistin. To circumvent this problem, a group contribution method was applied to estimate genistin's fusion enthalpy. According to the study of Dannenfelser and Yalkowsky [17–19], the total entropy of melting ($\Delta_{fus}S_m$) was estimated by a semiempirical equation (equation (2)) with two molecular descriptors, the molecular rotational symmetry number (σ) and molecular flexibility number (Φ).

$$\Delta_{fus}S_m = 50 - R\ln\sigma + R\ln\Phi, \tag{2}$$

where R is the gas constant. Rotational symmetry number (σ) is defined as the number of positions into which a molecule can be