Journal of Molecular Liquids 255 (2018) 43–50

Contents lists available at ScienceDirect

Journal of Molecular Liquids

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

# Solubility, thermodynamic properties and solute-solvent molecular interactions of luteolin in various pure solvents

Faiyaz Shakeel *, Nazrul Haq, Sultan Alshehri, Mohamed A. Ibrahim, Ehab M. Elzayat, Mohammad A. Altamimi, Kazi Mohsin, Fars K. Alanazi, Ibrahim A. Alsarra

*Kayyali Chair for Pharmaceutical Industries, Department of Pharmaceutics, College of Pharmacy, King Saud University, P.O. Box 2457, Riyadh 11451, Saudi Arabia*

ARTICLE INFO

*Article history:*
Received 20 December 2017
Received in revised form 23 January 2018
Accepted 25 January 2018
Available online xxxx

*Keywords:*
Activity coefficient
Bioflavonoid
Dissolution thermodynamics
Luteolin
Solid state characterization
Solubility

ABSTRACT

The objective of this study was to evaluate solubility, thermodynamic properties and solute-solvent molecular interactions of poorly water-soluble bioactive compound luteolin (LUT) in various pure solvents namely water ($H_2O$), ethylene glycol (EG), propylene glycol (PG), polyethylene glycol-400 (PEG-400), methanol (MeOH), ethanol (EtOH), isopropanol (IPA), 1-butanol (1-BuOH), 2-butanol (2-BuOH), ethyl acetate (EA), dimethyl sulfoxide (DMSO) and Transcutol® (THP) at temperatures "$T = 298.2$ K to 318.2 K" and pressure "$p = 0.1$ MPa". The solubility values of LUT at equilibrium were determined using a shake flask technique with the help of ultra-performance liquid chromatography technique at 348 nm. The solid state of LUT in pure and equilibrated form was characterized using differential scanning calorimetry (DSC) and powder X-ray diffraction (PXRD) techniques. The solubilities of LUT measured by shake flask method were correlated with "van't Hoff and Apelblat models". The DSC and PXRD spectra of pure and equilibrated LUT were similar, suggesting no transformation of LUT during equilibrium. Good correlation was recorded between experimental and calculated solubilities of LUT. The solubility values of LUT (mole fraction) were obtained maximum in PEG-400 ($1.27 \times 10^{-1}$) followed by DMSO ($7.15 \times 10^{-2}$), THP ($4.01 \times 10^{-2}$), 2-BuOH ($5.53 \times 10^{-3}$), 1-BuOH ($5.36 \times 10^{-3}$), EA ($5.11 \times 10^{-3}$), IPA ($4.14 \times 10^{-3}$), PG ($3.89 \times 10^{-3}$), EtOH ($3.08 \times 10^{-3}$), EG ($2.38 \times 10^{-3}$), MeOH ($8.16 \times 10^{-4}$) and $H_2O$ ($4.17 \times 10^{-6}$) at "$T = 318.2$ K" and similar trend was obtained at each temperature studied. The physical values of activity coefficients were determined with the help of ideal solubilities of LUT. Based on the physical values of activity coefficients, maximum solute-solvent interactions were observed in LUT-PEG-400, LUT-DMSO and LUT-THP. Apparent thermodynamic analysis suggested endothermic and entropy-driven dissolution of LUT in each pure solvent studied.


## 1. Introduction

Luteolin (LUT) [molecular structure: Fig. 1; IUPAC name: 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-4H-chromen-4-one; molecular formula: $C_{15}H_{10}O_6$; molar mass: 286.24 g $mol^{-1}$ and CASRN: 491-70-3) occurs as a "light yellow crystalline" solid [1,2]. It is a bioactive compound belongs to bioflavonoid category and present in celery, perilla leaf, chamomile tea and green pepper [3,4]. It shows various therapeutic activities such as antioxidant [5], anti-inflammatory [6–8], anti-allergic [8], anti-amnestic [9], hepatoprotective [10], cardioprotective [1], neuroprotective [6,9] and anticancer [11–14] activities in literature. The main problem associated with dosage form design of LUT is its very poor solubility in water [15]. Various formulation approaches such as cyclodextrin inclusion complexation [16,17], phospholipid complexation [10,18], cyclosophoraoses complexation [19], copolymer micelles [15] and solid-lipid nanoparticles [20] have been studied in order to enhance the solubility, dissolution rate, therapeutic activity and bioavailability of LUT in literature. The solubilities of LUT in mole fraction in eight different organic solvents namely methanol (MeOH), ethanol (EtOH), 1-propyl alcohol, isopropyl alcohol (IPA), 1-butyl alcohol (1-BuOH), acetone, hexane and dimethyl sulfoxide (DMSO) at temperature "$T = 273.15$ K to 333.15 K" and pressure "$p = 0.1$ MPa" have been reported elsewhere [1]. The mole fraction solubilities of LUT in binary {EtOH + water ($H_2O$)} systems at "$T = 273.15$ K to 323.15 K" and "$p = 0.1$ MPa" have also been reported elsewhere [21]. The equilibrium solubility of LUT in $H_2O$ at "$T = 310.2$ K" is also available in literature [15]. The equilibrium solubilities of LUT in various imidazole-based ionic liquids are also available in literature [2,22]. Nevertheless, the solubilities of LUT in pure solvents such as ethylene glycol (EG), propylene glycol (PG), polyethylene glycol-400 (PEG-400), 2-butyl alcohol (2-BuOH) and Transcutol® (THP) are not yet available in literature. Hence, the objective of present study was to evaluate solubility, thermodynamic properties and solute-solvent molecular interactions of LUT in various pure solvents namely $H_2O$, EG, PG, PEG-400, MeOH,

* Corresponding author at: Kayyali Chair for Pharmaceutical Industries, Department of Pharmaceutics, College of Pharmacy, King Saud University, Riyadh, Saudi Arabia.
*E-mail addresses:* faiyazs@fastmail.fm, fsahmad@ksu.edu.sa (F. Shakeel).

https://doi.org/10.1016/j.molliq.2018.01.155
0167-7322/

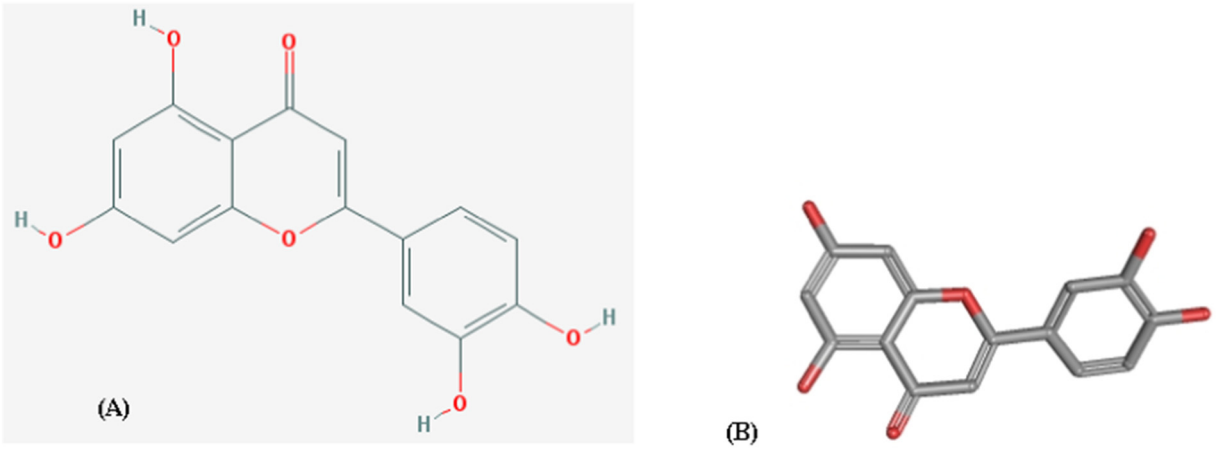


**Fig. 1.** Molecular structure of LUT; (A) 2D structure and (B) 3D structure.

EtOH, IPA, 1-BuOH, 2-BuOH, EA, THP and DMSO at "$T = 298.2$ K to 318.2 K" and "$p = 0.1$ MPa". The solubility data of LUT recorded in this study could be useful in "extraction/separation, purification, recrystallization, drug discovery and dosage form design" of LUT.

## 2. Experimental

### 2.1. Materials

LUT and THP [IUPAC name: 2-(2-ethoxyethoxy) ethanol] were obtained from "Beijing Mesochem Technology Co. Pvt. Ltd. (Beijing, China)" and "Gattefosse (Lyon, France)", respectively. MeOH (IUPAC name: methanol), EtOH (IUPAC name: ethanol), IPA (IUPAC name: isopropanol), 1-BuOH (IUPAC name: 1-butanol) and 2-BuOH (IUPAC name: 2-butanol) were obtained from "E-Merck (Darmstadt, Germany)". EG (IUPAC name: 1,2-ethanediol), PG (IUPAC name: 1,2-propanediol), PEG-400 (IUPAC name: polyethylene glycol-400), EA (IUPAC name: ethyl ethanoate) and DMSO (IUPAC name: dimethyl sulfoxide) were obtained from "Sigma Aldrich (St. Louis, MO, USA)". $H_2O$ (conductivity < 1.0 μS $cm^{-1}$) was obtained from "Milli-Q Water Purification Unit". The purity, source and method of analysis of these materials are presented in Supplementary Table 1 (Table S1).

### 2.2. Analysis of LUT using UPLC-UV method

"Waters Acquity® H-class Ultra-Performance Liquid Chromatography (UPLC)" system coupled with a Waters diode-array-ultra-violet detector (DAD-UV) by Aqcuity "UPLC (Waters, MA, USA)" was used for the analysis of LUT in solubility samples. Briefly, separation employed reverse-phase isocratic elution using "Acquity® UPLC BEH $C_{18}$ column (2.1 × 50 mm, 1.7 μm)" obtained from "Waters (Waters Inc., Bedford, MA, USA)" and a mobile phase consisting of a mixture of 0.1% formic acid and acetonitrile (70:30, v/v). The mobile phase was run at flow rate of 0.3 ml/min with injection volume of 1 μl. PDA detector was set to acquire 3D data from 210 to 400 nm while the 2D channel was recording at 348 nm. The column temperature was set at "$T = 313.2$ K". The peak of LUT was resolved at 1 min in a total run time of 1.2 min. The "Masslynx software" was used for data acquisition and processing. The calibration curve was constructed between the concentration of LUT and measured UPLC area. This calibration curve of LUT was recorded on mass/mass basis and obtained as linear in the range of (0.1 to 20.0) μg $g^{-1}$ with coefficient of determination ($R^2$) of 0.9993. The equation for regression line was recorded as $y = 128.09x + 48.24$; in which $y$ is the measured UPLC area of LUT and $x$ is the concentration of LUT (μg $g^{-1}$). The proposed analytical technique was validated well for "linearity, accuracy, precision, robustness, sensitivity, reproducibility and specificity" and found to be suitable for the analysis of LUT contents in solubility samples.

### 2.3. Solid state characterization of LUT

The characterization of solid state of LUT in both pure and equilibrated form was performed using "Differential Scanning Calorimetry (DSC)" and "Powder X-ray Diffractometric (PXRD)" techniques in order to evaluate crystallinity and possible transformation of LUT during equilibrium. DSC analysis of pure and equilibrated LUT was performed with the help of "DSC-8000 Instrument (Perkin Elmer, MA, USA)". The DSC assembly was equipped with chiller ($T = 253.2$ K) and autosampler. The calibration was carried out with the help of pure indium at "$T = 283.2$ K to 773.2 K". A mass of around 4.60 mg of pure LUT and 5.60 mg of equilibrated LUT was accurately weighed using digital balance and placed into an aluminium pan. Each aluminium pan was hermetically sealed. DSC thermograms of pure and equilibrated LUT were obtained under a nitrogen purge of 20 ml $min^{-1}$ with heating rate of 10.0 K $min^{-1}$ at "$T = 298.2$ K to 673.2 K".

The PXRD spectra of pure and equilibrated LUT were recorded using "Ultima IV Diffractometer (Rigaku Inc. Tokyo, Japan)" over the 2θ range of 3–80° at a scan speed of 0.5°/min. The tube anode used was "Cu with Ka = 0.1540562 nm mono chromatized with a graphite crystal (Rigaku Inc., Tokyo, Japan)". The spectra of pure and equilibrated LUT were obtained at tube voltage and tube current of 40 kV and 40 mA, respectively in step scan mode (step size 0.02°, counting time 1 s/step).

### 2.4. Determination of LUT solubility

The solubility values of LUT in various pure solvents namely $H_2O$, EG, PG, PEG-400, MeOH, EtOH, IPA, 1-BuOH, 2-BuOH, EA, DMSO and THP was measured using a shake flask method [23]. This method was chosen in this study due to its frequent application in achieving solid-liquid equilibrium (SLE) [23–26]. The solubility of LUT was measured at "$T = 298.2$ K to 318.2 K" and "$p = 0.1$ MPa". The excess quantity of crystalline LUT was added in known quantities of each pure solvent in triplicates. Each LUT-solvent mixture was vortexed for 10 min and transferred to the "OLS 200 Grant Scientific Biological Shaker (Grant Scientific, Cambridge, UK)" maintained at 100 rpm for 72 h. After 72 h, each mixture was removed from the shaker and allowed to settle LUT particles for 24 h [27]. After 24 h settling of LUT solid particles, the supernatants were carefully withdrawn, diluted (wherever required) and subjected