Measurement and Modeling Phase Equilibria of Coumarin and Its Derivatives in Supercritical Carbon Dioxide

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Abstract

The solubility of coumarin and its derivatives bonded by methyl-, methoxy- or hydroxy groups in supercritical CO_2 were measured in the ranges of 308.15-323.15 K and 8.5-25 MPa using a flow-type apparatus. The measured data were modeled by a new approximate lattice equation of state(EOS) proposed by the present authors. Coumarin and its 7-methyl- and 7-methoxy- derivatives showed higher solubility than 4- and 7-hydroxycoumarin. Also, coumarin derivatives with two functional groups(i.e., 6,7-dihydroxycoumarin, 7-hydroxycoumarin-4-acetic acid and 7-methoxy-coumarin-4-acetic acid, etc.) were almost insoluble in supercritical CO_2.

Keywords : experimental method, model, supercritical carbon dioxide, solubility, coumarin, coumarin derivatives

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1. Introduction

Supercritical fluid extraction(SFE) can be an efficient tool for extracting bioactive substances from natural plants[1,2]. However, SFE extracts from natural products usually contain many compounds and numerous derivatives. Therefore, if one intends to obtain a desired high-purity substance from a SFE total extract, it is essential to have quantitative knowledge of the phase equilibria between the solute and the supercritical

solvent. However, experimental as well as thermodynamic modeling studies on phase equilibria of biosubstances in supercritical fluids are extremely limited due to an inherently complex nature. In particular, for most biosubstances, necessary intrinsic properties such as critical constants, vapor pressure and molar density data are scarce.

Since the coumarin and its derivatives known to us for their strong bioactivities such as antibiotic, antitumor, vasodilatory and anticoagulant properties[3], researchers, have focussed on the extraction of them from natural plants using supercritical CO_2[4-10]. However, systematic measurement and thermodynamic modeling studies are not yet available. In this regard, the present work reports experimental and thermodynamic modeling of coumarin and its derivatives in supercritical carbon dioxide over a wide range of equilibrium conditions.

Among numerous types of coumarins, we selected a few types of coumarin with emphasis on the derivatives with fixed positions(e.g., C-4 and C-7 positions as shown in Fig. 1).

2. Experimental

Chemicals. Reagent-grade (<98% purity) coumarin and its derivatives(7-hydroxy-, 7-methoxy-, 7-methyl-, and 4-hydroxy-coumarins, 6,7-hydroxy-coumarin, 7-hydroxycoumarin-4-acetic acid and 7-methoxy-coumarin-4-acetic acid, etc.) were purchased from Aldrich Co(Milwaukee, WI, USA) and were used without further purification. CO_2 (<99.95%) was obtained from Seoul Gas Co(Seoul, Korea). Other solvents of HPLC-grade used in chromatographic analysis were purchased from J. T. Baker Co(ST. Louis, MO, USA).

Solubility Measurements. As shown elsewhere [8,10], a flow-type micro-scale apparatus was used. The volume of the equilibrium cell was 60 mL. Pressure was

controlled by a gas booster and Heise gauge. Temperature in an air-bath was controlled by PID controller and held constant within ±1 K. The effluent coumarin solutes dissolved in supercritical CO_2 were collected by a 2-step methanol trap and the extract remained in the equipment lines and valves were rinsed three times after each experimental run by methanol, acetone and chloroform, respectively.

The effluent flow rate was 200 ml/min at an ambient conditions of 298.15 K, and 1 atm by a metering valve. A mass flowmeter was used to measure the amount of CO_2 consumed. For each solute, experiments were repeated five times to ensure the reliability of the measured data. The solubility of the solutes was measured at three isotherms (308.15, 313.15 and 323.15 K) and at each isotherm, pressure was varied by 8.5, 10, 15, 20 and 25 MPa, respectively.

***Solubility Determination of Coumarins*.** Each coumarin tested in this study had its own chromophore and was identifiable by UV detector. We, therefore, used HPLC instead of gas chromatography. The mobile phase used in the HPLC was a 40:60 ratio of methanol: 5% formic acid [8]. For each solute, a calibration curve was prepared and it was used as the reference for assaying each sample.

3. Thermodynamic Modeling

Pure Physical Properties Estimation. For coumarin derivatives, like other natural biosubstances, almost no information of physical constants are reported in the literature. Thus, the necessary pure physical properties were estimated based on methods presented in a recent databook[11]. From it, the Lyderson-Forman-Thodos method of determining boiling point and critical temperature, and the Miller method for determining critical pressure and volume were used. Also, the molar volumes and vapor pressures were estimated using the Bhirud method and the Lee-Kestler correlation, respectively[11]

SCF Solubility Correlation by a Lattice EOS. To model the solubility of coumarins, a new equation of state(EOS), developed recently by the present authors, and based on the nonrandom lattice-hole theory, was utilized[12]. The EOS is written for a general mixture by

$$P = \frac{1}{\beta V_H}\left\{\frac{z}{2}\ln\left[1+\left(\frac{q_M}{r_M}-1\right)\rho\right]-\ln(1-\rho)+\frac{z}{2}\sum_{i=1}^{c}\theta_i\left(\frac{\tau_{0i}}{\sum_{k=0}^{c}\theta_k\tau_{ki}}-1\right)\right\}; \quad (1)$$

where, $q_M = \Sigma x_i q_i$, $r_M = \Sigma x_i r_i$, $\rho_i = N_i r_i / N_r$, $\rho = \Sigma \rho_i$ and x_j is the mole fraction of species i. We set the coordination number, z at 10 and the unit lattice cell volume, V_H, equal to 9.75 cm^3mol^{-1}. If we set the subscripts i=1 and j=0, the EOS becomes specific for pure fluids.

The two molecular parameters in the EOS for pure fluids; V_1^* and ε_{11} were determined from the estimated molar volumes and vapor pressures of pure components[11], and they were fitted by the following empirical expressions:

$$\varepsilon_{11} / k = E_a + E_b(T - T_0) + E_c(T\ln T / T_0 + T - T_0) \quad (2)$$

$$V_1^* = V_a + V_b(T - T_0) + V_c(T\ln T / T_0 + T - T_0); \quad (3)$$

where the reference temperature, T_0 is 273.15K.

The estimated values of coefficients for Eqns. 2 and 3 are summarized in Table 1 for CO_2 and coumarin derivatives.

For a 1-2 binary mixture, the cross interaction energy, ε_{12} is based on the combining rule defined by

$$\varepsilon_{12} = (\varepsilon_{11}\varepsilon_{22})^{0.5}\left(1-\lambda_{12}^{(0)}-\lambda_{12}^{(1)}T\right); \quad (4)$$

where the temperature-dependent binary adjustable parameters, $\lambda_{12}^{(0)}$ and $\lambda_{12}^{(1)}$ fitted using experimental values from the coumarin/CO_2 systems, are listed in Table 2.

4. Results and Discussion

The solubility of coumarin and its derivatives(i.e., 7-hydroxy-, 7-methyl-, 7-methoxy-, 4-hydroxy-coumarin, 6,7-dihydroxycoumarin, 7-hydroxycoumarin-4-acetic acid and 7-methoxy-coumarin-4-acetic acid, etc.) in supercritical CO_2 were measured by a flow-type equilibrium cell in the ranges of 308.15, 315.15, and 323.15K and 8.5, 10, 15, 20 and 25MPa. The experimental data were quantitatively predicted by a lattice-hole EOS formulated recently by the present authors(Table 1 and 2).

In Fig. 3, the effect of temperature(i.e., 308.15, 313. 15 and 323.15 K) and pressure(i.e., 8.5-25MPa) on the solubility of coumarin derivatives in CO_2 were shown. The phenomenon of crossover of solubility in the high pressure region was detected. In this fig. 3, calculated results from the EOS were shown in which the binary interaction energy parameter, l_{12} was calculated as a function of temperature(Table 2). In Figs. 4-7, the measured and calculated solubility of 7-hydroxycoumarin, 7-methylcoumarin, 7-methoxycoumarin, and 4-hydroxy-coumarin in CO_2 was summarized. In general, the EOS fit the data reasonably well.

Finally, effect of different functional groups in positions 4 and 7 on the respective solubility at 323.15K are shown in Fig. 8. We found that the basic coumarin, 7-methylcoumarin and 7-methoxycoumarin showed significantly higher solubility than 7-hydroxycoumarin and 4-hydroxycoumarin. Similar trends in solubility were detected for other isotherms(i.e., 301.15 and 313.15 K) In general, coumarin derivatives with hydroxy group show lower solubility than methyl- and methoxy-coumarins.

Furthermore, derivatives made up by the substitution of two functional groups such as 6,7-dihydroxycoumarin, 7-hydroxycoumarin-4-acetic acid and 7-methoxycoumarin-

4-acetic acid showed almost no solubility at all (y_2 lower than 10^{-6}).

In summary, the effect of functional groups on the solubility of coumarin derivatives in supercritical CO_2 was measured. Furthermore, the data can be quantitatively modeled by an EOS proposed by the present authors.

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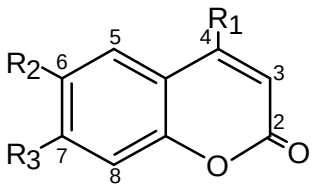
Table 1. Coefficients in energy and volume parameters defined by Eq. (2) and (3)

	E_a	E_b	E_c	V_a	V_b	V_c
carbon -dioxide	85.9130	-.1029	-.3656	34.2861	.0143	-.0131
coumarin	183.4034	-.1294	-.1537	103.2913	.0285	-.0255
7-hycroxycoumarin	89.1369	.4229	1.0499	38.3586	-.0652	-.5902
7-methylcoumarin	158.6453	.5264	5.0472	118.4976	.0528	.2024
7-methoxycoumarin	78.9319	.2531	.1591	51.6904	.0186	-.2123
4-hydroxycoumarin	89.1312	.5251	2.0559	38.1493	-.1137	-1.0664

Table 2. Binary interaction energy parameters defined by Eq. (4)

	$l_{12}^{(0)}$	$l_{12}^{(1)} \times 10^3$
CO_2-coumarin	-0.783116	2.83829
CO_2- 7-hycroxycoumarin	-2.20586	3.69143
CO_2-7-methylcoumarin	0.0319718	0.2280
CO_2-7-methoxycoumarin	-1.66545	2.5707
CO_2-4-hydroxycoumarin	-1.76904	2.41686



$R_1 = R_2 = R_3 = H$	coumarin
$R_1 = OH, R_2 = R_3 = H$	4-hydroxy coumarin
$R_1 = R_2 = H, R_3 = OH$	7-hydroxy coumarin
$R_1 = H, R_2 = R_3 = OH$	6,7-dihydroxy coumarin
$R_1 = R_2 = H, R_3 = OCH_3$	7-methoxy coumarin
$R_1 = R_2 = H, R_3 = CH_3$	7-methyl coumarin
$R_1 = R_3 = H, R_2 = CH_3$	6-methyl coumarin
$R_1 = CH_2COOH, R_2 = H, R_3 = OH$	7-hydroxy coumarin 4-acetic acid
$R_1 = CH_2COOH, R_2 = H, R_3 = OCH_3$	7-methoxy coumarin 4-acetic acic

Fig. 1. Coumarin derivatives and their measured solubilities in supercritical carbon dioxide.

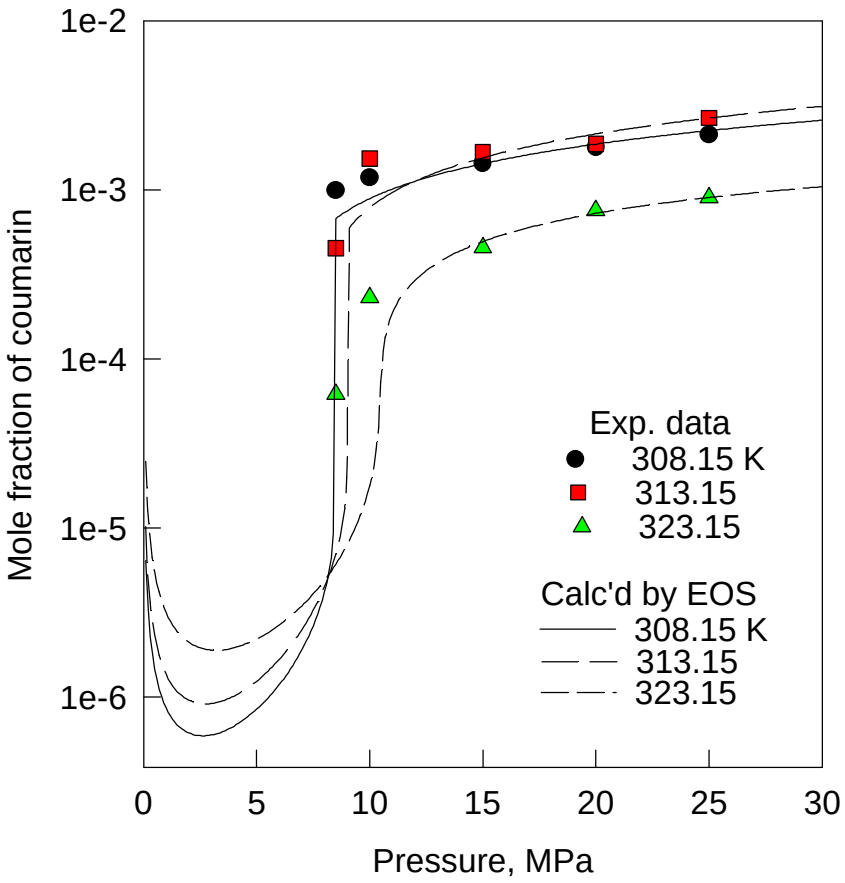


Fig. 2. Measured and calculated solubility of coumarin in supercritical CO_2

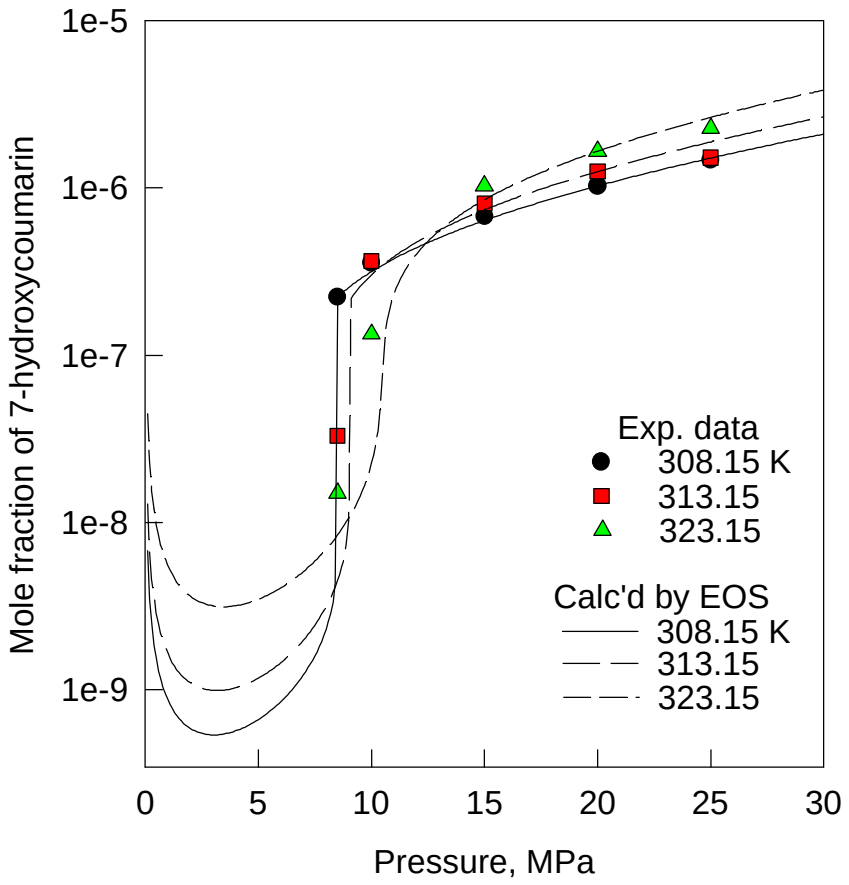


Fig. 3. Measured and calculated solubility of 7-hydroxycoumarin in supercritical CO_2

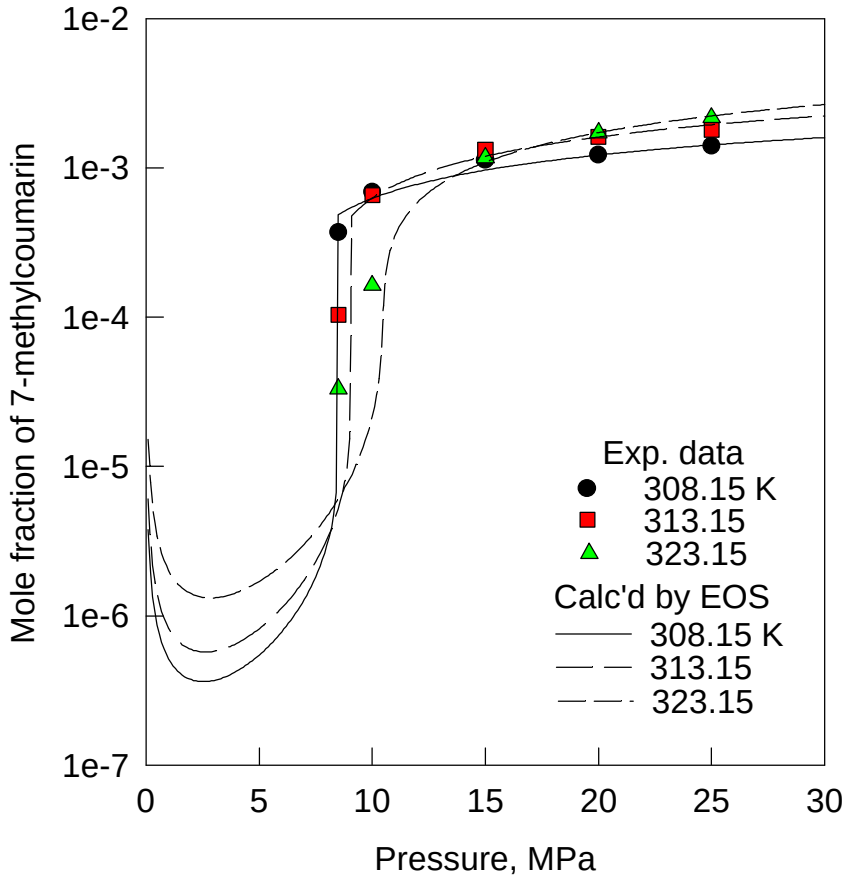


Fig. 4. Measured and calculated solubility of 7-methylcoumarin in supercritical CO_2

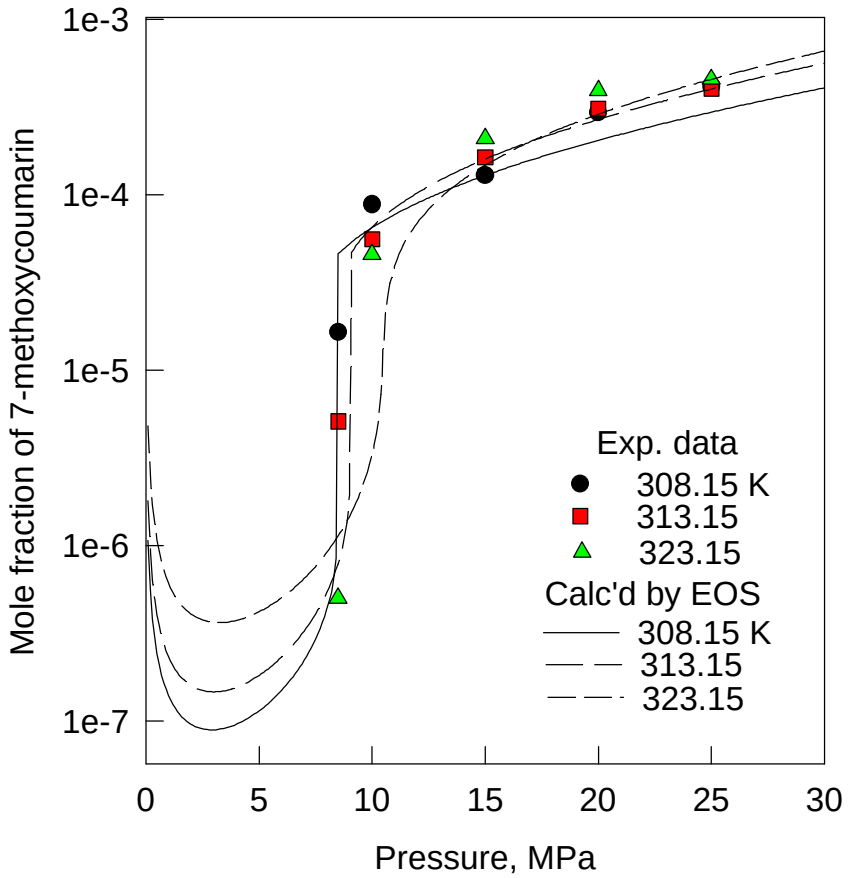


Fig. 5. Measured and calculated solubility of 7-methoxycoumarin in supercritical CO_2

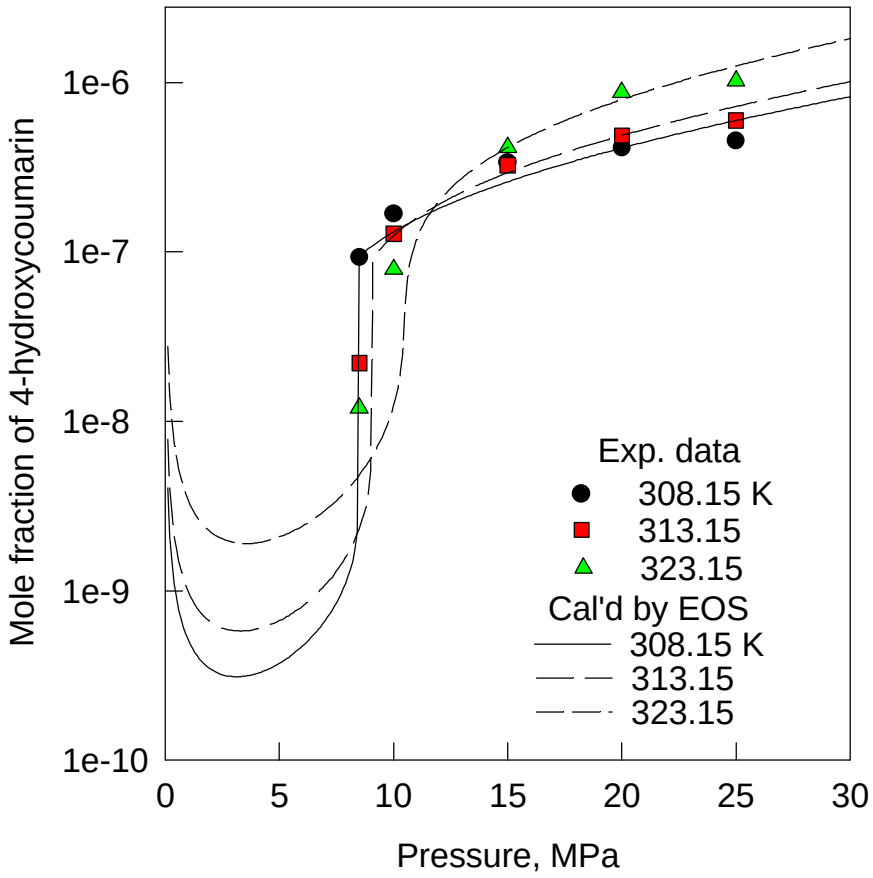


Fig. 6. Measured and calculated solubility of 4-hydroxycoumarin in supercritical CO_2

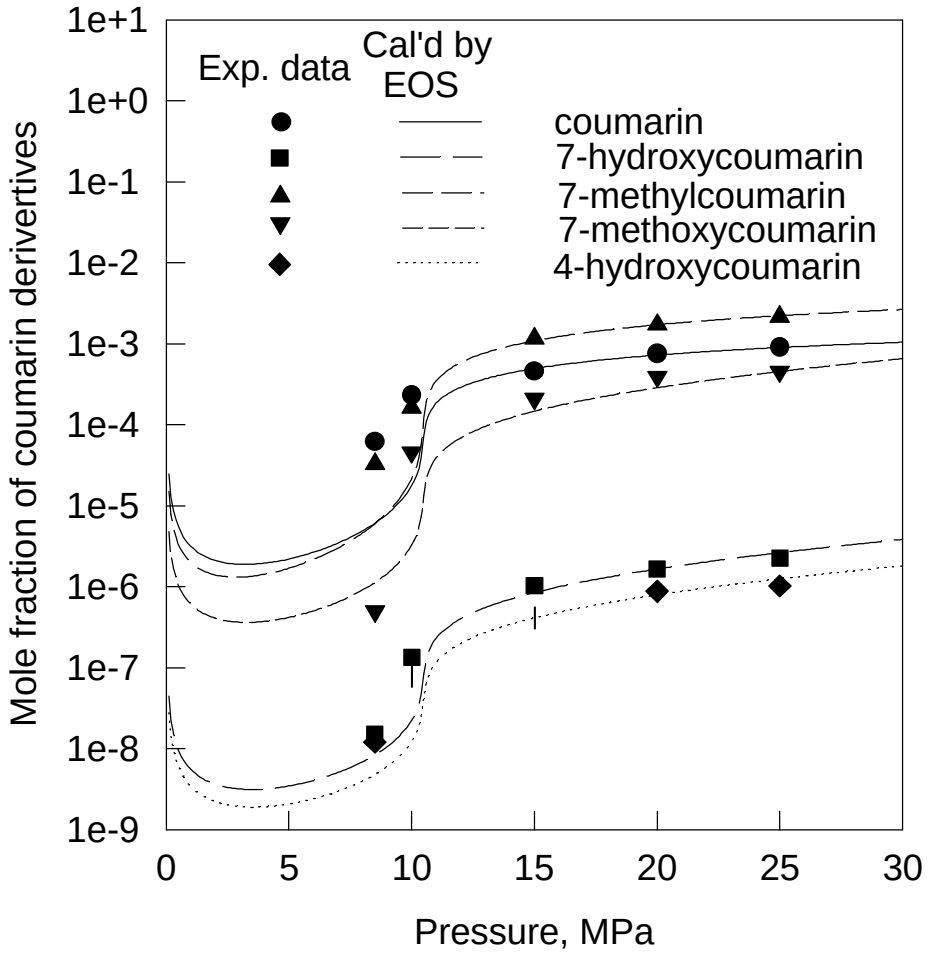


Fig.7. Effect of functional groups on the solubilities of coumarins in CO_2 at 323.15 K