

A Dynamic Model for Ultrasonic – assisted Extraction of Bio-active Compounds from Natural Products

Trung Kien Tran ^{*,1}

Lan Huong Phung ¹

Hoai Nga Le ¹

Thi Thu Huyen Nguyen ¹

Xuan Son Nghiem ²

Van Thiem Pham ³

¹ Department of Chemical Engineering, Hanoi University of Science and Technology (HUST), No. 1, Dai Co Viet Str., Hanoi, Vietnam.

² Department of dynamic and engineering equipment of plant, School of Process Sciences, Technische Universität Berlin, No. 135, 17. Juni Street, 10623 Berlin, Germany.

³ Bachkhoa Consultancy & Technology Transfer One Member Co., Ltd. (BKContech Co.,Ltd.), HUST, No. 1 Dai Co Viet Str., Hanoi, Vietnam.

*e-mail : kien.trantrung1@hust.edu.vn

Ultrasonic technique has been applied for extraction processes, especially for separation of bio-active substances from natural organic products because of the short time requirement, energy saving and easy operating. The ultrasonic-assisted extraction (UE) therefore becomes a promising option. However, a major available obstacle was the lack of appropriate models for designing technological process.

This paper presents a dynamic model for the UE process, application of separation of bio-active substances from natural products such as Curcumin from rhizome of *Curcuma Longa L.*, Epigallocatechin from green tea leaf of *Camellia Sinensis* and Rutin from bud of *Sophora Japonica*. The agreement between data from experiment and calculated ones with the model indicates that, this dynamic model is suitable for prediction of the UE process and for serving as the base for further investigation into scale-up the extraction process.

Keywords: Ultrasonic, Extraction, Curcumin, Epigallocatechin, Rutin.

INTRODUCTION

Natural organic compounds are getting more attention from both scientific and industrial perspectives as an alternative for

synthetic compounds (Colegate S. M. *et al.*, 2008, Sarker S. D. *et al.*, 2006). In this category, instead of reaction section, liquid-solid extraction is usually considered

as the heart of the processing plant. In order to handle the variety of raw materials, effective and flexible extraction techniques are needed. While conventional techniques are widely applied to large-volume productions such as vegetable oil, the advanced techniques target the small-volume but big-profit applications in pharmaceutical and food industries. Ultrasonic-assisted extraction (UE) process is a promising option since ultrasonic boost up the extraction rate without requiring high temperature or pressure, which raise the production expense as well as degrade the product quality in many cases. More details regarding the advantages of UE process can be found elsewhere (Gu X. *et al.*, 2007, Hitoshi K. *et al.*, 2007). In this paper, a model for single-stage batch UE is proposed and experimentally validated. The experiments were carried out with three raw materials: rhizome of *Curcuma Longa L.*, leaf of *Camellia Sinensis* and bud of *Sophora Japonica L.* The validated model can also be easily applied for multi-stage batch processes, which have more applications in industry. In the rest part of this paper, detailed modelling approach of this process is presented. Then the experimentations used for validation of the model are reviewed and the results are reported and discussed. The paper is concluded by focusing on its potential for being used in scale-up duties.

MODELLING

The extraction process modelled in this study is considered to be a simple mass transfer process without chemical reaction. The model development is based on the

following assumptions:

- All particles are of the same size and shape, and therefore, have a constant area;
- The extraction process is isothermal;
- The amount of solvent is kept constant during the extraction process;
- The mass and pore structure of the insoluble materials are assumed to be constant.

Furthermore, by assuming the mass transfer resistance to be localized only in the films, the unsteady-state mass transfer rate is derived as:

$$\frac{dq}{dt} = K_{eq}F(C^* - C) \quad (1)$$

where $\frac{dq}{dt}$ is the unsteady-state mass transfer rate, K_{ex} is extraction rate constant, F is the area of solid-liquid interface, C^* is the equilibrium concentration, and C is the solute concentration in the bulk of the solution.

Since the amount of solvent is kept constant, the solute concentration in the bulk of the solution is proportional to the content of solute in the bulk of the solution:

$$C = \frac{q}{V} = \frac{q}{mK_v} \quad (2)$$

where V is the volume of solvent, m is the mass of insoluble material, and K_v is the liquid-solid ratio.

The equilibrium concentration is proportional to the content of solute which remains in solid phase:

$$C^* = K_{eq} \frac{Q - q}{m} \quad (3)$$

where Q is total amount of solute, and K_{eq} is equilibrium constant.

For a specific raw material, the initial content of solute in solid phase and the area of solid-liquid interface F are proportional to the mass of insoluble material:

$$Q = K_c m \quad (4)$$

$$F = K_a m \quad (5)$$

where K_c is the content of solute per unit mass of solid phase at the beginning of extraction process and K_a is the area of solid-liquid interface per unit mass of solid phase.

Substituting equations (2) - (5) into equation (1) gives:

$$\frac{dq}{dt} = K_{ex} K_a m \left(K_{eq} K_c - \frac{K_{eq} K_v + 1}{K_v m} q \right) \quad (6)$$

By integrating between time limits 0 to t , equation (6) becomes:

$$q = K \left(1 - e^{-\frac{t}{T}} \right) \quad (7)$$

where K is the maximum content of solute in liquid phase:

$$K = \frac{K_{eq} K_v}{K_{eq} K_v + 1} K_c m \quad (8)$$

and T is the required time for the actual content to reach 63.21 % of the maximum value, i.e. when t equal to T then q is 63.21 % of K :

$$T = \frac{K_v}{K_{eq} K_v + 1} \frac{1}{K_{ex} K_a} \quad (9)$$

The extraction efficiency is:

$$H = \frac{K_{eq} K_v}{K_{eq} K_v + 1} \left(1 - e^{-\frac{K_{ex} K_a (K_{eq} K_v + 1)}{K_v} t} \right) \quad (10)$$

The model parameters (K_c , K_{eq} , K_{ex} , K_a) are estimated based on experimental data with two different liquid-solid ratios using least square method. The accuracy of the models can be evaluated by comparison between simulation and experimental results with another liquid-solid ratio.

This model is similar to typical models for conventional extraction methods (Harker J. H. *et al.*, 2002). However, with these methods, assumptions made above are often incorrect. Attempts to apply this type of model to conventional extraction lead to high tolerances (Sonawane S. S. and Patil V. S., 2008). In case of UE of bio-active compounds, the effect of ultrasonic help agitate both inside and outside of particle, increase mass transfer through cell membrane without permanently destroying it and these assumption can be accepted (Nguyen T. T. H. *et al.*, 2010). The model, thus, fit the experiment data with higher accuracy. More details about ultrasonic-assisted extraction of curcumin and the modelling approach can be found in (Tran T. K. *et al.*, 2007 and 2008).

EXPERIMENTS

Three extraction processes were studied in this research:

- Extraction of Curcumin from rhizome of *Curcuma Longa L.*
- Extraction of Epigallocatechin from leaf of *Camellia Sinensis*
- Extraction of Rutin from bud of *Sophora Japonica L.*

Materials and chemicals

For all these three case studies, the plants were collected from different provinces of Vietnam: Curcuma Longa L. from Bac Giang, Camellia Sinensis from Thai Nguyen, Sophora Japonica L. from Thai Binh. There are several suitable solvents such as ethanol, methanol, sodium hydroxide solution, etc. for extracting these bio-active substances. Among them, the ethanol-water mixture (96% vol.) was chosen because of the safety aspect, its availability and reasonable price.

Quantitative analyses were carried out using HPLC method. Standard Curcumin, Epigallocatechin and Rutin were used to develop the calibration curve. Solvents used as mobile phases consisted of acetic acid, acetonitrile, ethyl acetate, methanol, oxolane, phosphoric acid, sodium dihydrogen phosphate. All of them were at HPLC grade.

Experimental setups

a) For generating ultrasonic, the ultrasonic cleaner Model TPC-280, 30 kHz of Telsonic Company – Swiss was used as the extractor equipment.

- Three identical cylinder vessels were fixed at defined positions in an eight-probe ultrasonic tank.

- The volume of solution in each vessel was

set to 800 cm³ for Curcumin extraction, 600 cm³ for Epigallocatechin extraction and 200 cm³ for Rutin extraction. The vessels were kept closed during the extraction process to prevent possible solution evaporation. In order to change the liquid-solid ratio, the amount of solid feed will be changed instead of the volume of solution. Thus, lower liquid-solid ratio mean more solid feed and more extracted solute (but lower extract efficiency).

- The process was carried out at room temperature. Care was taken to ensure that no change in temperature occurred during the extraction process. This was achieved by circulating cold water during the process.

Overall the operating conditions were maintained constant throughout the experiment.

At each specific point of time, a one millilitre sample was taken and stored inside dark tubes. The total amount of sample is less than 2% of the total amount of solution (or 5% in the case of Rutin extraction). Thereby, the amount of solution is considered constant during the extraction process.

b) Equipment for analysis:

- HPLC Agilent 1100, Detector UV-VIS, column RP - C18 (250 mm x 4 mm): for Curcumin and Epigallocatechin analysis;

Table 1. Solvent composition and detection wavelength for analyses

Sample	Mobile phase (volume proportion)	Wavelength (nm)
Curcumin	CH ₃ OH : CH ₃ CN : CH ₃ OH/CH ₃ COOH (pH ≈ 4) = 25 : 50 : 25	425
Epigallocatechin	CH ₃ COOH 0.2 % : CH ₃ CN : CH ₃ OH : CH ₃ COOC ₂ H ₅ = 90 : 6 : 1 : 3	270
Rutin	CH ₃ OH : NaH ₂ PO ₄ /H ₃ PO ₄ (pH ≈ 3) : C ₄ H ₈ O = 10 : 70 : 20	254

- HPLC SCL-10A VP, Shimadzu, Detector UV, column C18 (150 mm x 4,6 mm): for Rutin analysis

The solvent compositions and detection wavelengths for all analyses are summarized in Table 1.

RESULTS AND DISSCUSION

The analysis results were used to calculate the solute concentration in the bulk of the solution at sampling time. The solute concentrations in the bulk of the solution versus time were fit into equations (7 - 9) to estimate the model parameters. The analysis results from validating experimental data were compared to the simulation results to assess the attained models.

Extraction of Curcumin from rhizome of *Curcuma Longa L.*

Two liquid-solid ratios were employed to estimate the model parameters: $K_V = 200$ and $K_V = 10$. The validated experiment was taken with the liquid-solid ratio $K_V = 100$.

With $K_V = 10$, the content of Curcumin in the saturated solution was 1.344 g. The estimated model parameters are summarized in Table 2, whereas the experimental results and the simulation results using $K_V = 200$ and $K_V = 100$ are presented in Figure 1.

The maximum error is 3.2% for $K_V = 200$ and 3.6% for $K_V = 100$. The following equation can be used to determine the extraction efficiency:

$$H = \frac{0.046K_V}{0.046K_V + 1} \left(1 - e^{-\frac{0.324(0.046K_V+1)}{K_V}t} \right) \quad (11)$$

Extraction of Epigallocatechin from leaf of *Camellia Sinensis*

Two liquid-solid ratios, $K_V = 100$ and $K_V = 10$, were employed to estimate the model parameters. The validated experiment was taken with the liquid-solid ratio $K_V = 40$. With $K_V = 10$, the content of Epigallocatechin in the saturated solution was 2.640 g. The estimated model parameters are summarized in Table 2.

The experimental results and the

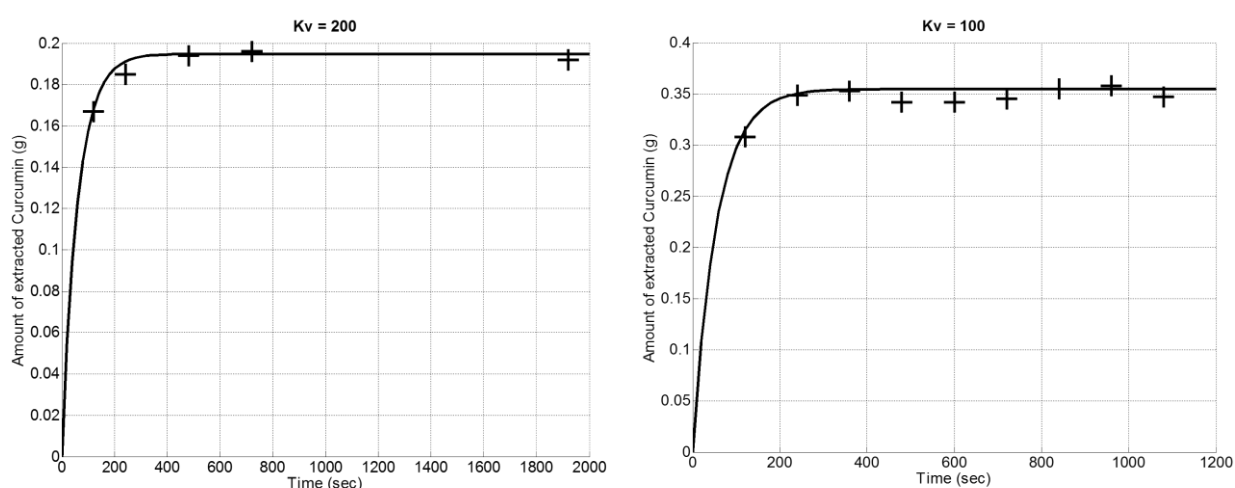


Fig. 1: Comparison simulation (line) and experiment (cross) for $KV = 200$ and $KV = 100$

Table 2. Estimated model parameters

Case study	K_C	K_{eq}	$K_{ex} \cdot K_a$
Curcumin / Curcuma Longa L.	0.054	0.046	0.324
Epigallocatechin / Camellia Sinensis	0.045	2.180	1.675e-3
Rutin / Sophora Japonica L.	0.355	0.247	1.417e-2

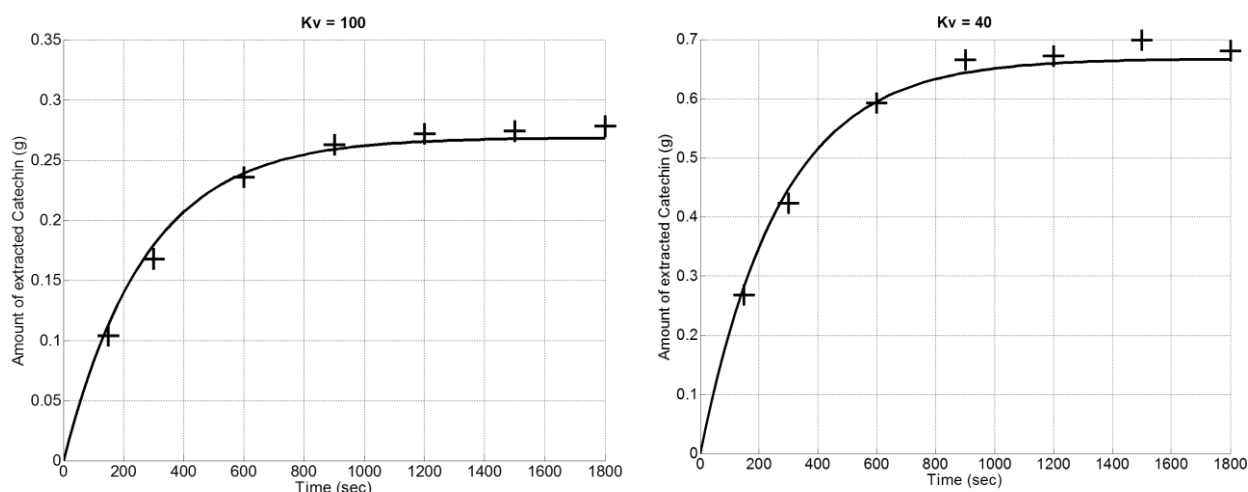


Fig. 2: Comparison simulation (line) and experiment (cross) for KV = 100 and KV = 40

simulation results with $K_V = 100$ and $K_V = 40$ are presented in Figure 2.

The maximum error is 3.2% and 5.6% for $K_V = 100$ and $K_V = 40$ respectively. The following equation can be used to determine the extraction efficiency:

$$H = \frac{2.180K_V}{2.180K_V + 1} \left(1 - e^{-\frac{1.675(2.180K_V+1)10^{-3}}{K_V}t} \right) \quad (12)$$

Extraction of Rutin from bud of Sophora Japonica L.

Two liquid-solid ratios were employed to estimate the model parameters: $K_V = 20$ and $K_V = 10$. The validated experiment was taken with the liquid-solid ratio $K_V = 15$. With $K_V = 10$, the content of Rutin in the saturated solution was 5.060 g. The estimated model parameters are summarized in Table 2. The experimental

results and the simulation results with $K_V = 20$ and $K_V = 15$ are presented in Figure 3.

The maximum error is 9.4% in case of $K_V = 20$ and 4.9% in case of $K_V = 15$. The following equation can be used to determine the extraction efficiency:

$$H = \frac{0.247K_V}{0.247K_V + 1} \left(1 - e^{-\frac{1.417(0.247K_V+1)10^{-2}}{K_V}t} \right) \quad (13)$$

Three case studies presented hereinabove were subjected to the proposed model considering different organs of different plants. The agreement between simulation and experimental results in all three cases ensure the wide applicability of the proposed model. The proposed model also predicts the maximum achievable extract efficiency for a specific material and fixed liquid-solid ratio. When the extraction time increases,

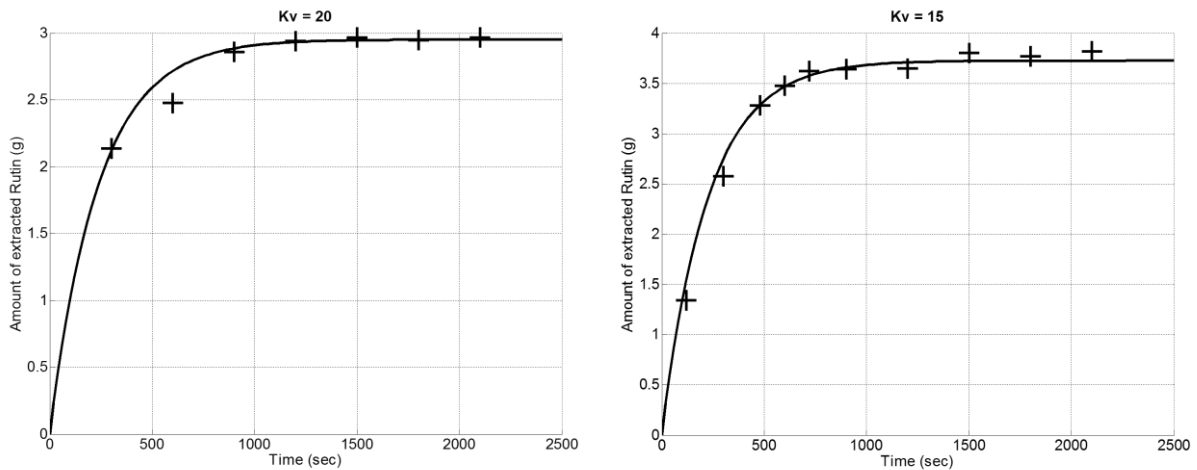


Fig. 3: Comparison simulation (line) and experiment (cross) for KV = 20 and KV = 15

the real efficiency approaches this value exponentially. Therefore, extraction process should not last longer than five times T (equation 9), when the efficiency is more than 99% of the maximum value. For multi-stage batch process, which is preferred in industry, this model can be applied for each single stage in order to calculate the extract time, extract efficiency, and solvent consumption of the whole process.

CONCLUSION

An efficient model was proposed to predict the dynamic behaviour of ultrasonic-assisted extraction process. The model reflects the effect of liquid-solid ratio and extraction time on the extraction efficiency and concentration of the extract. Since this model is independent of the scale of process, it can be used for simulating the ultrasonic-assisted extraction processes, and furthermore in large scale process design.

NOMENCLATURE

- C : Solute concentration in the bulk of the solution ($\text{g}\cdot\text{cm}^{-3}$)
- C^* : Solute concentration in the bulk of the solution in equilibrium with the content of solute in the solid phase ($\text{g}\cdot\text{cm}^{-3}$)
- F : Area of solid-liquid interface (cm^2)
- H : Extract efficiency
- K_a : Area of solid-liquid interface per unit mass of solid phase ($\text{cm}^2\cdot\text{g}^{-1}$)
- K_c : Content of the solute per unit mass of the solid phase at the beginning of the extraction process
- K_{ex} : Extraction rate constant ($\text{cm}\cdot\text{s}^{-1}$)
- K_V : Liquid-solid ratio ($\text{cm}^3\cdot\text{g}^{-1}$)
- M : Mass of insoluble material (g)
- W : Total amount of solute (g)
- Q : Content of the solute in solution (g)
- t : Time (s)
- V : Volume of solvent (cm^3)

REFERENCES

1. Colegate S. M., Molyneux R. J., (2008). *Bioactive Natural Products: Detection, Isolation, and Structural Determination*, 2nd ed., CRC Press, Raton.
 2. Gu X., Cai J., Zhang Z., Su Q. (2007). Dynamic Ultrasound-Assisted Extraction of Catechin and Caffeine in Some Tea Samples. *Annali di Chimica* 97: 321 – 330.
 3. Harker J. H., Richardson J. F., Backhurst J. R. (2002). *Chemical Engineering*, Vol. 2, 5th ed., Butterworth-Heinemann.
 4. Hitoshi K., Nobuyoshi M. (2007). Extraction of Catechins from Green Tea Using Ultrasound, *Japanese Journal of Applied Physics* 46 (7B): 4936 - 4938.
 5. Nguyen Thi Thu Huyen, Tran Trung Kien, Phung Lan Huong, Pham Van Thiem (2010). Research on extraction process for Rutin assisted by ultrasonic, microwave and stir bar. *Journal of science and technology*, 48 (6A): 296-304.
 6. Nguyen Thi Thu Huyen, Tran Trung Kien, Pham Van Thiem (2010). Applying the dynamic model of Ultrasonic assisted extraction process for study of rutin separation from bud *Sophora Japonica* L. *Journal of science and technology*, 48, (6A): 305-312.
 7. Sarker S. D., Latif Z., Gray A. I. (2006). *Natural Products Isolation*, 2nd ed., Humana Press Inc, New Jersey.
 8. Sonawane S. S., Patil V. S. (2008). Effect of Ultrasound on Leaching of Tannic Acid from Tea and its Modelling. *Chem. Eng. Technol.* 31 (9): 1304 – 1309.
 9. Tran T. K., Nghiem X. S., Phung L. H., Pham V. T. (2007). Study for isolation of Curcumin from *Curcuma Longa* L. using ultrasonic extraction, *Vietnam Journal of Chemistry* 45: 52 – 57.
 10. Tran T. K., Nghiem X. S., Phung L. H. (2008). Dynamic model of Curcumin isolation process from yellow turmeric using ultrasonic extraction, *Vietnam Journal of Chemistry* 46: 318-325
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