

Enhanced Microwave Extraction of Curcumin from Turmeric (*Curcuma longa* L.)

¹Tahmineh Ebadi, ²Ali Akbar Moghadamnia, ²Sohrab Kazemi and ¹Ghasem Najafpour Darzi

¹Biotechnology Research Lab, Faculty of Chemical Engineering,
Noshirvani University of Technology, Babol, Iran

²Department of Pharmacology and Physiology, School of Medicine,
Babol University of Medical Sciences, Babol, Iran

Abstract: Curcumin is a bioactive polyphenolic compound derived from the rhizomes of turmeric. It has wide application against many chronic diseases such as diabetes, Alzheimer, rheumatoid arthritis and large variety of cancers like breast, lung and blood. In this paper, the effective parameters in curcumin extraction using microwave irradiation such as particle size, modifier solvent for destruction of turmeric cell wall, organic solvent, contact time and solvent to solid ratio were investigated. Extraction of curcumin using microwave soaked in water with particle size of 0.2 mm was enhanced. Maximum curcumin extraction by acetone as organic solvent, for extraction time of 3 minutes and solvent to solid ratio of 15:1 (ml/g) was obtained. Based on HPLC analysis, the yield of purified curcumin was more than 70%. It was concluded that enhanced microwave extraction was high efficiency process. The process is low energy intensive with efficient use of solvent. The extraction and purification is economical while using less solvent and required short extraction time.

Key words: Curcumin • Turmeric • Microwave • Acetone • Curcumin yield

INTRODUCTION

As green chemistry in medical research is getting to be the most popular in past decades, application of medicinal plant is rapidly growing for the prevention and treatment of many diseases [1]. Polyphenols have significant role on protection of human health and treatment of illness. They are known as a great antioxidant and anticancer agent used for cancer therapy [2].

The rhizome of plant *curcuma longa* L. has wide application in pharmaceutical, cosmetic and food industries in present and past centuries [3]. About 3-5% of these rhizomes are bright yellow polyphenols called curcuminoids, consist of curcumin, demetoxy curcumin and bisdemetoxy curcumin [4, 5]. The extract of turmeric exerts several therapeutic properties including anti-inflammatory, antibacterial, antiviral, antifungal [6], anti HIV [7] and cancer chemopreventive performances [8]. It is also proved that curcuminoids lowered high blood sugar by inhibiting α -glucosidase activity [9]. Furthermore, turmeric powder traditionally uses for

treatment of rheumatism, anorexia, diabetic wounds, sinusitis, coryza, cough, hepatic disorders and binary disorders.

Curcumin is the main bioactive compound among curcuminoids and may be responsible for these medicinal effects. Curcumin has now been used to treat arthritis, diabetes, psoriasis, osteoporosis, cardiovascular diseases, Alzheimer and crohn's disease as well as different kind of cancers [10] and infection of immune system. It is interesting to state that recent researches demonstrate that curcumin can even prevent mental stress and depression related disorder [11] which has been also stated in Chinese traditional medicinal science [12].

Several extractive methods are reported for isolation of curcumin and other curcuminoids compounds; such as conventional extraction by solvent, use of alkaline solution and insoluble salt and also microwave assisted extraction [4].

Conventional methods for extraction of curcumin from natural sources are including use of organic solvents such as acetone, methanol, ethanol, ethyl acetate,

isopropyl alcohol and hexane. This method is based on correct selection of solvents and use of heat or mixing to increase the solubility of considered compounds and improving mass transfer. Soxhlet extraction is the most common type of extraction method and is still used as a standard in all cases [13].

Apart from its low extraction efficiency, conventional method is a time-consuming process and has harmful effects on environment due to use of large amount of organic solvent. Moreover, conventional method may cause degradation of target component which is sensitive to high temperature. Therefore, new methods have been reported for extraction of curcuminoids. Among these methods, ultrasound and microwave extraction have shown high efficiency and low energy and solvent consumption [14]. In many published papers, comparing of microwave extraction with other advanced and conventional methods of extraction, microwave extraction is accepted as a powerful and potential replacement for extracting organic compounds from plant materials [15].

The basic principles of microwave extraction process differ from conventional methods, because the extraction occurs due to changing in cell structure because of electromagnetic waves. In this way, process speed and high efficiency of extraction may be due to same direction gradient of two phenomena of mass and heat transfer [16].

This study aims to investigate some effective parameters in enhanced microwave extraction. These findings may help to improve our knowledge to apply this novel method in extraction of curcumin and increases its potential applications in manufacturing of curcumin- based drugs.

MATERIALS AND METHODS

Turmeric powder is used as raw material for curcumin extraction. The powder was supplied from local market. All solvents used for the extraction were analytical grade and purchased from Merck (Darmstadt, Germany). Standard curcumin was also supplied by Merck. The analytical methods were developed by means of spectrophotometer and HPLC analysis. Microwave apparatus was a commercial microwave for domestic use, Samsung from South Korea.

Experimental Procedure

Pretreatment of Turmeric: The rhizome of turmeric was ground and then the powder passed through sieves with different mesh size (0.177, 0.21, 0.25, 0.4 and 0.707 mm). It was necessary to determine the effect of particle

size in extraction process. A specified amount of different particle size of the powder was pretreated by irradiating samples at 100 W microwave power input for 4 minutes. Next step, all of these particle size were soaked in water, methanol, acetone and ethyl acetate (mass: solvent ratio of 1:3 w/v) and then irradiated like the previous samples. After drying samples in oven, small amounts of samples were dissolved in methanol and centrifuged to separate insoluble particles. Then, they were analyzed to determine their curcumin content by the use of standard calibration curve via spectrophotometric method.

Extraction

Type of Solvent: Known amount of dry turmeric powder was suspended in different solvents such as acetone, ethanol, methanol and 2-propanol (mass:solvent ratio 1:30 w/v) [17]. Then the samples were irradiated at 100W microwave power input for 4 minutes. After that the extracts were filtered through a 0.45 μ m filter, then filtered solids were dried, extracts were dissolved in methanol to prepare 300 mg/l curcumin solution. The amount of curcumin by the use of same method was investigated.

Solvent Concentration: The known amount of turmeric powder was suspended in the solvent which is defined from the last experiment in the proportion of 20-100% with water as a co-solvent. Then, samples were placed in microwave at the same situation to be irradiated.

Contact Time: A defined amount of dry powder of turmeric was suspended in the solvent; that was determined from the last experiment and then placed in microwave at power input of 100W for the exposure time of 1, 3, 5 and 7 minutes. The suitable microwave exposure time was defined by the measurements according to method previously explained.

Solvent to Feed Ratio: Turmeric powder was suspended in the solvent to make 10, 15, 20, 25 and 30 ml solvent per gram of turmeric powder. The power input in microwave set at 100W and the desired exposure time was identified. Then, maximum extraction for desired amount of solvent to feed at optimal conditions was defined.

Preparation of Standard Calibration Curve: The standard calibration curve for curcumin was prepared based on readings for absorbance of curcumin solution in the concentration ranged 1-6 mg/l. A stock solution was prepared in methanol, the absorbance was detected at wavelength of 420 nm. The calibration curve of curcumin

was plotted for concentration of curcumin versus absorbance. The concentration of curcumin is defined by the following equation and R^2 of for the plotted data is 0.999.

$$Y = 01531(\text{Absorbance reading}) + 0.0243$$

Curcumin Yield: Turmeric powder was placed in microwave for determination of curcumin yield at optimal condition. After that the mixture was filtered using 0.45 μ m filter and the extract was dried in oven 70°C. The amount of curcumin was calculated from standard calibration curve using HPLC analysis.

$$\text{Curcumin yield \%} = \frac{\text{curcumin extracted (g)}}{\text{turmeric used (g)}} \times 100$$

HPLC Method for Curcumin Analysis: For quantification of curcumin and curcuminoids, HPLC analysis was performed based on detection of concentration in the standard solution. The developed method was modified base on previous HPLC analysis reported in literature [14]. Samples were analyzed by HPLC using UV detector. The samples were injected and the elution was carried out with solvent flow rate of 1.0 ml/min at room temperature. The HPLC column used was C18 (250 \times 4.6mm). The mobile phase was acetonitrile and water (90:10 v/v). The detection wavelength was set at 420nm.

In order to determine the amount of curcumin extracted, pure curcumin with concentration 1.25, 2.5, 5, 10 and 20 ppm dissolved in methanol and then injected to HPLC after the certain retention time. The calibration curve of the HPLC peak from each standard solution is drawn. After that from the extracted powder, 5 ppm solution was prepared and injected to HPLC. With the aid of calibration curve the amounts of curcumin in the sample solutions were determined (Figure 1).

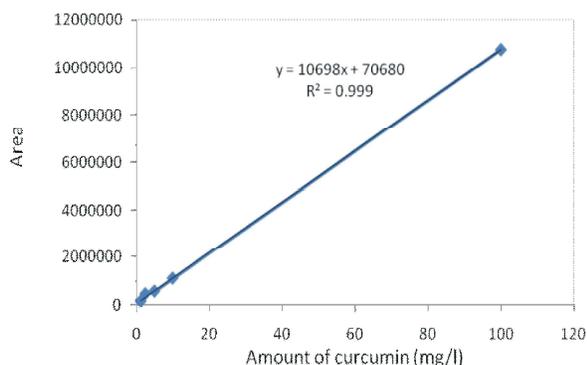


Fig. 1: Calibration curve of HPLC analysis of standard curcumin

RESULT AND DISCUSSION

Pretreatment: Figure 2 represents the effect of pretreatment on extraction of curcumin. It was demonstrated that when turmeric was soaked in water, more curcumin was released. It means cell disruption may easily happen when turmeric was pretreated in water. However, the same result did not happen when it was soaked in other solvents. The important factor in microwave assisted extraction is the solvent. In case of polar solvent, the ionic strength may assist solvent to absorb strongly microwave energy; that is most probably due to dipole moment. Another important property of microwave extraction would be the dielectric loss; which has the ability of solvent to absorb microwave energy and then dissipate energy to the surrounding molecules. In fact, water has higher dielectric constant than the other solvents. Increasing in dielectric constant usually causes more microwave energy absorption. Table 1 summarized dielectric constants and dissipation factors for several effective solvents used in extraction process.

A comparison between water and acetone shows that, water has much more ability to absorb microwave energy; however, its heat dissipation factor is less than acetone. That may cause overheating and even superheating of the product. When strong adsorption occurred, temperature inside the sample strictly increased; then cell rupture is resulted by the in situ water [18].

Wakte *et al.* [4] have reported that the soaked powder of turmeric in the extraction of curcumin by microwave technique was very effective. They represent curcumin extraction yield of turmeric powder which soaked in water was higher than the one soaked in ethanol [4].

About the particle size, it was found that small particle size used for extraction, curcumin content in extracts was much more than when larger particle size was used. Curcumin extraction for small particle size should be more effective than large particle; that was most probably due to more surfaces provided and exposed to solvent. The best extraction yield of curcumin was happened when the particle size was 0.2 mm.

Extraction Parameters

Effect of Solvent: Figure 3 illustrates the influence of solvent in the curcumin content. It shows acetone is the most desired solvent for curcuminoids extraction; because curcumin is a polyphenol which can be dissolved in organic and less polar solvent. Wakte *et al.* [4] have also showed that when ethanol used as extraction solvent, the curcumin content was 67%. In fact, curcumin contacted with acetone was used it became more [4].

Table 1: Dielectric constants and dissipation factors for solvents used in microwaveextraction

Solvent	Dielectric constant	Dissipation factor $\tan \delta (\times 10^{-4})$
Acetone	20.7	5555
Methanol	32.6	6400
Water	78.3	1570
Ethyl acetate	6.02	5316

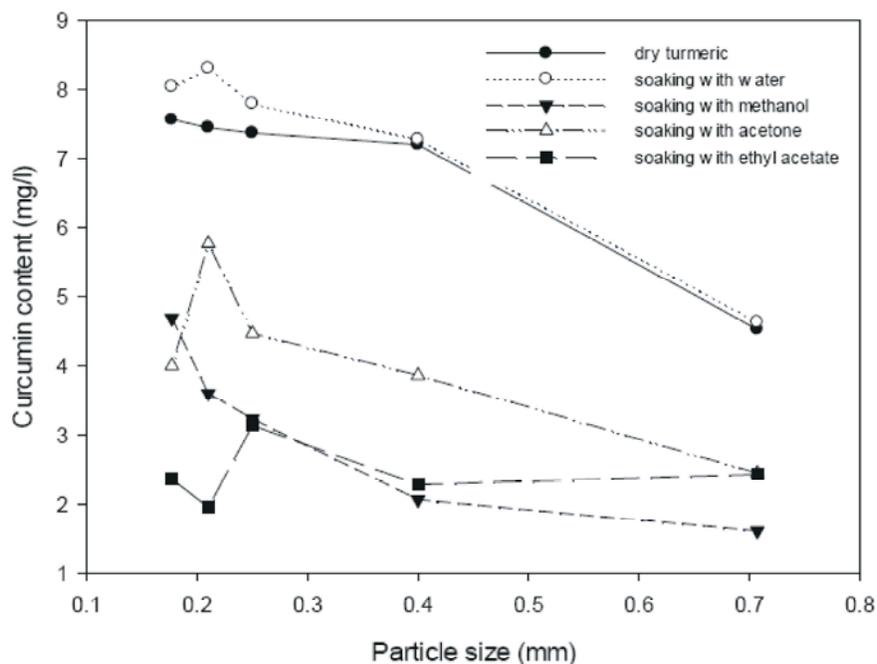


Fig. 2: Curcumin content with respect to particle size for dry and soaked turmeric, under microwave irradiation (4min) and power input (100W)

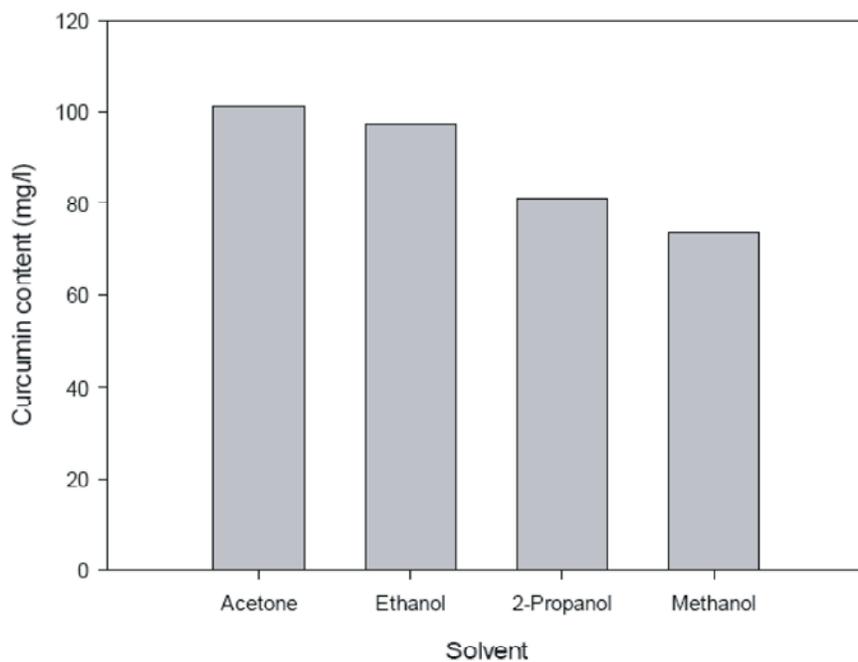


Fig. 3: Effect of solvent in extracted curcumin content

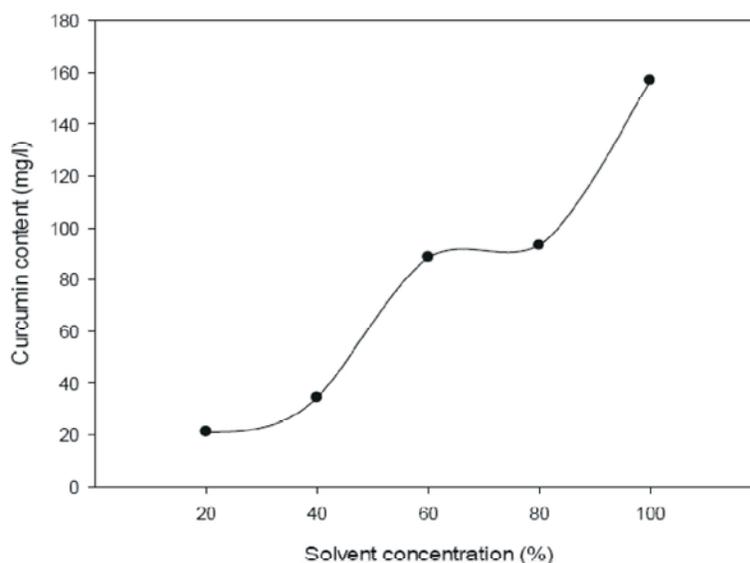


Fig. 4: Effect of solvent concentration in curcumin extraction

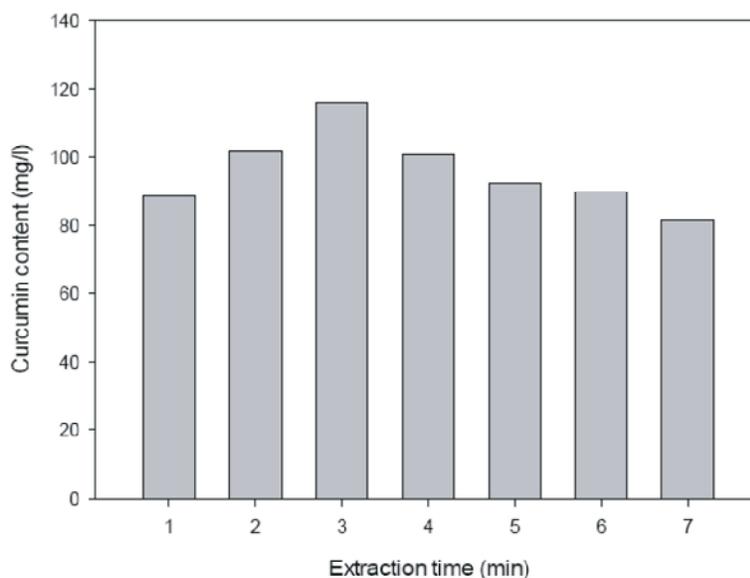


Fig. 5: Curcumin concentration with respect to extraction time

Effect of Solvent Proportion: Figure 4 shows curcumin concentration with respect to percentage of solvent. It seems that presence of water may not be useful in extraction of curcumin. Dahmoune *et al.* [17] reported that solvent property in microwave aided extraction may act as an important role in extraction of polyphenolic compounds from *Myrtus communis* L. leaves. In addition, it was noted that water and low concentration of ethanol can easily penetrate into cells. Use of plant cell pretreatment assisted cell disruption happens and then curcuminoids are freely available for solvent extraction of polyphenols in turmeric. Addition of water into solvent

can easily get access to cell; however extra amount of water may reduce solubility of the solvent.

Effect of Extraction Time: Figure 5 represents the effect of extraction of curcumin with respect to extraction time. The extraction efficiency increased with respect to time up to 3 minutes. However, it began to decline for additional time up to 7 minutes. That is probably due to destruction and denaturation of curcumin due to long period of irradiation. Besides extraction time, there is an important factor affecting on irradiation that is microwave power input (V).

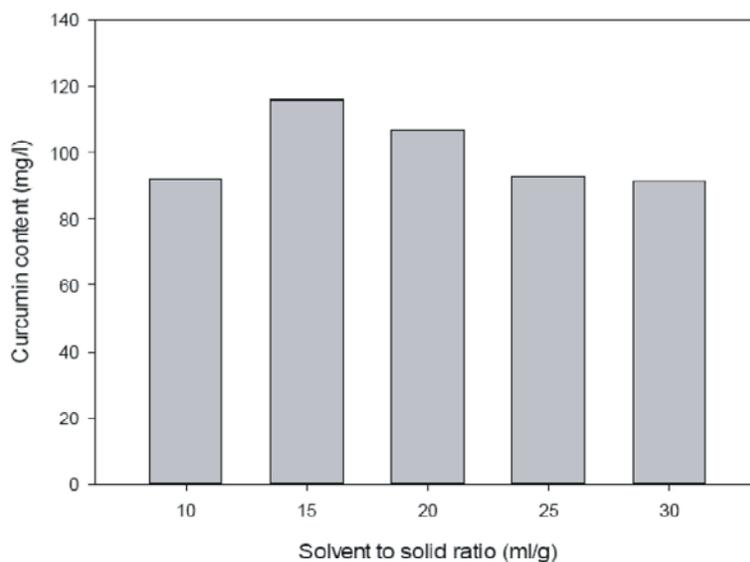


Fig. 6: Effect of solvent to solid ratio

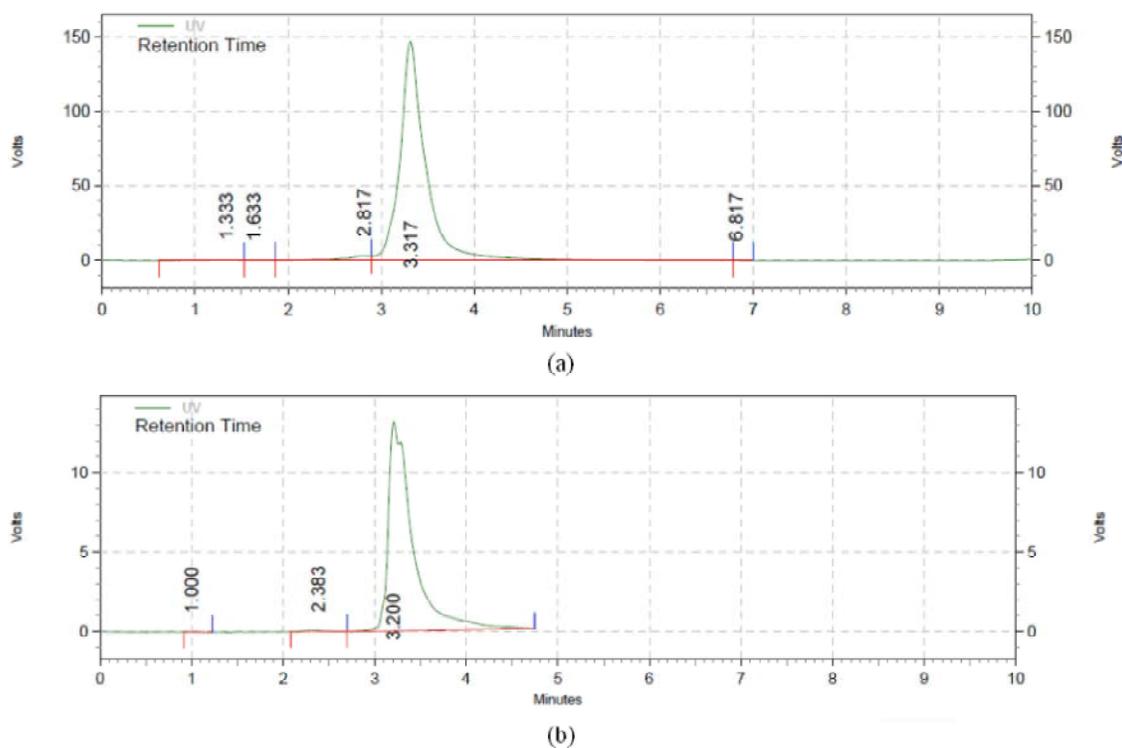


Fig. 7: HPLC curve a) 10 mg/l of standard curcumin b) 5 mg/l of extracted curcumin

Mandal *et al.* [19] have shown that in microwave assisted extraction of curcumin, when 1 minute exposure time was used for extraction, by increasing microwave power from 20 to 60% that can increase extraction efficiency. In contrary, for period of exposure, say 4 minutes extraction time, increasing in microwave power caused decrease in extraction efficiency.

Effect of Solvent to Solid Ratio: One of the effective parameter in extraction for the recovery of phenolic compounds is feed to solvent ratio. In our case, solvent volume per unit mass of turmeric was chosen. In an extraction process, it is important to maximize extraction yield, while minimizing consumption of solvent [17]. In the current study, the recovery of curcumin from turmeric was

enhanced by increasing solvent/solid ratio during extraction upto 15:1 (ml:g). Excess amount of solvent, solvent/solid ratio of 30:1(ml:g) caused to decline (Figure 6). Low solvent/solid ratio has promoted mass transfer barrier as the distribution of active compounds in concentrated regions caused interaction of intracellular compounds out of cell matrix [20]. However, high ratio of solvent/solid caused absorption of additional microwave power and then denaturation of compounds occurred.

Curcumin Yield: When all extraction parameters were optimized, the defined amount of turmeric powder with 0.2 mm particle size was experimented for extraction at optimum condition and placed under microwave radiation. Amount of curcumin was measured using HPLC method. Figure 7 is shown HPLC analysis of standard and extracted curcumin. Curcumin content in turmeric was obtained as 3.51%. Based on reported data, it was found that curcumin content in turmeric powder is in the range of 3-5% [4]. Therefore, the reported yield was satisfactory in compare to similar cases. If you take maximum level as ideal point say 5%, then current work turmeric had curcumin extraction efficiency of more than 70% and it can be consider as high efficiency.

CONCLUSION

In this research paper, we optimized the effective parameters acting on microwave-assisted extraction of curcumin from turmeric's rhizomes. The result showed water was a suitable modifier for pretreatment of turmeric due to its high dielectric constant. Among all organic solvents, acetone was chose the best organic solvent for curcumin extraction because of its low polarity. Irradiation of microwave for long exposure say 7 min increased the denaturation of curcumin. The curcumin yield was obtained more than 66.7%. In our work, microwave-assisted extraction is a cost-effective method for curcumin isolation due to its low solvent and less time consumption.

ACKNOWLEDGEMENT

Authors are gratefully acknowledged Biotechnology Research Lab., Noshirvani University of Technology (Babol, Iran) and Babol University of Medical Sciences provided and facilitated the present research.

REFERENCES

1. Pan, M.H., T.M. Huang and J.K. Lin, 1999. Biotransformation of curcumin through reduction and glucuronidation in mice. *Drug Metabolism and Disposition*, 27(4): 486-494.
2. Basnet, P. and N. Skalko-Basnet, 2011. Curcumin: an anti-inflammatory molecule from a curry spice on the path to cancer treatment. *Molecules*, 16(6): 4567-4598.
3. Hatcher, H., R. Planalp, J. Cho, F. Torti and S. Torti, 2008. Curcumin: from ancient medicine to current clinical trials. *Cellular and Molecular Life Sciences*, 65(11): 1631-1652.
4. Wakte, P., B. Sachin, A. Patil, D. Mohato, T. Band and D. Shinde, 2011. Optimization of microwave, ultra-sonic and supercritical carbon dioxide assisted extraction techniques for curcumin from *Curcuma longa*. *Separation and Purification Technology*, 79(1): 50-55.
5. Baumann, W., S.V. Rodrigues and L.M. Viana, 2006. Pigments and their solubility in and extractability by supercritical CO₂ - I: the case of curcumin. *Brazilian Journal of Chemical Engineering*, 17(3): 323-328.
6. Huang, L., J. Zhang, T. Song, L. Yuan, J. Zhou, H. Yin, T. He, W. Gao, Y. Sun and X. Hu, 2016. Antifungal curcumin promotes chitin accumulation associated with decreased virulence of *Sporothrix schenckii*. *International Immunopharmacology*, 34: 263-270.
7. Fischer, P., G. Karlsson, T. Butters, R. Dwek and F. Platt, 1996. N-butyldeoxyojirimycin-mediated inhibition of human immunodeficiency virus entry correlates with changes in antibody recognition of the V1/V2 region of gp120. *Journal of virology*, 70(10): 7143-7152.
8. Mosieniak, G., M.A. Sliwinska, D. Przybylska, W. Grabowska, P. Sunderland, A. Bielak-Zmijewska and E. Sikora, 2016. Curcumin-treated cancer cells show mitotic disturbances leading to growth arrest and induction of senescence phenotype. *The international journal of biochemistry & cell biology*, 74: 33-43.
9. Du, Z.Y., R.R. Liu, W.Y. Shao, X.P. Mao, L. Ma, L.Q. Gu, Z.S. Huang and A.S. Chan, 2006. α -Glucosidase inhibition of natural curcuminoids and curcumin analogs. *European Journal of Medicinal Chemistry*, 41(2): 213-218.

10. Naksuriya, O., S. Okonogi, R.M. Schiffelers and W.E. Hennink, 2014. Curcumin nanoformulations: a review of pharmaceutical properties and preclinical studies and clinical data related to cancer treatment. *Biomaterials*, 35(10): 3365-3383.
11. Chang, X.R., L. Wang, J. Li and D.S. Wu, 2016. Analysis of anti-depressant potential of curcumin against depression induced male albino wistar rats. *Brain Research*, 1642: 219-225.
12. Yu, Z., L. Kong and Y. Chen, 2002. Antidepressant activity of aqueous extracts of *Curcuma longa* in mice. *Journal of Ethnopharmacology*, 83(1): 161-165.
13. Fan, J.P., R.F. Zhang and J.H. Zhu, 2010. Optimization of microwave assisted extraction of total triterpenoid in *Diospyros kaki* leaves using response surface methodology. *Asian Journal of Chemistry*, 22(5): 3487.
14. Li, M., M.O. Ngadi and Y. Ma, 2014. Optimisation of pulsed ultrasonic and microwave-assisted extraction for curcuminoids by response surface methodology and kinetic study. *Food Chemistry*, 165: 29-34.
15. Li, Y., L. Han, R. Ma, X. Xu, C. Zhao, Z. Wang, F. Chen and X. Hu, 2012. Effect of energy density and citric acid concentration on anthocyanins yield and solution temperature of grape peel in microwave-assisted extraction process. *Journal of Food Engineering*, 109(2): 274-280.
16. Aguilera, J.M., C. Tzia and G. Liadakis, 2003. Solid-liquid extraction. *Extraction Optimization in Food Engineering*, pp: 35-55.
17. Dahmoune, F., B. Nayak, K. Moussi, H. Remini and K. Madani, 2015. Optimization of microwave-assisted extraction of polyphenols from *Myrtus communis* L. leaves. *Food Chemistry*, 166: 585-595.
18. Veggi, P.C., J. Martinez and M.A.A. Meireles, Fundamentals of microwave extraction, in *Microwave-assisted Extraction for Bioactive Compounds 2012*, Springer. pp: 15-52.
19. Mandal, V., Y. Mohan and S. Hemalatha, 2008. Microwave assisted extraction of curcumin by sample-solvent dual heating mechanism using Taguchi L 9 orthogonal design. *Journal of Pharmaceutical and Biomedical Analysis*, 46(2): 322-327.
20. Chan, C.H., R. Yusoff, G.C. Ngoh and F.W.L. Kung, 2011. Microwave-assisted extractions of active ingredients from plants. *Journal of Chromatography A*, 1218(37): 6213-6225.