

Evaluation of solvent efficiency for extraction of bioactive curcuminoids from turmeric (*Curcuma longa* L.)

Jana Klopchevska^{*1}, Aleksandar Chadikovski¹, Zoran Kavrakovski²,
Marija Srbinoska³, Vesna Rafajlovska¹

¹*Institute of Organic Technology, Faculty of Technology and Metallurgy, Ss. Cyril and Methodius University in Skopje, Rudjer Boskovic 16, 1000 Skopje, Republic of North Macedonia*

²*Institute of Applied Chemistry and Pharmaceutical Analysis, Faculty of Pharmacy, Mother Theresa 47, Ss. Cyril and Methodius University in Skopje 1000 Skopje, Republic of North Macedonia*

³*Scientific Tobacco Institute-Prilep, University "St. Kliment Ohridski"-Bitola, Kicevska bb, 7500 Prilep, Republic of North Macedonia*

Introduction

Turmeric (*Curcuma longa* L., fam. Zingiberaceae) is one of the most cultivated plant that is used since ancient times as a spice, food preservative, colouring and flavour agent. The turmeric is also recognized as a herbaceous medicinal plant widely used in folk medicine for a long time in curing diseases, due to the bioactive compounds such as non-volatile curcuminoids (curcumin, demethoxycurcumin, and bisdemethoxy-curcumin) and volatile essential oil (sesquiterpenoids and monoterpenoids) usually synthesized in the rhizomes as the commonly used plant parts. The curcuminoids are lipophilic polyphenol compounds with a bright orange-yellow color that possess a wide range of biological activities including antioxidant, antimicrobial, antiinflammatory, antidiabetic, antiaging, anticarcinogenic, antiinfective, cardio-protective, and neuroprotective effects (Yadav et al., 2013; Rolfe et al., 2020; de Oliveira Filho et al., 2021).

The production of herbal drug preparations includes several steps, such as procedures for the proper collection, drying, and grinding of plant materials, followed by extraction, fractionation, concentration, and isolation of extractable compounds. Identification and quantification of major bioactive compounds is also recommended. Solid liquid extraction conditions such as temperature, time of extraction, solvent type, extraction technique used, solid and liquid ratio, as well plant material characteristics were

studied to optimize yields, composition and properties of the extracts (Abubakar et al., 2020; Fongang et al., 2021).

The aim of this study was to evaluate the efficiency of the solvent type for extraction of curcuminoids from turmeric (*Curcuma longa* L.).

Materials and methods

Plant material. Turmeric powder purchased from BIOCOSMOS Aronija, Zdrava Hrana, DOOEL (Skopje, Republic of North Macedonia).

Extraction of plant material. The extraction of turmeric powder (5 g ± 0.1 mg) was carried out with Soxhlet method using 250 mL solvent. The solvents used: ethanol (96% v/v), acetone, methanol, diethyl ether and n-hexane were supplied from Merck (Darmstadt, Germany). After 300 min extraction, the solvent was removed at 40°C, 200 mPa using a rotary evaporator (Devarot Elektromedicina, Slovenia). The steps of drying, cooling, and weighing were repeated until the difference between two consecutive weights was smaller than 2 mg. The yield of the extract was calculated based on the dry matter weight (DM) of sample used. The extraction procedure was performed in duplicate under the same operating conditions.

Surface colour determination. The colour CIE Lab parameters of turmeric samples before and after extraction, L* (lightness/darkness), a* (red/green), b* (yellow/blue), chroma (C), and hue (H°) were determined by using Dr.

*jana@tmf.ukim.edu.mk

LANGE spectra colorimeter (Chelmsford, UK) at Illuminat D65 and 10° observer angle as reference. The standardized values for a white plate were $L^* = 95.68$, $a^* = -0.53$, and $b^* = 3.12$. The measurements in then replications were performed. Color difference (ΔE) was calculated by using Hunter-Scottfield's equation $\Delta E = (\Delta a^2 + \Delta b^2 + \Delta L^2)^{1/2}$ where $a = a^{*0} - a^{*1}$, $b = b^{*0} - b^{*1}$, and $L = L^{*0} - L^{*1}$. The subscript "0" and "1" indicates the turmeric sample before and after extraction.

TLC analysis. Compounds of turmeric extracts were separated on silica gel 60 F₂₅₄ plates in chloroform and methanol (95:5 v/v) as a mobile phase. The compounds were visualized under ultraviolet light at wavelength of 254 nm. Retention values (R_f) were calculated. Each separated spots was scraped, transferred in 1 mL ethanol, and centrifuged for 15 min at 5000 rpm (centrifuge type Heraeus Biofuge Fresco, UK). The obtained extract was transferred with ethanol in 5 mL volumetric flask. UV-Visible spectral characteristics were determined on a Varian Cary Scan 50 spectrophotometer (Switzerland).

Determination of total curcuminoid content. The turmeric extracts were dissolved in volumetric flask (25 mL) with ethanol (96% v/v). The absorbance was measured at wavelength of 460 nm in 1 cm quartz cells, at 25°C. The curcuminoid content was calculated using the extinction coefficient of the curcumin ($^{1\%}E_{427nm} = 61864$) in ethanol (Majhi et al., 2010).

Results and discussion

The color of turmeric extracts obtained with ethanol, methanol and acetone was dark yellow-orange, while extracts obtained by using non-polar diethyl ether and *n*-hexane were characterized with light yellow-orange and yellow colour, respectively. The yield of ethanolic turmeric extract was the highest (10.02 g/100 g DM) in comparison to the other used solvents i.e. methanol (9.08 g/100 DM), acetone (6.83 g/100 g DM), diethyl ether (6.33 g/100 g DM), and *n*-hexane (3.45% g/100 g DM). The determined values for the turmeric colorimetric were $L^*(29.24)$, $a^*(12.87)$, $b^*(43.34)$, $C(45.22)$, and $Ho(73.40)$. The highest ΔE value (30.15) was determined when turmeric was extracted with diethyl ether. At turmeric extraction with acetone was calculated the lowest value for color difference ($\Delta E = 22.36$). No spots were separated on the TLC plate with *n*-hexane extract. From the turmeric extracts obtained with ethanol, methanol, acetone, and diethyl ether on the TLC plates were separated three spots with R_f values that ranged between 0.622÷0.666, 0.738÷0.755, and 0.822÷0.827. The separated spots refer to curcumin ($R_f = 0.83$), dimethoxycurcumin ($R_f = 0.75$) and bismethoxycurcumin ($R_f = 0.66$) according to Majhi et al. (2005). In the visible spectral part, the absorption

maximum (λ_{max}) for the first separated spot was at 416 nm, while for the second and the third separated spot were at 422÷424 nm and 426÷432 nm, respectively. The turmeric extracts obtained with ethanol, methanol, acetone and diethyl ether show λ_{max} in the UV (195÷210 nm and 234÷237 nm) and the visible region (423 ÷427 nm). In the *n*-hexane turmeric extract, λ_{max} values were determined at 195÷199 nm, 237÷298 nm, and 344÷349 nm. The highest curcuminoid content expressed as curcumin (1560 mg/100 g extract) was obtained when extraction solvent diethyl ether was used. The curcuminoid content in the extracts obtained with ethanol, methanol and acetone was 500 mg/100 g, 598 mg/100 g, and 837 mg/100 g, respectively. Due to the low curcuminoid solubility in the aliphatic organic solvents such as *n*-hexane, small quantity of curcuminoids in the extract (12 mg/100 g) was determined.

Conclusion

At the turmeric extraction, the highest extract yield was obtained with ethanol, while the diethyl ether allows to obtain the extract richest in the biologically active curcuminoids.

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