

2018 IOP Masturi Optimization of condition extraction in quantification of total flavonoid content in the seeds of the Arummanis (*Mangifera indica* L.) mango from Indonesia

by Masturi Masturi

Submission date: 09-Sep-2020 01:59PM (UTC+0700)

Submission ID: 1382651679

File name: Masturi_et_al_2019__ICMSE2018.pdf (1.65M)

Word count: 3008

Character count: 15685

PAPER • OPEN ACCESS

Optimization of condition extraction in quantification of total flavonoid content in the seeds of the Arummanis (*Mangifera indica* L.) mango from Indonesia

4

To cite this article: Masturi *et al* 2019 *J. Phys.: Conf. Ser.* **1321** 022041

View the [article online](#) for updates and enhancements.

Recent citations

- [Identification of flavonoid compounds and total flavonoid content from biowaste of local durian shell \(*Durio zibethinus*\)](#)
Masturi *et al*

**IOP ebooks™**

Bringing together innovative digital publishing with leading authors from the global scientific community.

Start exploring the collection—download the first chapter of every title for free.

Optimization of condition extraction in quantification of total flavonoid content in the seeds of the Arummanis (*Mangifera indica* L.) mango from Indonesia

Masturi¹, D Alighiri^{2,*}, K Nuzulina¹, M Rodhiyah¹ and A Drastisianti³

¹ Physics Department, Faculty of Mathematics and Natural Sciences, Universitas Negeri Semarang, Indonesia

² Chemistry Department, Faculty of Mathematics and Natural Sciences, Universitas Negeri Semarang, Indonesia

³ Department of Chemistry Education, Faculty of Sciences and Technology, Universitas Islam Negeri Walisongo Semarang, Indonesia

*Corresponding author: dante_alighiri@mail.unnes.ac.id

Abstract. Mangoes are an abundant local fruit. Mango seed is a biowaste material that only becomes an environmental problem. Therefore, a study was conducted to utilize one of the compounds contained in mango seeds particularly the seeds of the Arummanis (*Mangifera indica* L.) from Indonesia, namely flavonoids. One of the most optimum methods is used for extraction with a variety of solvents, namely ethanol, methanol, and hexane. Determination of total flavonoid content is based on the $AlCl_3$ method with total flavonoids content expressed in QE (Quercetin equivalent) at a maximum wavelength of 425 nm. The results showed that the most optimal total flavonoid content was from ethanol extract with the reflux method of 3.234 mg QE/g extract.

14

1. Introduction

Mango (*Mangifera indica* L.) is one of the commercially important tropical fruits in the world. Mango is known to be the most important tropical fruit of Asia, grown commercially in more than 87 countries. This is due to mangoes possess the delicious taste, exotic aroma, and high nutritional value that many people like [1,2]. Mangoes are rich in water, sugar, fiber, minerals, vitamins, and antioxidants [2]. Among internationally traded tropical fruits, mangoes rank of the second-largest tropical fruit traded in the world only to banana both in quantity and value and fifth in total production among major fruit crops worldwide. World production in the mangoes is estimated to be over 42 million tones per year. India is the world's largest producer with a production of 1.525.000 tones per year followed by China, Kenya, Thailand, Indonesia and Mexico [3]. The mango production of Indonesia in 2005 reached 1.20 million tones with a harvest area of 176.000 ha and production centers in Indramayu Regency, Majalengka, Cirebon, Pemalang, Blora, Situbondo, Probolinggo, Pasuruan, Buleleng and Karangasem [4]. The most important mango varieties cultivated in Indonesia from two main species are *Mangifera indica*, namely, Arummanis, Golek, Gedong, Manalagi, and Cengkir and *Mangifera foetida*, namely, Kemang and Kweni.

Like most ways to consume mangoes, in Indonesia, mostly, the consumption of mangoes is also eaten directly in the form of fresh fruit. Other product types of mangoes are marketed in the form of canned fruit, concentrate, dried fruit, juice, and jam [5], from this the production process it is



Content from this work may be used under the terms of the [Creative Commons Attribution 3.0 licence](https://creativecommons.org/licenses/by/3.0/). Any further distribution of this work must maintain attribution to the author(s) and the title of the work, journal citation and DOI.

Published under licence by IOP Publishing Ltd

estimated that 35 to 60% is waste [6]. In particular, mango seeds, more than 1 million tons of mango seeds are produced as waste, and currently, the waste of these seeds is not used for any commercial [7]. Therefore, the seeds of the mango only become an environmental problem.

Based on the many benefits obtained from the mangoes, we tried to examine more about the characteristics and extracts of the seeds of the mango. Several studies have been conducted on the benefits of fruit and mango skin, but research on mango seeds is limited, especially mango seeds from the type of Arummanis which is a local fruit of Indonesia. Like mango skin, mango seeds are also discarded as a by-product during the fruit processing industry. Depending on the variety, the kernel constitutes 45-85% of mango seeds and about 20% of all fruits [8-10]. During this time many mango seeds are wasted because there are no results or products based on the seeds of the mangoes themselves. Therefore, this study aims to find out one of the many ingredients found in plants, namely flavonoids.

Phenol compounds are sometimes considered out of date. However, there are many interesting topics about these compounds that lead to the emergence of discoveries continuously. This makes flavonoid compounds one of the branches of natural materials chemistry that continues to grow [11]. Moreover, the extraction methods of total flavonoid content in the seeds of the Arummanis (*Mangifera indica* L.) mango from Indonesia has not been reported. Thus, the best condition of extraction was obtained by comparing with the yield of total flavonoids of the seeds of the Arummanis (*Mangifera indica* L.) mango.

2. Methods

2.1. Material

The seeds of the Arummanis (*Mangifera indica* L.) mango was collected from Central Java province of Indonesia. The standard of quercetin was purchased from Sigma Aldrich. Aluminium chloride (AlCl_3), ethanol, methanol, and hexane were pure analytical reagent from Merk.

2.2. Sample preparation

The seeds of the Arummanis (*Mangifera indica* L.) mango was cleaned using water, then dried at room temperature (25-27 °C). The process of drying mango seeds was carried out for approximately one week. The dried mango seeds was pulverised using a blender. The powder of the mango seeds of was extracted using three variations of methods, i. e. maceration, reflux, and soxhlet with 96% ethanol solvents. Then, the most optimum method was used for other solvent variations, i.e. ethanol, methanol and hexane.

2.3. Determination of the maximum wavelength (λ_{max})

Determination of the maximum wavelength was carried out by using UV-Vis spectrophotometer and running with quercetin as reference solutions. 2 mL of quercetin solution and 1 mL of testing solution were accurately pipetted into 25 mL volumetric flask, respectively, with 3 mL of 0.1 mol/mL aluminium chloride (AlCl_3) solution separately. Then, the mixture was diluted to volume with 70% ethanol and shaken up. After 30 minutes, the absorption spectra of testing solution and reference solution were gained by wavelength scanning at 400–450 nm, showing a maximum absorption wavelength at 425 nm.

2.4. Standard curves preparation

Weighed as much as 50 mg of standard quercetin and dissolved in ethanol to a final volume of 50 mL. This solution was piped 0.25 mL, 0.5 mL, 0.75 mL, 1 mL, 1.25 mL, 1.5 mL, 1.75 mL and 2 mL, respectively. Then, diluted with distilled water to the final volume of 10 mL so that the concentrations of 25, 50, 75, 100, 125, 150, 175, 200 $\mu\text{g}/\text{mL}$ quercetin were produced. From each concentration, the standard quercetin solution was piped 2 mL. Then, 2 mL of 2% AlCl_3 and 2 mL of potassium acetate were added to 120 mM. Samples were incubated for one hour at room temperature. Absorbance was measured at a maximum wavelength of 425 nm by using the UV-Vis spectrophotometer against an appropriate blank solution [12]. Using the absorbability as ordinate and the concentration as abscissa, the specification curve was obtained. The calibration curve presented a linear response within the

concentration range of 25 – 200 µg /mL. The regression equation was $y = 0.0029x - 0.0008$, $R^2 = 0.9922$.

2.5. Determination of total flavonoid content

Weighed 100 mg of extract, dissolved in 10 mL ethanol, to obtain a concentration of 10 mg/mL. From the solution was pipetted 2 mL then added 2 mL of 2% $AlCl_3$ solution and 2 mL of 120 mM potassium acetate. Samples were incubated for 24 hour at room temperature. Absorbance was measured at a maximum wavelength of 425 nm by using the UV-Vis spectrophotometer. The extraction yield of flavonoids was calculated from standard curve. The concentration of flavonoids was read (mg/mL) on the standard curve. The total flavonoid content in extra was expressed in equivalent quercetin provisions (mg QE/g extract) [12]. Determination of the total flavonoid content of the extract with other solvents was carried out by the same method and composition, which distinguishes the solvent used to dissolve the extract in sample preparation.

2.6. Optimization of extraction condition

Extract method selection: to select efficient extract method, maceration, reflux, and soxhlet extraction were compared. The result showed that reflux could provide the highest extraction yield in quantification of total flavonoid content (Table 2.).

Extract solvent selection: to select efficient extract solvent, methanol, ethanol, and hexane were compared. The result showed that ethanol could provide the highest extraction yield in quantification of total flavonoid content (Table 3.).

3. Result and Discussion

3.1. Optimization of methods in determining total flavonoid content

To determine efficient method, the effect of three main factor including solvent, extraction time, material/solvent ratios were studied. The extraction process was carried out aimed at taking the chemical compounds contained in the sample. The principle of extraction is based on the displacement of the component period of the substance dissolved into the solvent so that displacement occurs in the interface layer and diffuses into the solvent [13]. The extraction methods used in this study were maceration, reflux, and soxhlet. The optimum of extraction time in maceration method was carried out within 24 hours while reflux and soxhlet were carried out for six hours. The three extraction processes were carried out three repetition by using 96% ethanol solvent. The extract obtained was concentrated with the evaporator until a thick brownish-red extract was obtained.

Quantitative analysis of total flavonoids content using UV-Vis spectrophotometer was carried out to find out how much the total flavonoid content contained in the ethanol extract of Arummanis (*Mangifera indica* L.) mango seeds. Flavonoid analysis was carried out using UV-Vis spectrophotometry because flavonoids contained a conjugated aromatic system that showed strong absorption bands in the spectrum of ultraviolet light and visible light spectrum [13].

Table 1. The absorbance of the standard quercetin solution at a maximum wavelength of 425 nm on the ethanol extract of Arummanis (*Mangifera indica* L.) mango seeds

Concentration (ppm)	Absorbance (y)
25	0.073
50	0.136
75	0.233
100	0.280
125	0.364

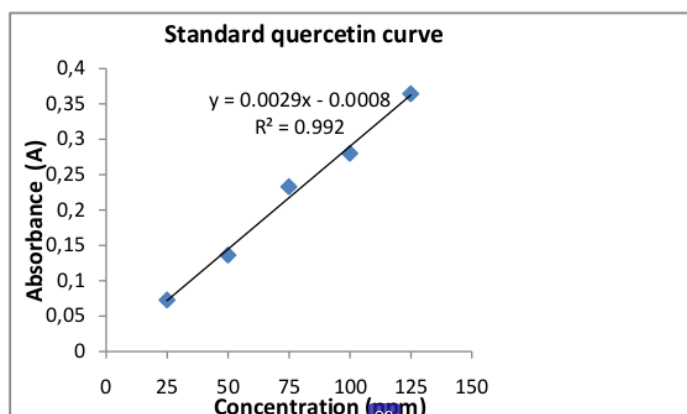


Figure 1. Quercetin calibration curve at a maximum wavelength of 425 nm

In this study to determine the total flavonoids content in the sample used quercetin as a standard solution with a series of concentrations of 25, 50, 75, 100, and 125 ppm. Concentration series are used because the method used in determining the levels is a method that uses the standard curve equation, to make a standard curve first several concentration series are made to obtain a linear equation that can be used to calculate the percent level. Quercetin was used as a standard solution because quercetin is a flavonoid of the flavonol group that has a keto group at C-4 and has a hydroxyl group on neighboring C-3 or C-5 atoms of flavones and flavonols [14]. Measurement of maximum wavelength absorption is run from wavelength 400–450 nm. The running results show the maximum wavelength of the standard quercetin at a wavelength of 425 nm. The maximum wavelength used to measure the absorption of samples of the ethanol extract Arummanis (*Mangifera indica* L.) mango seeds shown in Table 1.

From these measurements, it can be concluded that the higher the concentration used, the higher the absorbance obtained. The raw results of quercetin obtained are plotted between the levels and absorbance so that the linear regression equation was obtained $y = 0.0029x - 0.0008$ with the R^2 value was obtained 0.9922, and the R-value was obtained 0.996. The quercetin calibration curve equation can be used as a comparison to determine the total concentration of flavonoids in the sample extract.

Testing of quantitative analysis with UV-Vis spectrophotometer used a blank solution as a control that functions as a check compounds that do not need to be analyzed though multiplying zero number in UV-Vis spectrophotometer instrument [15].

Table 2. Variation methods extraction in determining of the total flavonoid content of ethanol extract of Arummanis (*Mangifera indica* L.) mango seeds

Methods	Total Flavonoid Content (mg QE/g)
Maceration	0.391 ± 0.003
Reflux	3.234 ± 0.002
Soxhlet	0.455 ± 0.005

Treatment means of the ANOVA test

Values were expressed as the mean ± standard deviation of three replications

The mean difference is significant at the $p \leq 0.050$

* Highly significant, $p = 0.000$

In measuring the total flavonoid compounds, the sample solution was added by $AlCl_3$, which can form complexes, so that the wavelength shifted to visible direction with the solution produced a yellower color [16]. The incubation treatment for 1 hour before the measurement was intended so that

the reaction runs perfectly and resulting color intensity is more maximal. According to the results of this study, the total flavonoid content of the ethanol extract of Arummanis (*Mangifera indica* L.) mango seeds shown in Table 2.

From the results in Table 2., Show that the most optimum results are extraction using the reflux method. The reflux extraction process is carried out by heating in a shorter time but obtaining higher results than the other methods.

3.2. Optimization of solvents in determining total flavonoid content

The variation of the solvent used in this optimization is 96% ethanol and 96% methanol by reflux method. According to the results of this study, we can see the total flavonoid content of Arummanis (*Mangifera indica* L.) mango seeds are shown in Table 3.

Table 3. Variation of solvent in the extraction process of determining of the total flavonoid content of extract of Arummanis (*Mangifera indica* L.) mango seeds

Solvent	Total Flavanoid Content (mg QE/g)*
methanol	0.863 ± 0.003
ethanol	3.234 ± 0.004
hexane	0.325 ± 0.006

Treatment means of the ANOVA test

Values were expressed as the mean ± standard deviation of three replications

The mean difference is significant at the $p \leq 0.050$

* Highly significant, $p = 0.000$

From the results in Table 2. showing the optimum total flavonoid content of the reflux method using ethanol solvent. In terms of traceability, ethanol has advantages compared to water and methanol. Chemical compounds capable of being extracted with more ethanol than methanol and water runners. So that, the optimum of the total flavonoid content of extract of Arummanis (*Mangifera indica* L.) mango seeds were obtained from ethanol extract by the reflux method of 3.234 mg QE/g.

4. Conclusion

This result provided the reliable experimental basis for industrial production to extract the total flavonoids in this crude drug. Using quercetin as standard substance and aluminum chloride ($AlCl_3$) solution as chromogenic agent, the total flavonoids content was measured by UV-Vis spectrophotometer. The confirmation test indicated the optimal method was reproducible, accurate, and feasible. Under this condition, the results represented that the amount of total flavonoids in the sample was obtained from the ethanol extract of Arummanis (*Mangifera indica* L.) mango seeds from Central Java province of Indonesia by the reflux method of 3.234 mg QE/g extract.

13

Acknowledgment

This research was funded by The Ministry of Research, Technology and Higher Education Republic of Indonesia.

Reference

- [1] Engels C, Gänzle M G and Schieber A 2010 *J. Agric. Food Chem* **58** 775
- [2] Tharanathan R N, Yashoda H M and Prabha T N 2006 *Food Rev. Int.* **22** 95
- [3] FAOSTAT 2015 *FAO Statistical Pocketbook World Food and Agriculture* (Rome: FAO)
- [4] Kementerian Pertanian 2007 *Mentan Panen Mangga dan Anggur di Probolinggo* (Jakarta: Direktorat Budidaya Tanaman Buah, Kementerian Pertanian) (Unpublished)
- [5] Masibo M and He 2009 *Food Rev. Int.* **23** 346
- [6] Shea N O, Arendt E K and Gallagher E 2012 *Innov. Food Sci. Emerg. Technol.* **16** 1

- [7] Leanpolchareanchai J, Padois K, Falson F, Bavovada R and Pithayanukul P 2014 *Molecules* **19** 17107
- [8] Arogba S S 1997 *J. Sci. Food Agric.* **73** 321
- [9] Hemavathy J, Prabhakar J V and Sen D P 1988 *Asian Food J.* **4** 59
- [10] Solís-Fuentes J A and Durán-de-Bazúa M C 2011 *Mango (Mangifera indica L.) seed and its fats. In V. Preedy, R. R. Watson, & V. B. Patel (Eds.), Nuts and Seeds in health and disease prevention (pp. 741–748) Chapter 88* (San Diego: Academic Press)
- [11] Luo H, Cai Y, Peng Z, Liu T and Yang S 2014 *Chem. Cent. J.* **8** 1
- [12] Stankovic M S 2011 *Kragujevac J. Sci.* **33** 63
- [13] Harborne J B 1987 *Metode Fitokimia; Penuntun Cara Modern Menganalisa Tumbuhan, Terbitan Kedua, Terjemahan Kosasih Padmawinata dan Iwang Soediro* (Bandung: ITB)
- [14] Azizah D N, Kumolowati E and Faramayuda F 2014 *Kartika Jurnal Ilmiah Farmasi* **2** 45
- [15] Basset J, Denney R C, Jeffrey G H and Mendhom J 1994 *Buku Ajar Vogel Kimia Analisis Kuantitatif Anorganik* (Jakarta: EGC)
- [16] Chang C, Yang M and Chern J G H 2002 *J. Food Drug Anal* **10** 178

2018 IOP Masturi Optimization of condition extraction in quantification of total flavonoid content in the seeds of the Arummanis (*Mangifera indica* L.) mango from Indonesia

ORIGINALITY REPORT

17%

SIMILARITY INDEX

13%

INTERNET SOURCES

14%

PUBLICATIONS

9%

STUDENT PAPERS

PRIMARY SOURCES

1	publications.aston.ac.uk Internet Source	2%
2	www.neliti.com Internet Source	2%
3	vdocuments.site Internet Source	1%
4	Submitted to Sriwijaya University Student Paper	1%
5	china.iopscience.iop.org Internet Source	1%
6	Submitted to Universitas Diponegoro Student Paper	1%
7	Mital J. Kaneria, Sumitra V. Chanda. "The Effect of Sequential Fractionation Technique on the Various Efficacies of Pomegranate (<i>Punica granatum</i> L.)", Food Analytical Methods, 2012 Publication	1%

8	pubs.rsc.org Internet Source	1%
9	www.impactjournals.us Internet Source	1%
10	journals.plos.org Internet Source	1%
11	ifst.pericles-prod.literatumonline.com Internet Source	1%
12	Submitted to Lovely Professional University Student Paper	1%
13	link.springer.com Internet Source	1%
14	Ajila, C.M.. "Valuable components of raw and ripe peels from two Indian mango varieties", Food Chemistry, 2007 Publication	1%
15	www.pharmacognosy.in Internet Source	1%
16	www.ucsiuniversity.edu.my Internet Source	<1%
17	journal.unnes.ac.id Internet Source	<1%
18	Submitted to TAR University College Student Paper	<1%

19

Md. Jahurul Haque Akanda, Norazlina M.R., Azzatul F.S., Sharifudin Shaarani et al. "Hard Fats Improve the Physicochemical and Thermal Properties of Seed Fats for Applications in Confectionery Products", Food Reviews International, 2019

Publication

<1%

20

ei-ado.aciar.gov.au

Internet Source

<1%

21

Kang, Suk-Nam, Young-Min Goo, Mi-Ra Yang, Rashid Ibrahim, Jae-Hyeon Cho, Il-Suk Kim, and Ok-Hwan Lee. "Antioxidant and Antimicrobial Activities of Ethanol Extract from the Stem and Leaf of *Impatiens balsamina* L. (Balsaminaceae) at Different Harvest Times", Molecules, 2013.

Publication

<1%

22

Rajesh, K.P., H. Manjunatha, V. Krishna, and B.E. Kumara Swamy. "Potential in vitro antioxidant and protective effects of *Mesua ferrea* Linn. bark extracts on induced oxidative damage", Industrial Crops and Products, 2013.

Publication

<1%

23

Aktsar Roskiana Ahmad, Juwita Juwita, Siti Afrianty Daniya Ratulangi. "Penetapan Kadar Fenolik dan Flavonoid Total Ekstrak Metanol Buah dan Daun Patikala (*Etilingera elatior* (Jack)

<1%

R.M.SM)", Pharmaceutical Sciences and Research, 2015

Publication

24

Fariba Ebrahimi, Somayeh Rastegar.
"Preservation of mango fruit with guar-based edible coatings enriched with *Spirulina platensis* and Aloe vera extract during storage at ambient temperature", *Scientia Horticulturae*, 2020

<1%

Publication

Exclude quotes On

Exclude matches < 4 words

Exclude bibliography On