Optimization of drying temperature and water extraction time of Monascus-fermented durian seed for the Monacolin K content using Response Surface Methodology

by Ignatius Srianta

International Food Research Journal 21(1): 73-75 (2014)
Journal homepage: http://www.ifrj.upm.edu.my

Optimization of drying temperature and water extraction time of *Monascus*-fermented durian seed for the Monacolin K content using Response Surface Methodology

[1*]Srianta, I., [1]Nugerahani, I., [1]Sutedja, A. M. and [2]Widharna, R. M.

[1]*Department of Food Technology, Faculty of Agricultural Technology, Widya Mandala Surabaya Catholic University, Jalan Dinoyo 42-44 Surabaya 60625, Indonesia*
[2]*Faculty of Pharmacy, Widya Mandala Surabaya Catholic University, Jalan Dinoyo 42-44 Surabaya 60625, Indonesia*

Article history

Received: 1 August 2013
Received in revised form: 27 August 2013
Accepted: 28 August 2013

Keywords

Optimization
Monascus
Monacolin K
Drying
Extraction
Response surface methodology

Abstract

Optimization of the drying temperature and hot water extraction time of *Monascus*-fermented durian seed for the monacolin K content in functional drink for the reduction of blood cholesterol was investigated in this study. *Monascus*-fermented durian seed was produced by inoculating the spore suspension of *Monascus* sp. KJR2 into boiled durian seed cuts and then incubated at room temperature (30°C) for 14 days. The experimental design for optimization of drying temperature of the *Monascus*-fermented durian seed (MFDS) and extraction time was carried out using the central composite of Response Surface Methodology. The products were then extracted with distilled water (95°C) at various extraction time. The extracts were analyzed for the monacolin K content by using HPLC. The results showed that the optimum conditions of the product containing monacolin K were drying temperature of 35°C and extraction time of 1 minute.

Introduction

Many people in the world suffer from elevated plasma cholesterol (hypercholesterolemia), a condition occurring when the LDL-cholesterol content of the blood is above the normal level. Hypercholesterolemia is major cause of artherosclerosis, coronary heart disease, stroke and other diseases related to blood circulation (Endo, 1988; Lin *et al.*, 2008). *Monascus*-fermented products have been traditionally used in treatment to reduce blood cholesterol. Cholesterol is synthesized from acetyl-CoA, the precursor, through a series of enzyme reactions. In humans, most of the cholesterol in the body is produced in the liver. Hepatic cholesterol synthesis is regulated by the activity of the enzyme hydroxymethylglutaryl-CoA (HMG-CoA) reductase, which catalyzes the conversion of HMG-CoA to mevalonate. Many researchers reported that *Monascus* fungi produce monacolin K, a HMG-CoA reductase inhibitor (Endo, 1980; Li *et al.*, 1998; Seraman *et al.*, 2010; Venkateswaran and Vijayalakshmi, 2010). Our previous study showed that *Monascus*-fermented durian seed contains monacolin K, an antihypercholesterolemic agent (Srianta *et al.*, 2012).

Some researchers reported that monacolin K is heat sensitive (Ou *et al.*, 2009; Jirasatid *et al.*, 2013). As the monacolin K is sensitive to temperature, this study objective was to optimize the drying temperature and hot water extraction time in the production of the MFDS functional drink-containing Monacolin K. The optimum levels with highest monacolin K content were determined by response surface methodology (RSM).

Materials and Methods

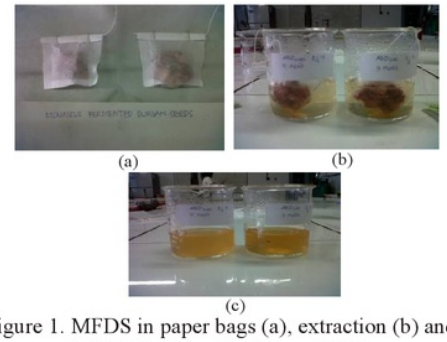
Culture

Monascus sp. KJR2 was obtained from the Center for Food and Nutrition Research, Widya Mandala Surabaya Catholic University. It was maintained on Saboraud's Dextrose Agar (SDA) slant, preserved at 4°C and subcultured monthly. After *Monascus* sp. KJR2 was grown on SDA slants at room temperature (30°C) under static conditions for 14 days, 10 mL of sterile distilled water was added and the spores were scraped under aseptic conditions. 0.1 mL of the spore suspension was inoculated into Saboraud's Dextrose Broth (SDB) and then was incubated at room temperature (30°C) for 10 days. It was used as starter culture to produce *Monascus*-fermented durian seed.

Production of MFDS and MFDS functional drink

Durian seeds were obtained from local durian seller. Durian seeds were stored in a freezer (-4°C) until used. Durian seeds were boiled in a $CaCO_3$ solution of 5% w/v for 10 min. After the seed coat

*Corresponding author.
Email: *srianta_wm@yahoo.com*
Tel: +6231 5678478 ext 110



(a) (b) (c)

Figure 1. MFDS in paper bags (a), extraction (b) and MFDS functional drink model (c)

were peeled off, the seeds were cut into small size of 1 cm x 1 cm x 1 cm. A 50 g of small cut durian seed was transferred into 300 mL flask, mixed thoroughly, autoclaved at 121°C for 15 min, then left to cool to room temperature, inoculated with the spore suspension of *Monascus* sp. KJR2 and incubated at room temperature (30°C) for 14 days in static conditions (with manual shaking daily). *Monascus*-fermented durian seed were dried in an oven at various temperatures i.e. 35, 40, 45, 50 and 55°C for 24 hours. The MFDS functional drink model is the hot water extract of MFDS (Figure 1). It was produced by extraction 2.5 g of the MFDS packed in paper bag with 25 mL of distilled water (95°C) for various extraction time i.e. 1, 3, 5, 7 and 9 minutes. The extracts were analyzed for the monacolin K content by using HPLC.

Determination of monacolin K content

Monacolin K content of the MFDS extracts were analyzed by using High Performance Liquid Chromatography (HPLC) according to Wang *et al.* (2006) with slight modification. The extracts were filtered through filter paper, followed by filtration through a 0.45 µm milipore filters and then the filtrate was analyzed by HPLC (model LC-20A Prominence, Shimadzu, Japan). Chromatographic separation was conducted on a Shim-Pack ODS C18 column (250 mm x 4.6 mm i.d.). Acetonitrile-phosphoric acid solution (pH 2.5), 65:35 v/v was used as the mobile phase. The solvents were filtered through 0.45 µm milipore filters prior to use.The eluent was pumped at a flow rate of 1.0 mL/min. Monacolin K was detected by SPD 20-A UV-Vis detector at 238 nm. Monacolin K from Sigma-Aldrich was used as the standard.

Experimental design and data analysis

Experimental design using Central Composite with 2 factors, 1 replication, total runs is 13 with cube points 4, center points in cube 5 and axial points 4, and alpha 1,41421. Table 1 shows the experimental design used in this study. The obtained data were analyzed by using Minitab 16 Software

Table 1. Experimental design of central composite with 2 factors

Run	A	B
1	0.00000	0.00000
2	0.00000	0.00000
3	0.00000	0.00000
4	-1.00000	1.00000
5	0.00000	-1.41421
6	-1.00000	-1.00000
7	0.00000	1.41421
8	0.00000	0.00000
9	1.41421	0.00000
10	0.00000	0.00000
11	1.00000	1.00000
12	-1.41421	0.00000
13	1.00000	-1.00000

Note: A: drying temperature (°C) and B: extraction time (min)

Results and Discussion

Figure 2 is the chromatograms of Monacolin K standard and a representative of the samples being analysed. Monacolin K retention time on those HPLC analytical conditions was 14.4 min. The Monacolin K content of 13 runs samples were shown on Table 2. Figure 3 show the result of the analysis of the data obtained. The optimum conditions for the extraction of the Monacolin K are drying temperature of 35°C and extraction time of 1 minute. The results reflect that the Monacolin K was stable at the lowest drying temperature and the shortest extraction time. Our observation concur with the results reported by other researchers who studied the effect of the temperature on the Monacolin K content, that the Monacolin K is heat sensitive. Li *et al.* (2005) reported that monacolin K were dehydrated during storage of red yeast rice powder at 60 and 80°C. Ou *et al.* (2009) also reported that monacolin K in red yeast rice solution was easily degraded under thermal treatment at 85 - 121°C. Heating at 121°C for 90 minutes reduce almost 50% of monacolin K. Under thermal treatment, monacolin K degrade to dehydromonacolin K. It does not only reduce therapeutic effects of monacolin K, but also leads to unnecessarily adsorption of degraded products into human body (Chairote *et al.*, 2008).

The air-exposed during drying process did in this study might also contribute to the monacolin K degradation. Recent study on kinetic degradation of monacolin K in red rice powder showed that vaccum and atmospheric conditions of red rice powder indicate different degradation mechanisms (Jarasatid *et al.*, 2013). They proposed that under the influence of atmospheric thermal condition, monacolin K acid form will degrade into oxidized product other than dehydromonacolin K acid form.

Further studies on *in vitro* and *in vivo* evaluation of the MFDS functional drink model obtained from

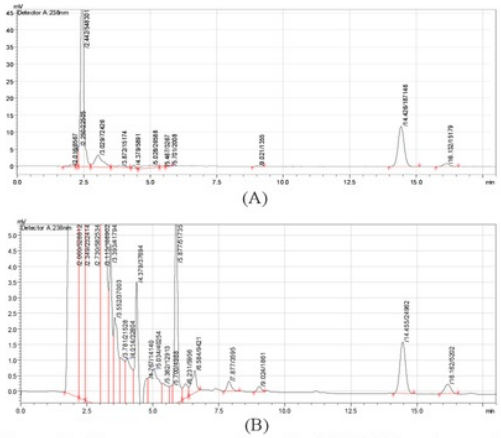


Figure 2. Chromatogram of standard MK (A) and sample (B)

Table 2. MK content of 13 runs samples

Run	Drying Temperature (°C)	Extraction Time (Min)	MK Content (mg/L)
1	45	5	0.0168
2	45	5	0.0168
3	45	5	0.0168
4	40	7	0.0000
5	45	1	0.2202
6	40	3	0.1042
7	45	9	0.0252
8	45	5	0.0168
9	55	5	0.2128
10	45	5	0.0168
11	50	7	0.0000
12	35	5	0.1594
13	50	3	0.0000

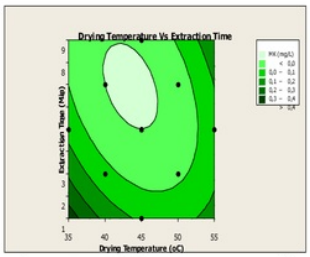


Figure 3. Graph contour of the response surface of drying temperature and extraction time

optimum conditions are needed.

Conclusion

The optimum process conditions for developing functional drink based on *Monascus*-fermented durian seed containing monacolin K were drying temperature of 35°C and water extraction time of 1 minute.

Acknowledgements

Thanks to Kopertis VII and Directorate General of Higher Education, Ministry of National Education, Republic of Indonesia for the financial support through competitive research Penelitian Hibah Bersaing with contract number of 005/SP2H/PDSTRL/K7/KL/II/2013.

References

Chairote, Em.-on., Chairot, G., Niamsup, H. and Lumyong, S. 2008. The presence and the content of monacolins in red yeast rice prepared Thai glutinous rice. World Journal of Microbiology and Biotechnology 24: 3039–3047.

Endo, A. 1980. Monacolin K, a new hypocholesterolemic agent that specifically inhibits 3-hydroxy-3-methylglutaryl coenzyme A reductase. Journal of Antibiotics (Tokyo) 33: 334-336.

Endo, A. 1988. Chemistry, Biochemistry and Pharmacology of HMG-CoA Reductase Inhibitors. Klin Wochenschr 66:421- 427.

Jirasatid, S., Nopharatana, M., Kitsubun P. and Tongta, A. 2013. Degradation kinetics of monacolin K in red yeast rice powder using multiresponse modeling approach. Journal of Food Engineering 116: 436–443.

Li, C., Zhu, Y., Wang, Y., Zhu, J.S., Chang, J. and Kritchevsky, D. 1998, *Monascus Purpureus*-fermented rice (red yeast rice): a natural food product that lowers blood cholesterol in animal models of hypercholesterolemia. Nutrition Research 18(1): 71-81.

Li, Y.G., Liu, H. and Wang, Z.T. 2005. A validated stability-indicating HPLC with photodiode array detector (PDA) method for the stress tests of *Monascus purpureus* fermented rice, red yeast rice. Journal of Pharmaceutical and Biomedical Analysis 39: 82–90.

Lin, Y.L., Wang, T.H., Lee, M.H. and Su, N.W. 2008. Biologically active components and nutraceuticals in the *Monascus*-fermented rice: a review. Applied Microbiology and Biotechnology 77:965–973.

Ou, H.P., Wang, C.C.R. and Lai, L.S. 2009. Thermal degradation kinetics analysis of monacolin K in *Monascus*-fermented products. LWT Food Science and Technology 42: 292–296.

Seraman, S., Rajendran, A. and Thangavelu, V. 2010. Statistical Optimization of anticholesterolemic drug lovastatin production by the red mold *Monascus purpureus*. Food and Bioproducts Processing 88: 266-276.

Srianta, I., Hendrawan, B., Kusumawati, N. and Blanc, P.J. 2012. Study on durian seed as a new substrate for angkak production. International Food Research Journal 19(3): 941-945.

Venkateswaran, V. and Vijayalakshmi, G. 2010. Finger millet (*Eleusine coracana*) – an economically viable source for antihypercholesterolemic metabolites production by *Monascus purpureus*. Journal of Food Science and Technology 47(4): 426-431.

Optimization of drying temperature and water extraction time of Monascus-fermented durian seed for the Monacolin K content using Response Surface Methodology

ORIGINALITY REPORT

%17	%6	%11	%6
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

PRIMARY SOURCES

1	Jirasatid, Sani, Montira Nopharatana, Panit Kitsubun, and Anan Tongta. "Degradation kinetics of monacolin K in red yeast rice powder using multiresponse modeling approach", Journal of Food Engineering, 2013. Publication	%4
2	A. Endo. "Chemistry, biochemistry, and pharmacology of HMG-CoA reductase inhibitors", Klinische Wochenschrift, 05/1988 Publication	%4
3	Submitted to Universiti Sains Malaysia Student Paper	%3
4	203.64.245.61 Internet Source	%2
5	Wang, Jyh-Jye, Chung-Lin Lee, and Tzu-Ming Pan. "Modified Mutation Method for Screening Low Citrinin-Producing Strains of *Monascus purpureus* on Rice Culture", Journal of	%1

	Agricultural and Food Chemistry, 2004. Publication	
6	Submitted to Jawaharlal Nehru Technological University Student Paper	%1
7	www.ftb.com.hr Internet Source	%1
8	www.science.gov Internet Source	%1
9	Submitted to Higher Education Commission Pakistan Student Paper	%1
10	dspace.cc.tut.fi Internet Source	%1
11	spakten.net Internet Source	%1

EXCLUDE QUOTES	ON	EXCLUDE MATCHES	< 10 WORDS
EXCLUDE BIBLIOGRAPHY	ON		