Orally administered pressure-blanched white saffron (Curcuma mangga Val.) improves antioxidative properties and lipid profiles in vivo

Dwiyati Pujimulyani a,*, Umar Santosob, Sri Luwihana D a, Ali Marufc

a Faculty of Agroindustry, University of Mercu Buana Yogyakarta, Yogyakarta, 55753, Indonesia
b Faculty of Agricultural Technology, Gadjah Mada University, Yogyakarta, 55281, Indonesia
c Key Laboratory for Biorheological Science and Technology of Ministry of Education, State and Local Joint Engineering Laboratory for Vascular Implants, Bioengineering College, Faculty of Medicine, Chongqing University, Chongqing, 400030, China

ARTICLE INFO

Keywords:
Food science
White saffron
Pressurized blanching
Total phenols
Antioxidant activities
Lipid profiles

ABSTRACT

This research focused on studying the effects of orally administered pressure-blanched white saffron on the antioxidative properties and lipid profiles of wistar rats. White saffron was blanched in autoclave for 2.5, 5, 7.5, and 10 min at 100, 105, 110, 115, and 120 °C, which are equivalent to 14.17, 17.53, 20.79, 24.54, and 28.81 psia, respectively. A total of 30 male wistar rats aged four weeks were fed with a standard diet (N), oxidized peanut oil diet + unblanched white saffron (A), oxidized peanut oil diet + blanched white saffron (B), oxidized peanut oil diet + pressure-blanched white saffron (C), and oxidized peanut oil diet + aquadest (NC), for two weeks after pre-treatment with the standard diet for a week. In vivo study showed treatment with pressure-blanched white saffron could significantly improve SOD, Vitamin E, and HDL levels compared to the negative control (NC); 686.44 U/g Hb, 10.87 µg/mL, and 94.17 mg/dL versus 405.37 U/g Hb, 7.44 µg/mL, and 43.47 mg/dL, respectively. Meanwhile, treatment with pressure-blanched white saffron could significantly reduce MDA, total cholesterol, LDL, and triglyceride levels in the blood compared to the negative control (NC); 1.98 mmol/L, 108.74 mg/dL, 40.99 mg/dL, and 172.61 mg/dL, respectively. The results showed that pressurized blanching could significantly increase antioxidant levels of white saffron, and its dried form could improve antioxidative properties and lipid profiles in vivo.

1. Introduction

Antioxidants are usually obtained from natural resources and synthetic materials. Those from synthetic materials are more effective, but less safe health-wise. Blanching as preparatory heating is conducted to deactivate enzymes that play a role in polyphenol degradation and to increase the antioxidant levels of agricultural products. At 100 °C, pressurized blanching on wheat could increase total wheat flour phenols (Cheng et al., 2006). Corn blanched in autoclave showed an increased total phenol level (Randhir et al., 2008). On black beans, pressurized blanching showed higher ORAC (Oxygen Radical Absorbance Capacity) value than unpressurized (Xu and Chang, 2008b). Likewise, steaming of lentils at 15 psi for 15 min could significantly improve the ORAC value due to Maillard reaction (Xu and Chang, 2008a).

Furthermore, pressurized tea extract, heated fruits, and heated vegetables showed an increased antioxidant activity (Manzocco et al., 2000; Nicoli et al., 1999). In addition, heated bilberry extract had higher antioxidant activity than its unheated form due to the degradation of glycosides to aglycons and sugars (Yue and Xu, 2008). According to Sadilova et al. (2006), anthocyanins (glycoside compounds) could hydrolyze into anthocyanidins under acidic conditions.

White saffron as a major source of antioxidants is very easy to cultivate and do not require special treatment. Interestingly, the rhizomes can be processed into different kinds of food products, such as instant powder, syrup, and dry and dried sweets. Pujimulyani et al. (2013) have studied the effects of blanching on the antioxidative activity and phenolic contents of white saffron. Blanching in 0.05% citric acid media at boiling temperature for 5 min could escalate total phenolics of white saffron from 1.01 ± 0.04 to 1.33 ± 0.15 mg/g dried extract, with a significant rise in catechin, epigallocatechin, and epigallocatechingallat (EGCG) levels. In addition, the epicatechin and gallocatechingallat levels were similar to those of the unblanched white saffron, while the gallic acid level of the blanched white saffron was slightly lower than that of the unblanched.

* Corresponding author.
E-mail address: dwiyati2002@yahoo.com (D. Pujimulyani).

https://doi.org/10.1016/j.heliyon.2020.e04219
Received 24 November 2019; Received in revised form 19 March 2020; Accepted 11 June 2020
2405-8440/© 2020 The Author(s). Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).
This research was focused on examining the effects of pressure-blanched white saffron on the antioxidative properties and lipid profiles of oxidized peanut oil-treated wistar rats. Initially, blanched white saffron with appropriate levels of total phenols, EGCG, 2,2,1-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging, and ferric reducing antioxidant power (FRAP) was chosen for in vivo research. Theoretically, it was assumed that administration of pressure-blanched white saffron to the oxidized peanut oil-treated wistar rats could improve their superoxide dismutase (SOD), Vitamin E, and high-density lipoprotein (HDL) levels, while reducing the malondialdehyde (MDA), total cholesterol, low-density lipoprotein (LDL), and triglyceride levels.

2. Materials and methods

2.1. Materials

The plant, white saffron (Curcuma mangga Val.), was freshly harvested from Sedayu, Bantul, in Yogyakarta. The reagents were ethanol, methanol, HCl, acetate buffer, FeCl3·6H2O, Na2CO3, Na3NO2, AlCl3·6H2O, NaOH, acetone, acetic acid, vanillin, H3PO4, N2 gas, CH3CN, ethyl acetate (E Merck), aquabidestilata (Ika Pharmindo), EDTA, ketamine, and distilled water (aquadest).

Some instruments, such as autoclave, vacuum rotary evaporator (Heidolph VV, 2000), UV-Vis spectrophotometer (Genesys-20), incubator, vacuum filter, centrifuge, 0.45-μm-millex filter, microfator, sartorius scale, homogenizer, blender, High-Performance Liquid Chromatography (HPLC) Knauer with C18 column, Photodiode Array Detector (DAD) UV 6000LP, Smartline pump, and ChromGate 3.1.6 software, were used in this study.

2.2. Preparation of pressure-blanched white saffron and animal study

White saffron rhizomes were washed, peeled, and blanched in autoclave at different temperatures (100, 105, 110, 115, and 120 °C; equivalent to 14.71, 17.53, 20.79, 24.54, and 28.81 psia, respectively) for 2.5, 5, 7.5, and 10 min. Then, it was extracted, evaporated, and freeze-dried to get the dried extract (Figure 1).

The dried white saffron powder with the highest total phenols and EGCG and the appropriate DPPH and FRAP values was chosen to feed wistar rats. Figure 2 showed the schematic diagram of the research design.

A total of 30 male wistar rats, aged four weeks, were fed with the American Institute of Nutrition (AIN) standard diet (N), oxidized peanut oil diet + unblanched white saffron (A), oxidized peanut oil diet + blanched white saffron (B), oxidized peanut oil diet + pressure-blanched white saffron (C), and oxidized peanut oil diet + aquadest (NC) for two weeks after pre-treatment with the standard diet for a week. The composition of the oxidized peanut oil diet has been adapted to the AIN standard (Reeves et al., 1993) (Table 1).

The animal experiments were conducted in UGM Center for Food and Nutrition Studies (PSPG), Gadjah Mada University, Indonesia, from March to October 2012, in compliance with relevant laws and approved by the ethical committee led by Prof. Dr. Ir. Endang Sutriswati Rahayu, M.S. The experimental animals were separately kept in a cage at room temperature, the wistar rats were anesthetized with ketamine (60 mg/kg) and their blood was drawn through the orbital sinus, given ethylendiaminetetraacetic acid (EDTA) anticoagulant, and the liver, kidney, and testes were harvested and weighed accordingly. To analyze the antioxidative properties and lipid profiles, blood samples were taken before and after treatments with the extract (Figure 3).
concentrations of gallic acid ranging from 31.875 to 510 mg/L with \( R = 0.99 \). Total phenols were determined as mg gallic acid equivalent (GAE) per gram of dried extract.

2.5. Determination of EGCG level

The quantification of EGCG (mg/g dried extract) was performed using HPLC (Monagas et al., 2007). Briefly, 500 \( \mu \)L of extract solution (100 mg dried extract dissolved in 3 mL methanol:HCl with a ratio of 1000:1) was evaporated using \( N_2 \) followed with the addition of 1 mL of \( \text{H}_3\text{PO}_4 \), filtered using millex filter 0.45 \( \mu \)m, and then injected to the HPLC with C18 column (4.6 \( \times \) 250 mm, dp 5 \( \mu \)m). Quercetin-3-rutinoside was used as a standard (\( \lambda \text{ of 256 nm, } 50 ^\circ \text{C} \)), and a solution of \( \text{H}_3\text{PO}_4:\text{CH}_3\text{CN:acetic ethyl} \) with a ratio of 84:12:4 was used as the eluent.

2.6. Determination of FRAP value

Antioxidant’s capability to reduce \( \text{Fe}^{3+} \) was determined using FRAP method (Benzie and Strain, 1996). Initially, the FRAP reagent was prepared by gently adding 300 mM acetic buffer to as prepared FRAP reagent. It was then swirled for 1 min and left for 4 min. The \( \lambda \text{ of 593 nm} \) was used to measure the absorbance. Thereafter, FRAP value was calculated in mg ferro equivalent/g dried extract using a calibration curve of \( \text{Fe}^{2+} \) (from 4.3 to 137.5 mg ferro/L, \( R = 0.99 \)).

2.7. Determination of SOD level

Initially, blood was drawn from the rat’s orbital sinus, given EDTA anticoagulant and through centrifugation at 3,000 rpm for 10 min, the red blood cells were isolated from the plasma. Thereafter, 50 \( \mu \)L of hemolysate with a ratio of 1:200 (v/v) of red blood cells and distilled water (equivalent to about 75 \( \mu \)g \( \text{Hb} \)) was prepared to measure the SOD level in erythrocytes by the RANSOD Kit (Randox Laboratories, Ltd., Crumlin, UK), following the manufacturer instructions. The SOD level was expressed as unit per gram of hemoglobin (U/g \( \text{Hb} \)), in which one unit was defined as the amount of protein that inhibits the rate of 2-para (iodophenyl)-3 (nitrophenyl)-5(phenyl) tetrazolium chloride (INT) reduction by 50%.

2.8. Determination of vitamin E level

Blood plasma (20 \( \mu \)L) was mixed with 100 \( \mu \)L of extract solution (5 mg of BHT/mL in ethanol:butanol, 50:50 v/v). The mixture was centrifuged at 12,000 rpm for 5 min. The supernatant (20 \( \mu \)L) was injected to the HPLC (flowrate of 1 mL/min) with C18 column, and detected using UV-Vis at \( \lambda \text{ of 292 nm} \) with the retention time of 3–6 min.

2.9. Determination of MDA level

MDA activity as an indicator of free radicals (highly reactive molecules) generation or lipid peroxidation was determined by quantifying the concentration of thiobarbituric acid (TBA) reactive substances (TBARS). Briefly, 750 \( \mu \)L of phosphoric acid was pipetted into a 13-ML polypropylene tube and 50 \( \mu \)L of plasma was then added. The solution was gently mixed and 250 \( \mu \)L of 40 mM TBA solution was subsequently added. Finally, 450 \( \mu \)L of distilled water was added and covered tightly, and was heated for 1 h. After cooling into an ice bath, the solution was subsequently mixed gently and applied to Sep-Pak C18 column (pre-washing was conducted with 5 mL of methanol and double distilled water (dd \( \text{H}_2\text{O} \)). The TBARS was eluted from the column with 4 mL of methanol. The absorbance was measured spectrophotometrically at 532 nm using tetaethoxypropane (TEP) as the external standard and the level of lipid peroxides was indicated as nmol of MDA.

2.10. Determination of lipid profiles

Total cholesterol, LDL, HDL, and triglycerides were measured from blood plasma using DiaSys Diagnostic Systems GmbH kits (Germany), with an enzymatic photometric method (Cholesterol FS, “CHOD-PAP”; LDL Precipitant, “CHOD-PAP”; HDL Precipitant, “CHOD-PAP”), and a colorimetric enzymatic method (Triglyceride FS, “glycerol-3-phosphate-oxidase (GPO)”). To measure the cholesterol level, 1 mL of the enzymatic triglyceride reagent was pipetted into test tubes and added with 10 \( \mu \)L of plasma or glycerol standard, and mixed gently. The mixtures were then incubated at 37 °C for 15 min, and the absorbance was detected at 500 nm within 60 min, against the reagent blank in which distilled water was substituted for the sample. The absorbance of the sample was divided by the standard and multiplied by its concentration to obtain total cholesterol in the blood (similar calculation for HDL, LDL, and triglycerides).

To measure the HDL level, 200 \( \mu \)L of plasma or the standard was added and mixed gently with 500 \( \mu \)L of precipitation reagent, prepared from phosphotungstic acid and magnesium chloride. Thereafter, it was incubated for 15 min at room temperature and then centrifuged at 2500 g
for 20 min. Within 2 h after the centrifugation, 100 μL of the clear supernatant or the standard was mixed with 1 mL of cholesterol reagent. The mixtures were then incubated at 37 °C for another 5 min, and the absorbance was detected at 500 nm within 45 min, against the reagent blank.

A similar procedure with HDL level measurement was used to measure the LDL level. Briefly, 100 μL of serum or the standard was added with 1 mL of precipitation reagent (prepared from heparin and sodium citrate). The mixtures were then incubated at room temperature for 15 min and then centrifuged at 2500 g for 20 min. Within 1 h after the centrifugation, 100 μL of the clear supernatant or the standard was mixed with 1 mL of cholesterol reagent. The mixtures were then incubated at 37 °C for another 5 min, and the absorbance was detected at 500 nm within 45 min, against the reagent blank.

Finally, to measure the triglyceride level, 10 μL of plasma or the standard was added with 1 mL of as prepared reagent containing Good's buffer, 4-Chlorophenol, ATP, Mg²⁺, glycerol kinase, peroxidase, lipoprotein lipase, 4-Aminoantipyrine, and GPO. Thereafter, the mixtures were then incubated at 37 °C for 10 min, and the absorbance was detected at 500 nm within 60 min, against the reagent blank in which distilled water was substituted for the sample. Total cholesterol, LDL, HDL, and triglycerides were expressed in mg/dL.

2.11. Statistical analysis

Statistical analysis was conducted using SPSS software (version 16). The data were analyzed by Randomized Complete Block Design (RCBD). Furthermore, Duncan's multiple range test was involved to determine a significant difference between the sample means (p ≤ 0.05).

3. Results and discussions

3.1. In vitro study of pressure-blanching white saffron

In an in vitro study conducted, white saffron was blanched at different temperatures and time points to select the optimum condition of blanching, as seen in Figure 4.A-D. In general, blanching in autoclave at 120 °C for 7.5 min showed the optimum condition for increasing total phenols, DPPH value, and EGCG level of white saffron, thereby being selected for the pressure-blanching treatment (C). However, the FRAP value was much lower than blanching in lower temperatures at the same time (7.5 min). It was notable that the total phenols of white saffron after pressurized blanching obviously increased compared to that of fresh ones (Figure 4B). The increase might be due to the degradation of glycosides into aglycons and complex phenolic compounds into simple substances. The result was similar to the study by Nickel et al. (2016) and Geetha et al. (2018) that examined pressurized blanching in Chenopodium quinoa.
Grains and tomatoes, respectively. In addition, Aisyah et al. (2014) examined the antioxidant activity of blanched vegetables, which could increase the yield of extraction due to the easy separation of antioxidants from the cell matrices.

Similarly, the DPPH value of white saffron increased noticeably compared to that of fresh ones (Figure 4A). A minimum temperature of 110 °C was required to increase free radical scavenging ability (RSA) of white saffron. The increased antioxidant activity was suspected due to the release of phenolic compounds. Furthermore, the EGCG level of pressure-blanched white saffron rose significantly compared to that of fresh ones (Figure 4C). This catechin-based compound was very important due to its potential role in managing oxidative stress, as proven by the high FRAP value of white saffron during blanching under pressure (Figure 4D).

A correlation analysis was conducted based on the data from Figure 4A-D. The results showed that at blanching temperature of 120 °C, total phenols and DPPH value would rise as the increased time of blanching (R = 0.93). Meanwhile, at 10 min, they would rise as the increased temperature of blanching (R = 0.87) (Table 2). In addition, at 120 °C, total EGCG level and FRAP value would increase as the increased time of blanching (R = 0.82), while there was no significant correlation between the two variables at blanching time of 10 min with the increased temperature of blanching (R = 0.42) (Table 3).

However, regardless of the beneficial effects of pressurized blanching, the duration should be controlled up to the optimal time, because long exposure showed a significant reduction in DPPH, total phenols, EGCG, and FRAP value of white saffron after reaching their peaks (Figure 4A-D).

### 3.2. In vivo study of antioxidative properties

After two weeks of treatment, SOD, Vitamin E, and MDA levels of wistar rats were evaluated for their antioxidative properties, shown in Figure 5. In vivo study of antioxidative properties. A) SOD, B) Vitamin E, and C) MDA levels of rats treated with the standard diet for a week followed with a two-week treatment of the standard diet (1), oxidized peanut oil diet + unblanched white saffron (2), oxidized peanut oil diet + blanched white saffron (3), oxidized peanut oil diet + pressure-blanched white saffron (4), and oxidized peanut oil diet + aquadest (5). Data are presented as mean ± SD. Different notations indicate a significant difference, p ≤ 0.05.
Figure 5A-C. SOD is one of the biological antioxidants in humans or animals, and its high level in blood indicates the body is in healthy condition. In vivo study revealed that the SOD level of wistar rats fed with pressure-blanched white saffron was much higher than that of the negative control (NC), and slightly higher than that of unblanched (A) and blanched (B) (Figure 5A). The result was in accordance with Ahsan (2013), that studied the effect of pressurized blanching on the inhibition of peroxide formation. In another study, water blanching could also increase the antioxidant activity of white saffron as measured by 2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) method, with IC₅₀ value of 24.23 ± 2.77 µg/mL (Pujimulyani et al., 2018). The presence of antioxidants increases the activity of SOD enzymes to suppress the generation of reactive oxygen species (ROS) (Palido-Moran et al., 2016).

Vitamin E is an essential antioxidant required by the human body. Figure 5B showed Vitamin E level of pressure-blanched white saffron (C) was much higher than that of blanched (B), unblanched (A), and the negative control (NC), respectively. Its increase in the blood was related to the high amount of curcumin after blanching. According to Rai et al. (2010), curcumin could induce its production and prevent deoxyribonucleic acid (DNA) damage by reducing oxidative stress.

Interestingly, the MDA level of pressure-blanched white saffron (C) was similar to that of the positive control (N) and much lower than that of the negative control (NC), indicating the swelling of livers and kidneys due to oxidation. Meanwhile, the testicles' weight of the negative control (NC) was much smaller than those of pressure-blanched white saffron (C) and the positive control (N), indicating the swelling of livers and kidneys due to oxidation. In addition, the uric acid level of pressure-blanched white saffron (C) was examined before and after treatment, and it was twice lower than that of the negative control (NC) (Figure 7).

3.3. In vivo study of lipid profiles

Blood lipid profiles of oxidized peanut oil-treated wistar rats, including total cholesterol, LDL, HDL, and triglyceride, were evaluated after treatment as shown in Figure 6A-D. Pressure-blanched treatment (C) effectively suppressed the cholesterol levels as compared to the negative control (NC) (more than twice) (Figure 6A). According to Pujimulyani et al. (2010), water blanched white saffron showed higher antioxidant activity than unblanched, as measured by DPPH and FRAP assay. The bioactive compounds in pressure-blanched white saffron might reduce oxidation-induced cholesterol accumulation.

It is believed that a high level of LDL in the blood can cause harm to the body. However, the LDL level of wistar rats fed with pressure-blanched white saffron (C) was thrice lower than that of the negative control (NC) (Figure 6B). This might be due to the presence of curcumin (Pujimulyani and Sutardi, 2003), polyphenols (Pujimulyani et al., 2010), and dietary fibers (Rezki, 2017) in white saffron. Curcumin-rich diet could reduce the LDL level in the blood (Su et al., 2017). Furthermore, a study by Gani et al. (2013) showed that polyphenol compounds and dietary fibers in red gedi could also reduce the LDL level in the blood.

Conversely, the HDL level of wistar rats fed with pressure-blanched white saffron (C) was twofold higher than that of the negative control (NC) (Figure 6C). This might be due to the presence of curcumin in white saffron (Ganaji et al., 2017). Regarding its effect on HDL function, the results of Syaefudin et al. (2016) showed that feeding Peking broiler ducks with black saffron flour could increase their HDL level. The active substances of essential oils and curcumin could help improve intestinal peristalsis (Rositawati et al., 2010) and facilitate loss of bile salts in the duodenum, which subsequently triggers livers to produce more bile salts. Bile salt's production requires cholesterol, therefore when it is not sufficient, the production of HDL will increase to help the cholesterol transport from tissues to the liver (Hartoyo et al., 2005). In addition, the presence of quercetin in white saffron (Pujimulyani et al., 2012) could increase the HDL level (Made Harumi et al., 2015). Furthermore, this antioxidant could further inhibit atherosclerosis progression through the improvement of blood transport due to reduction of cholesterol levels (Fernandes-Silva et al., 2012).

On the other hand, the triglyceride level of wistar rats fed with pressure-blanched white saffron (C) was twice lower than that of the negative control (NC) (Figure 6D). Again, this might be due to the presence of curcumin. According to Su et al. (2017) and Mohammadi et al. (2013), curcumin-rich diet could effectively reduce the triglyceride level as studied in vivo. Furthermore, the uric acid level of pressure-blanched white saffron (C) was examined before and after treatment, and it was twice lower than that of the negative control (NC) (Figure 7).

Surprisingly, livers' and kidneys' weight of the negative control (NC) were much higher than those of pressure-blanched white saffron (C) and the positive control (N), indicating the swelling of livers and kidneys due to oxidation. Meanwhile, the testicles' weight of the negative control (NC) was much smaller than those of pressure-blanched (C) and the

Figure 6. In vivo study of lipid profiles. A) Total cholesterol, B) LDL, C) HDL, and D) triglyceride levels of wistar rats treated with the standard diet for a week followed with a two-week treatment of the standard diet (1), oxidized peanut oil diet + unblanched white saffron (2), oxidized peanut oil diet + blanched white saffron (3), oxidized peanut oil diet + pressure-blanched white saffron (4), and oxidized peanut oil diet + aquadest (5). Data are presented as mean ± SD. Different notations indicate a significant difference, p ≤ 0.05.
positive control (N). This might be due to the oxidation and cell damages, leading to the shrinking of testicles' weight (Figure 8).

Results of the in vivo study showed antioxidants induced the improvement of lipid profiles of oxidized peanut oil-treated wistar rats. This might be correlated to the antioxidant metabolic pathways of Vitamin E and SOD. Primarily, Vitamin E metabolism is initiated with a CYP4F2/CYP3A4-dependent ω-hydroxylation cycle followed by five cycles of subsequent β-oxidation, forming water-soluble metabolites, including long-chain metabolites (e.g., carboxymethyldeciylhydroxychromanol (CDMDHC, 11’-COOH) and carboxymyloctylhydroxychromanol (CDMOHC, 9’-COOH)); intermediate-chain metabolites (e.g., carboxymethylhexylhydroxychromanol (CDHHC, 7’-COOH) and carboxymethylbutylhydroxychromanol (CMBHC, 5’-COOH)); and short-chain metabolites (e.g., carboxyethylhydroxychromanol (CEHC, 3’-COOH)), excreted through urine or feces (Schmölz et al., 2016). In blood circulation, Vitamin E is transported by the plasma lipoproteins and erythrocytes. In erythrocytes, it can be mediated by either passive diffusion or receptor-mediated transport. Interestingly, it has been studied in vivo for its role in up-regulating the ATP-binding cassette transporters ABCG5/ABCG8, and ABC1, which are responsible for maintaining cholesterol influx and efflux for balancing the cholesterol level in the body (Rogi et al., 2011). Furthermore, a clinical study showed the effects of vitamin E as a natural antioxidant combined with fish oil in improving lipid profiles by reducing total cholesterol and LDL levels and reducing anti-oxidized LDL autoantibodies by protecting LDL against oxidation to prevent cardiovascular-related diseases, especially atherosclerosis (Alves Luzia et al., 2015; Maruf et al., 2019). Another recent clinical study showed that vitamin E supplementation led to a significant reduction in mortality of 29,092 patients with cardiovascular, heart, stroke, cancer, and respiratory diseases, with a risk reduction of 30% (Huang et al., 2019).

On the other hand, SOD is an important enzyme in the body that can catalyze the dismutation of superoxide radical into oxygen and hydrogen peroxide, leading to the modulation of oxidative stress. Mondola et al. (2002) studied the effects of different types of SODs on cholesterol metabolism. The result showed that all forms of SODs affected cholesterol metabolism independently by decreasing microsomal enzyme 3-hydroxy-3-methylglutaryl CoA (HMG-CoA) reductase activity and its protein levels, leading to protein kinase C-mediated reduction of cholesterol synthesis. Additionally, a clinical study showed that an imbalance in SOD and thioredoxin reductase (TrxR-1) activities was significantly correlated to LDL oxidation, a marker of oxidative stress (Augusti et al., 2012). Therefore, SOD plays a pivotal role in maintaining low levels of oxygen metabolites in tissues and in defense against oxidative stress.

4. Conclusions

In conclusion, white saffron blanched under pressure showed the ability to improve SOD, Vitamin E, and HDL levels, while reducing MDA, total cholesterol, LDL, and triglyceride levels in the blood. Initially, the optimal blanching condition was at 120 °C for 7.5 min, which was equivalent to the pressure of 28.81 psia, giving the highest total phenols, EGCG, and DPPH of 47.35 mg GAE/g dried extract, 0.3317 mg/g dried extract, and 12.04 mg trolox/g dried extract, respectively. In vivo study showed that treatment with the pressure-blanching white saffron could significantly improve SOD, Vitamin E, and HDL levels in the blood compared to the negative control (NC); 686.44 U/g Hb, 10.87 µg/mL, and 94.17 µg/dL versus 405.37 U/g Hb, 7.44 µg/mL, and 43.47 mg/dL, respectively. Meanwhile, it could significantly reduce MDA, total cholesterol, LDL, and triglyceride levels compared to the negative control (NC); 1.98 mmol/L, 108.74 mg/dL, 40.99 mg/dL, and 78.06 mg/dL versus 8.54 mmol/L, 232.46 mg/dL, 149.17 mg/dL, and 172.61 mg/dL, respectively. The results showed that pressurized blanching could significantly increase the antioxidant levels of white saffron and its dried form could improve antioxidative properties and lipid profiles in vivo.

Declarations

Author contribution statement

D. Pujimulyani: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

U. Santoso: Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

S. Luwihana D.: Performed the experiments; Contributed reagents, materials, analysis tools or data.

A. Maruf: Analyzed and interpreted the data; Wrote the paper.

Funding statement

This work was supported by the Directorate General of Higher Education, the Ministry of Research, Technology and Higher Education of the Republic of Indonesia (Hibah Fundamental [0541/023-04.1/00/2012]).
Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

References

Ahlan, M.U., 2013. Pengaruh Waktu Blanching Bertekanan pada Suhu 120 °C terhadap Kemampuan Penghambatan Pembentukan Peroksidasi Kunir Putih (Curcuma mangga Val.). Faculty of Agroindustry, University of Mercu Buana Yogyakarta. Thesis.

Aiyah, Y., Radiansyah, R., Muhamin, M., 2014. Pengaruh pemanasan terhadap aktivitas antioksidan pada beberapa jenis sayuran. J. Teknol. Dan. Ind. Pertan. Indonesia. 6, 2.

Alves Luzia, L., Mendes Aldrighi, J., Teixeira Damasceno, N.R., Rodrigues Sampaio, G., Aparecida Manoilo Soares, R., Tande Silva, I., et al., 2015. Fish oil and vitamin e change lipid profiles and anti-LDL-antibodies in two different ethnic groups of women transitioning through menopause. Nutr. Hosp. 32, 165-174.

Augusti, P.R., Ruviaro, A.R., Quatrín, A., Somacal, S., Conterato, G.M., Vicentini, J.T., et al., 2012. Imbalance in superoxide dismutase/thiorodoxin reductase activities in hypercholesterolemic subjects: relationship with low density lipoprotein oxidation. Lipids Health Dis. 11, 79.

Benzie, I.F.F., Strain, J.J., 1996. The ferric reducing ability of plasma (FRAP) as a measure of 'antioxidant power' the FRAP assay. Anal. Biochem. 239, 70-76.

Cheng, Z., Su, L., Moore, J., Zhou, K., Luther, M., Yin, J.J., et al., 2006. Effects of postharvest treatment and heat stress on availability of wheat antiwheat. J. Agric. Food Chem. 54, 5625-5629.

Fernandes-Silva, M.M., Carvalho, V.O., Guimaraes, G.V., Bocal, F., Bocchi, E.A., 2012. Physical exercise and microRNAs: new frontiers in heart failure. Arq. Bras. Cardiol. 98, 459–466.

Gani, N., Momuat, L.I., Pitoi, M.M., 2013. Prospek pengembangan kunir putih (Curcuma xanthoriza Will.) sebagai sumber nutrisi. J. Food Technol. Industry 29, 8-17.

Ganjali, S., Blesso, C.N., Banach, M., Pirro, M., Majeed, M., Sahebkar, A., 2017. Effects of curcumin and its synthetic analogs on inflammatory cytokines and coagulation factors in patients with nonalcoholic fatty liver disease. J. Clin. Lipidol. 11, 1031-1052.

Gani, N., Momuat, L.I., Pitoi, M.M., 2013. Prophylaxis lipida plasma tiuk wistar yang pertoleransi lemak jenuh pada piberbenetan gedi merah (Abduhsamus manihot L.). J. MIPA UUrsat. Online 2, 44-49.

Ganjali, S., Blesso, C.N., Banach, M., Pirro, M., Majeed, M., Sahebkar, A., 2017. Effects of curcumin on HDL functionality. Pharmacol. Res. 119, 208–218.

Geetha, K., Hulamani, S., Shivaleela, H.B., 2018. Effect of cooking on total antioxidant activity, polyphenols and flavonoid content in commonly consumed vegetables. Int. J. Curr. Microbiol. Appl. Sci. 7, 1459-1466.

Hartoyo, B., Raharjo, S., Marsono, Y., Santoso, U., 2013. The phenolic substances and antioxidant activities of white turmeric (Curcuma mangga Val.). J. Food Sci. 73, C491–C499.

Hartoyo, B., Raharjo, S., Marsono, Y., Santoso, U., 2012. Effect of blanching on antioxidant activity and glycosides of white saffron (Curcuma mangga Val.). Int. Food Res. J. 19, 617-621. https://ijrif.upm.edu.my/19%20(02)%202012%20%20/IJRFU%202012% 20%20wiyani.pdf.

Nickel, J., Spanier, L.P., Botelho, F.T., Gultepe, M.A., Helbig, E., 2016. Effect of different types of processing on the total phenolic compound content, antioxidant capacity, and amination content of Campophium quinoa Wild grains. Food Chem. 209, 139-143.

Nicolli, M.C., Anese, M., Parpinel, M., 1999. Influence of processing on the antioxidant properties of fruit and vegetables. Trends Food Sci. Technol. 10, 94-100.

Palido-Moran, M., Moreno-Fernandez, J., Ramirez-Tortosa, C., Ramirez-Tortosa, M., 2016. Curcumin and health. Molecules 21, 264.

Pujimulyani, D., Sutardi, 2003. Curcuminoïd content and antioxidative properties on white saffron extract (Curcuma mangga Val.). In: Proceeding International Conference on Redesigning Sustainable Development on Food and Agricultural System for Developing Countries.

Pujimulyani, D., Raharjo, S., Marsono, Y., Santoso, U., 2010. Aktivitas antioksidan dan kadar senyawa fenolik pada kunir putih (Curcuma mangga Val.) sejar dan setelah blanching. Agri. 30, 2.

Pujimulyani, D., Raharjo, S., Marsono, Y., Santoso, U., 2012. The effect of blanching on antioxidant activity and glycosides of white saffron (Curcuma mangga Val.) as affected by blanching methods. World Acad. Sci. Eng. Technol. 7, 947–950.

Pujimulyani, D., Yulianto, W.A., Setiyowati, A., Arumawardana, S., Amalia, A., Kusuma, H.S.W., et al., 2018. Amylase inhibition and free radical scavenging activities of white turmeric extract and fractions. J. Food Technol. Industry 29, 10-18.

Rai, B., Kaur, J., Jacobs, R., Singh, J., 2010. Possible action mechanism for curcumin in pre-cancerous lesions based on serum and salivary markers of oxidative stress. Int. J. Oral Sci. 52, 251–256.

Randhir, R., Kwon, Y., Shetty, K., 2008. Effect of thermal processing on phenolics, antioxidant activity and health-relevant functionality of select grain sprouts and seedlings. Innovat. Food Sci. Emerg. Technol. 9, 355–364.

Reeves, P.G., Nielsen, F.H., Fahey Jr., G.C., 1993. AIN-93 purified diet for laboratory rodents: final report of the American Institute of Nutrition ad hoc writing committee on the reformulation of the AIN-76A rodent diet. J. Nutr. 123, 1959–1951.

Rezki, R., 2017. Pengaruh Peningkatan Bahan Segar terhadap Aktivitas Antioxidan IC50 dan Kadar Serat Kasar Kunir Putih (Curcuma mangga Val.). Faculty of Agroindustry, University of Mercu Buana Yogyakarta. Thesis. https://eprints.mercubuana-yogyakarta.ac.id/id/eprint/1486.

Syaefudin, A.A., Murwani, R., Isroli, I., 2010. Upaya peningkatan performa itik Mojosari periode starter melalui penambahan temulawak (Curcuma xanthoriza Val.) dalam ransum memperbaiki produktivitas itik Mojosari. J. Biologi. 16, 34-38.

Sofoh, S., Ebing, C., Barke, O., 2006. Thermal degradation of acetylated and nonacetylated anthocyanins. J. Food Sci. 71, C504–C512.

Schmölzer, L., Birringer, M., Lorkovská, S., Wallert, M., 2016. Complexity of vitamin E metabolism. World J. Biol. Chem. 7, 14–43.

Singleton, V.L., Orthofer, R., Lamuela-Raventos, R.M., 1999. [14] Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-cioicelau reagent. Methods Enzymol. 299, 152–178.

Su, L.Q., Wang, Y.D., Chi, H.Y., 2017. Effect of curcumin on glucose and lipid metabolism, antioxidative activities of white turmeric extract and fractions. J. Food Technol. Industry 29, 408–416.

Fayeh Jr., G.C., 1993. AIN-93 purified diets for laboratory rodents: final report of the American Institute of Nutrition ad hoc writing committee on the reformulation of the AIN-76A rodent diet. J. Nutr. 123, 1959–1951.

Rostiwati, I., Sifinat, N., Muhariyan, 2010. Upaya peningkatan performa itik Mojosari periode starter melalui penambahan temulawak (Curcuma xanthoriza Val.). In: Proceeding International Conference on Redesigning Sustainable Development on Food and Agricultural System for Developing Countries.

Mondola, P., Seré, R., Santillo, M., Damiano, S., Bifulco, M., Lezza, C., et al., 2002. Effect of Cu/Zn superoxide dismutase on cholesterol metabolism in human hepatocarcinoma (HepG2) cells. Biochem. Biophys. Res. Commun. 295, 603–609.

D. Pujimulyani et al. Heliyon 6 (2020) e04219