



## Research article

# Stabilization of rice bran using a continuous microwave oven

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## Abstract

The effects of continuous microwave heating and storage temperature on free fatty acids (FFA) contents, peroxide values (PV) and 2-thiobarbituric acid values (TBA) in rice bran stabilization were examined. Raw rice bran (RRB) was adjusted to 21% moisture content on a wet basis and heated in a continuous microwave oven at 6,400 W and 2,450 MHz for 15.75 min. Then, it was packed in zipper-topped bags and stored at 4°C and 25°C for 16 wk. After 16 wk, the FFA contents of the RRB stored at 4°C and 25°C increased from 4.42% to 35.78% and 53.24%, respectively. Overall, there was no significant change in the continuous microwaved rice bran (MWRB) during storage. The PV and TBA values of RRB stored at 4°C and 25°C significantly increased and were higher than for MWRB. The results showed that a hydrolytic reaction and oxidative rancidity of rice bran can be prevented using continuous microwave heating. The heating affected the hydrolytic reaction and oxidative rancidity when the solutions were packed in zipper-topped bags and stored at 4°C for up to 16 wk.

## Introduction

Rice bran is a byproduct during the milling process of brown rice with the proportion of rice bran being between 7% and 8.5% of the total grain (Henderson and Perry, 1976). Rice bran is rich in functional nutrients and contains 15% to 23% oil, while crude rice bran oil contains 3% to 4% wax and 4% unsaponified lipid and the mineral content in rice bran varies based on the availability of nutrients in the soil and also contains iron, aluminum, calcium, sodium, chlorine, potassium, magnesium, manganese, phosphorus, silicon and zinc (Lu and Luh, 1991). After the milling process is completed, the lipids are decomposed to FFA and glycerol by enzymatic reaction of lipase. Due to the breakdown of the lipids, the rice bran has a short shelf life and reduced extraction yield. In addition, the lipoxygenase enzyme also generates an oxidation reaction between oxygen and the rice bran oil which in turn, generates oxidative rancidity and degrades the oil quality making it unsuitable for human consumption (Frankel,

1984; Saunders, 1985; Barnes and Galliard, 1991). Microwave heating is a new method to heat rice bran and denature the mentioned enzymes (Ramezanzadeh et al., 1999a; Ramezanzadeh et al., 1999b; Ramezanzadeh et al., 2000). Microwave heating mechanisms can be either ionic polarization or dipolar rotation. The main advantages of microwave processing are high temperature, fast heating, and a reduction in the processing time and energy consumption. Since microwave heating is both internal and volumetric, it speeds up the way in which food is cooked (Calay et al., 1995), as with microwave energy being used to cook food takes only 2–7 min to heat. Many researchers have investigated the stabilization of rice bran using a home microwave oven (Tao, 1989; Yoshida et al., 1991; Malekian, 1992). Therefore, the current research focused on using a continuous microwave oven to stabilize rice bran for commercial production. The objective was to study the effect of the continuous microwave heating and storage temperature on the extraction yield and quality of oil.

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## Materials and Methods

### *Rice bran collection*

Commercial rice bran was acquired from the Wattanawanit rice mill (Nakhon Pathom, Thailand). The rice bran was sieved using a 60-mesh sieve (Endcotts Ltd., London, UK) to remove foreign impurities such as broken rice and husk. The rice bran was kept at -20°C until the day of sample preparation.

### *Microwave stabilization*

The frozen rice bran was defrosted at room temperature. By adding tap water, the moisture content of the raw rice bran (RRB) was adjusted from 11.0% to 21.0% on a wet basis. A sample was mixed using a rotating mixer to completely distribute the water (Tao, 1989; Malekian, 1992; Ramezanzadeh et al., 1999a; Ramezanzadeh et al., 1999b; Ramezanzadeh et al., 2000). Then, the sample was spread on 14 Teflon trays to a thickness of 1 cm with 250 g per tray and then heated in a continuous microwave oven (V1, PrimAsia Technology, Bangkok, Thailand) at 6,400 W and 2,450 MHz at a very low belt speed (4.75 cm/min) resulting in a heating time of 15.75 min. Additionally, the highest temperature of the sample after heating in the continuous microwave oven, was  $125 \pm 0.5^\circ\text{C}$ . Thereafter, the sample was cooled to room temperature (approximately  $25^\circ\text{C}$ ) and packed in zipper-topped bags. Finally, approximately 30 kg of the rice bran was used for this experiment in a completely randomized design.

### *Packaging and storage of rice bran*

The continuous-microwave-heated RRB samples were weighed at 250 g per bag. The samples were packed in aluminum foil zipper-topped bags. The bags were stored for sampling after a few days and then in the 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, 8<sup>th</sup>, 12<sup>th</sup> and 16<sup>th</sup> weeks of storage. Half the bags were stored at  $4^\circ\text{C}$  and the remainder at  $25^\circ\text{C}$ .

### *Extraction of rice bran oil*

Rice bran oil was extracted using hexane at a ratio of rice bran:hexane of 1:3 (weight per volume). The extractions took place for 1 hr at room temperature. In addition, the samples were vacuum filtered (110 mm diameter filter paper). The extract solution was evaporated using a rotary evaporator (R-300 EL, Buchi, Switzerland). For further analysis, the rice bran oil was stored at -20°C. Oil extraction yields were defined as grams per 100 g on a dry basis.

### *Free fatty acids determination*

FFAs were determined using the method of Association of Oil Chemists' Society (1997), with modifications in the size of samples and concentration. In the current study, 5g of rice bran oil was added

into a 125 mL volumetric flask with 50 mL of neutralized alcohol and 2 mL of phenolphthalein, and then titrated using 0.1 N sodium hydroxide until a pink color developed and the percentage of FFA was determined using Equation 1:

$$\text{Percentage of FFA} = (\text{milliliters of alkali} \times N \times 28.2) / (\text{grams of rice bran oil}) \quad (1)$$

where N is the normality of the sodium hydroxide solution.

### *Peroxide value determination*

Peroxide values (PVs) were determined using the method of Association of Oil Chemists' Society (1997), with modifications in the size of the samples. PV determinations were tested by adding 1 g of rice bran oil into a 125 mL volumetric flask, followed by 6 mL of 3:2 acetic acid:chloroform solution, 0.1 mL of saturated potassium iodide and 6 mL of distilled water. The solution was kept in the dark for 1 min. Afterwards, 1 mL of starch indicator solution was added. Titration using 0.01 N of sodium thiosulfate was halted when the solution became clear. PV was measured in units of milli equivalent per kilogram (meq/kg) and determined using Equation 2:

$$\text{PV} = [(S - B) \times 1,000] / \text{grams of rice bran oil} \quad (2)$$

where B is the volume of titrant measured in milliliters of blank, S is the volume of titrant measured in milliliters of sample and N is the normality of the sodium thiosulfate solution.

### *2-Thiobarbituric acid value*

2-Thiobarbituric acid (TBA) values were determined using the method of Pearson (1976), with modification in the size of sample. First, 10 g of rice bran oil was added into a test tube followed by 2.5 mL of 4 N hydrochloric acid and then boiled for 30 min. Second, 5 mL of sample solution was added into the test tube, followed by 5 mL of TBA reagent and the test tube was placed in a water bath at  $100^\circ\text{C}$  for 35 min. Finally, absorbance of the reaction solution was measured in the cuvette at 538 nm and determined using Equation 3:

$$\text{TBA value} = [7.8 \times (A - B)] \quad (3)$$

where A is the absorbance of the sample and B is the absorbance of the blank.

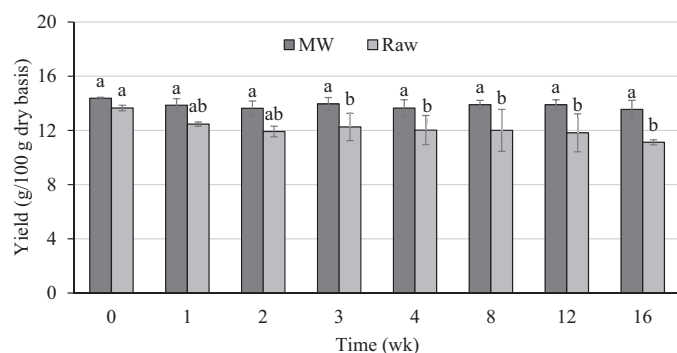
### *Statistical analysis*

Data were analyzed using analysis of variance (ANOVA). Means were compared using Duncan's multiple range test facilitated by the SPSS program version 16.0 (SPSS Inc.; Chicago, IL, USA).

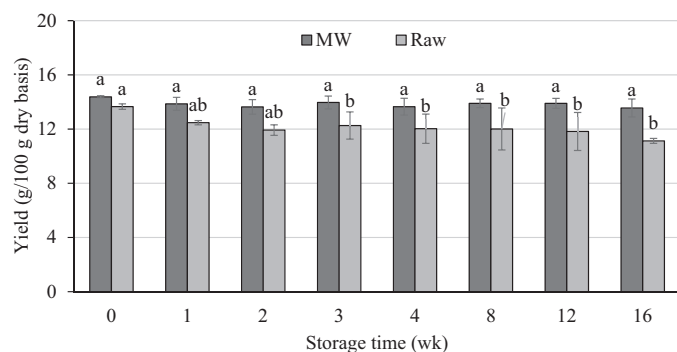
## Results and Discussion

### Extraction yields of rice bran oil

The continuous microwave oven heating and storage temperature affected the yields of oil as shown in Fig. 1 and Fig. 2. Altogether, the yields of rice bran oil in RRB significantly decreased from 13.68% to 13.66% and from 12.56% to 11.13% at 4°C and 25°C, respectively. Because the enzymes (lipase and lipoxygenase) were still in the active form, the cold temperature slowed down the activity of the enzyme. The yields of rice bran oil after being heated using continuous microwaves decreased slightly from 14.40% to 14.38% and from 14.14% to 13.56% at 4°C and 25°C, respectively. Furthermore, the yields of rice bran oil after stabilization using the continuous microwave treatment were higher than the yields of raw rice bran oil. In addition, the highest yield of rice bran oil resulted microwave heating destroyed most of the enzymes and the cooler temperature slowed down the activity of the remaining enzymes, which was consistent with other plant oil extraction studies. For example,



**Fig. 1** Yield of rice bran oil after stabilization using continuous microwave heating in zipper-topped bags and storage at 4°C; histograms represent mean  $\pm$  SD; Different lowercase letters above histograms indicate significant different ( $p < 0.05$ ); MW = continuous microwave rice bran; Raw = raw rice bran

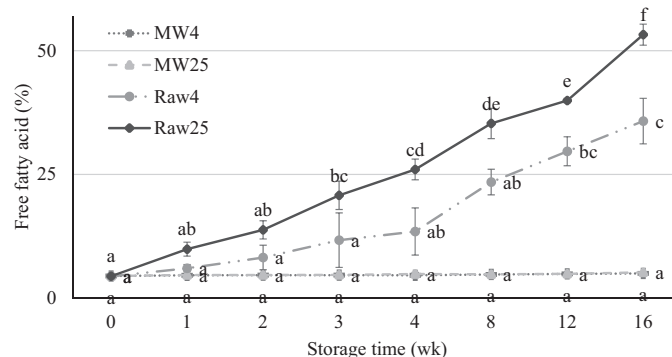


**Fig. 2** Yield of rice bran oil after stabilization using continuous microwave heating in zipper-topped bags and storage at 25°C; histograms represent mean  $\pm$  SD; Different lowercase letters above histograms indicate significant different ( $p < 0.05$ ); MW = continuous microwave rice bran; Raw = raw rice bran

Azadmard-Damirchi et al. (2010) reported that pretreatment of rape seed using microwave heating, could enhance the oil extraction yield compared to the solvent extraction method and the oil extraction yield from hazelnut seed increased with microwave heating (Ramezazadeh, 2000 and Thanonkaew et al., 2012). In summary, the yield of rice bran oil increased following stabilization using microwave heating when compared to roasting and steaming (Ramezazadeh, 2000 and Thanonkaew et al., 2012) and the yields of rice bran oil stored at 4°C were higher than at 25°C after 16 wk of storage.

### Free fatty acid of rice bran oil

The effect of continuous microwave oven heating on rice bran stability was studied by monitoring the change in the FFA content (Fig. 3). The FFA in RRB increased rapidly from an initial value of 4.42% to 35.78% and 53.24%, during 16 wk of storage at 4°C and 25°C, respectively. Additionally, the FFA in MWRB ranged from 4.47% to 5.00% and 5.21% during the 16 wk of storage at 4°C and 25°C, respectively. However, there were no significant differences during the 16 wk of storage at 4°C compared to 25°C. The continuous microwave heating inhibited lipase activity. Lastly, the hydrolytic reaction reduced the moisture content of rice bran to a safe level (less than 13%) as defined by Tao (1989) and Malekian (1992). This result was in agreement with other researchers. Tao (1989) and Malekian (1992) reported that the FFA content of microwave-heated rice bran from long grain and medium grain varieties increased only slightly during 4 wk of storage at 25°C and that the FFA content following microwaving and then storage in a refrigerator showed very little change over 8 wk. Ramezazadeh (1999a) reported that there was no significant difference in the FFA content of rice following microwave heating between storage at room temperature and being chilled in the refrigerator over 16 wk. Storage at 4°C resulted in a lower FFA content during 16 wk of storage compared to at 25°C.



**Fig. 3** Free fatty acids in raw rice bran oil and rice bran oil after stabilization using continuous microwave heating and packed in zipper-topped bags and stored at 4° and 25°C; values are the mean  $\pm$  SD; Different superscript letters above error bars of each treatment indicate significant different ( $p < 0.05$ ) among time points; MW4 = continuous-microwave-heated rice bran stored at 4°C; MW25 = continuous-microwave-heated rice bran stored at 25°C; Raw4 = raw rice bran stored at 4°C; Raw25 = raw rice bran stored at 25°C.

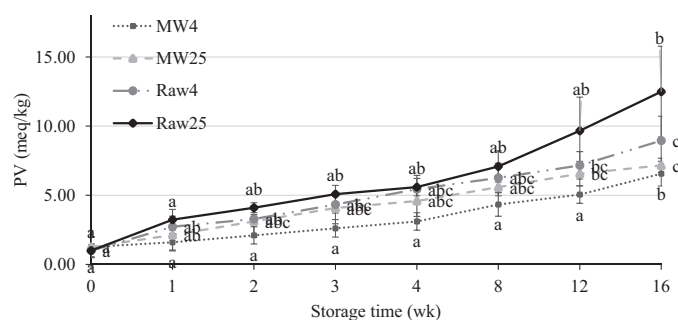
### Peroxide values of rice bran oil

The effects of continuous microwave heating on rice bran stability were studied by measuring the change in PV (Fig 4). The PV in RRB increased rapidly from 0.99 meq/kg to 8.95 meq/kg and 12.49 meq/kg during 16 wk of storage at 4°C and 25°C, respectively. Furthermore, the PV in MWRB ranged from 1.26 meq/kg to 6.56 meq/kg and 7.16 meq/kg during 16 wk of storage at 4°C and 25°C, respectively. There was no significant difference during the 16 wk of storage at 4°C. From the many tests conducted, continuous microwaving destroyed most of the lipoxygenase enzyme and reduced the oxidative reaction. Due to atmospheric packaging, there was oxygen in the package that could react with the oil; therefore, the PV slowly increased. In summary, vacuum packaging is necessary for long-term storage. Storage at 4°C resulted in a lower PV during 16 wk of storage compared to at 25°C.

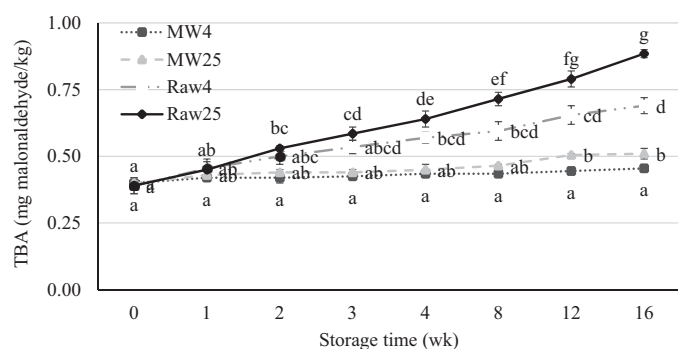
### 2-Thiobarbituric acid values of rice bran oil

The effects of continuous microwave oven heating on rice bran stability were studied by measuring the change in TBA (Fig 5). The TBA in RRB increased rapidly from an initial value of 0.39 mg/kg to 0.72 mg/kg and 0.90 mg/kg during 16 wk of storage at 4°C and 25°C, respectively. The TBA value in MWRB ranged from 0.40 mg/kg to 0.44 mg/kg and 0.53 mg/kg during 16 wk of storage at 4°C and 25°C, respectively. There was no significant difference during 16 wk of storage at 4°C. Continuous microwaving slowed down the reaction which retarded rancidity. Esaka et al. (1986; 1987) suggested that microwave heating was more effective in inactivating lipoxygenase in winged bean seeds, and in soybeans with a high moisture content. They concluded that microwave heating inactivates the lipoxygenase of winged bean seeds and soybeans in much less time than does conventional heating. Following microwave heating, the lipoxygenase in the high moisture soybean sample was completely inactivated. The temperature of soybeans was 100°C (Wand and Toledo, 1987). This confirmed that not only is the temperature important for lipoxygenase inactivation but also the moisture content of the sample, because of the high microwave energy absorption. In the current study, continuous microwave heating was able to inactivate most of the lipoxygenase enzyme and the moisture content in the rice bran samples was adjusted from an initial 11% to 21% before the continuous microwave heating. Overall, the temperature in the rice bran samples reached  $125 \pm 0.5^\circ\text{C}$  during the continuous microwave heating. Storage at 4°C resulted in a lower TBA level during 16 wk of storage compared to at 25°C.

Therefore, the recommended storage conditions for up to at least 16 wk for the prevention of a hydrolytic reaction and oxidative rancidity for rice bran that has been heated using continuous microwave ovens were the use of zipper-topped bags and storage at 4°C. Heating using a continuous microwave oven could increase the capacity of production compared with using a home microwave oven. Lastly, this knowledge can be applied in commercial production.



**Fig. 4** Peroxide value (PV) in raw rice bran oil and rice bran oil after stabilization using continuous microwave heating and packed in zipper-topped bags and stored at 4° and 25°C; values are the mean  $\pm$  SD; Different superscript letters above error bars of each treatment indicate significant different ( $p < 0.05$ ) among time points; MW4 = continuous-microwave-heated rice bran stored at 4°C; MW25 = continuous-microwave-heated rice bran stored at 25°C; Raw4 = raw rice bran stored at 4°C; Raw25 = raw rice bran stored at 25°C.



**Fig. 5** 2-Thiobarbituric acid (TBA) in raw rice bran oil and rice bran oil after stabilization using continuous microwave heating and packed in zipper-topped bags and stored at 4° and 25°C; values are the mean  $\pm$  SD; Different superscript letters above error bars of each treatment indicate significant different ( $p < 0.05$ ) among time points; MW4 = continuous-microwave-heated rice bran stored at 4°C; MW25 = continuous-microwave-heated rice bran stored at 25°C; Raw4 = raw rice bran stored at 4°C; Raw25 = raw rice bran stored at 25°C.

The rice bran could be stabilized by heating in a continuous microwave oven. The moisture content should be adjusted to 21% on a wet basis. The time of heating was 15.75 min using 6,400 W and 2,450 MHz. The heating could destroy lipase and lipoxygenase enzymes and the rice bran could be stored for up to 6 wk.

### Conflict of Interest

The authors confirm that there is no known conflict of interest associated with this publication and there has been no substantial financial support for this work that could have influenced its outcome.

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