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Study on the stabilization effect of continuous microwave on wheat germ

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ABSTRACT

The purpose of this research is to study the stabilization effect of microwave on wheat germ using continuous microwave processing equipment. Results from the study indicated that microwave technique had a significant stabilization effect on wheat germ. When the on-line processing capacity of wheat germ was 20 kg/h, the optimal conditions for wheat germ stabilization were microwave power of 4 kW, the processing time of 8 min and ventilation rate of 60 N m^3/h. Under such conditions, the residual lipase activity decreased to 14.01% of that of the raw wheat germ. The lipoxygenase lost its activity completely under such intensity microwave processing. After 60 days' accelerated storage, the acid value increased only 6.56% of that of the stabilized wheat germ before storage. In addition, microwave could cause the death of harmful microorganisms. It was clear that the continuous microwave stabilization process was practicable for wheat germ stabilization.

1. Introduction

Wheat germ is a by-product obtained from wheat milling. It contains nutrients which are good for human consumption (Brandolini and Vidal, 2012). About 2 million tons of wheat germ has not been fully exploited in China and other parts of the world; it is mostly blended with bran and used as a kind of feed for animals. The reason for this is the physiological activity of wheat germ is the strongest among all parts of a wheat seed since it contains the highest amount of fat and water. When wheat germ is separated from the whole wheat grain, the fatty acids in wheat germ increase quickly within a short period of time due to the action of lipase and lipoxygenase, which will eventually lead to rancidity. Also because of the high nutritional content, microorganisms do act on the wheat germ thereby proliferating causing it to agglomerate, to become mouldy or ferment. Rapid spoilage process of wheat germ has been a key factor in restricting it for further processing.

Currently, the most common stabilization method for preventing wheat germ from spoilage is heat treatment, which includes dry heating and steaming (Srivastava et al., 2007; Yöndem-Makascıoğlu et al., 2005). However, the existing dry heating methods do not perfectly inactivate lipase because enzyme renaturation occurs easily. The steaming methods lead to high moisture content in wheat germ, and it also requires huge investment for processing facilities, which limits its industrial application (Ferrara and Benson, 1991). Research has indicated that microwave stabilization combining thermal and non-thermal effects works more efficiently than traditional heating methods (Rodríguez-López et al., 1999; Wu et al., 2008). Moreover, microwave has several advantages including its high efficiency, lack of nutrient loss, and above all it ensures sterilization while drying.

Recently, microwave technology has been widely used for the processing of agricultural products. Wheat germ stabilization studies were conducted using a domestic microwave oven. However, in these studies, stabilization effects of wheat germ enzymes were investigated by batchwise mode (Kermasha et al., 1993; Vetrimani et al., 1992). It was evident that the data was different from the use of industrial processing equipment, which uses the continuous microwave processing. Reports on wheat germ stabilization by means of continuous microwave mainly dealt with the stabilization effects rather than its mechanism and the manufacturing process (Klingler, 1994; Zwingelberg and Fretzdorff, 1996). In this paper, self-designed continuous microwave processing equipment was used to investigate wheat germ stabilization process and the mechanism involved in wheat germ stabilization by determining lipase activity, moisture content, acid value, sensory quality, etc., aiming at offering a theoretical basis for microwave stabilization techniques and related equipment development.

2. Materials and methods

2.1. Raw materials

Wheat germ flakes, which were separated from commercially available hard red winter wheat, were purchased from Shandong Yongle Food Co. in China. It was stored at a temperature of

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−20 °C in a freezer. Besides, linoleic acid (99% pure) was purchased from Fluka Chemical Corp. (Ronkonkoma, NY). Olive oil was purchased from Mueloliva (Priego de Cordoba, Spain). All other chemicals were of analytical grade.

2.2. Stabilization test system

The continuous microwave processing equipment (Dong et al., 2007) consists of six parts: namely, the control system, microwave generator system, transmission system, material transport system, temperature control system and air control system (Fig. 1). The frequency of the microwave processing equipment was 2450 ± 50 MHz; the microwave power was within a range of 0.0–6.0 kW (adjustable); the drum rotary speed was controlled by electromagnet (stepless, adjustable). 97FLJ2WYD4-2F Centrifugal fan was adopted as dehumidifier, and the max air output was 80 N m^3/h. The wheat germ processing capacity of the equipment was 15–50 kg/h.

2.3. Water activity and moisture content analysis

Wheat germ water activity and moisture content were measured before and after microwave processing using a water activity meter (AquaLab Series 3TE, Decagon Devices, Pullman, WA, USA) and a Halogen Moisture Analyzer (Model HB43- S, Mettler Toledo International Inc., Switzerland).

2.4. Lipase activity determination

The method is based on that of Rose and Pike (2006) with modification. Wheat germ was first homogenized and 2 g of them (accurate to 0.001 g) was transferred into two centrifuge tubes labeled (A_f) and (A_i) respectively, where (A_f) and (A_i) refers to the sample and the blank. Olive oil of 1.5 mL was added to the sample tube (A_f) to mix with wheat germ sample. The sample tube was incubated at 45 °C for 4 h. The reaction product was then extracted by 30 mL n-hexane. The extraction was further evaporated on a rotary vacuum evaporator at 40 °C, and the residue was dissolved in 4 mL isooctane. After 4 h, the ground wheat germ in the blank tube (A_i) was also mixed with 1.5 mL olive oil and treated in the same way as that in the sample tube. Moreover, 2 mL 5% (W/V) copper acetate (adjusted pH to 6.1 with pyridine) was added to the sample tube as well as that of the blank tube and were vigorously shaken. After centrifugation at 1000 g (BR4, Jouan, Winchester, VA, France) for 3 min, the upper organic phase were transferred and determined at 715 nm using a UV–Vis spectrophotometer (model UV 9600, Rayleigh, Beijing, China). Finally the lipase activity (U/g) was calculated according to the following calculation:

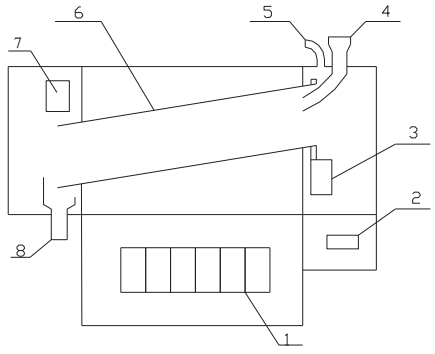

Fig. 1. Sketch map of continuous microwave processing equipment. 1 Microwave heating pipe; 2 controller; 3 transmission device; 4 feed-in port; 5 air outlet; 6 roller; 7 far infrared thermoscope; 8 outlet port.

$$LA = 1000\frac{(4+v)(A_f - A_i)}{\varepsilon t l s}$$

where 1000 = conversion ratio from mol/L to μmol/mL; 4 = volume of isooctane (mL); v = volume of olive oil (mL); A_f = absorbance of testing sample wheat germ at 715 nm; A_i = absorbance of blank sample wheat germ at 715 nm; ε = molar absorbance of oleic acid at 715 nm (M^{-1} cm^{-1}); t = reaction time (h); l = thickness of cuvette (cm), s = dry weight of sample (g).

2.5. Lipoxygenase activity determination

Lipoxygenase activity was determined by measuring the increase in absorbance at 234 nm through the hydroperoxidation of linoleic acid on a DU 800 spectrophotometer (Beckman Coulter, Inc., Fullerton, CA, USA) at 25 °C. Firstly, 10 g of ground (sifted by 30 mesh sieve) wheat germ sample was transferred into 100 mL cold acetic buffer (0.1 mol/L, pH 4.5) and was stirred for 30 min at 4 °C. The slurry was then centrifuged at 12000*g* (BR4, Jouan, Winchester, VA, France) for 15 min at 4 °C with the supernatant being the raw lipoxygenase enzyme solution (Shiiba et al., 1991). The substrate was Oleic acid and absolute ethyl alcohol mixture [the concentration of oleic acid and tween-20 were 2.53×10^{-3} mol/L and 0.08% (w/v), respectively]. The entire reaction system constituted 2.0 mL phosphate buffer solution, 200 μL substrate solution and 50 μL raw lipoxygenase solution (Xu et al., 2012). The blank solution was prepared by mixing 200 μL of substrate solution with 2.0 mL of buffer and 50 μL of inactive lipoxygenase solution. The reaction mixture was stirred for 10 s after which absorbance was recorded by computer at intervals of 1 s for 300 s. The initial reaction velocity was calculated by linear regression on data set between 30 s and 210 s of the reaction curve. One unit of lipoxygenase activity was defined as an increase in absorbance of 0.01 at 234 nm per minute per mg of protein under assay conditions.

2.6. Accelerated storage test

The accelerated storage test was done at a constant temperature oven of 50 °C for 60 days. The microwave-treated wheat germ and the raw wheat germ (control) were all packed in polyethylene bags (250 g each). Acid value, lipase activity, lipoxygenase activity and microorganism content were assayed at regular intervals. Free fatty acid content of wheat germ oil was determined according to the recommended methods of National Standard of the People's Republic of China (GB 5009.37-2003). Total colonies and mould counts of the wheat germ samples were determined according to the recommended methods of National Standard of the People's Republic of China (GB 4789.2-2010, GB 4789.15-2010) and the content of total colonies and mould were calculated as CFU/g wheat germ.

2.7. Single factor test

Continuous microwave processing equipment was used in this section. Feed-in port of the equipment was adjusted to ensure the flow rate of wheat germ at 20 kg/h. The microwave power, processing time and ventilation rate were optimized using three single factor tests as follows:

(1) Microwave power was set at 1.0, 2.0, 3.0, 4.0, 5.0 kW, microwave processing time 8 min, ventilation rate 80 N m^3/h.
(2) Microwave processing time was set at 2, 4, 6, 8, 10 min, microwave power 3.0 kW, ventilation rate 80 N m^3/h.
(3) Ventilation rate was set at 0, 20, 40, 60, 80 N m^3/h, microwave power 3.0 kW, microwave processing time 4 min.