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Research Article

PREPARATION AND EVALUATION OF RICE BRAN OIL MASK

JIRAPORNCHAI SUKSAEREE*, CHAOWALIT MONTON, LAKSANA CHAROENCHAI, PATHAMAPORN PATHOMPAK

Faculty of Pharmacy and Sino-Thai Traditional Medicine Research Center (Cooperation between Rangsit University, Harbin Institute of Technology, and Heilongjiang University of Chinese Medicine), Rangsit University, Pathum Thani 12000, Thailand.
Email: jirapornchai.s@rsu.ac.th

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ABSTRACT

Objective: This work was to prepare rice bran oil mask which prepared from composition of chitosan blended with hydroxypropyl methylcellulose E5 or polyvinyl alcohol. They comprised of glycerine, propylene glycol, polyvinyl pyrrolidone K30, and tween 80 as ingredients in formulations.

Methods: The 2 g chitosan was dissolved in 100 mL distilled water and 0.5% acetic acid was added. It was mixed with another ingredients and rice bran oil by traditional beaker method. They were characterized the appearance, pH, and viscosity values and were analyzed the γ-oryzanol content by HPLC method.

Results: Their pH and viscosity were in the range of 6.184-6.130. The F1 formula had significantly higher viscosity values compared to F2 formula. The formulations were found to be safe when applied on the skin. The physical appearance of bran mask was slightly yellow colored. The F1 formula had stable formulation after storage at three conditions. They found the γ-oryzanol ranged of 92.45 – 97.51% and 87.10 – 91.28% for F1 and F2 formula, respectively.

Conclusion: Thus, F1 formula was suitable for future development and usage of rice bran oil mask formulation with good viscosity.

Keywords: Rice Bran Oil, Rice (*Oryza sativa* L.), Mask, Formulation.

INTRODUCTION

Thailand, India, and southern China are where rice, or *Oryza sativa* L., which is a genus of perennial grass in the Poaceae or grass family, originated. In ancient times, rice was domesticated and diversified. Now, it is cultivated in wet tropical, semi-tropical, and warm temperate areas around the world for the production of its cereal grain [1-2]. It is therefore the most important cereal crop for human consumption in the world.

Hom-Pathum rice as known as Khaw Hom-Pathum is the one types of rice species which is developed and planted in paddy fields in Pathum Thani and other provinces in the central parts of Thailand. Hom-Pathum rice is less fragrant and less photosensitive. Hom-Pathum rice is soft and tender. It is a year round crop and can be grown in an easier manner. It has a natural aroma depending on its age and the areas it was planted in. However, Hom-Pathum comprised of various nutritional substances such as vitamin B1, B2, niacin, carbohydrates, protein, and minerals such as iron, calcium, and phosphorous [3]. Moreover, both their rice bran coats are full of valuable nutrients.

Nowadays, rice bran oil has become widely interesting, and is quickly growing as an important application in the health and nutritional industry. This is due to rice bran oil potentially providing benefits to health, in its natural state, as it contains several beneficial constituents. Rice bran oil has high smoke point and delicate flavor which can be produced into an excellent cooking and salad oil. γ-Oryzanol is an important component of crude rice bran oil [4]. Basically, rice bran oil contains 3-5% unsaponifiable matter, and micronutrients including γ-oryzanol (0.9-2.1%), phytosterols and tocotrienol (355-685 ppm), sterols (2-3%), and steryl ester (2.8-3.1%) [1-3]. Many publications report that rice bran oil can reduce harmful cholesterol, (also known as low density lipoprotein, LDL) without reducing the "good" cholesterol, known as high density lipoprotein or HDL [4-7]. γ-Oryzanol has been shown to possess antioxidant, anti-inflammatory, anti-tumor, and hypocholesterolemic activities [8]. In addition, rice bran oil has high levels of free fatty acids (FFA). The fatty acid composition of rice bran oil is palmitic acid (14-22%), stearic acid (0.9-2.5%), oleic acid (38-46%), linoleic acid (33-40%), and linolenic acid (0.2-2.9%), as reported in the codex standard and other publications [4, 8-11]. Specifically, the linoleic and linolenic acids expresses as ω-6 and ω-3 fatty acid, respectively. They appear to reduce the risk of health effects such as cancer, cardiovascular disease, inflammation, developmental disorders, and cognitive aging [10-15]. However, the diverse contents of various compositions of rice bran oil are mainly due to differences between varieties of rice: the origin, environmental factors, and genotype, as well as solvent extraction conditions. This work was to prepare rice bran oil mask which prepared from composition of chitosan blended with hydroxypropyl methylcellulose E5 (HPMC) or polyvinyl alcohol (PVA). They comprised of glycerine, propylene glycol (PG), polyvinyl pyrrolidone K30 (PVP), and tween 80 as ingredients in formulations. They were characterized the appearance, pH, and viscosity values and were analyzed the γ-oryzanol content by HPLC method.

MATERIALS AND METHODS

Materials

A standard mixture of γ-oryzanol (analytical grade) was purchased from TCI, Tokyo, Japan. Hom-Pathum rice bran samples were provided by a local milling company in Thailand. The rice bran samples were passed though sieve number 20 and immediately extracted under cold press conditions. This sample was obtained from the mill as liquid oil prepared in the laboratory [16] (Fig. 1). Chitosan was purchased from Seafresh, Thailand. PVP, HPMC, PVA, glycerine, PG, tween 80, paraben concentrate (methyl paraben:propyl paraben 10:2 adjusted to 100 mL with PG) were purchased from Sigma, USA. Other chemicals were pharmaceutical or analytical grade.

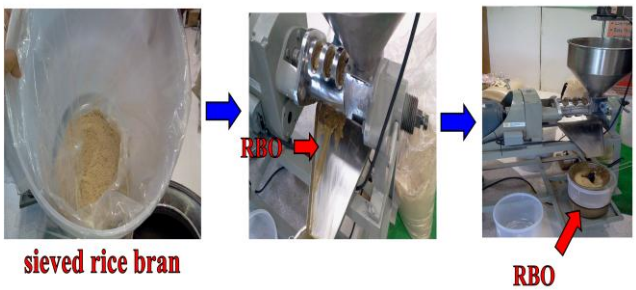

Fig. 1: The process of rice bran oil from rice bran of Hom-Pathum rice [16]

Rice bran oil mask preparation

The 2 g chitosan was dissolved in 100 mL distilled water and 0.5% acetic acid was added. Different formulations of rice bran oil mask were prepared by traditional beaker method are presented in Table 1. The formed rice bran oil mask was kept in well-closed container at ambient temperature for further evaluation.

Physical appearance and physical properties of the formulations

The appearances of these formulations were visually observed by the researcher. The pH value was measured using a pH meter (Mettler Toledo, Germany). Viscosity was measured using a programmable viscometer (Brookfield, Brookfield Engineering Laboratories Inc., USA) with a spindle LV 4.

Table 1: The ingredients of rice bran oil mask formulations

Ingredients	Formulations (g)	
	F1	F2
Rice bran oil	0.5	0.5
2% w/w Chitosan	20	20
HPMC	2	-
PVA	-	2
Glycerine	1	1
PG	1	1
PVP K30	1	1
Tween 80	1	1
Paraben concentrate	1	1

Preparation of rice bran oil mask

The sample of rice bran oils mask were accurately weighed 1 g in a volumetric flask (size 25 ml) and the volume was diluted with isopropanol. The samples were filtered through a membrane filter (0.45 µm) before HPLC analysis. The experiments were carried out in triplicate.

Preparation of γ-Oryzanol standard solutions

γ-Oryzanol standard was accurately weighed to 25 mg in a volumetric flask (size 25 ml) and the volume was adjusted to 25 ml with isopropanol. The stock solution was serial two-fold diluted to six concentrations and filtered through a membrane filter (0.45 µm) before HPLC analysis [17].

Isolation of individual γ-oryzanol [17]

The four major of steryl ferulates (Fig. 2) were isolated from standard γ-oryzanol mixture by HPLC. A 50 mg of γ-oryzanol mixture was dissolved in isopropanol 10 ml. An aliquot of 30 µl was separated on HPLC (Agilent Technologies, USA) with a Poroshell 120 EC-C18 column (3.0×150 mm, 2.7µm). Elution was achieved by using a solvent mixture of acetonitrile and methanol (60:40 v/v), with a flow rate of 0.8 ml/min. Four peak were collected (peak 1 t_R 10.6 min; peak 2 t_R 12.2 min; peak 3 t_R 13.8; peak 4 t_R 15.9 min). The isolated compounds were used as an external standard for HPLC quantitative analysis.

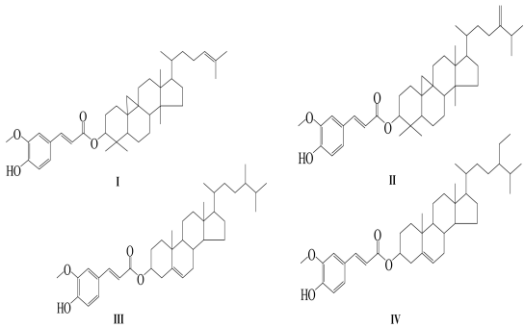

Fig. 2: Chemical structures of the steryl ferulates identified from the commercial standard γ-oryzanol mixture (I) cycloartenyl ferulate; (II) 24-methylenecycloartanyl ferulate; (III) campesteryl ferulate; (IV) and β-sitosteryl ferulate [17].

***HPLC conditions* [17]**

HPLC analysis was carried out using the Agilent 1200 series equipped with an Agilent 1200 series photodiode-array detector (PDA) and autosampler. Data analysis was performed using OpenLAB CDS EZChrom software (Agilent, USA). Separation was achieved at 25ºC on a Poroshell 120 EC-C18, 3.0×150 mm, 2.7 µm (Agilent Technologies, USA). The mobile phase consisted of acetonitrile:methanol (60:40 v/v) and was pumped at a flow rate of 0.8 ml/min. The injection volume was 10 µl. The quantitation wavelength was set at 325 nm. The validation of the analytical method was followed by the guidelines of the International Conference on Harmonization of Technical Requirement for the Registration of Pharmaceuticals for Human Use (ICH). The calibration curve was linear over the concentration range of 500-15.125 µg/ml. Cycloartenyl ferulate, 4-methylenecycloartanyl ferulate, campesteryl ferulate, and β-sitosteryl ferulate exhibited linearity over the evaluated ranges with correlation coefficients 0.9996. Both intra-day and inter-day precision were estimated by the relative standard deviation were less than 2% and 5%, respectively. Recoveries in the range of 100.1-101.9% were observed for all compounds [17].

RESULTS AND DISCUSSION

Their pH and viscosity are presented in Table 2. The pH values of the formulations were in the range of 6.184-6.130. The F1 formula had significantly higher viscosity values compared to F2 formula because the HPMC had high viscosity than PVA polymer. Thus, when the chitosan solution was mixed with HPMC, this formula had highly viscosity value. However, the formulations were found to be safe when applied on the skin [18]. The physical appearance of different rice bran mask formulations is shown in Fig. 3. They were slightly yellow colored. The F1 formula had stable formulation after storage at three conditions compared to F2 formula. Thus, F1 formula was suitable for future development and usage of rice bran oil mask formulation with good viscosity.

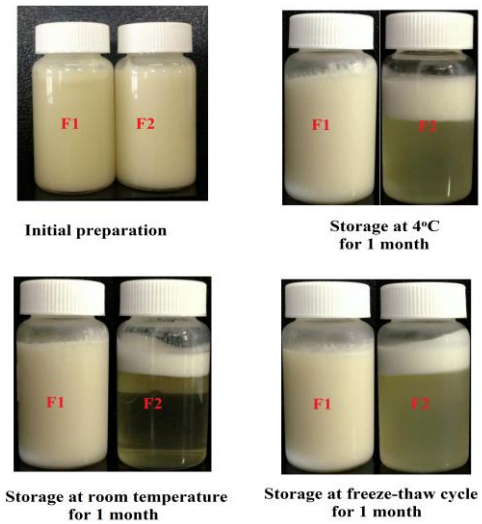

Fig. 3: Physical appearance of different rice bran oil mask formulations for initial preparation and after storage at different condition for 1 month

The rice bran oil content mask formulation

F1 and F2 formulas were extracted and diluted with isopropanol, and then they were analyzed with HPLC to detect the γ-oryzanol compound (contained compound I-IV, Fig. 2). The initial formulation weight, initial rice bran oil, initial γ-oryzanol content, and γ-oryzanol content found showed in Table 3-4 for F1 and F2 formula, respectively. They found the γ-oryzanol ranged of 92.45 – 97.51% and 87.10 – 91.28% for F1 and F2 formula, respectively. This due to the F1 formula had high viscosity, it might be highly stored the γ-oryzanol more than the F2 formula. In addition, the chromatogram of standard γ-oryzanol, F1 formula, and F2 formula showed in Fig. 4 – 5.

Table 2: The pH and viscosity of rice bran oil mask formulations

Formulas	pH	viscosity		
		50 rpm	60 rpm	100 rpm
F1	6.184±0.048	36706.67±936.91	32554.67±2405.54	28207.33±820.92
F2	6.130±0.001	780.00±46.48	763.33±56.15	732.00±49.53

Table 3: γ-oryzanol in rice bran oil mask F1 formula analyzed by HPLC method

	Initial formulation weight (g)	Initial Rice bran oil (mg)	Initial γ-oryzanol content (mg)	γ-oryzanol content found (mg)	γ-oryzanol content found (mg)	γ-oryzanol content found (%)	γ-oryzanol content found (%)
F1 formula							
Initial preparation							
1	1.05	19.37	354.52	397.73±9.39	356.76±27.31	112.19±2.65	97.51±9.78
2	1.12	20.66	378.15	329.20±2.10		87.05±0.55	
3	1.09	20.11	368.03	343.36±3.15		93.30±0.86	
Storage at 4◦C for 1 month							
1	1.05	19.41	355.16	390.03±10.03	347.71±28.21	110.02±2.83	95.05±9.98
2	1.06	19.48	356.44	320.39±4.28		84.73±1.13	
3	1.10	20.36	372.58	332.71±5.32		90.40±1.44	
Storage at room temperature for 1 month							
1	1.05	19.43	355.60	388.74±9.39	347.59±27.43	109.65±2.65	95.01±9.76
2	1.08	19.90	364.24	321.32±2.10		84.97±0.55	
3	1.03	19.06	348.75	332.71±3.15		90.40±0.86	
Freeze-thaw 6 cycles							
1	1.01	18.69	341.99	373.73±9.39	338.30±23.62	105.42±2.65	92.45±8.65
2	1.04	19.20	351.31	315.71±2.10		83.49±0.55	
3	1.03	19.09	349.29	325.45±3.15		88.43±0.86	

Table 4: γ-oryzanol in rice bran oil mask F2 formula analyzed by HPLC method

	Initial formulation weight (g)	Rice bran oil (mg)	Initial γ-oryzanol content (mg)	γ-oryzanol content found (mg)	γ-oryzanol content found (mg)	γ-oryzanol content found (%)	γ-oryzanol content found (%)
F2 formula							
Initial preparation							
1	1.09	20.11	368.03	345.31±0.99	338.01±4.86	93.83±0.27	91.28±3.21
2	1.19	21.96	401.79	337.26±8.77		86.47±3.87	
3	1.03	19.00	347.77	331.47±5.40		93.54±2.26	
Storage at 4◦C for 1 month							
1	1.06	19.53	357.36	336.29±0.99	327.38±5.94	91.38±0.27	88.40±3.15
2	1.06	19.49	356.65	326.37±8.77		83.68±3.74	
3	1.09	20.05	366.91	319.48±5.40		90.16±2.17	
Storage at room temperature for 1 month							
1	1.0312	19.03	348.17	332.69±0.99	324.91±5.19	90.40±0.27	87.75±3.26
2	1.043	19.24	352.16	323.18±9.37		82.86±3.71	
3	1.0015	18.48	338.14	318.84±6.87		89.98±2.16	
Freeze-thaw 6 cycles							
1	1.07	19.73	361.07	326.67±0.99	322.47±2.80	88.76±0.27	87.10±3.15
2	1.03	19.01	347.84	321.28±8.77		82.37±3.69	
3	1.04	19.18	350.94	319.47±5.40		90.15±2.17	

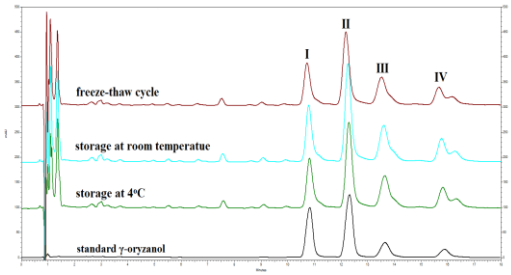

Fig. 4: HPLC chromatograms of standard γ-oryzanol ((I) cycloartenyl ferulate, (II) 4-methylenecycloartanyl ferulate, (III) campesteryl ferulate, and (IV) β-sitosteryl ferulate) and rice bran oil mask (F1 formula)

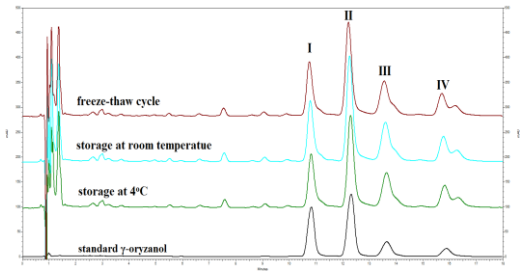

Fig. 5: HPLC chromatograms of standard γ-oryzanol ((I) cycloartenyl ferulate, (II) 4-methylenecycloartanyl ferulate, (III) campesteryl ferulate, and (IV) β-sitosteryl ferulate) and rice bran oil mask (F2 formula)

CONCLUSION

We have successfully prepared rice bran oil mask formulations from chitosan blended with different polymer types: HPMC or PVA. The pH and viscosity values of these formulations indicated that they were safe for direct application to the skin; therefore, they were suitable for preparing rice bran oil mask. Moreover, F1 formula could be further developed for cosmetic applications.

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