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Ali W. Nofan \*

Ethar Z. Naji

Department of Food Sciences.,  
College of Agriculture,  
University of Tikrit, Tikrit,  
Iraq

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**Extraction Conditions determination for Oryzanol  
extracted from Rice bran**

**ABSTRACT**

The extracted Oryzanol under the optimum conditions were determined , Five temperatures were used which are 30, 40, 50, 60, and 70 degrees Celsius and the results showed that the ideal temperature for extraction is 40 C° and Five heating times were used which are 10, 20, 30, 40, and 50 minutes and the results showed that the ideal heating time for extraction is 20 minutes . As for the volume of the solvent mixture used in the extraction, five sizes were used, which are 15, 35, 55, 75, and 95 ml, and it was found that the best volume for extraction is 95 ml in addition five ratios of Solvent Hexane : Isopropanol were used for extraction 1:1 , 1:3 , 3:1 , 3.5:0.5 , 0.5:3.5 V/V and the ideal ratio for mixing the hexane solvents: isopropanol 1: 3 volume : volume.

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**INTRODUCTION**

Rice is a staple food for about 70% of the world's population. Rice bran are a byproduct of the rice manufacturing process, which is 10-12% of the weight of the bean, and includes bean wrappings, skullcaps and an iron layer in addition to the fetus (Lacerda et al , 2010). Rice bran in the world are used mainly as feed, in the production of rice oil, in the preparation of functional foods, in baking products, and Snak food products, in addition to that they do not affect the susceptibility and storage acceptance of these products.

Numerous studies have indicated the importance of the nutritional value of rice bran , as it contains protein, ash, vitamins, minerals, and biological materials. It is a good source of dietary fiber, as it contains approximately 21-27%, and 1.9% soluble dietary fiber (Lilitchan et al , 2008),

\* Corresponding author: E-mail: [Ali.w.nofan@gmail.com](mailto:Ali.w.nofan@gmail.com)

and About 12-15% protein (Renuka et al , 2007) The protein content of rice bran depends on the degree of bleaching of brown rice (Sumantha et al , 2006).

Gama Oryzanol is a natural compound derived or extracted from rice bran oil , where the rice bran contains a mixture of ferric acid, sterols, and triple terrain alcohol (Alexander et al., 2006). In 1997, Shin and others (1997) prepared a study of its content that oryzanol isolated from rice lizard oil (RBO) has nutritional effects on animals, and there are several studies that explain the benefits of oryzanol , its effectiveness and stability.

Extraction is the first and crucial step in the study of natural antioxidants from plants. Many extraction factors play important roles in extraction efficiency, such as type, concentration, extraction solvent, extraction temperature, extraction time and PH. Among them, the solvent is one of the most influencing factors. Several solvents have been used to extract antioxidants from food and medicinal plants. The choice of solvents depends on the nature and chemical polarity of the antioxidant compounds to be extracted. Most phenols, flavanoids and anthocyanins are water-soluble antioxidants. Polar and medium polar solvents, such as water, ethanol, methanol, propanol, acetone and their aqueous mixtures, are widely used for extraction (Belwal et al., 2016) carotenoids are fat soluble antioxidants, and common organic solvents, such as hexane mixtures with acetone, ethanol, methanol or ethyl acetate mixture with Use of acetone, ethanol, and methanol (Sharmila et al., 2016).

## MATERIALS AND METHODS

A sample of Rice bran from Mashkhab 2 was obtained from Al Amir Flour Mill in Najaf. The Oryzanol was extracted from the rice bran under study, by adding 50 ml of distilled water to 10 g of rice bran in a glass beaker, then adding 2 g of ascorbic acid (97% purity), after which the mixture was mixed using a magnetic mixer for 30 minutes with Heat at a temperature of 30 ° C., then add to the mixture 55 ml of the solvent mixture hexane: isopropanol in a ratio of 1: 1 (volume: volume) and continue mixing for 30 seconds, after which the sample was centrifuged at a speed of 3000 r / min for 15 minutes, then The leachate was separated from the precipitate and the residual precipitate was recovered twice using the same solvent mixture, after which the leachate samples were collected from the steps Previously, the solvent was evaporated using a rotary evaporator, and a pale white crystalline precipitate was formed representing the Oryzanol.

**Temperature:** the extract oryzanol from the rice bran under study using different temperatures for heating included (30, 40, 50, 60, 70) for a period of 30 minutes and with a solvent size, then estimate the extracted oryzanol using spectroscopy at a wavelength of 320nm and the adoption of the standard curve in calculating the extracted oryzanol

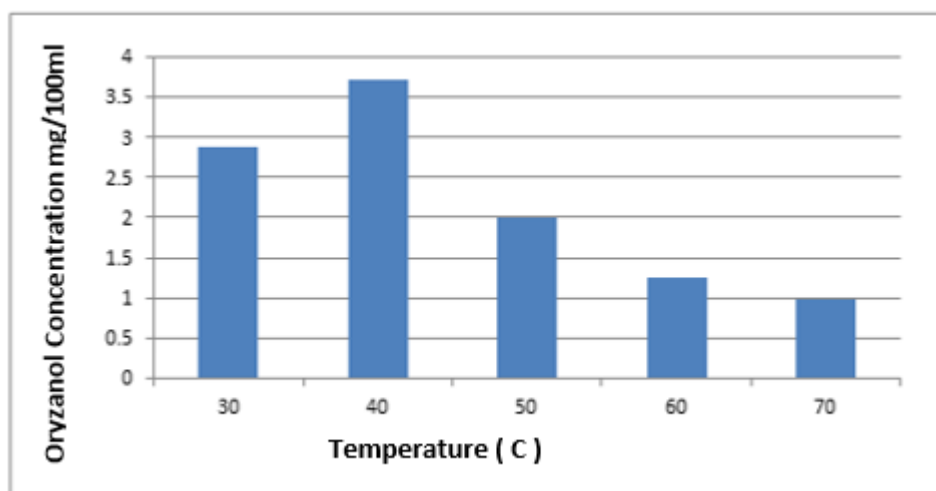
**Heating Time :** The oryzanol was extracted from the rice bran under study by heating at 40 ° C. for different periods, including (10, 20, 30, 40, 50) minutes, with confirmation of other extraction conditions.

**Solvent Value:** The mixture of hexane and isopropanol was used in a ratio of 1: 1 (volume: volume) of different sizes and (15,35,55,75,95) ml was used to extract the oryzanol from the rice bran under study by heating at a temperature of 40 ° C. for 20 minutes. And estimate the proportion of oryzanol extracted using the standard curve.

**Hexane : Isopropanol Ratio:** 95 ml of hexane mixture was used: oryzanol and in various proportions included (0.5: 3.5, 1: 1, 1: 3, 3: 1, 3.5: 0.5) volume: volume in the extract of oryzanol extracted from the rice bran under study with heating at 40 M for 20 minutes, then adopt the standard curve in calculating the percentage of the oryzanol extracted.

## RESULT AND DISCUSSION

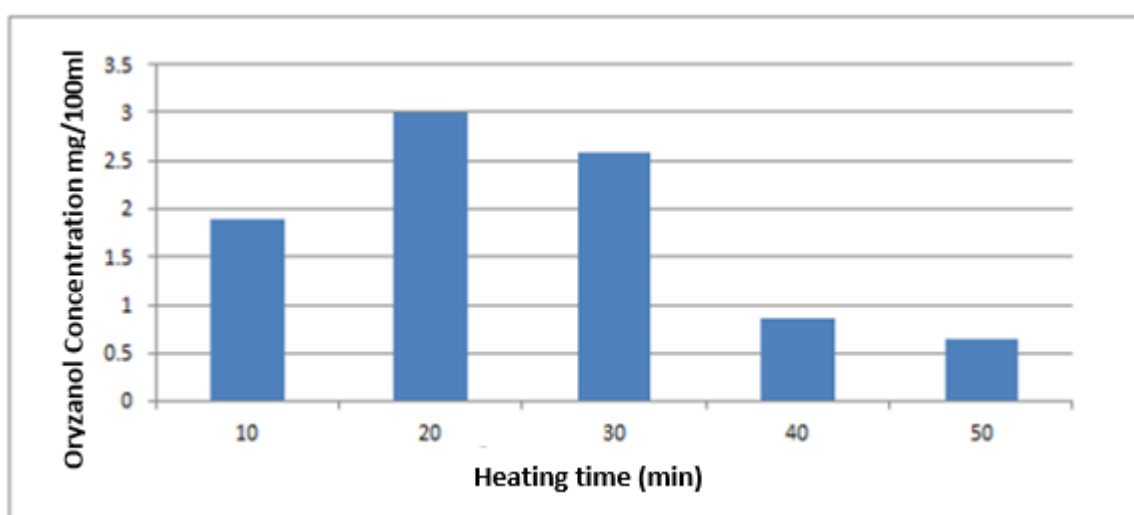
It is observed from Figure (1) the concentrations of the extracted oryzanol samples at 5 degrees of temperature, as it was found that the concentration of the oryzanol was 2.88 mg / 100 g when the mixture was heated at a temperature of 30 ° C, and it reached 3.72 mg / 100 g at a temperature of 40 ° C. Using a temperature of 50 ° C. The concentration of oryzanol at this temperature was 2.01 mg / 100 g, while at a temperature of 70 ° C. the concentration of the oryzanol extracted was 0.98 mg / 100 g.



**Figure 1:** Effect of heating temperature on oryzanol extraction

The reason for the low concentrations of oryzanol when the temperature rises is due to the high temperatures leading to the evaporation of the solvents and thus the efficiency of extraction decreases as the boiling point of hexane is less than the boiling point of isopropanol, as these temperatures lead to the extraction of oil in addition to the antioxidant of oryzanol. These high temperatures affect the composition of the antioxidant oryzanol.

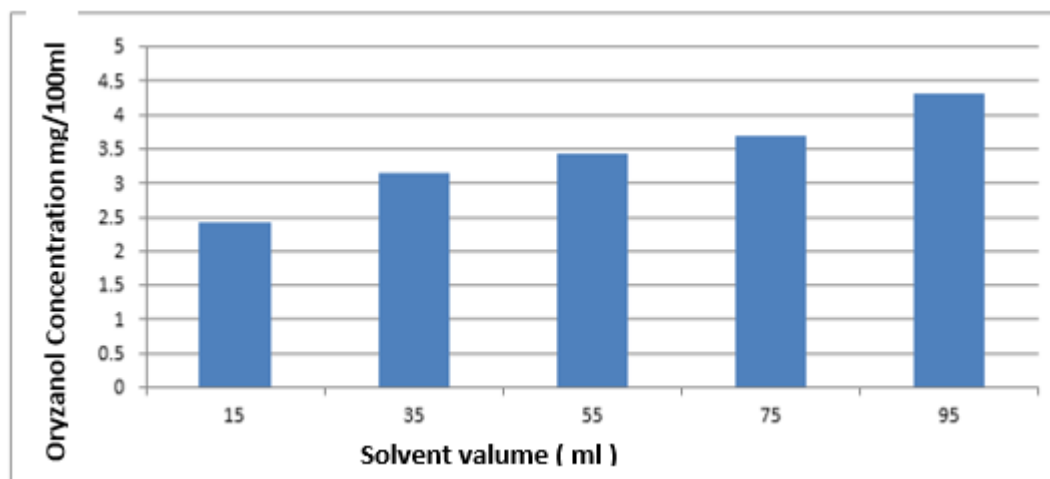
Five heating times were used to extract the antioxidant oryzanol as shown in Figure (2) . The concentration of the extracted oryzanol at a temperature of 40 ° C. for 10 minutes was 1.89 mg / 100g while the concentration of the oryzanol at the same temperature for 20 minutes reached 2.99 mg / 100 g. The concentration of the extracted oryzanol heated for 30 minutes was 2.68 mg / 100 g at the same temperature, while the concentration of the extracted oryzanol reached 0.87 mg / 100 g and 0.64 mg / 100 g for durations of 40 and 50 minutes, respectively.



**Figure 2:** Effect of heating time on oryzanol extraction.

Reduced concentrations by increasing the heating time can be attributed to the quality of the solvents used and their volatility, as the more solvent the time of extraction the greater the chance of volatilization and thus reduce the efficiency of extraction, (Lilitchan et al., 2008).

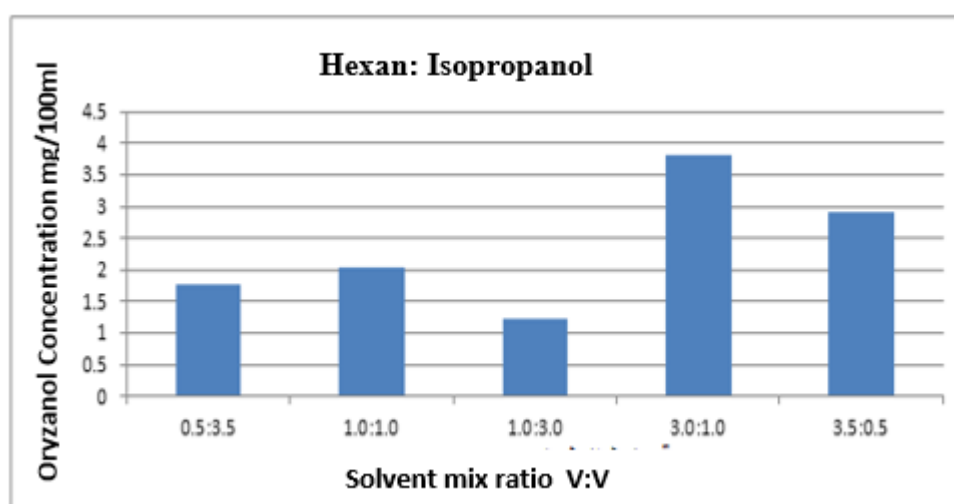
Figure (3) shows the concentrations of the anti-oxidant oryzanol extracted from (Rice bran), as it appears that the concentration of oryzanol when using 15 ml of solvent at a temperature of 40 ° C and for a 30-minute heating period reached 2.42 mg / 100 g either when using The volume of 35 ml on the same extraction conditions amounted to 3.15 mg / 100 g, and when increasing the volume to 55 ml, it was found that the concentration was 3.44 mg / 100 g, and when the volume increased to 75 ml the concentration of the oryzanol extracted was 3.68 mg / ml and the concentration of the extracted oryzanol when increasing volume to 95 ml 4.31 mg / 100 g.



**Figure 3:** Effect of solvent volume on oryzanol extraction.

The reason for increasing the concentration by increasing the volume of the solvent mixture can be attributed to the fact that the opportunity to volatilize the solvents was reduced, thus maintaining the extraction efficiency for as long as possible.

Figure (4) shows the concentration of the antioxidant oryzanol extracted using five ratios of the solvent mixture. It was found that the concentration of the oryzanol reached 1.76 mg / 100 g in extracting ratio of 3.5: 0.5 hexane: isopropanol volume: volume, but when using equal proportions of solvents 1 : 1 hexane: isopropanol volume: volume so the concentration reached 2.04 mg / 100 g, and the concentration of the oryzanol extracted was 1.22 mg / 100 g when using a mixture ratio 3: 1 hexane: isopropanol volume: volume, and the highest concentration of the extract oryzanol reached 3.83 mg / 100 g when using a ratio of 3: 1 hexane: isopropanol volume: volume, and when using a ratio of 3.5: 0.5 hexane: isopropanol volume: volume It was found that the concentration of the extracted oryzanol was 2.91 mg / 100 g.



**Figure 4:** effect of solvent mix ratio of Isopropanol :Hexan on oryzanol extraction.

These results are consistent with what Bemvenuti et al. (2012) stated, that Extraction increases with an increase in the ratio of isopropanol solvent in the isopropanol mixture with hexane. It is known that absorption is directly proportional to concentration and the reason for increased absorption is due to the fact that isopropanol is more stable and less volatile than hexane and thus Extraction efficiency is very high when there is a high proportion of isopropanol compared to hexane.

These results are also consistent with what was mentioned by Xu, Godbar (2000), Zulliaikah and others (2009), as their studies showed a high absorbance of the oryzanol extracted with this mixture, and the reason for the appearance of this percentage of absorbance compared to the previous mixture may be attributed to the effectiveness of the mixture more than effective Use of each solvent separately, as confirmed by Bemvenuti et al. (2012).

However, when using an equal mixture of solvents 1: 1 volume: volume it was observed that the concentration of oryzanol is less than the previous results and this is consistent with what Azrina and others (2008) stated, where they stated that the equal proportions of solvents in the mixture give a better result, because the oryzanol is generally recognized as having High stability in isopropanol compared to hexane. Therefore, when using a solvent mixture of the same size, hexane loses or evaporates during preparation, which makes the gravity of the extraction on isopropanol and thus the quality of extraction is greater.

When a mixture of solvents was used with a greater amount of hexane, it was found that the concentrations of oryzanol at the hexane ratio: isopropanol 1: 3 and 0.5: 3.5 volume: volume reached 12.51 mg / 100 g and 9.77 mg / 100 g. This supports what Lilitchan and others have found. (2008) where they showed in their studies less absorption and less extraction rate for this mixture, and the reason may be due to the quality of the solvents present in the mixture and the volatility of these solvents since isopropanol is less volatile than hexane, but despite the strength of the volatilization of hexane to the large amount in which it is present A comparison with isopropanol makes it the dominant mixture and hence the frying production Because Oryzanol is more persistent in isopropanol compared to hexane.

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### تحديد الظروف المثلى لاستخلاص الاوريزانول من سحالة الرز

علي وليد نوفان واينار زكي ناجي<sup>1</sup>

1 قسم علوم الأغذية- كلية الزراعة- جامعة تكريت- العراق

#### المستخلص

تم تحديد الظروف المثلى لاستخلاص الاوريزانول ، إذ استخدمت خمس درجات حرارة وهي 30 و 40 و 50 و 60 و 70 درجة مئوية وأظهرت النتائج أن درجة الحرارة المثالية للاستخلاص هي 40 م° وخمس اوقات تسخين تم استخدامها 10 و 20 و 30 و 40 و 50 دقيقة وأظهرت النتائج أن وقت التسخين المثالي للاستخلاص هو 20 دقيقة. أما حجم خليط المذيب المستخدم في الاستخلاص فقد تم استخدام خمسة أحجام وهي 15 و 35 و 55 و 75 و 95 مل، ووجد أن أفضل حجم للاستخلاص هو 95 مل ، بالإضافة إلى نسب خليط المذيبات هكسان: ايزوبروبانول (حجم:حجم) 1:1 ، 1:3 ، 3:1 ، 1:3 ، 0.5:3.5 ، 3.5:0.5 ، 0.5:3.5 حجم:حجم واظهرت النتائج ان النسبة المثالية هي 1:3 حجم:حجم .

الكلمات المفتاحية : سحالة الرز ، اوريزانول ، استخلاص ، ظروف الاستخلاص .

