FA Monoalkylesters from Rice Bran Oil by *in situ* Esterification

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ABSTRACT: Extraction and *in situ* esterification of rice bran oil with ethanol were investigated by studying the effects of rice bran oil FFA content and water content of ethanol. Ethyl ester formation in the ethanol phase increased as FFA content increased. Neutral oil solubility in this phase fell considerably, resulting in a high ethyl ester content. The decrease of the water content in ethanol led to an increase in neutral oil solubility in ethanol and promoted the equilibrium of reaction to ethyl-ester formation, resulting in lower FFA content of the product. The main factor that affected yield and monoester content when using high-acidity bran and various monohydroxy alcohols was the solubility of neutral oil in alcohol. The highest monoester content was obtained with methanol.

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FA monoalkyl esters produced by esterification of FA with monohydroxy alcohols are widely used in industry for their lubricating and softening properties. Monoesters, also defined as biodiesel, are a promising alternative diesel fuel and are commercially produced or used around the world, including the United States, Austria, the Czech Republic, France, Germany, Italy, Malaysia, and Sweden (1). Greater use of FA esters as a lubricant has occurred in the last decade (2). It was predicted that in Germany that the market share of this kind of biodegradable lubricating oil would grow from 2 to 3% in 1994 up to 15% by the year 2000 (3).

The production of FA monoesters by alcoholysis of refined TG using alkali catalysis has been studied for several decades, and a major amount of industrial production has been achieved with this method. The process is relatively simple, and both continuous- and batch-operating plants have been developed (4). However, the high production cost of biodiesel relative to petrodiesel has been a major disincentive. The cost of vegetable oils is approximately 80% of the biodiesel production cost (4,5). Therefore, more inexpensive materials, such as unrefined high-acidity oils, soapstocks, and discarded oils are being studied as oil alternatives to reduce production cost (4,6–8).

The production of FA monoesters under alkaline conditions from high-FFA oils is more complicated than when using neutral TG oils because FFA reacts immediately with alkali. Since the acid-catalyzed alcoholysis rate of TG is very low, processing must be in two successive steps of esterification and alcoholysis (4,6,8,9).

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Another approach to produce monoesters from high-acidity oils is *in situ* esterification, i.e., simultaneous oil extraction and acidic esterification or alcoholysis. In this process, alcohol acts as an extraction solvent for oil components and as a reagent to esterify these components (10–12). By using this process the production cost of biofuels may be further reduced since the oil extraction step in the conventional process will be omitted as well. In previous work, *in situ* esterification of rice bran oil with methanol was investigated in detail (12). In this study *in situ* esterifications of rice bran oil with ethanol (96.0 and 99.1%), *n*-propanol, isopropanol, and *n*-butanol were performed to determine the effects of FFA content of rice bran oil, the water content of ethanol, and the chain length of alcohol on *in situ* esterification. In addition to *in situ* esterifications, extractions of rice bran with the same alcohols were investigated in order to understand the mechanism of *in situ* esterification and so to produce pure, cheap alkyl esters in high yield.

EXPERIMENTAL PROCEDURES

Materials. Commercial rice bran, provided by a rice mill (Bereket Celtik, Istanbul, Turkey), was sieved to a particle size of <0.6 mm to remove foreign materials, hull fragments, and broken kernels. The rice bran was stored in a sealed container at room temperature (5–25°C) for 5 mon in order to increase the FFA content of the oil by enzymatic action. Another part of the rice bran was stored at 4°C, also in a sealed container, to inhibit TG hydrolysis.

All the reagents used were of analytical grade (Merck, Darmstadt, Germany) except 96% fermentation-derived ethanol, which was purchased from Turkish State Monopolies (Istanbul, Turkey). Dehydration of ethanol was achieved by drying over calcium oxide (11). The moisture content of dried and distilled ethanol was determined to be 0.9% by Karl Fischer titration (13).

Effects of FFA content of rice bran oil on extraction and in situ *esterification with 96% ethanol.* Samples were taken from the stored rice bran to provide a 14.5–81.0% range of FFA contents. The total bran oil content was determined in duplicate by Soxhlet extraction of the bran with hexane prior to *in situ* esterification. The *in situ* esterification was done by mixing rice bran (50 g) with 200 mL ethanol and 5 mL of concentrated sulfuric acid. The mixture was refluxed for 1 h with magnetic stirring. The mixture was vacuum-filtered, and the filter cake was washed with 100 mL alcohol and dried overnight at room temperature. The residue was reextracted in a Soxhlet apparatus with hexane to obtain the residual bran oil. For the extraction experiment, the same procedure was repeated in duplicate without using sulfuric acid for catalysis. The ratio of residual bran oil to the total amount of oil originally in the bran was calculated as a percentage for both the *in situ* esterification and extraction experiments. The percentage of oil dissolved in alcohol was determined from the difference between the amounts of the original and residual oil samples. The FFA contents of the rice bran oil and the residual oil were determined by standard acid-alkali titration (14), and FFA calculated as oleic acid. By assuming that the oil is composed mainly of TG, partial glycerides, unsaponifiable matter, and FFA, the difference between the amount of oil and the amount of FFA can be calculated as the amount of neutral oil. According to these results, the distributions of the original oil, FFA, and neutral oil between the residual oil and the oil dissolved in alcohol were calculated for both *in situ* esterification and extraction experiments.

Water (100 mL) was added to the filtrate obtained from *in situ* esterification, and the solution was then extracted three times with hexane (3 × 50 mL). The combined extracts were washed with water, dried over sodium sulfate, and evaporated to give the esterified product or crude ester. The ethyl ester content of the crude ester was determined by column chromatography (12). TLC investigations of all the samples obtained during this study, such as original rice bran oil, residual bran oil, oil dissolved in alcohol, and crude ester, were also performed to observe the distribution of oil components between phases (12).

Effect of the water content of ethanol. In situ esterification (5 mL sulfuric acid) and extraction experiments were repeated twice for 1 h using 99.1% ethanol with low- (13.5% FFA) and high-acidity (85.6% FFA) bran samples as described above.

In situ *esterification and extraction of rice bran oil with various monohydroxy alcohols. In situ* esterification (7.5 mL sulfuric acid) and extraction experiments were performed with bran samples containing about 74–80% FFA by using methanol, ethanol (96 and 99.1%), *n*-propanol, isopropanol, and *n*-butanol as described above.

RESULTS AND DISCUSSION

Effect of FFA content of rice bran oil. Table 1 shows the effect of varying bran FFA on the composition of the residual oil after extraction and *in situ* esterification and the compositions of crude ethyl ester obtained. For example, a rice bran sample containing 64.6% FFA was extracted with 96% ethanol under the conditions described above, and after extraction 7.31% of the initial oil remained in the bran, whereas 92.7% dissolved in ethanol. The FFA content of the residual bran oil was 45.5%. After the extraction, 5.15% of the initial FFA was in bran, and 95.5% was dissolved in the ethanol phase; 11.2% of the neutral oil, i.e., total oil components other than FFA, remained in bran and 88.8% was dissolved in ethanol.

When using the same bran for *in situ* esterification, 12.8% of the initial oil, 1.62% of the initial FFA, and 33.2% of the initial neutral oil remained in the brans. This means that more FFA and less neutral oil dissolved in the ethanol phase in *in situ* esterification relative to extraction. These differences are clearly seen in Table 2 where the results were normalized for 100 g of initial rice bran oil. In previous research with *in situ* alcoholysis of soybean oil with 96% ethanol, the reaction proceeded through successive dissolution and alcoholysis of TG, and the overall reaction rate was determined from the extraction and alcoholysis rates (11). This means that as ethyl esters were formed, more TG dissolved in ethanol. In an earlier study of *in situ* esterification of rice bran with methanol (12), the methanol solubility of neutral oil was reduced by the presence of FAME. These two observations seem contradictory, but the data shown in Tables 1 and 2 indicate that ethyl ester formation led to an increase in the amount of FA dissolved in ethanol through dissolution and esterification, and the solubility of neutral oil components was reduced. It is thus possible that *in situ* esterification and *in situ* alcoholysis reactions depend mainly on the solubility of each component of oil in alcohol, which is affected by the presence and concentration of the other components in the medium. The esterification and alcoholysis reaction rates also seem to depend on initial oil and product compositions.

The decrease of neutral oil solubility in ethanol (96%) is not so high as that in methanol in *in situ* esterification of rice bran oil (12). These decreases were found to increase the selectivity of FA, i.e., the ratio of the amount of FA dissolved in alcohol to the amount of neutral oil dissolved in alcohol. For example, after the extraction of rice bran oil having 45.2% FFA with

TABLE 1
Effect of Rice Bran Oil FFA Content on Extraction and *in situ* Esterification with 96% Ethanol

Procedure	Initial content of FFA in oil (%)	FFA content of residual oil in bran (%)	Residual bran: Oil (%)	Residual bran: FA (%)	Residual bran: Neutral oil (%)	Crude ethyl ester: FFA content (%)	Crude ethyl ester: Ester content (%)
Extraction	14.5	3.7	31.5	8.02	35.5		
	25.0	11.6	18.3	8.49	21.6		
	44.0	22.5	14.4	7.36	19.9		
	64.6	45.5	7.31	5.15	11.2		
	81.0	54.6	6.58	4.43	15.7		
In situ esterification	14.5	3.44	33.3	7.86	37.6	7.44	36.6
	25.0	11.20	14.8	6.63	17.5	7.46	48.4
	44.0	6.48	12.9	1.90	21.5	7.87	50.0
	64.6	8.18	12.8	1.62	33.2	9.77	73.8
	81.0	9.21	9.7	1.10	46.4	8.70	78.0

TABLE 2
Amount of Oil, FA, and Neutral Oil Dissolved in 96% Ethanol According to the FFA Content of Rice Bran Oil

	FFA content of initial oil (%)	Oil dissolved in ethanol (g)			
		Oil	FA	Neutral oil	FA/neutral oil
Extraction	14.5	68.5	13.3	55.2	0.24
	25.0	81.7	22.9	58.8	0.39
	44.0	85.6	40.8	44.8	0.91
	64.6	92.7	61.3	31.4	1.95
	81.0	93.4	77.4	16.0	4.84
In situ esterification	14.5	66.7	13.3	53.4	0.25
	25.0	85.2	23.3	61.9	0.38
	44.0	87.1	43.2	43.9	0.98
	64.6	87.2	63.5	23.7	2.68
	81.0	90.3	80.1	10.2	7.85

methanol, the selectivity of FA was 1.88, whereas after *in situ* esterification, the selectivity was 4.87 (12). After the extraction of rice bran oil having 44.0% FFA with 96% ethanol, the selectivity was 0.91, while the same value was 0.98 after *in situ* esterification (Table 2). Thus, to obtain a product having high ethyl ester content, it is necessary to work under conditions that provide the highest FA selectivity. As can be seen in Table 1, the ethyl ester content of crude ester increased with increasing FFA content of rice bran oil, since the FA selectivity also increased with the increasing FFA content of rice bran oil.

Effect of water content of ethanol on extraction and in situ *esterification.* Table 3 shows the results obtained from extractions and *in situ* esterifications of rice brans (13.5 and 85.6% FFA) with 99.1% ethanol when compared with those obtained from rice brans having similar FFA contents and 96% ethanol (Table 1). The distributions of the oil components, which constituted 100 g of initial rice bran oil, in the alcohol phases are given in Table 4. For low-acidity oils, when the water content of ethanol was decreased, the amount of total oil and neutral oil dissolved in alcohol increased considerably even though the amounts of FA dissolved were nearly the same for both extraction and *in situ* esterification. This result indicates that selectivity of FA in 96% ethanol is higher than the selectivity of FA in 99.1% ethanol. It may be possible to produce a crude ester containing higher ethyl ester content with 96% ethanol, since the solubility of neutral oil is lower. However, the ethyl ester content of crude ester obtained with 99.1% ethanol was higher than that obtained with 96% ethanol. Table 3 shows that the FFA content of the crude ester from 99.1% ethanol was 4.92%, whereas the FFA content of crude ester from 96.0% ethanol was 7.44%, since the equilibrium of the esterification reaction shifted to the right in the lower water content medium. From the results for high-acidity oils in Table 3 and 4, we concluded that the crude ester obtained with 99.1% ethanol had higher ethyl ester and lower FFA contents not only because of the equilibrium condition but also because of the decrease in the solubility of neutral oil in ethanol when the medium contained higher amounts of ethyl ester.

In situ *esterification and extraction of rice bran oil with various monohydroxy alcohols.* The results of extractions and *in situ* esterifications of rice brans having high acidity (74–80% FFA) with different alcohols are shown in Table 5. More than 90% of FFA in bran was dissolved in alcohol during extraction regardless of the M.W. of alcohol, and the solubilities of neutral oils in alcohol increased with an increase in the M.W. of the alcohols. The difference between the amounts of neutral oil dissolved in *n*-propanol and isopropanol depends on the difference in their dielectric constants. Comparison of extraction and *in situ* esterification results indicates that the *in situ* esterification reaction proceeds through dissolution and esterification of FFA. As monoesters form, dissolution of neutral oil components is reduced considerably. Since the mono-

TABLE 3
Effect of Ethanol Concentration on Extraction and *in situ* Esterification of Rice Bran Oil

	Initial content of FFA in oil (%)	Alc. conc. (%)	FFA content of residual oil in bran (%)	Residual bran			Crude ethyl ester	
				Oil (%)	FA (%)	Neutral oil (%)	FFA content (%)	Ester content (%)
Extraction	14.5	96.0	3.70	31.5	8.03	35.5		
	13.5	99.1	7.26	17.0	9.14	18.2		
	81.0	96.0	54.6	6.58	4.43	15.7		
	85.6	99.1	80.5	5.66	5.33	7.6		
In situ esterification	14.5	96.0	3.44	33.3	7.86	37.6	7.44	36.6
	13.5	99.1	6.00	10.1	4.49	11.0	4.92	42.5
	81.0	96.0	9.21	9.70	1.10	46.4	8.70	78.0
	85.6	99.1	7.10	7.60	0.63	49.0	6.24	80.8

TABLE 4
Amount of Oil, FA, and Neutral Oil Dissolved in Ethanol (96 and 99.1%) According to the FFA Content of Rice Bran Oil

Procedure	Initial FFA content of oil (%)	Alcohol conc. (%)	Oil dissolved in ethanol (g) Oil	FA	Neutral oil	FA/Neutral oil
Extraction	14.5	96.0	68.5	13.3	55.2	0.24
	13.5	99.1	83.0	12.2	70.8	0.17
	81.0	96.0	93.4	77.4	16.0	4.84
	85.6	99.1	94.3	81.0	13.3	6.10
In situ esterification	14.5	96.0	66.7	13.3	53.4	0.25
	13.5	99.1	89.9	12.9	77.0	0.17
	81.0	96.0	90.3	80.1	10.2	7.85
	85.6	99.1	92.4	85.1	7.3	11.58

TABLE 5
Extraction and *in situ* Esterification of Rice Bran Oil with Various Monohydroxy Alcohols

Procedure	Alcohol	Temperature (°C)	Initial content of FFA in oil (%)	FFA content of residual oil in bran (%)	Residual bran Oil (%)	Residual bran FA (%)	Residual bran Neutral oil (%)	Crude ethyl ester FFA content (%)	Crude ethyl ester Ester content (%)
Extraction	Methanol	65	74.4	34.0	12.10	5.52	31.2		
	Ethanol (96%)	78	79.9	34.9	6.68	2.92	21.6		
	Ethanol (99.1%)	78	78.4	60.0	5.46	4.18	10.1		
	Isopropanol	82	74.7	69.7	6.18	5.77	7.4		
	n-Propanol	97	74.7	71.9	9.98	9.60	11.1		
	n-Butanol	117	77.1	76.3	6.84	6.77	7.1		
In situ esterification	Methanol	65	74.4	3.31	25.20	1.12	95.2	2.70	85.8
	Ethanol (96%)	78	79.9	12.6	8.89	1.40	38.7	8.80	76.4
	Ethanol (99.1%)	78	78.4	11.8	6.71	1.01	27.4	6.91	78.3
	Isopropanol	82	74.7	14.6	7.05	1.38	23.8	11.9	69.7
	n-Propanol	97	74.7	10.0	14.90	1.99	53.0	5.20	74.8
	n-Butanol	117	77.1	10.5	12.80	1.91	49.5	7.83	68.0

ester content of the crude ester fraction depends mainly on neutral oil solubility, the highest monoester content can be obtained from *in situ* esterification of the highest-acidity rice bran with methanol. In contrast, the monoester content of crude ester increased with increasing of monoalcohol M.W. from *in situ* alcoholysis of soybean oil (11).

By *in situ* esterification of high-acidity rice bran oil, which cannot be employed directly in foods, the oil can be recovered from the bran as crude FA alkyl esters in a one-step process.

REFERENCES

1. Knothe, G., Historical Perspectives on Vegetable Oil-Based Diesel Fuels, *inform 12*:1103–1107 (2001).
2. Kohashi, H., Application of Fatty Acid Esters for Lubricating Oil, *Proceedings of World Conference on Oleochemicals into the 21st Century,* edited by T.H. Applewhite, American Oil Chemists' Society, Champaign, 1990, pp. 243–250.
3. Wu, X., X. Zhang, S. Yang, C. Chen, and D. Wang, The Study of Epoxidized Rapeseed Oil Used as a Potential Biodegradable Lubricant, *J. Am. Oil Chem. Soc. 77*:561–563 (2000).
4. Murayama, T., Evaluating Vegetable Oils as a Diesel Fuel, *inform 5*:1138–1145 (1994).
5. Cuperus, F.P., G. Boswinkel, and J.T.P. Derksen, The Processing of New Oilseed Crops—An Economic Evaluation, *J. Am. Oil Chem. Soc. 73*:1635–1640 (1996).
6. Haas, M.J., and K.M. Scott, Combined Nonenzymatic-Enzymatic Method for the Synthesis of Simple Alkyl Fatty Acid Esters from Soapstock, *Ibid. 73*:1393–1401 (1996).
7. Nelson, L.A., T.A. Foglia, and W.N. Marmer, Lipase-Catalyzed Production of Biodiesel, *Ibid. 73*:1191–1195 (1996).
8. Ghosh, S., and D.K. Bhattacharyya, Utilization of Acid Oils in Making Valuable Fatty Products by Microbial Lipase Technology, *Ibid. 72*:1541–1545 (1995).
9. Choo, Y.M., K.Y. Cheah, A.N. Ma, and A. Halim, Conversion of Crude Palm Kernel Oil into Its Methyl Esters on a Pilot Plant Scale, *Proceedings of World Conference on Oleochemicals into the 21st Century,* edited by T.H. Applewhite, American Oil Chemists' Society, Champaign, 1990, pp. 292–295.
10. Özgül, S., and S. Türkay, *In situ* Esterification of Rice Bran Oil with Methanol and Ethanol, *J. Am. Oil Chem. Soc. 70*:145–147 (1993).
11. Kıldıran, G., S. Özgül-Yücel, and S. Türkay, *In situ* Alcoholysis of Soybean Oil, *Ibid. 73*:225–228 (1996).
12. Özgül-Yücel, S., and S. Türkay, Variables Affecting the Yields of Methyl Esters Derived from *in situ* Esterification of Rice Bran Oil, *Ibid. 79*:611–614 (2002).
13. Mitchell, J., Aquametry, in *Encylopedia of Industrial Chemical Analysis,* edited by F.D. Snell and C.L. Hilton, Interscience Publishers, New York, 1966, Vol. 1, pp. 142–147.
14. Paquot, C., and A. Hautfenne, *Standard Methods for the Analysis of Oils, Fats and Derivatives,* 7th edn., Blackwell Scientific Publications, Oxford, 1970, pp. 73–77.

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