Journal of Oleo Science
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J. Oleo Sci. ***59****, (5) 229-233 (2010)*

Chemical Synthesis and Characterization of Palm Oil-Based Difatty Acyl Thiourea

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Abstract: Difatty acyl thiourea (DFAT), which has biological activities as antibiotics and antifungal, has been synthesized from palm oil and thiourea using sodium ethoxide as catalyst. Ethyl fatty ester (EFE) and glycerol were produced as by-products. The synthesis was carried out by reflux palm oil with thiourea in ethanol. In this process, palm oil converted to about 81% pure DFAT after 11 hour and molar ratio of thiourea to palm oil was 6.0: 1 at 78℃. Elemental analysis, Fourier transform iInfrared (FTIR) spectroscopy and ^{1}H nuclear magnetic resonance (NMR) technique were used to characterize both DFAT and EFE.

Key words: palm oil, thiourea, ethyl fatty esters, sodium ethoxide

1 INTRODUCTION

Thiourea and its derivatives have attracted much attention due to their antifungal activities, antibacterial and insecticidal properties particularly against tubercle bacillus[1,2]. They were also used as surfactants, lubricants, detergents and antifoams in industrial applications[3,4].

Benzoyl thiourea derivative of chitosan was prepared and investigated its antifungal efficacy against sugar-beet pathogens, and showed that the antifungal activity of the derivative was much better than that of native chitosan[5]. Synthesis of heterocyclic skeletons by the reaction of N1-(2-Cyanophenyl)-benzimidoyl chloride with thioamides was reported by Walid *et al*. (2002)[6]. 5-substituted 3-aminothiophene-2- thioamides[5] from 3-amino-2,4- pentadienthioamides[4] by [C-C-C-C] + [-S]-addition were synthesized and charactrized[7].

Three different acyl thiourea derivatives of chitosan (CS) were synthesized and their structures were characterized by FT-IR spectroscopy and elemental analysis the degree of grafting of the acyl thiourea group in the derivatives was related to antifungal activity; higher substitution resulted in stronger antifungal activity[8]. Our literature search shows that no information regarding synthesis of difatty acyl thiourea directly from palm oil.

In this study, difatty acyl thiourea has been synthesized using palm oil with thiourea in presence of sodium ethoxide as a catalyst. The palm oil is a mixture of triacylglycerides, just like any ordinary fat, which are esters of glycerol with different saturated and unsaturated fatty acids. Malaysia is currently the world's largest producer and exporter of palm oil and it is the major source of vegetable oil for industrial application[9].

2 EXPERIMENTAL

2.1 Materials

Palm oil was obtained from Ngo Chew Hong Oils and Fats (M) Sdn. Bhd., Malaysia. Thiourea, ethanol and sodium metal were purchased through local suppliers from Merck, Germany.

2.2 Synthesis of DFAT

In a 250 mL round-bottomed flask fitted with a reflux condenser and a magnetic stirrer, finely cut sodium (173 mmol) was dissolved in 100 mL of super-dried ethanol. After all the sodium has reacted, palm oil was then added into this solution followed by dried thiourea (which was obtained by drying at 60℃ for 4 h) dissolved in 50 mL of hot (70℃) ethanol at different molar ratio of substrates presented in Table 2. After mixing by shaking, the mixture was refluxed for 11 h on an oil bath at 78℃ . The contents of the flask were cooled to room temperature, transferred into a separatory funnel and allowed to settle over night.

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Accepted December 7, 2009 (received for review November 8, 2009)
Journal of Oleo Science ISSN 1345-8957 print / ISSN 1347-3352 online
http://www.jstage.jst.go.jp/browse/jos/

The bottom layer comprised of glycerol was removed. The top layer which comprised of the products, was poured into a beaker, and mixed with 100 mL of hot distilled water (60℃) and 10 mL of concentrated hydrochloric acid, and stirred for 15 min. Difatty acyl thiourea solution was separated from the white mass of ethyl fatty ester by filtering. The clear solution containing difatty amide was then cooled in an ice bath. The pale yellow product was collected on a Buchner funnel and washed with 50 mL of cold water and then dried in a vacuum desiccator over phosphorous pentoxide. The conversion of palm oil into DFAT was about 81%. The preparation reaction is shown in the Scheme 1.

The proposed mechanism of palm oil into DFAT and EFE is shown in Scheme 2. By using the base catalyst (sodium ethoxide), the first step leads to proton withdrawing from (-NH_2) in thiourea molecule, as proposed in our system, (Scheme 2A). As illustrated in Scheme 2B, the thiourea ion attacks one carbonyl of the triglyceride group and forms N-carbamoyl fattythioamide which attacks the second carbon thioamide in the glycerides group after proton withdrawing from it by base catalyst. After two consecutive reactions, 2,3-dihydroxypropyl fatty ester, 1,3 dihydroxypropan-2-fatty ester (as intermediates) and difatty acyl thiourea are formed[10]. In the last step (Scheme 2C), the alkoxy group attacks the third carbonyl of the triglyceride group and forms ethyl fatty ester and glycerol[11].

2.3 Characterizations

Qualitative analysis of amide groups of DFAT was carried out by observing the colored complex formed after the ethanolic solution of the DFAT reacted with copper (II). The presence of amide in DFAT was also determined by FTIR spectra. FTIR spectra in the range 4000-280 cm^{-1} were obtained using Perkin-Elmer 1650 infrared fourier transform spectrometer by KBr pellet technique.

Elemental analyzer (LECO CHNS-932) was used for quantitative analysis of nitrogen contents. The determination was carried out under O_2 and N_2 atmospheric conditions using sulfamethazine as a standard.

The presence of amide (CO NH) proton in DFAT was determined by ^1H Nuclear Magnetic Resonance (Joel, Japan) using $CDCl_3$ as a solvent.

3 RESULTS AND DISCUSSION

3.1 Identification of DFAT and EFE

3.1.1 FTIR spectra

Characteristic bands of palm oil were observed at 3012, 2924, 2858, 1741, 1465, 1163, and 722 cm^{-1} resulting from C-H stretching of CH=CH, C-H asymmetric stretching of CH_2, C-H symmetric stretching of CH_2, C=O stretching of ester (glyceride), CH_2 scissoring, O-C-C stretching, and CH_2 rocking, respectively[12] DFAT spectra show new bands at 3349, 1626, 1049 cm^{-1} and 946 cm^{-1}attributed to N-H stretching, C=O stretching, C-N stretching of amide, C=S stretching, respectively. The disappearance of peaks at 1745 and 1163 cm^{-1} and appearance of peaks at 3349, 1626 and 1049 cm^{-1} indicate that fatty amides have been formed[13]. EFEs spectra show the presence of the major absorption bands of palm oil with disappearance of peaks which correspond to amide.

3.1.2 ^1H NMR spectra of DFAT

(400MHz) ($CDCl_3$): δ 0.88 (t, J = 8.8 Hz, 6H, 2 x CH_3), 1.24 (m, H, CH_2), 1.53 (4H, 2 x CH_2 CH_2 C=O NH), 2.19 (4H, 2 x CH_2 CH = CH), 2.33 (t, J = 10.9 Hz, 2H, 2 x CH_2 CO NH), 5.87 (2H, CH = CH), 10.8 (br, s, 2H, CO NH).

3.1.3 ^1H NMR spectra of EFEs

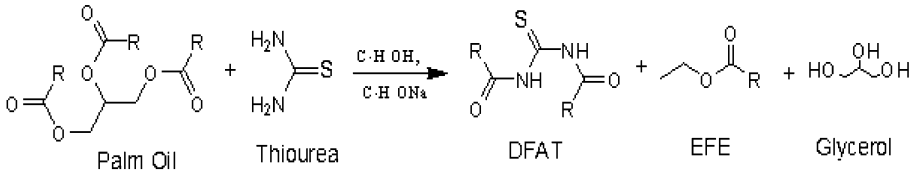
(400MHz) ($CDCl_3$): δ 0.88 (t, 3H CH_3), 1.27 (m, H, CH_2), 1.28 (t, 3H CH_3), 1.64 (2H, CH_2 CH_2 C=O), 2.19 (4H, 2 x CH_2 CH = CH), 2.34 (t, 2H, CH_2 C=O), 4.14 (q, 2H, CH_2 -O), 5.83 (2H, CH = CH).

3.1.4 Elemental analysis

Based on the average fatty acid composition for palm oil reported by Tan *et al*. (1981)[14], the theoretical nitrogen content of the DFAT is 5.12%. The nitrogen content of the DFAT was 5.01% analyzed by elemental analysis while there was no significant presence of nitrogen in EFE.

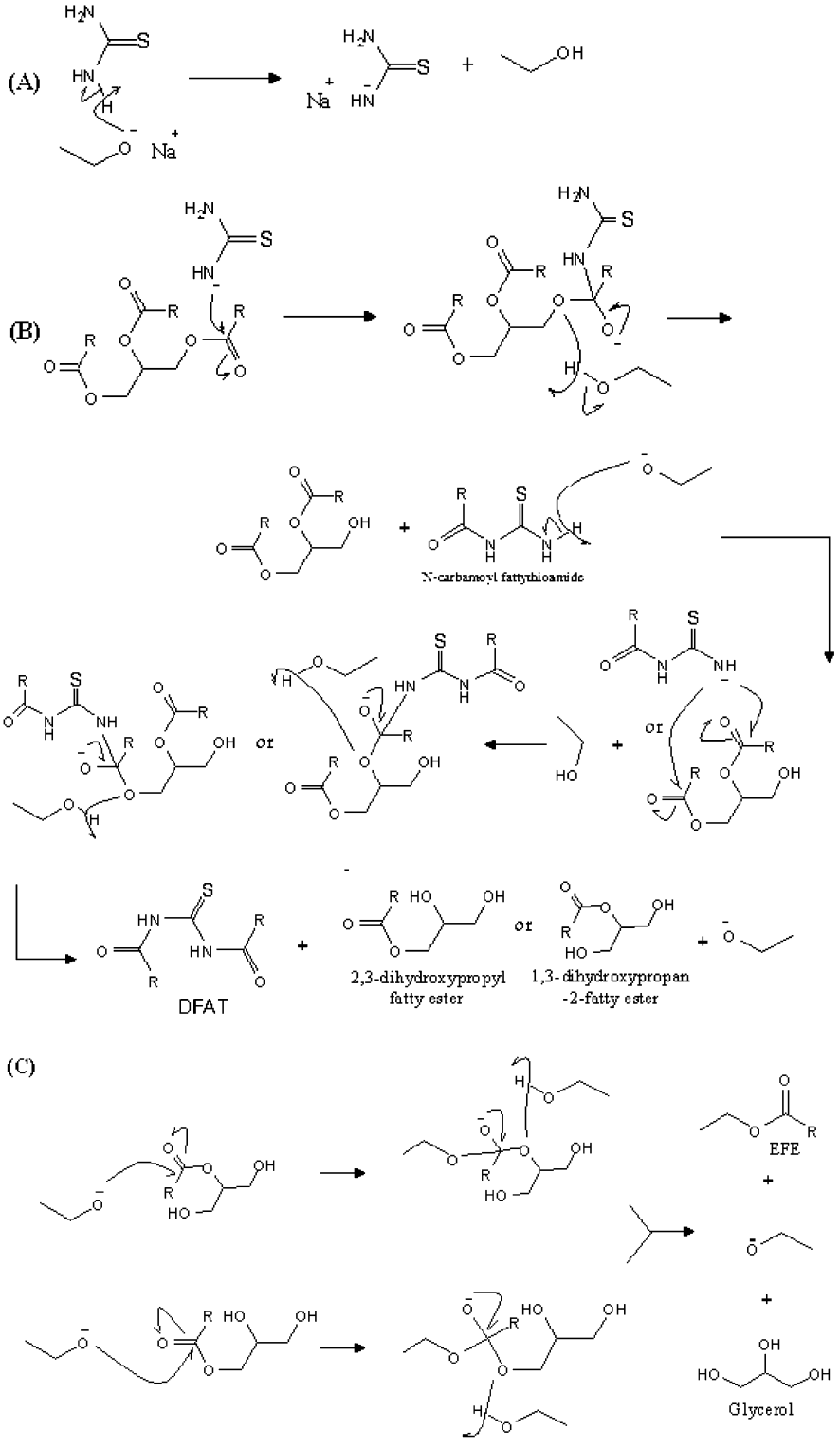
3.2 Optimum reaction conditions

Difatty acyl thiourea was prepared with different molar ratios of thiourea and palm oil. The effect of molar ratio is given in Table 1. Molar ratio of thiourea to palm oil as 6.0:1.0 was considered optimum. The reaction time was optimized from 1 h to 15 h. The effect of reaction time is shown in Table 2, in this table, it is found that 11 h is the appropriate time for highest conversion of palm oil into difatty acyl thiourea. In this reaction, catalyst plays an important role, the percentage of catalyst (sodium ethoxide) to palm oil were studied. Table 3 illustrates the effect



R= C_{12}: 0, C_{14}: 0, C_{16} : 0, C_{18} :0, C_{18} : 1, C_{18} : 2, C_{18} : 3, C_{20} : 0.

Scheme 1 DFAT Synthesis from Palm Oil



Scheme 2 Proposed Catalytic Cycle of Sodium Ethoxide in the Amidation and Transesterification of Palm Oil: (A) Proton Withdrawing from Thiourea, (B) Amidation and (C) Transesterification.

Table 1 Conversion of Palm Oil as a Function of Reaction Time: Reaction Conditions: Thiourea/Palm Oil Molar Ratio=6.0:1.0, and Reaction Temperature=78˚C

Reaction Time (h)	Conversion of palm oil into difatty acyl thiourea (%)
1	6.23
2	17.41
3	23.92
4	30.26
5	36.17
6	41.13
7	51.68
8	58.44
9	69.27
10	74.15
11	81.46
12	81.92
13	82.25
14	82.78
15	82.95

Table 2 Conversion of Palm Oil as a Function of Thiourea/Palm Oil Molar Ratio. Reaction conditions: reaction time=11 h and reaction temperature=78˚C

Thiourea/Palm oil (mmol:mmol)	Conversion of palm oil into difatty acyl thiourea (%)
1:1	10.63
2:1	23.91
3:1	41.53
4:1	59.22
5:1	67.42
6:1	81.15
7:1	78.23
8:1	75.41

of catalyst amount to palm oil. Molar ratio of catalyst to palm oil as 5.0: 1.0 gave the maximum conversion.

4 CONCLUSION

This study shows that difatty acyl thiourea was successfully synthesized by refluxing palm oil, ethanol and thiourea in the presence of sodium ethoxide as catalyst. In addition to glycerol, ethyl fatty esters were produced as the side products. The best suitable conditions of reaction were investigated. It was found that at 78℃ temperature, 6.0: 1.0 molar ratio of thiourea to palm oil, 5.0: 1.0 molar ratio of catalyst (sodium ethoxide) to palm oil and 11 h reaction time were the best suitable conditions of reaction. Qualitative analysis of complex formation, FTIR spectroscopy, ^{1}H NMR technique and elemental analysis results confirm the formation of difatty acyl thiourea.

Table 3 Effect of Catalyst Loading on Conversion of Palm Oil. Reaction conditions: Thiourea/palm oil molar ratio=6.0:1.0, reaction time=11 h and reaction temperature=78˚C

Thiourea/Palm oil (mmol:mmol)	Conversion of palm oil into difatty acyl thiourea (%)
1:1	9.18
2:1	21.42
3:1	46.13
4:1	68.36
5:1	81.92
6:1	81.31
7:1	80.52
8:1	80.11

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