Fractionation of Fatty Acid in Palm Kernel Oil using Supercritical CO₂ at Different Pressures and Temperatures

On-line reference number: 1035

Md Zaidul I.S.¹, Nik Norulaini N.A², Ariffin A.³, Mohd Omar, A.K.³, Hiroshi Inomata¹, *Richard L. Smith¹

¹Department of Chemical Engineering, Research Center of Supercritical Fluid Technology, Tohoku University Aoba-ku, Aramaki Aza, Aoba-04, Sendai 980-8579, Japan
² School of Distant Education, ³School of Industrial Technology, Universiti Sains Malaysia, 11800 P. Pinang, Malayisa
*Tel. & Fax: +81-(22) 217-5863; E-mail: smith@scf.che.tohoku.ac.jp

ABSTRACT

Application of supercritical carbon dioxide (SC-CO₂) to fractionate shorter-medium (C8-C14) and longer chain (C16-C18:2) triglycerides in terms of fatty acids constituent in palm kernel oil (PKO) was studied for formulating possible cocoa butter replacer blends. The extraction of dehulled ground PKO was carried out at 40 and 80°C and pressures ranging from 20.7 to 48.3 MPa with a flow type apparatus. The PKO was extracted successively into four fractions. At higher temperatures (80 °C) the total yield of PKO increased with pressure. The highest yield (99.6%) was obtained at 48.3 MPa and 80°C with minimum CO₂ used. Much shorter chain C8, C10, C12 fatty acids constituents were extracted fraction 1. Longer chain 16, C18:0, C18:1 and C18:2 fatty acid constituents were extracted in fraction 4. The slip melting points were reduced to 6 - 8 °C for fractionated PKO in the later fractions. The latter fractions had composition and slip melting points close to that of cocoa butter replacer blends.

KEYWORDS: Cocoa butter replacers blend, fatty acid constituents, palm kernel oil, supercritical fractionation
INTRODUCTION

Palm kernel oil (PKO) is regarded as food-grade oil that is of high quality. Its application could be greatly extended if PKO components could be fractionated or modified to have properties that are closer to those of cocoa butter replacers (CBRs), which have a very high value in the confectionary market. Fractionation of PKO is made possible by solubility differences between the triglycerides component. A convenient and environmental friendly solvent for reducing the shorter and medium chain (C8-C14), and increasing the longer chain (C18:0-C18:2) fatty acid constituents in PKO is CO2 (Zaidul, 2003, Norulaini et al., 2004a, 2004b). Some studies have been reported on fractionation of PKO using SC-CO2. Hassan et al. (2000, 2001), Norulaini et al. (2004a, 2004b) and Zaidul (2003) studied the fractionation of PKO using SC-CO2 at high pressures (<50 MPa) and a temperatures from 40 to 80 °C. Ragunath et al. (1992, 1993) also fractionated fatty acids and triglycerides in palm kernel oil based on their carbon number using SC-CO2 at temperature range of 40–80 °C and pressures up to 30 MPa. They separated fatty acids and triglycerides of different carbon numbers of C12, C18:1. According to Taylor (1996) supercritical fluids are of particular interest to the fats and oil analyst because of the high solubility exhibited by lipid material in supercritical CO2. Given that nutrition labeling is becoming common in many countries regarding total saturated and unsaturated fat content in packaged foods, modified that can be achieved with CO2 is of high industrial interest.

Vegetable oils consist mainly of triglycerides, triacylglycerols of fatty acids. Triglycerides are characterised by their carbon number, which is total number of carbon atoms in the three constituent fatty acids. Triolein (glyceroltriuoleate) with carbon number 54 is their typical constituent, because C18:1 constituent is the most abundant acid in vegetable oils and it is very good for confectionary fat like cocoa butter fat due to its lower melting point. On the contrary, some shorter and medium chain fatty acids constituents degrade this type of confectionary fat and thus, some type of fractionation or separation is needed to modify edible fats or to formulate CBR blends.

CBRs are those fats, which replace cocoa butter partially or wholly in chocolate products. CBRs are further classified as i) cocoa-butter-equivalents, which behave like cocoa butter and are able to blend in cocoa butter in any proportion without altering the physico-chemical characteristics of cocoa butter; ii) Cocoa-butter-substitute, which are fats, and can be blended with cocoa butter to a limited extent, without significantly altering its physico-chemical characteristics.
In cocoa butter, the C12 and C14 fatty acid constituents are present as a trace or very low amounts, whereas the C16, C18:0 and C18:1 constituents are high. On the contrary, in PKO the C12 constituent is high, and C18:0 and C18:1 constituents are relatively low compared with cocoa butter. Since CO2 has been used to fractionate various oils, it may be possible to use this solvent to decrease the C12 content and to increase the C18:0-C18:1 content among the various fractions. The objective of this study is to fractionate the triglycerides of PKO in terms of fatty acid components using SC-CO2 to achieve fractions that might be suitable for use in formulations for CBR blends.

MATERIALS AND METHODS

MATERIALS

Palm kernels were collected from Malpom Sdn. Bhd., Nibong Tabal, Penang. Commercial liquefied carbon dioxide (99.9% purity), carrier gas (nitrogen, 99.9% purity), and auxiliary gases hydrogen (99.9% purity) and compressed air (free from organic impurities) were purchased from Malaysian Oxygen Ltd. (Penang). n-Hexane, AR Grade obtained from R & M Marketing U.K. Reference standards of methyl ester of caprylic acid (C8), capric acid (C10), lauric acid (C12), myristic acid (C14), palmitic acid (C16), stearic acid (C18), oleic acid (C18:1) and linoleic acid (C118:2) with the purity of 99% of each, and sodium methoxide were obtained from Sigma, USA. TC-Wax column (30 m × 0.25 mm) was bought from Science Inc., Tokyo.

METHODS

Preparation of Palm Kernel for Experiment

The palm kernels were prepared for the experiments by dehulling and grinding according to a procedure developed by Zaidul (2003) as briefly described here. About 500 g of palm kernels were soaked into 500 ml 12 M hydrochloric acid and then heated to 90°C for 20 minutes. After heating the kernels for 30 minutes, they were kept for another 10 min at room temperature and then washed with water to remove the acid before minimum pressure was applied to the kernels to remove the testa. The dehulled palm kernels were ground into small pieces 0.5-1.5 mm.
Extraction of Palm Kernel Oil using Soxhlet Method

Soxhlet extraction used for comparison with supercritical extraction was carried out in duplicate for 15 g of palm kernel with 200 mL n-hexane for 6 hours. The sample was then dried in the oven at 103 ± 1 °C for 2 hours after which it was cooled in desiccators before reweighing.

Characterisation of Palm Kernel

The determination of moisture content was carried out according to PORIM test method, no. p5.2, 1985. The moisture content of dehulled ground palm kernel was found to be 4.87%. The total oil determined in the dehulled ground palm kernel was found to be 50.12 ± 0.2% (g oil extracted/100 g of dehulled ground palm kernel) on dry basis by Soxhlet extraction.

Supercritical Fluid Extraction

The experimental set-up for the supercritical fluid extraction process is shown in Figure 1. It consisted of a pump (American Lewa, Holistic, Massachusetts) with a maximum capacity of 68.9 MPa (10000 psi), an oven (S.C.S.I. Instrument System), a chiller (B/L-730, Yih Der, Taipei, Taiwan) and a 50 cm extraction cell with 13 mm diameter and 320 mm height, and a wet gas meter (WNK-1A, Sinagawa Corp., Tokyo).

Liquid carbon dioxide was pumped into the heated extraction cell loaded with approximately 20 g (dry basis) of dehulled ground palm kernel, of less than 2 mm diameter in size. Pressures ranging from 20.7 to 48.3 MPa and temperatures of 40 and 80°C were used. At each temperature-pressure combination, the extraction was run continuously for a total of 40 minutes. The oil was collected at the end of every 10 minutes giving a total of 4 fractions. The CO₂ flow was varied in order to maintain the desired pressure and temperature of the extractor and the volume of CO₂ passed through the extraction cell was recorded at atmospheric pressure and temperature at the end of each 10 minutes run using a wet gas meter. The yields of oil were defined in percent on 100g dry palm kernel basis).
Figure 1. Schematic diagram of supercritical fluid extraction of palm kernel oil

\[
\text{Total oil yield (g) of four successive fractions by SC-CO}_2 \times 100% \\
= \frac{\text{Total oil yield extracted (g) by Soxhlet}}{\text{Total oil yield extracted (g) by Soxhlet}} 
\]

\[
\text{Fractional oil yield (%)} = \frac{\text{Oil yield (g) of 10 min. interval by SC-CO}_2}{\text{Total oil yield (g) extracted by Soxhlet}} \times 100% 
\]

The density of CO\textsubscript{2} at different temperature and pressure was referred to NIST database and the total CO\textsubscript{2} expressed in dm\textsuperscript{3} (liter). Flow rate = CO\textsubscript{2} used (dm\textsuperscript{3}) / minute. The flow rate of the CO\textsubscript{2} used ranged from 1.5 to 3.5 dm\textsuperscript{3} / min at 40 °C and 1.2 to 3.81 dm\textsuperscript{3} / min at 80 °C.
Analysis of Oil Components

Each fraction of oil was analysed to determine triglycerides in term of determining fatty acid components. A 100 µL of the test sample was thoroughly mixed with 1 ml n-hexane and 1 µL sodium methoxide in a 2 ml vial, and then shaken vigorously. The clear upper layer of methyl ester was pipetted off and injected into a GC using external standard method, PORIM test method, no. p3.4, 1995. A gas chromatograph (Model, G-3000, HITACHI) was used to determine the fatty acids profile. The oven temperature was set at 190°C, the detector and injector temperature held constant at 250 °C. Carrier gas flow rate was 1 ml/min with a split ratio of 1:100. Chromato-Integrator (HITACHI, D-2500) and an auto injector (AI-1000) were used according to PORIM test method, no. p3.5, 1985. Melting points of fractional oils were determined by PORIM test method, no. p4.2, 1995.

RESULTS AND DISCUSSION

Table 1 shows the fractional and total oil yield in percent at different pressures and temperatures in 10 min intervals.

Table 1: Fractional and Total Oil Yield (%) Versus Time for Successive Fractions (10 min intervals), and CO₂ Required at Different Pressures and Temperatures

<table>
<thead>
<tr>
<th>Pressure (MPa)</th>
<th>20.7</th>
<th>27.6</th>
<th>34.5</th>
<th>41.4</th>
<th>48.3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temp. (°C)</td>
<td>40</td>
<td>80</td>
<td>40</td>
<td>80</td>
<td>80</td>
</tr>
<tr>
<td>Fraction No.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>9.8</td>
<td>8.4</td>
<td>26.2</td>
<td>19.3</td>
<td>37.1</td>
</tr>
<tr>
<td>2</td>
<td>8.8</td>
<td>8.3</td>
<td>22.5</td>
<td>16.9</td>
<td>18.4</td>
</tr>
<tr>
<td>3</td>
<td>8.7</td>
<td>8.1</td>
<td>12.3</td>
<td>14.9</td>
<td>16.9</td>
</tr>
<tr>
<td>4</td>
<td>8.6</td>
<td>8.5</td>
<td>10.2</td>
<td>15.1</td>
<td>8.9</td>
</tr>
<tr>
<td>Total oil yield (%)</td>
<td>35.9</td>
<td>33.3</td>
<td>71.2</td>
<td>66.2</td>
<td>81.3</td>
</tr>
<tr>
<td>Total CO₂ used (g)</td>
<td>134</td>
<td>155.5</td>
<td>120</td>
<td>144</td>
<td>112</td>
</tr>
</tbody>
</table>
The total oil yield was calculated from the total of four successive fractions of total 40 min. Total oil yield (Table 1) was the lowest at 20.7 MPa and highest at 48.3 MPa. An increase in pressure increased the extraction yield at a given temperature. Table 1 also shows that the fraction 1 generally had the highest yield for all pressures. The yield gradually decreased with time due to higher concentration of the longer chain components. This trend was observed for all the pressures except 20.7 MPa, where it was found to be almost same for each fraction (Table 1). On the other hand, for pressures ranging from 27.6 MPa to 48.3 MPa, yield varied greatly between the fractions. The solubility of fatty acids in SC-CO$_2$ extraction depends on the length of the hydrocarbon chain and the presence of functional groups, as well as on the effect of the extraction parameters, pressure and temperature (Stahl et al., 1988). At 80 °C relatively higher yields were obtained in the fractions 1 and 4. In Table 1, it was observed at constant temperature that the total CO$_2$ used decreased with increasing pressure and the same trend was observed for both temperatures. The rate of the reduction of CO$_2$ was found to be the highest at 48.3 MPa and 80 °C. High yields could thus be achieved with small amounts of CO$_2$ for the highest pressures.

Figure 2 shows the effect of pressure and temperature on the total oil yield obtained from the total of four successive fractions. The yield increased with pressure for both temperatures. However, yields at 80 °C were higher than those at 40 °C at the higher pressures. The highest yield (99.6%) was obtained at 80 °C and 48.3 MPa. Bisunadan (1993) carried out supercritical extraction of palm oil from fruits and obtained the highest yield when the fruits were extracted at 50 MPa and 80 °C. At high pressures and temperatures, density and the solubility increased exponentially due to the exponential increase in the vapour pressure of the oil (Bisunadan, 1993).

Figure 3 shows a comparison of triglycerides in terms of fatty acids in the Soxhlet extracted PKO, commercial cocoa butter (CB) and PKO extracted in the fraction 1 via SC-CO$_2$ at 80 °C. The C8, C10, C12 and C14 fatty acid constituents were found to increase with increasing pressure. The C16, C18:0, C18:1 and C18:2 fatty acid constituents decreased with increase in pressure. As the pressure became greater, more C8 to C14 fatty acids were extracted in fraction 1. Conversely, fatty acids such as C16 to C18:2 yields were low from 34.5 to 48.3 MPa. At higher pressures (34.5 to 48.3 MPa) large differences were found in the fatty acid constituents.

Figure 4 shows the triglycerides composition in terms of fatty acids of PKO extracted in fraction 4 at 48.3 MPa and 80 °C. The fatty acids of C8 to C14 reduced drastically, and C16 and C18:2 increased remarkably with increase in pressure and temperature. As the temperature and pressure of the extraction increased, the solubility
of the longer chain (C16-C18:2) fatty acids increased on the other hand the solubility of shorter and medium chain (C8-C14) fatty acids decreased simultaneously. The increase in the amount of long chain fatty acids present in the extract was likely due to the increased solubility with time and influenced by the extraction conditions. Thus, the shorter chain fatty acids seemed to have high extractability for all conditions used in the extractions.

Figure 2. Total oil yield (%) extracted in four successive fraction at 40 °C and 80 °C at different pressures
Figure 3. Comparison of fatty acid composition in commercial CB (Kheiri, 1982) and PKO Soxhlet, and SC-CO₂ extracted in the 1st fraction (10 min) at various pressures and 80 °C

In comparing results between fraction 1 (Figure 3) and fraction 4 (Figure 4), it is important to note that the C12 constituent yield in the fraction 4 was markedly reduced, while that of C18:1 exhibited a large increase in yield. This trend is closer to the fatty acid composition of cocoa butter. Although the short and medium chain fatty acids (C8-C14) were still present in SC-CO₂ extracted PKO at pressure higher than 34.5 MPa and 80 °C, further studies could be conducted to reduce these short and medium chain fatty acids by blending with other oils that contain more unsaturated fatty acid constituents.
The slip melting points (SMP) of fraction 1, fraction 2, fraction 3 and fraction 4 were found to be 28.6, 27.2, 21.5 and 20.3, respectively at 48.3 MPa and 80 °C. The SMP reduction in the later fraction was most likely due to increase of unsaturated fatty acid constituents. Since fraction 4 possessed the lowest SMP, it is more suitable for blending to formulate CBRs blends. CBRs of good quality should have a sharp melting point like cocoa butter but be hard and brittle at room temperature (Kheiri, 1982).

Figure 4. Comparison of fatty acid composition in commercial CB (Kheiri, 1982) and PKO Soxhlet, and SC-CO₂ extracted in the last fraction (40 min) at various pressures and 80 °C
CONCLUSIONS

Supercritical carbon dioxide can be used to fractionate palm kernel oil to decrease C8-14 and concentrate C16-C18:2 fatty acid constituents. This means that these fractions can possibly be used for blending to formulate CBRs blends. Higher pressures ranged from 34.5 to 48.3 MPa and at 80 °C were found to be effective to fractionate shorter-medium (C8-C14) and longer (C16-C18:2) chain fatty acid constituents. High yields (99.6%) were obtained at 48.3 MPa and 80 °C. For fraction 4, compared with Soxhlet extraction, the C12 constituent content could be reduced to 56.3% and C18:1 constituent content could be increased to 97.1%. Moreover, fraction 4 constituents had the lowest melting point among all other fractions and the constituents matched those of possible cocoa butter replacers blends. Palm kernel oil can be fractionated with supercritical carbon dioxide to produce suitable triglycerides in terms of fatty acid constituents for cocoa butter replacers blends. Further research will probably be conducted for blending the last fraction of palm kernel oil extracted with supercritical carbon dioxide to formulate cocoa butter replacer fats. Other unsaturated constituents or non C12 constituent content oil could be used to reduce the remaining C12 constituents so as to match the physico-chemical properties of cocoa butter.

REFERENCES


Palm Oil Research Institute of Malaysia (PORIM) test methods; Bangi, Selangor, Kuala Lumpur, 1995


