

Partial hydrogenation of fatty acid methyl esters at supercritical conditions

Maj-Britt Macher^a, Johan Högberg^a,
Poul Møller^b, and Magnus Härröd^a

Under supercritical or near-critical conditions propane is a very good solvent for both lipids and hydrogen. Thus, it is possible to generate an essentially homogeneous phase, in which the transport resistances for the hydrogen are eliminated. Therefore, the hydrogen concentration at the catalyst surface can be greatly increased, resulting in extremely high reaction rates and products having low *trans* fatty acid contents.

In this study we present results from hydrogenation of rapeseed fatty acid methyl esters under near-critical and supercritical conditions. Temperature, residence time, hydrogen pressure, and catalyst life were varied systematically, using a statistical experimental design, in order to elucidate reaction rate and *trans* fatty acid formation as functions of the above variables. The experiments were carried out in a microscale fixed-bed reactor, using a 3% Pd-on-aminopolysiloxane catalyst. At 92 °C, a hydrogen pressure of 4 bar, and a residence time of 40 ms we obtained a *trans* content of $3.8 \pm 1.7\%$ at a iodine value of 70. Our results support the findings from traditional processes that at a constant iodine value (IV) the *trans* content decreases with decreasing temperature, increasing p_{H_2} , and increasing residence time. The reaction rate at our best conditions was roughly 500 times higher than in traditional batch hydrogenation.

Partielle Hydrierung von Fettsäuremethylestern bei überkritischen Bedingungen. Im überkritischen oder nahe-kritischen Zustand besitzt Propan sehr gute Lösungseigenschaften für sowohl Fette als auch Wasserstoff. Dies ermöglicht es, eine im wesentlichen homogene Reaktionsmischung zu erzeugen, in der die Transportwiderstände für den Wasserstoff eliminiert sind. Dadurch kann die Wasserstoffkonzentration an der Katalysatoroberfläche wesentlich erhöht werden, was zu extrem hohen Reaktionsgeschwindigkeiten führt. Außerdem können auf diese Weise Produkte mit einem niedrigen Gehalt an *trans*-Fettsäuren erzielt werden.

In der vorliegenden Studie präsentieren wir die Ergebnisse der partiellen Hydrierung von Rapsöl-Fettsäuremethylestern bei nahe-kritischen und überkritischen Bedingungen. Unter Verwendung eines statistischen Versuchsdesigns wurden Temperatur, Aufenthaltszeit, Wasserstoffpartialdruck und Katalysatorlebensdauer systematisch variiert, um die Reaktionsgeschwindigkeit und die Bildung von *trans*-Fettsäuren als Funktionen dieser Variablen darzustellen. Die Versuche wurden in sehr kleinem Maßstab in einem Festbettreaktor durchgeführt. Der verwendete Katalysator bestand aus 3% Pd auf einem Aminopolysiloxan-Träger. Bei 92 °C, einem Wasserstoffdruck von 4 bar und einer Aufenthaltszeit von 40 ms erreichten wir einen *trans*-Gehalt von $3,8 \pm 1,7\%$ bei einer Jodzahl von 70. Unsere Ergebnisse bestätigen die für den traditionellen Prozeß gefundenen Trends, daß bei konstanter Jodzahl der *trans*-Gehalt abnimmt, wenn die Temperatur gesenkt, der Wasserstoffdruck erhöht, bzw. die Aufenthaltszeit verkürzt wird. Die höchsten von uns erzielten Reaktionsgeschwindigkeiten waren rund 500-fach höher als in herkömmlichen Batch-Prozessen.

1 Introduction

Partial hydrogenation is the standard process to convert liquid oils into semi-solid fats and to improve the oxidation stability of an oil [1]. Using this technique a certain melting point and profile are achieved and a substantial proportion of the native *cis* bonds is changed into *trans* bonds (a *trans* content of 30–50% is normal for an oil partially hydrogenated to a iodine value (IV) of 70). Lately, *trans* fatty acids (in the following simply called *trans*) have become the subject of many health discussions related to ingestion of fats and oils [2].

The total annual production of hydrogenated oils is about 25 million tons [3, 4]. Most of them are produced in the original way, i.e. as a batch process in large slurry reactors ($5\text{--}20\text{ m}^3$), at high temperatures, low pressures, and long reaction times (140–200 °C, 1–3 bar, 2 h) [2]. In these reactors the reaction rates are limited by the low concentration of hydrogen at the catalyst surface. This is due to the low solubility of hydrogen in the oil and to transport resistances for the hydrogen from the gas phase to the catalyst surface. Furthermore, insufficient hydrogen concentration at the catalyst surface promotes the formation of *trans* fatty acids. The half-hydrogenation theory predicts that *trans* formation will de-

crease when the hydrogen concentration at the catalyst surface is increased [5–7]. This has been verified in several studies [e.g. 8, 9].

Considerable research has been concentrated on the development of new catalysts and new reactor designs to improve the mass transfer. However, it has not been possible to produce a partially hydrogenated oil having a low *trans* content (i.e. *trans* < 5% at IV 50–80) [1]. Today there are two alternatives for achieving this product quality while keeping the *trans* content below 5% (a level consistent for butter [2]). One is the transesterification of a native oil with a fully hydrogenated oil. The other is to use tropical oils, such as palm oil, which are solid at room temperature and do not contain any *trans* fatty acids. Both of these routes are more expensive than partial hydrogenation, but their use is increasing because they solve the *trans* fatty acid problem.

It would be a big step forward, if the partial hydrogenation process could be improved, in the sense of increasing the hydrogen concentration at the catalyst surface and in that way reducing the risk for *trans* formation. The addition of propane to the oil can improve the solubility of hydrogen in the oil [10]. Furthermore, with supercritical propane it is possible to go even further: At certain conditions, one can bring the whole reaction mixture to a supercritical state and generate a substantially homogeneous phase, in which the differences between gas and liquid are eliminated. Thus, the transport resistances for hydrogen between different phases are avoided, and the hydrogen concentration at the catalyst

^a Chalmers University of Technology, Department of Food Science, c/o SIK, PO Box 5401, 40 229 Göteborg, Sweden.

^b Marselis Boulevard 38¹⁴, 8000 Aarhus, Denmark.

surface can be controlled to a great extent. The principles behind the hydrogenation at supercritical conditions (concentration profiles, phase diagrams etc.) are in detail described elsewhere [11] and a patent has been filed [12].

For our experiments we chose propane as solvent because of its superior solvent properties for both oils and hydrogen [11], and because it is allowed for unlimited use in food applications [13]. Recently, propane has been applied in a full scale plant for de-oiling of lecithins for food and pharmaceutical applications (SKW Trostberg AG, Germany) [14]. The handling of propane in industrial scale is well known from the petrochemical industry and the introduction of propane does not increase the risks in a hydrogenation plant. Such a plant has to be explosion-proof already because of the hydrogen used. In the following we will present some results from a systematic investigation of the partial hydrogenation of rapeseed FAME using propane as a solvent to achieve extremely favourable reaction conditions.

2 Materials and Methods

2.1 Materials

The substrate used was methylated rapeseed oil (*Larodan*, Sweden). The composition of the fatty acid methyl esters (FAMES) was as follows: 3.5% saturated; 63.0% *cis*; 23.5% *cis* + *cis*; and 10.0% *cis*+*cis*+*cis*. This composition corresponds to a iodine value (IV) of 114.5. The catalyst was 3% Pd on aminopolysiloxan, in the form of granules with a particle size of 0.5–1.0 mm, which was prepared at the Department of Inorganic Chemistry at the University of Erlangen-Nürnberg, Germany. Hydrogen (hydrogen plus 4.5) and propane (instrument quality 1.6) were purchased from AGA (Lidingö, Sweden).

2.2 Experimental procedure

A flow sheet for the continuous reactor used is shown in Fig. 1. The amounts of hydrogen and oil were controlled according to the methods described in literature [15]. Propane, hydrogen, and oil were mixed at room temperature (M). The mixture was heated to the desired reaction temperature (Temp) and brought into the reactor. This reactor was an HPLC tube (inner diameter 2.1 mm, length 50 mm) filled with a given amount of catalyst. The height of the catalyst bed in the different runs ranged from 2 to about 20 mm. After the reactor, samples were collected on-line by means of a *Rheodyne* switching valve (*Model 7000*) (A) [16]. Heptane was used to flush and dissolve the samples from the sampling loop, which could then be injected into the HPLC without further preparation. After the sample valve, the pressure was allowed to expand to atmospheric pressure in a pressure reduction valve (P), whereupon the product separated from the gases (Sep). This valve (P) (again a *Rheodyne* valve) was used simultaneously to regulate the propane flow, in order to

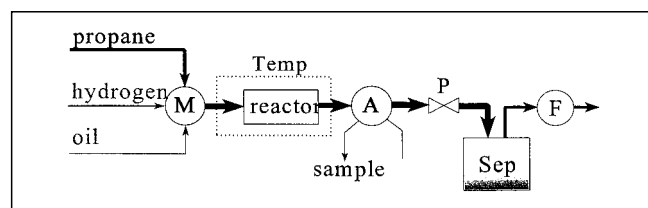


Fig. 1. Flow sheet for the reactor used. M – Mixer. Temp – Temperature control. A – Sample valve. P – Pressure reduction valve. Sep – Separation gas/oil. F – gas flowmeter.

Tab. 1. Experimental design.

Variables		Levels				
		Very low	Low	Average	High	Very high
Temperature	[°C]	48	70	100	130	152
Hydrogen pressure	[bar]	1.3	2.0	3.0	4.0	4.7
Residence time	[ms]	60	100	200	400	664
Catalyst life	[kg oil/ g Pd]	1.3	1.3	2.6	5.0	5.0

maintain a constant residence time and composition in the reactor. After the separation, the gas flow (propane and hydrogen) was measured (F). To control the composition of the reaction mixture and to estimate the reaction times, the densities of hydrogen and propane were required. We calculated them from the *Peng-Robinson* equation of state [17].

The experiments were carried out according to a central composite experimental design (2^4 factorial design + star (2×4) + center point = 25 experiments) [18]. The variables (see Tab. 1) were temperature (T), hydrogen pressure (p_{H_2}), residence time (t), and catalyst life (which is defined as the amount of oil processed per gram of active catalyst, i.e. kg_{FAME}/g_{Pd} in our case). The total pressure, the lipid concentration and the total flow rate were kept constant, at 100 bar, 0.35 mol-% (= 2.4 wt-%), and 48 mmol/min, respectively.

The samples were analyzed using an HPLC (*Shimadzu*, Tokyo, Japan) equipped with a silver-ion column (*Chrom-Spher Lipids*, 5 μ m, 4.6 \times 250 mm, *Chrompack* Ltd., Middelburg, The Netherlands). The method we used was a gradient elution, which has been developed in our department from an isocratic method [19, 20]. The isomeric form (*cis* or *trans*) and composition of the fatty acids were determined using individual calibration curves for each fatty acid. From these data we calculated iodine value (IV) [21] and *trans* content. Subsequently, we determined equations for these variables as functions of temperature (T), hydrogen pressure (p_{H_2}), residence time (t), and catalyst life. The data processing was done using the software programme SYSTAT [22].

The results are presented as contour plots to facilitate the interpretation. In the graphs, the significance of the results is expressed by SEE (= Standard Error of Estimate, expressed in the same unit as the goal function). A change in a variable is considered significant, when it causes a change larger than $k \cdot SEE$ in the goal function. The factor k is used to define the range that covers 100% of the variation in the experiments.

3 Results and Discussion

The most important factor for us was to make sure that the reaction mixtures (FAME/propane/hydrogen) in our experiments really were in a homogeneous phase. Therefore, we chose to work in a very dilute system. In Fig. 2 the compositions of our reaction mixtures are illustrated with a ternary phase diagram of the system FAME/propane/hydrogen. The stoichiometric need for full hydrogenation of rapeseed-FAME is roughly 1.5 mol of hydrogen per mol of FAME. Thus, all experimental points should lie to the right of the dashed line in Fig. 2 to make complete hydrogenation possible. The total pressure (100 bar) was chosen to ensure a good solubility for both FAME and hydrogen. This pressure,

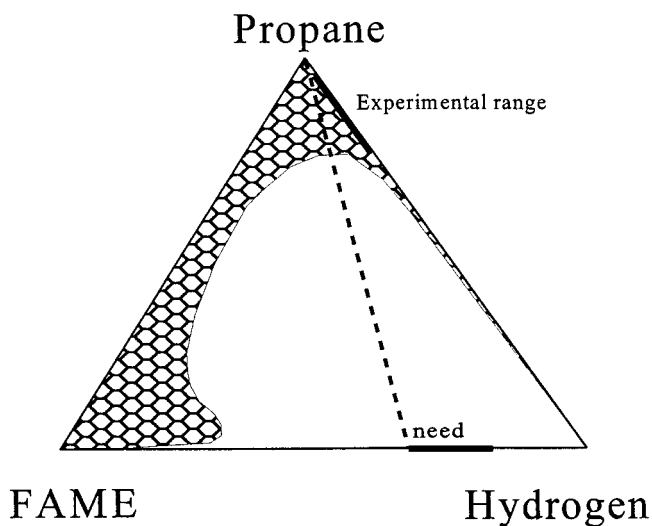


Fig. 2. Schematic phase diagram for the system FAME, propane, and hydrogen. The shaded area represents the estimated single phase region. The composition of the reaction mixtures used in our experiments is indicated with (*). The dashed line indicates the stoichiometric amount of hydrogen needed for complete hydrogenation of the substrate.

as well as the lipid concentration and the total flow, were kept constant throughout the experiments. Therefore, all mixtures used are located on a line parallel to the propane-hydrogen axis in Fig. 2.

As long as the whole reaction mixture is in a homogeneous phase, the hydrogen concentration can be chosen freely because the transport resistance for the hydrogen is minimized. This implies good access to hydrogen at the catalyst surface throughout the whole reactor. Therefore, extremely high space-time yields and very low *trans* fatty acid contents are achieved. The catalyst life had no significant effect on either reaction rate or *trans* fatty acid formation, which means that the catalyst did not deactivate within the experimental range. However, our experiments were only run to relatively low levels of catalyst life (i.e. life $\leq 5 \text{ kg}_{\text{FAME}}/\text{g}_{\text{Pd}}$), so the behaviour of the catalyst after longer running times (i.e. at higher values of life) needs to be further investigated.

3.1 Reaction rate and iodine value (IV)

Fig. 3 shows the IV as a function of temperature and hydrogen pressure. A rise in temperature increases the reaction rate and, consequently, lowers the IV. This can be explained with the increased activity of the catalyst at higher temperature. The effect of p_{H_2} is depending on the temperature. At high temperature, an increase in p_{H_2} increases the reaction rate (e.g. at 130°C , an increase in p_{H_2} from 2 to 4 bar lowers the IV from 65 to 25), but this effect decreases with decreasing temperature. This behaviour can also be explained with the changing activity of the catalyst at different temperatures: At high temperature the catalyst activity is high, thus, if more hydrogen is added, this results in a higher reaction rate. At low temperature the catalyst activity is lower; the catalyst is saturated with hydrogen already at low p_{H_2} and cannot convert additional hydrogen. Therefore, an increase in p_{H_2} has a low effect at low temperature.

In Fig. 4 the IV is plotted as a function of temperature and residence time. Again, the accelerating effect of increasing temperature is clearly shown. At 130°C , a residence time of 140 ms is sufficient to reach an IV of 60, which is roughly

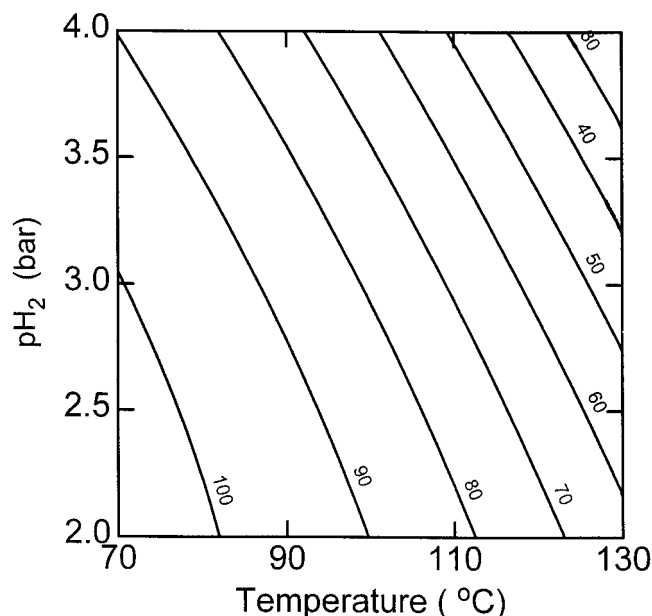


Fig. 3. Iodine value (IV) as a function of temperature and hydrogen pressure. Residence time = 400 ms, catalyst life = 5 kg oil/g Pd, $1.7 \cdot \text{SEE} = 17.5$.

our goal value. However, this IV can also be reached at lower temperatures, if the residence time is increased (compare 100°C and 400 ms in Fig. 4). This should be kept in mind, because this fact will become very important when discussing product quality.

At 100°C , 4 bar hydrogen and 400 ms we reached an IV of 60 (see Fig. 3 and 4). The reaction rate at these conditions, expressed as Lipid Hourly Space Velocity (LipidHSV, i.e. the amount of processed oil per reactor volume and time), was $50\,000 \text{ kg}_{\text{FAME}}/\text{m}^3_{\text{Reactor}} \text{ h}$. This is roughly a factor 125 higher than traditional triglyceride hydrogenation in batch processes ($400 \text{ kg}_{\text{oil}}/\text{m}^3_{\text{Reactor}} \text{ h}$), although our experiments were mainly designed as a screening of reaction conditions, and cannot be considered as optimized. However, we have

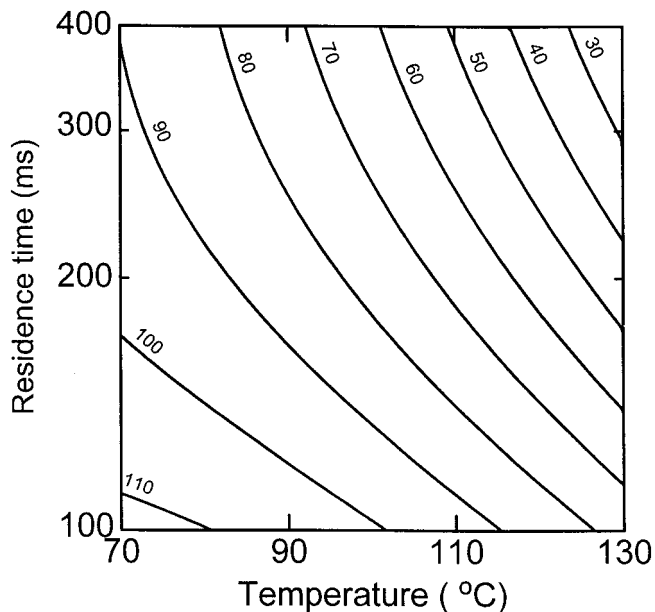


Fig. 4. Iodine value (IV) as a function of temperature and residence time. Hydrogen pressure = 4 bar, catalyst life = 5 kg oil/g Pd, $1.7 \cdot \text{SEE} = 17.5$.

also done a successful test on hydrogenation at higher lipid loadings, which shows the great potential for this technology. In this experiment we reached an IV of 70 at a lipid concentration of 15 wt-% (at 100°C, 23 bar hydrogen, and 400 ms residence time). The corresponding LipidHSV was 213 000 kg_{FAME}/m³_{Reactor} h, i.e. roughly 530 times higher than traditional.

3.2 Product quality – *trans* fatty acids

In addition to obtaining a satisfactory IV, achieving a low *trans* content was the other important goal in our experiments. The best results were obtained at 92°C, 4 bar hydrogen, and a residence time of 400 ms. We reached an IV of 70 at a *trans* concentration of 3.8 ± 1.7%, which is considerably lower than the levels obtained in an optimized research batch reactor [9]. In the modelling of the data with respect to *trans* content we excluded the data points having an IV < 50. This was for the following reason (see Fig. 5): In the beginning of a hydrogenation, i.e. while the IV is still high, the *trans* content is low. As the IV decreases, the *trans* concentration increases and reaches a maximum roughly at IV = 50. From IV = 50 down to IV = 0 (i.e. full hydrogenation), the *trans* content decreases, due to the formation of more and more saturates. This means, it is possible to get the same *trans* content at two different IV, and our model is unable to follow this curvature because of its limited complexity. Therefore, and because the IV-range below 50 was of little interest in this partial hydrogenation study, we decided to “cut off” the lower part (this concerns three data points). Therefore, all *trans* lines in the following plots end at IV = 50.

The following figures, Fig. 6 and 7, show the formation of *trans* fatty acids at different conditions. We have added the (for this study) most interesting IV-lines to make it easier to see the area that combines a suitable IV with a low *trans* concentration. To facilitate the interpretation of these figures, we will first give a short example of how to read these plots: In Fig. 6, starting at pH₂ = 3.5 on the H₂-axis and moving from low to high temperature, the IV decreases, while the *trans* content increases. However, from the point of passing IV = 50 the *trans* model is no longer valid. As shown in Fig. 5, the *trans* content will not continue to increase below IV = 50; it will decrease, due to the increasing hydrogenation to saturates. Another example: if you follow the line for *trans* = 8 from the temperature axis and upward in Fig. 6, it is valid until it reaches IV = 50, then it ends because the *trans*

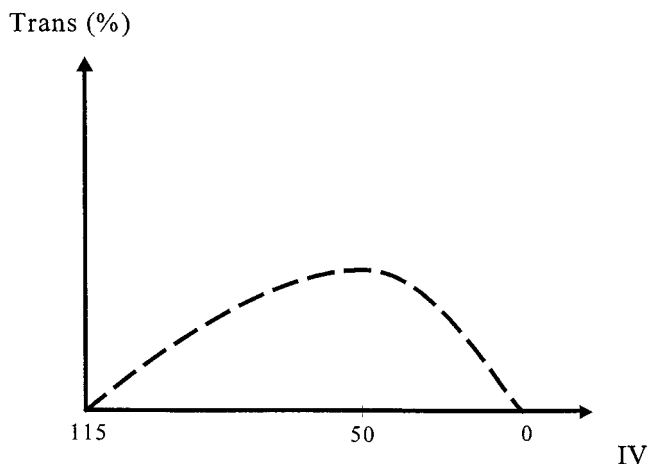


Fig. 5. Schematic picture of the change in *trans* formation with decreasing IV during hydrogenation. Below IV = 50 the *trans* content decreases due to increased formation of saturates.

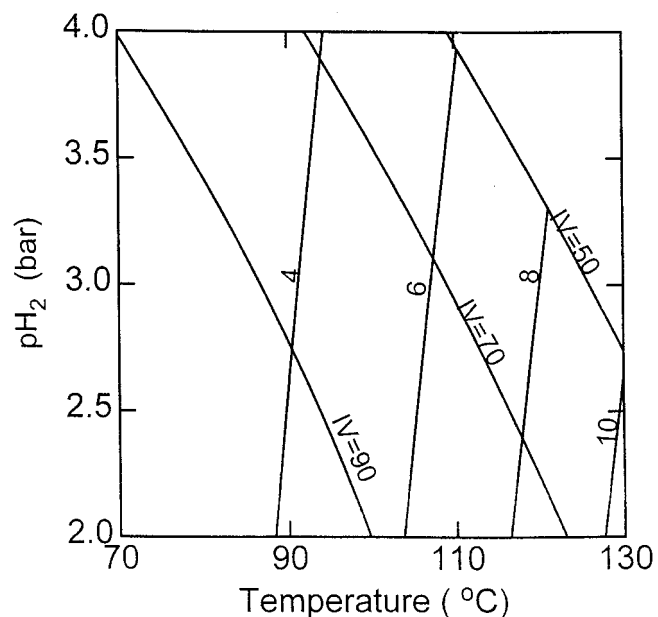


Fig. 6. *Trans* fatty acid content as a function of temperature and hydrogen pressure. Residence time 400 ms, catalyst life = 5 kg oil/g Pd, 2.0*SEE = 1.7. The added IV-lines are taken from Fig. 3, 1.7*SEE = 17.5.

model is no longer valid there (due to the excluded *trans* data at IV below 50).

Fig. 6 confirms the well-known fact that, at constant IV and constant residence time, the *trans* formation decreases with decreasing temperature and increasing pH₂. For example, if you follow the line for constant IV = 70, a decrease in temperature from 120 to 92°C at an increase in pH₂ from 2.0 to 4.0 bar lowers the *trans* concentration from 9 to 3.8%. Another possibility to lower the *trans* concentration at a given IV is shown in Fig. 7. This plot shows the effect of temperature and residence time on *trans* formation. IV = 70 can be achieved by a reduction in temperature from 130 to 90°C to-

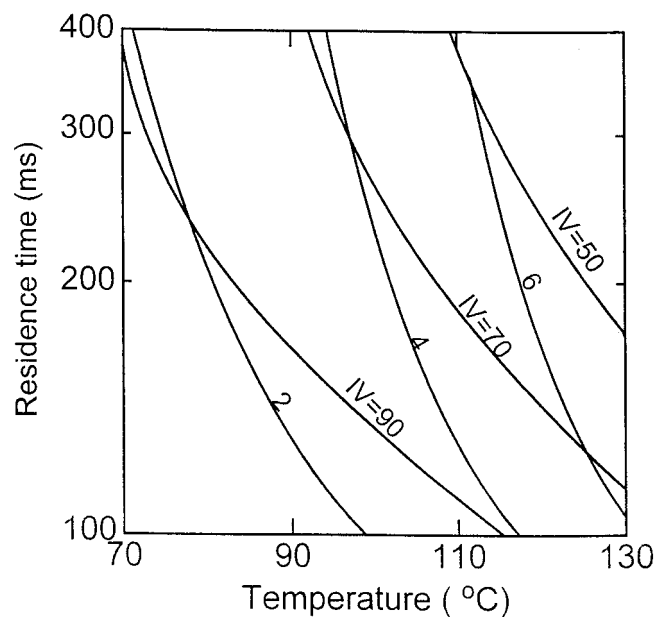


Fig. 7. *Trans* fatty acid content as a function of temperature and residence time. Hydrogen pressure = 4 bar, catalyst life = 5 kg oil/g Pd, 2.0*SEE = 1.7. The added IV-lines are taken from Fig. 4, 1.7*SEE = 17.5.

gether with an increase in residence time (from 110 to 400 ms). In this case, the *trans* concentration is reduced from 6.2 to 3.8%.

The trends in Fig. 6 and 7 indicate that the *trans* formation can be further reduced, if we lower the temperature and increase the residence time. Since the effect of hydrogen pressure was not significant at low temperatures, this indicates that low *trans* contents can perhaps even be obtained at lower hydrogen pressures.

3.3 Conclusions and outlook

In this study we have presented a completely new way for hydrogenation of oils. In supercritical hydrogenation, the whole reaction mixture is in a homogeneous phase, which eliminates the transport resistances for hydrogen. Thus, we can control the hydrogen concentration at the catalyst surface in a new way and increase it to extremely high levels, which leads to very high reaction rates. The reaction rate in our best experiments was a factor 530 higher than in standard batch hydrogenation. Furthermore, the formation of *trans* fatty acids in partial hydrogenation is dramatically reduced. At an IV around 70, the *trans* concentration of our product was only 3.8%, which is far below the values obtained in current industrial batch processes.

Since the most important criteria for a partial hydrogenation process relate to product quality rather than reaction rate, *trans* content and melting profile are crucial for partially hydrogenated products like margarine. Based on our very promising results in FAME hydrogenation, we are currently focusing on triglyceride hydrogenation to optimize reaction conditions and product quality. Another topic that requires further investigation is the performance of the catalyst at higher lipid loadings, i.e. to determine the borders for the homogeneous phase area that allows these extremely fast and selective reactions. This, as well as the life of the catalyst, are subjects to ongoing research.

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References

- [1] *Y. H. Hui* (ed.), *Bailey's Industrial Oil and Fat Products*, Vol. 4, 5th edn., John Wiley & Sons Inc., New York, NY, 1996.
- [2] *K. W. J. Wahle* and *W. P. T. James*, *Isomeric Fatty Acids and Human Health*, *Eur. J. Clin. Nutr.* **47** (1993), 828.

- [3] *S. Mielke*, *Oil Demand Forecast to Rise 32% during Decade*, *INFORM* **3** (1992), 695.
- [4] *B. Fitch Haumann*, *Tools: Hydrogenation, Interesterification*, *INFORM* **5** (1994), 668.
- [5] *I. Horiuti* and *M. Polanyi*, *Exchange Reactions of Hydrogen on Metallic Catalysts*, *Trans. Faraday Soc.* **30** (1934), 1164.
- [6] *R. R. Allen* and *A. A. Kiess*, *Isomerization during Hydrogenation, 1. Oleic Acid*, *J. Am. Oil Chem. Soc.* **32** (1955), 400.
- [7] *R. R. Allen*, *Hydrogenation Research*, *J. Am. Oil Chem. Soc.* **63** (1986), 1328.
- [8] *N. Hsu*, *L. L. Diosady*, and *L. J. Rubin*, *Catalytic Behaviour of Palladium in the Hydrogenation of Edible Oils II. Geometrical and Positional Isomerization Characteristics*, *Ibid.* **66** (1989), 232.
- [9] *P. H. Berben*, *P. J. W. Blom*, and *J. C. Sollie*, *Palladium and Platinum Catalyzed Oil Hydrogenation: Effects of Reaction Conditions on *trans*-Isomer and Saturate Formation*, *Proceedings of AOCs meeting 1995*.
- [10] *H. Schiemann*, PhD thesis, University of Erlangen-Nürnberg, Germany, 1993.
- [11] *M. Härröd*, *M. Macher*, *S. van den Hark*, and *P. Møller*, unpublished work.
- [12] *M. Härröd* and *P. Møller*, PCT patent application, 1994, WO 96/01304.
- [13] EC, 1984, EC directive 84/344/EEC.
- [14] *J. Heidlas*, *De-oiling of lecithins by near-critical fluid extraction*, *Agro-Food-Industry Hi-Tech* **1** (1997), 9.
- [15] *K. H. Pickel*, *Proceedings of 2nd Internat. Symp. on Supercritical Fluids*, Ed. *M. McHugh*, Johns Hopkins University, Baltimore, MD, 1991, p. 457.
- [16] *M. Härröd* and *I. Elfman*, *Enzymatic Synthesis of Phosphatidylcholine with Fatty Acids, Isooctane, Carbon Dioxide, and Propane as Solvents*, *J. Am. Oil Chem. Soc.* **72** (1995), 641.
- [17] *R. Dohrn*, *Berechnungen von Phasengleichgewichten*, Vieweg, Braunschweig/Wiesbaden, Germany, 1994.
- [18] *G. Box*, *W. G. Hunter*, and *J. S. Hunter*, *Statistics for Experimenters. An Introduction to Design, Data Analysis and Model Building*, John Wiley & Sons Inc., New York, NY, 1978.
- [19] *I. Elfman*, *S. van den Hark*, and *M. Härröd*, *Gradients of *n*-Heptane and Acetonitrile in Silver Ion HPLC Analyses of *cis* and *trans* Bonds in Lipids*, *J. Am. Oil Chem. Soc.* (1997), **74** (1997), 1177.
- [20] *R. O. Adlof*, *Separation of *cis* and *trans* Unsaturated Fatty Acid Methyl Esters by Silver Ion High-Performance Liquid Chromatography*, *J. Chromatogr. A* **659** (1994), 95.
- [21] *D. Firestone*, *Official methods and recommended practices of the American Oil Chemists' Society*, 4th edn., American Oil Chemists' Society, Champaign, 1996, Cd 1–25.
- [22] *L. Wilkinson*, *SYSTAT 6.0*, SYSTAT Inc., Evanston, IL, 1996.

Addresses of the authors: MSc *Maj-Britt Macher*, MSc *Johan Högerberg*, PhD *Magnus Härröd*, Chalmers University of Technology, Department of Food Science, c/o SIK, P.O. Box 5401, 40229 Göteborg, Sweden. MSc *Poul Møller*, Marselis Boulevard 38¹⁴, 8000 Aarhus C, Denmark.

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