Transesterification of Trimethylolpropane and Rapeseed Oil Methyl Ester to Environmentally Acceptable Lubricants

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ABSTRACT: Biodegradable trimethylolpropane [2-ethyl-2-(hydroxymethyl)-1,3-propanediol] esters of rapeseed oil fatty acids were synthesized by transesterification with rapeseed oil methyl ester both by enzymatic and chemical means, both in bench and pilot scales. Nearly complete conversions were obtained with both techniques. A reduced pressure of about 2 to 5 kPa, to remove the methanol formed during transesterification, was critical for a high product yield. The quantity of added water was also critical in the biocatalysis. *Candida rugosa* lipase was used as biocatalyst and an alkaline catalyst in chemical transesterifications. In biocatalysis the maximum total conversion to trimethylolpropane esters of up to 98% was obtained at 42°C, 5.3 kPa, and 15% added water. The maximum conversion of about 70% to the tri-ester was obtained at the slightly higher temperature of 47°C. The reaction time was longer in the biocatalysis, but considerably higher temperatures were required in chemical synthesis. In the chemical synthesis tri-ester yields increased when the temperature was first held at 85 to 110°C for 2.5 h and subsequently increased to up to 120°C for 8 h. The trimethylolpropane esters obtained were tested as biodegradable hydraulic fluids and compared to commercially available hydraulic oils. The hydraulic fluids based on trimethylolpropane esters of rapeseed oil had good cold stability, friction and wear characteristics, and resistance against oxidation at elevated temperatures.
JAOCS 75, 1557–1563 (1998).

KEY WORDS: Biocatalysis, 2-ethyl-2-(hydroxymethyl)-1,3-propanediol, hydraulic oil, lipase, lubricant, rapeseed oil, trimethylolpropane, transesterification.

Interest in the production of biodegradable, environmentally acceptable esters for biodiesel, lubricants, solvents, surface active agents, and the like by lipase (triacylglycerol acylhydrolase, EC 3.1.1.3) biocatalysis from vegetable oils has increased markedly during the last few years (1,2). For example, butyl oleate (3) may be used as biodiesel additive, PVC plasticizer, water-resisting agent, and in hydraulic fluids. Rapeseed oil fatty acid esters of 2-ethyl-1-hexanol (4) can be used to replace conventional organic solvents in some detergent applications such as in car shampoos, and as a solvent for printing ink. Biodegradable polyesters (5) are of interest, for example, as surgical implants and agricultural plastic films.

Furthermore, the interest in biodiesel and in environmentally acceptable biodegradable lubricating base oils has rapidly increased recently. It has been estimated that, in Germany, for instance, the market share of biodegradable lubricating oils will grow from 2 to 3% in 1994 up to 15% by the year 2000 (6). Biodegradable lubricants were first developed for two-stroke outboard engines in the early 1980s, with the main base fluid composed of neopentylpolyol esters of branched-chain fatty acids (7). Eychenne and Mouloungui (8) have recently reviewed the developments in environmentally friendly lubricating oils based on neopentylpolyols such as neopentyl glycol, pentaerythritol, and trimethylolpropane. In the mid-1980s, biodegradable chainsaw oils based on natural esters of rapeseed oil were introduced on the market (9). Biodegradable trimethylol propane esters of sunflower oil (10) or rapeseed oil (11,12) fatty acids can be used in the production of hydraulic fluids. Trimethylolpropane esters of low viscosity and high temperature stability have long been used as lubricants for aircraft, and a method has been patented to produce esters of the high thermal stability required for the lubricants used in high-performance jet turbine engines (13). It has also been shown that a mixture of trimethylolpropane esters having both a sufficiently high viscosity and low pouring point for car engine oils can be prepared (14,15). A few years later, novel gas turbine engine oils based on monopentaerythritol and trimethylolpropane esters, with a reduced tendency to form deposits, were patented (16).

Fats and oils are conventionally modified by chemically catalyzed transesterification. The current worldwide annual production of rapeseed oil methyl ester for biodiesel has been estimated at about one million tonnes (17). Lipase-catalyzed transesterification has been proposed for the modification of fats and oils (18), and for the production of biodegradable solvents (4) and polymers (2). Osada *et al*. (19) and Monot *et al.* (20) have demonstrated that hydrophilic polyols can be esterified by lipase in the presence of an organic solvent such as di-*n*-butyl ether or tetrahydrofuran. In this paper we describe both chemical and enzymatic transesterification between trimethylolpropane and rapeseed oil methyl ester in high

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yields, without any additional organic solvent, as an environmentally acceptable alternative to chemical synthesis of trimethylolpropane esters (21,22).

MATERIALS AND METHODS

Materials. Finnish rapeseed oil was obtained from Raisio Group, Oil Milling Division (Raisio, Finland). The average fatty acid composition of rapeseed oil (average molar mass 880 g/mol) was: oleic acid 57%, linoleic acid 22%, linolenic acid 12%, palmitic acid 4%, eicosaenoic acid 2%, stearic acid 1%, erucic acid 1%, others 1%. Trimethylolpropane, 2-ethyl-2-(hydroxymethyl)-1,3-propanediol (molar mass 134,18 g/mol), was obtained from E. Merck (Darmstadt, Germany). Sodium hydroxide was from E. Merck, and sodium ethoxide and sodium methoxide were from J.T. Baker (Deventer, The Netherlands). All other reagents were of analytical grade, unless otherwise indicated. *Candida rugosa* lipase powder, hydrolytic activity 8000 U/g (Biocatalysts, Pontypridd, United Kingdom) was used throughout this work without further purification. The carriers tested for lipase immobilization were: Celites R-630, R-640, and R-626 (Manville, United Kingdom), Diatomite (Kenite, Witco, Japan), Duolites ES-561 and ES-762 (neutral adsorption resin, Dia-Prosim, Vitry Chauny, France), GCC (Cultor, Finland), GDC 200 (weak alkaline anion exchange resin, Cultor, Finland), HPA 25 (strong alkaline anion exchange resin, Mitsubishi Kasei, Tokyo, Japan), Kieselgel 60 (Merck), and WA 30 (weak anion exchange resin, Mitsubishi Kasei).

Synthesis of rapeseed oil methyl ester. Rapeseed oil methyl ester was synthesized chemically as follows: rapeseed oil (0.3 mol) was weighed into a 500 mL three-necked flask equipped with a thermometer, condenser, stirrer, and sample adapter, and 2.0 mol methanol was added under stirring. The reaction mixture was then heated to 60°C, and 0.5% (w/w) alkaline catalyst was added. After the reaction was completed in 4 h as determined by thin-layer chromatography (TLC), the reaction mixture was washed with acidic water. Glycerol formed was separated, and the excess alcohol was distilled off. The rapeseed oil methyl ester (average molar mass 884 g/mol) content of the product varied from 95 to 99%, as determined by highperformance liquid chromatography (HPLC).

Enzymatic synthesis of rapeseed oil trimethylolpropane ester. Transesterification for the synthesis of trimethylolpropane tri-ester of rapeseed oil fatty acids (average molar mass 922 g/mol) was carried out either at ambient pressure, in capped or open 13-mL Kimax test tubes, or at a reduced pressure (2.0 to 13.3 kPa), in 25-mL round-bottomed flasks equipped with a vertical condenser (cooling water temperature 6°C), typically as follows: trimethylolpropane (0.607 g, 4.5 mmol) was dissolved in 0.7 mL (15% w/w of total mass of the substrates) of distilled water, after which rapeseed oil methyl ester (4.00 g, 13.6 mmol) and solid lipase preparation (40% w/w; immobilized lipase 50% w/w) were added. Reaction was usually carried out at a temperature between 37 and 47°C under a reduced pressure usually of about 2 to 16 kPa with magnetic stirring at 250 rev/min (or 150 to 200 rev/min for the round-bottomed flasks), and the ratio of trimethylolpropane and rapeseed oil methyl ester varied between 1:3 and 1:4.5. The total sample was extracted twice with acetone, after which the enzyme precipitate was removed by centrifuging (3500 rev/min, relative centrifugal force $1900 \times g$). The supernatant was transferred into a 1.5 mL Eppendorf tube and stored at –20°C for later analysis.

Chemical synthesis of rapeseed oil trimethylolpropane ester. Rapeseed oil methyl ester (565.8 g, 0.64 mol or 15 kg, 17.1 mol) was weighed into a reaction vessel equipped with a thermometer, condenser, and efficient stirrer. The methyl ester was heated to 60°C and trimethylolpropane (25 g, 0.2 mol or 716 g, 5.3 mol) was added. After the trimethylolpropane had melted, sodium methylate (2.95 g, 0.5% w/w) was added as catalyst under adequate stirring. Unless otherwise mentioned, the mixture was refluxed (in the small pilot-scale experiment the mixture was first heated up to 85°C, after which the temperature was gradually increased to 110°C during a period of 2 h) under a reduced pressure (3.3 kPa) for a total of about 8 h, with samples taken at certain intervals for analyses. The mixture was neutralized with acidic water, washed with warm (50°C) water, and dried over anhydrous sodium sulfate.

Analytical. The reaction was monitored by semiquantitative TLC, using Kieselgel 60 F_{254} TLC plates (Merck) and ethyl acetate/*n*-heptane (4:96 vol/vol) as solvent. The plates were developed for 45 min, sprayed with a mixture of acetic acid/sulfuric acid/anisaldehyde (100:2:1, vol/vol/vol), dried for 10 min at room temperature, and heated at 105°C for 5 min.

Quantitative analyses were carried out with high-performance gel permeation chromatography (HPGPC), using Perkin-Elmer (Norwalk, CT) HPLC equipment with PE 7500 professional computer Chromatographics 3 program, SI-316 satellite integrator, PE Series 4 pump, HP1047A RI-detector (Hewlett-Packard, Palo Alto, CA), and Ultrastyragel 500 Å and 100 Å columns (Waters, Milford, MA) with a 0.45 µm filter (Rheodyne, Cotati, CA, USA) in the front of the columns. Acetone was evaporated off from 1000 µL samples in 4 h in a vacuum oven at 2.6 kPa, after which 1000 µL of tetrahydrofuran (THF) was added. The chromatograms were developed with HPLC-grade THF (Rathburn, Walkerburn, United Kingdom). The components were eluted on the basis of their molar mass in descending order. The rapeseed oil conversion was reported as % trimethylolpropane tri-ester or as a total conversion in % to trimethylolpropane esters (mono-, di-, and tri-esters of trimethylolpropane).

Infrared (IR) spectra were run using a Perkin-Elmer 883 or FTIR 16 PC spectrophotometer and either a 0.025-mm sodium chloride cuvette or KBr pellet.

Viscosity was determined according to the ASTM D445 standard (23), filterability according to the CETOP RP 124-H-standard (24), particle purity by calculating microscopically particles larger than 5 and 15 µm according to the ISO 4406 standard, cold stability according the VTT standard (25), cold viscosity behavior and viscosity according to the

ASTM D445 standard, foaming according to the ASTM D892 standard, color according to the ASTM D1500 standard, pour point according to the ASTM D97 standard, friction and wear according to the fourball test (ASTM D2783 standard), oxidation stability according to the ASTM D525, DIN 51586, and DIN 51554 standards, corrosion stability according to the Cincinnati-Milacron test (26), and biodegradability according to CEC-L-33-A-93 (27) or OECD-301 standard.

RESULTS AND DISCUSSION

All transesterifications were carried out without any additional organic solvent. When the lipase-catalyzed transesterification between rapeseed oil methyl ester and trimethylolpropane was carried out in capped Kimax test tubes the conversions to trimethylolpropane tri-ester were discouragingly low. The highest conversion of 11.2% was obtained in 120 h with 40% (w/w) *C. rugosa* lipase, at 37°C and 15% (w/w) added water. Under these conditions the relative quantity of the tri-ester of trimethylolpropane was always only about one-half of that of the di-ester, and about one-half of the rapeseed oil methyl ester remained unreacted. At ambient pressure, an increase in temperature up to 58°C did not improve conversion, apparently owing to methanol formed during the transesterification. Osada *et al*. (19) synthesized polyol esters using THF as a solvent and *Rhizopus delemar* lipase as biocatalyst at ambient pressure, but in this case also the yields were low with, for example, only 40% pentaerythritol monocaproate in 72 h at 30°C.

If the quantity of lipase was doubled and the system was equipped with a reflux condenser, conversion to trimethylolpropane tri-ester increased to about 34%, but part of the rapeseed oil methyl ester still remained unreacted. Also, Monot *et al*. (20) observed in an organic solvent system that conversion to the tri-ester of trimethylolpropane increased markedly with an increase in lipase quantity. Nevertheless, it soon became clear that satisfactory conversions could not be obtained at ambient pressure.

When lipase biocatalysis was performed in a round-bottomed flask equipped with a vertical condenser under reduced pressure but otherwise identical conditions, the conversion to trimethylolpropane tri-ester at 37°C increased to about 18% in 24 h at 12.0 kPa, to about 44% at 5.3 kPa, and to about 66% at 2.0 kPa (Table 1). When the temperature was increased to 47°C, the yield was of the same order of magnitude as at 37°C, 2.0 kPa. Consequently, most of the subsequent experiments were done under vacuum at 5.3 kPa, 47°C to minimize oxidation and to remove methanol formed during the reaction.

In lipase-catalyzed transesterifications a certain quantity of water is necessary in the reaction mixture for the enzyme to function, although too much water may promote reverse product hydrolysis (18). In the present case, at the initial stage a certain minimum quantity of water was also necessary to solubilize the substrate trimethylolpropane. When the quantity of added water was increased from 8 to 30% with 40% (w/w) lipase, 37°C, 12.0 kPa, and the ratio of trimethylolpropane and rapeseed oil methyl ester was 1:3.5, the total conversion to trimethylolpropane esters in 24 h increased with the increase in added water amount from about 35% with 8% added water to nearly 80% with 30% added water (Fig. 1). However, the conversion to the trimethylolpropane tri-ester increased only slightly, to about 30%. We have also previously demonstrated that lipase-catalyzed esterification can take place in unexpectedly high yields in the presence of an excess of water (28). Also, McNeil and Berger (29) have shown that under certain conditions lipases can catalyze ester synthesis with diols in spite of a large excess of water.

TABLE 1
Effect of Pressure on the Conversion of Trimethylolpropane[a] to Trimethylolpropane Tri-Ester (molar ratio of trimethylolpropane and rapeseed oil methyl ester 1:3, *Candida rugosa* lipase 40%, w/w; added water 15%, w/w; 37°C; 24 h)

Time	Conversion (%)			
(h)	Ambient pressure	12.0 kPa	5.3 kPa	2.0 (kPa)
1	2	2	7	12
3	4	2	14	25
5	7	4	22	24
7	7	6	23	24
9	8	9	26	39
11	8	8	36	51
24	8	18	44	66

[a]Trimethylolpropane, 2-ethyl-2-(hydroxymethyl)-1,3-propane-diol.

Table 2 shows the effect of the ratio of trimethylolpropane and rapeseed oil methyl ester on the total conversion to trimethylolpropane esters in 24 h with 15% added water at 37°C, with maximum total conversion of more than 90% with the substrate ratio of 1:3.5. If the ratio of trimethylolpropane and rapeseed oil methyl ester was changed to 1:4.5 and the temperature was increased to 47°C, the total conversion to trimethylolpropane esters of nearly 90% was obtained with 8% added water in 68 h, with little further increase with up to 30% added water (Fig. 1). Still further increases in water content had an adverse effect on transesterification. Monot *et al*. (20) generally employed a two- to threefold molar excess of acid in order to drive the reaction toward completion. However, from a practical application point of view, a small amount of the di-ester is a much smaller problem than a large excess of unreacted acid or rapeseed oil methyl ester. Al-

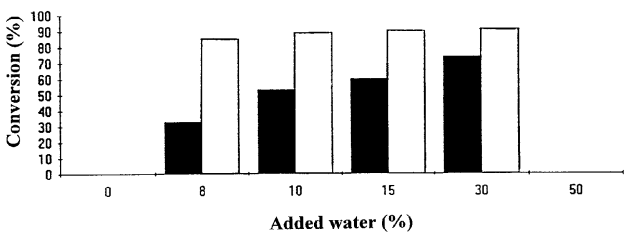

FIG. 1. Effect of the quantity of added water on the total conversion of trimethylolpropane [2-ethyl-2-hydroxymethyl)-1,3-propanediol] to trimethylolpropane esters (■ molar ratio of trimethylolpropane and rapeseed oil methyl ester 1:4.5; *Candida rugosa* lipase 40%, w/w; 37°C, 12 kPa, 24 h; □ molar ratio of 1:3.5; *C. rugosa* lipase 40%, w/w; 47°C, 5.3 kPa, 68 h)

TABLE 2
Effect of the Molar Ratio of Trimethylolpropane and Rapeseed Oil Methyl Ester on the Total Conversion of Trimethylolpropane to Trimethylolpropane Esters[a] (*C. rugosa* lipase 40%, w/w; added water 15%, 37°C, 5.3 kPa, 24 h)

	Conversion (%)	
Molar ratio	Enzymatic	Chemical
1 : 1.8	55	
1 : 2.0	65	
1 : 2.2	67	
1 : 2.3	68	
1 : 2.6	75	
1 : 3.0	80	82
1 : 3.1		84
1 : 3.2		86
1 : 3.3		80
1 : 3.4		77
1 : 3.5	**95**	76
1 : 3.6		74
1 : 3.8		73
1 : 4.0		72
1 : 4.5	85	
1 : 9.0	70	

[a]For abbreviations see Table 1.

though lipase-catalyzed esterification is slower than hydrolysis, according to Monot *et al.* (20) trimethylolpropane tricaprylate is very difficult to hydrolyze enzymatically. This may, at least in part, explain the relatively high transesterification rate and yield even at fairly high water concentrations.

Under elevated temperatures rapeseed oil tends to oxidize and decompose to free fatty acids, reducing the yield even when the reaction is carried out under nitrogen atmosphere. The reaction temperature was chosen to be high enough to melt trimethylolpropanol properly and to allow efficient mixing, yet low enough to minimize oxidation and thermal inactivation of lipase. In chemical transesterification the temperature was still higher so that all fatty acid esters could react. When the temperature was increased to 47°C in enzymatic transesterification, with a further reduction of pressure to 5.3 kPa, and 15% added water, about 64% conversion to trimethylolpropane tri-ester was reached in 24 h and 68% in 68 h, with no by-products nor any residual rapeseed oil methyl ester (Fig. 2). Under these conditions, the total conversion to trimethylolpropane esters increased to about 75% in 24 h and 90% in 68 h. No enzymatic hydrolysis of trimethylolpropane tri-ester could be observed under these conditions. Although the maximal conversion of about 70% to the tri-ester under the operating conditions used was obtained at 47°C, the maximal total conversion of about 98% to trimethylolpropane esters was obtained at 42°C. A decrease in the lipase quantity from 40 to 20% (w/w) under otherwise similar conditions had little effect, with a total conversion of 76% obtained in 24 h. This can be explained by improved mixing and thus by a better contact between the solid enzyme preparation and substrates. The product mixture also contained some trimethylolpropane di-ester and trimethylolpropane mono-ester but, under these conditions, no residual rapeseed oil methyl ester could be detected.

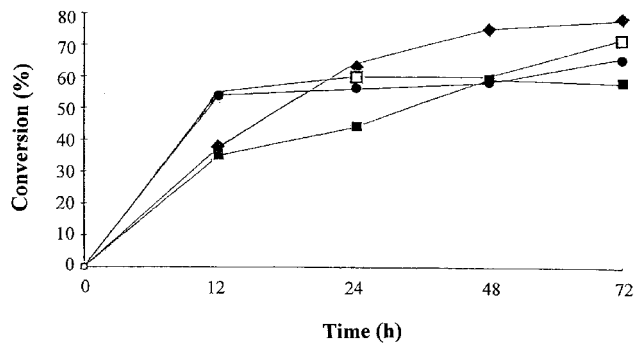

FIG. 2. Time courses of trimethylolpropane ester synthesis at various temperatures (molar ratio of trimethylolpropane and rapeseed oil methyl ester 1:4.5; *C. rugosa* lipase 40%, w/w; added water 15%, w/w; 5.3 kPa; ■ 37°C, □ 42°C, ◆ 47°C, ● 52°C). For abbreviations see Figure 1.

If the ratio of trimethylolpropane and rapeseed oil methyl ester was changed to 1:4.5, a 68% (72%, when calculated on the basis of the limiting substrate trimethylolpropane) yield of trimethylolpropane tri-ester was obtained in 20 h, and the yield was further increased to about 70% when the pressure was reduced to 2.0 kPa. In this case, however, a considerable amount of the excess rapeseed oil methyl ester was left in the product mixture. A high total conversion of 95% was obtained with the molar ratio of 1:3.5 at 5.3 kPa with little or no residual substrates present, a result preferred from an industrial application point of view over the maximum conversion to the tri-ester obtained with an excess of rapeseed oil methyl ester.

In the chemical transesterification only a slight excess of rapeseed oil methyl ester was preferred, with highest conversions obtained using the molar ratio of 1:3.1–1:3.2 (Table 2). Under these conditions total conversions of the order of 86% or higher could be easily obtained. When the reaction temperature was held at 80 to 120°C, optimally at 110–120°C, total conversion to trimethylolpropane esters was up to 99.0% in about 10 h. No residual rapeseed oil methyl ester could be found under these conditions. In chemical catalysis, markedly lower catalyst quantities could be used. When the catalyst quantity was varied within the range of 0.1 to 2.0% (w/w), the total conversion was always at least 70%. The highest conversions of at least 85% were obtained with 0.7% (w/w) catalyst.

Because preliminary experiments (22) had clearly suggested that the conversion of rapeseed oil methyl ester to the desired trimethylolpropane esters might increase with the use of an immobilized lipase, several carrier materials for the *C. rugosa* lipase employed were investigated (Fig. 3). The highest total conversions of about 95% to trimethylolpropane esters were obtained with lipase immobilized on Celite R-630 (40%, w/w, 47°C, 5.3 kPa, 42 h). This compares with about 70% yield of the tri-ester with 20% (w/w) and 55% yield with 10% (w/w) of the original solid lipase preparation in 68 h. With Duolites ES-561 and ES-762, GDC 200, GCC and HPA 25 as carriers, typically about 70% total conversion was obtained. With the use of silica gel the reaction did not proceed

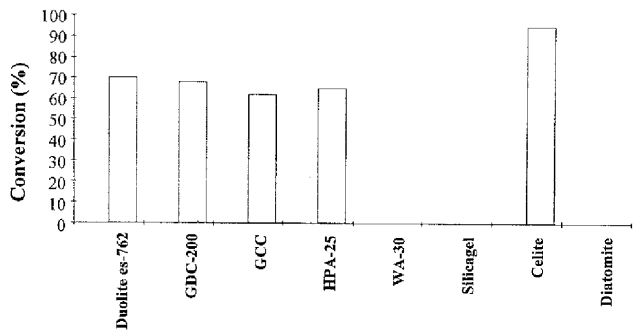

FIG. 3. Effect of the type of carrier on the total conversion of trimethylolpropane to trimethylolpropane esters (*C. rugosa* lipase 40%, w/w; added water 15%, 47°C, 5.3 kPa, 42 h). For abbreviation see Figure 1.

beyond the di-ester stage, and with WA 30 and Diatomite no conversion could be obtained.

Figure 4 illustrates example time courses of the formation of trimethylolpropane esters both with the *C. rugosa* lipase immobilized on Duolite ES-561 (Fig. 4A) and with the commercial immobilized *Rhizomucor miehei* lipase Lipozyme IM 20 (Fig. 4B). With the *C. rugosa* lipase about 70% conversion to the trimethylolpropane tri-ester was reached in 68 h at 47°C with 15% added water, while an equal conversion was reached with the Lipozyme IM 20 at 58°C with no added water in 24 h, 84% conversion in 48 h, and 90% in 66 h. With Lipozyme IM 20 a total conversion of as high as 92.5% to trimethylolpropane esters was obtained in 22 h. Lipozyme IM 20 tolerates higher operating temperatures which, in part, explains the result. It should also be noted that the ester synthesis and transesterification activities of different lipases do not necessarily correlate with the respective hydrolytic (lipase) activities (30), and that the relative hydrolytic activity of Lipozyme IM 20 was, in this case, only about 5% of that of the same quantity of the solid *C. rugosa* lipase preparation used. Stepwise addition of lipase did not markedly improve the result. The commercial immobilized *C. antarctica* lipase Novozyme 435 behaved quite differently. This enzyme was much more sensitive to the water content of the reaction mixture than Lipozyme IM 20, and typically only mono- and di-esters were formed in 66 h with at most traces of trimethylolpropane tri-ester.

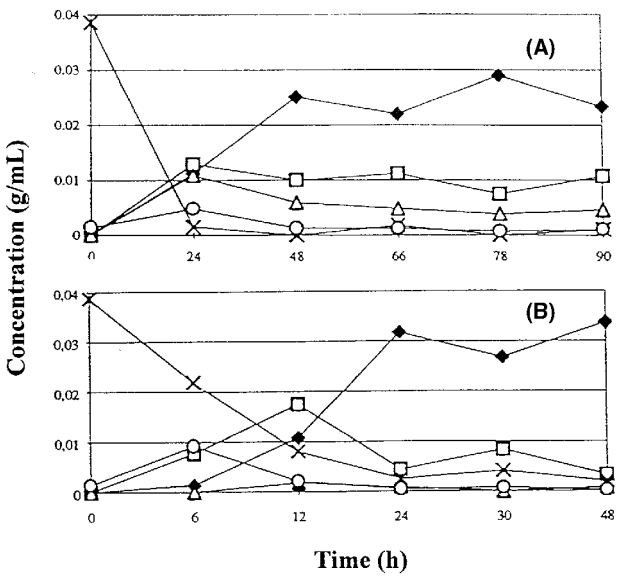

FIG. 4. Typical time courses of transesterification between trimethylolpropane and rapeseed oil methyl ester, catalyzed by (a) *C. rugosa* lipase immobilized in Duolite ES 561 (47°C, 5.3 kPa, biocatalyst 40%, w/w; water 15% of substrates) and (b) immobilized *R. miehei* lipase Lipozyme IM 20 (58°C, 5.3 kPa, biocatalyst 40%, w/w, no added water); ◆ trimethylolpropane tri-ester; □ trimethylolpropane di-ester; △ trimethylolpropane mono-ester; × rapeseed oil methyl ester; ○ unknown).

Hydraulic fluid prepared from the trimethylpropanol esters produced in the present work was characterized, tested, and compared both with pure comparable vegetable oil-based and commercial synthetic ester-based hydraulic fluids, using a commercial mineral-oil-based hydraulic fluid as the reference. The main purpose for a hydraulic fluid is to reduce friction and to decrease wear. Owing to the high oxygen content of the ester-based hydraulic fluids they can form a very stable, unimolecular film over a metal surface. They are also nontoxic, nonbioaccumulating, and biodegradable. The hydraulic fluid composition prepared in this work had a viscosity grade of 32, and contained (w/w) 0.5–2.5% antioxidant, 1.0–5.0% pour-point depressor, 0.4–2.0% antiwear agent, and 0.1–0.5% antifoam agent. Table 3 shows some of the properties of the hydraulic fluid prepared on the basis of the trimethylolpropane esters produced in this work. The viscosity index calculated on the basis of the ASTM D2270 standard illustrates temperature stability, and was 220 in the present work as compared with 187 for the reference mineral oil. With a filterability of 92%, the hydraulic fluid prepared compared well with commercial preparations and was superior to the reference mineral oil (72% filterability). Small particles can cause serious wear problems in fine hydraulic systems. The hydraulic fluid prepared had an ISO 4406 purity class of 13/8, which differed little from the comparable commercial products and was better than the reference mineral oil (14/10). Cold stability is very important for machines working out-of-doors during the winter season. The pour point, according to the ASTM D97 standard, was –41°C, while that of the standard mineral oil was –40°C. Only a commercial synthetic ester-based fluid had a higher pour point of –48°C. The friction, wear, and oxidation and corrosion stability were also comparable to the values of the reference products. The set limit for wear according to the ASTM D2783 and D4172 standards is 0.5 mm. All, with the exception of the mineral oil reference sample, passed this limit. The biodegradability in all cases but the mineral oil exceeded 90%, with the OECD 301 standard set limit of 70%.

In conclusion, it was demonstrated that lipase-catalyzed transesterification of trimethylolpropane with methyl ester of rapeseed oil fatty acids to biodegradable trimethylolpropane tri-ester is possible in a relatively high yield. With *C. rugosa* lipase, at a reduced pressure of 5.3 kPa, at 47°C and 15% of

TABLE 3
Characteristics of the Trimethylpropane-Based Hydraulic Fluid as Compared with a Number of Commercial Products

Characteristic[a]	Hydraulic fluid[b]				
	a	b	c	d	e
Viscosity [mm^2/s, 40°C]					
ASTM D445	32.9	33.8	50.4	29.3	32.6
Viscosity stability index					
ASTM D2207 [–]	220	220	190	242	187
Filterability					
CETOP RP1244 [%]	92	96	—	—	72
Purity class					
ISO 4406	13/8	13/8	13/8	15/10	14/10
Foaming					
ASTM D892 [s]	175	120	151	40	150
[mm]	33	20	28	8	30
Color					
ASTM D1500 (scale 1–20)	4–	3+	4	4	—
Pour point					
ASTM D97 [°C]	–41	–39	–36	–39	–40
Cold viscosity, –10°C [mm^2/s]	1030	829	1430	2160	496
Friction and wear					
ASTM D2783 and D4172 w car [mm]	0.40	0.48	0.46	0.41	0.54
Oxidation stability					
ASTM D525 [▽ psi]	42	30	39	29	39
DIN 51586 [▽ visc. %]	12.4	28.8	20.3	24.1	15.8
Corrosion stability					
Cincinnati-Milacron [mg][c]	–0.3	0.5	0	1.2	0.4
Biodegradability					
CEC-L-33-A-93 [%]	>90	>90	>90	>90	—
OECD 301 F [%]	>85	>85	>85	—	—

[a]Standards for each characteristic are specified in the Materials and Methods section.
[b]a, Present oil; b, Raisio Bio Safe 32, new vegetable-oil-based product; c, Raisio Bio Safe 46, synthetic ester-based product; d, commercial synthetic ester-based product; e, commercial mineral oil-based product.
[c]Weight change of steel rod.

added water, 64% of the trimethylolpropane was converted to the tri-ester in 24 h. A total conversion to the trimethylolpropane esters of about 98% was obtained with the molar substrate ratio of 1:3.5 at a reduced pressure of 2.0 kPa at 42°C in 72 h, and of about 95% at 5.3 kPa at 47°C in 68 h. In chemical transesterification using sodium methylate as the catalyst, up to 99% total conversion to trimethylolpropane esters was obtained in about 10 h, but the temperature required was as high as 120°C. The trimethylolpropane esters produced appeared to meet the requirements as a basic fluid for hydraulic oils quite well. The laboratory testing showed that, especially at higher temperatures, trimethylolpropane esters were superior to vegetable oil-based hydraulic fluids and comparable to commercially available synthetic ester-based fluids. The pour point was lower, the friction and wear resistance was excellent, and the cold, oxidation, and viscosity stability was better than those of the reference fluids.

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[Received June 15, 1998; accepted August 25, 1998]