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Synthesis of Deuterated and Protiated Triacylglycerides by Using 1,1′-Carbonyldiimidazole Activated Fatty Acids

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Synthetic model triacylglyceride oils are important compounds for applications in pharmaceutical and food chemistry. Herein, a practical and highly efficient methodology for the synthesis of saturated and unsaturated triacylglycerides utilizing saturated and unsaturated fatty acids activated by 1,1′-carbonyldiimidazole (CDI) has been developed and applied in the synthesis of deuterated medium chain triglyceride (MCT) oil for studies of plant-based and diary food emulsions. The deuterium labelled compounds were used to gain new insight into the mechanism of the reaction, which was confirmed by density-functional theory (DFT) calculations.

1. Introduction

Triacylglycerides are widely used components in the food and pharmaceutical sector.[1,2] The employed triacylglycerides are often composed of either unsaturated or saturated fatty acids depending on nutritional and applicative aspects.[2–4] Natural oils like coconut oil, milk fat and palm kernel oil,[4] as well as pure synthesized medium chain triglyceride (MCT) oils contain mainly saturated fatty acids, while sunflower, olive, or rapeseed oil[5] contain significant amounts of unsaturated fatty acids.

Commonly, the synthesis of triacylglyceride oils is performed either with a chemical catalyst like sulfuric acid or *p*-toluenesulfonic acid at high reaction temperatures, or with a biological catalyst like lipase (Scheme 1a).[2,4] However, both synthetic ways have major disadvantages especially when working with expensive starting materials such as synthetic or isotope labelled fatty acids. The classical chemical synthesis under acidic conditions usually requires an excess of fatty acids and is rich in by-products which need to be removed, causing an overall loss of expensive starting materials.[6] In recent years an additional methodology using *N*,*N*′-dicyclohexylcarbodiimide (DCC) in the presence of 4-dimethylaminopyridine (DMAP) has been developed and even utilized in the synthesis of isotope labelled triacylglycerides.[7] However, this method suffers from the formation of quantitative amounts of dicyclohexylurea (DCU) as a side product, which makes the purification of the final product particularly challenging.

On the other hand, the classical biological synthesis using enzymes is highly sensitive to the structure of the fatty acids. This makes the use of unnatural, or isotope labelled fatty acids in enzyme reactions particularly challenging.[4]

The use of 1,1′-carbonyldiimidazole (CDI) is very well established in peptide synthesis for the formation of amide bonds.[8] In addition, it is frequently utilized to convert amines into ureas or carbamates.[9] The application of CDI in ester formation is less common since cheaper reagents like thionyl chloride or DCC (*Steglich* esterification) are generally preferred. To our knowledge, the use of CDI in the formation of triacylglycerides has not been reported yet. However, when working with expensive isotope labelled starting materials the higher cost of CDI compared to alternative reagents can be outmatched by an increase in yield and purity.

For our research regarding plant-based and dairy emulsions we were interested in obtaining deuterated triacylglycerides. Varying the isotopic composition with deuterated solvents such as D_2O is a common technique in NMR spectroscopy[10] and is particularly important in neutron scattering as it allows to vary the contrast between different parts of a sample. This can be used to resolve complex structural details.[11] It is therefore important for the field of food and pharmaceutical science to provide also the often-used triacylglycerides in well-defined quality regarding the degree of deuteration.

Therefore, we explore a practical approach of chemical synthesis via CDI activated fatty acids to increase the yield and purity of deuterated as well as protiated triacylglycerides with either un- or saturated fatty acids of varying chain lengths (Scheme 1b). Our main goal is the synthesis of protiated and deuterated MCT oils as well as a synthetic plant oil mimic containing mostly unsaturated fatty acids.

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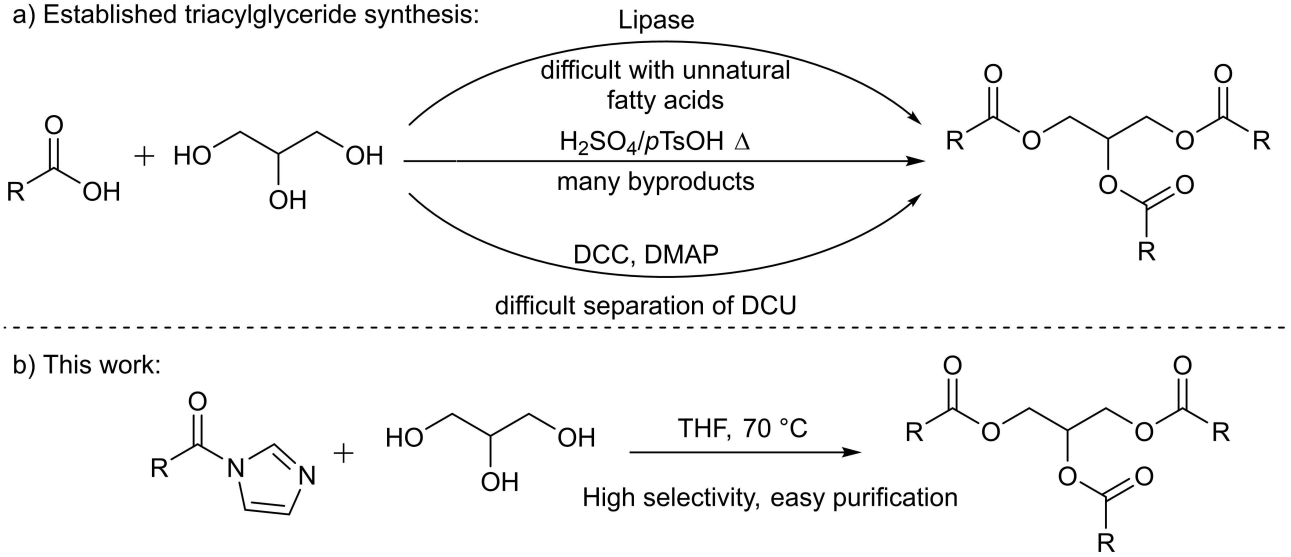
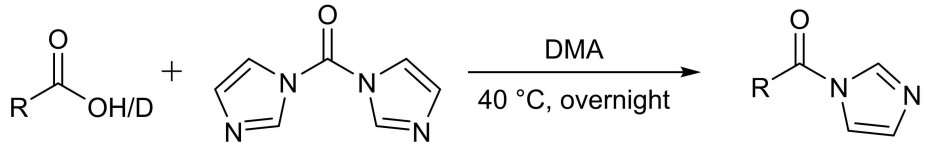


Scheme 1. Different methodologies for the synthesis of triacylglycerides.

2. Results and Discussion

2.1. Activation of the Fatty Acids with 1,1′-Carbonyldiimidazole

Initially, we systematically explored the activation of fatty acids with various chain lengths, isotopic composition, and degree of unsaturation with CDI. First, we explored the reaction of octanoic (**1 a**) and decanoic acid (**1 b**) with CDI (**2**, 1.5 mol. equiv.) in dimethylacetamide (DMA) at 40 °C overnight (Scheme 2). Then the activated fatty acids were precipitated in water to obtain 95–97% of the crude products **3 a** and **3 b** as amorphous powders. Recrystallization from *n*-hexane resulted in the formation of colourless needles in 82–85% yield. The products showed signs of hydrolysis after a few weeks of storage in a fridge at 2 °C but could be stored inside an argon filled glovebox for multiple months without any signs of decomposition. When the reaction was repeated with the perdeuterated fatty acids **1 c** and **1 d**, the corresponding products **3 c** and **3 d** were obtained in slightly lower yields of 91–94%. As NMR-analysis showed a high purity of the crude products after a simple precipitation in water, we decided to proceed without any additional recrystallization to minimize the loss of material. However, in both cases we observed partial back exchange of the deuterium atoms in the α-position of the amide groups (0.61 and 0.56 protons respectively as a mixture of α-CHD and α-CH_2 compounds), thus reducing the deuteration degree of that position to 70%. We assumed that the incorporated protons originated from the non-deuterated carboxylic acid groups. Therefore, we exchanged the carboxylic protons in both perdeuterated (**1 e**) and protiated octanoic acid (**1 f**) with deuterium by stirring the acids with an excess of D_2O and repeated the reactions with CDI. In the case of the perdeuterated octanoic acid **1 e** this did indeed prevent the undesired exchange, with the deuteration degree of the product **3 e** matching that of the starting material (97%). In the



1a: C_7H_{15}-COOH
1b: C_9H_{19}-COOH
1c: $C_7D_{14.5}H_{0.5}$-COOH (97% D)
1d: $C_9D_{18.6}H_{0.4}$-COOH (98% D)
1e: $C_7D_{14.5}H_{0.5}$-COOD (97%D)
1f: C_7H_{15}-COOD
1g: $C_{15}H_{31}$-COOH
1h: tech. grade linoleic acid

2 (1.5 equiv.)

3a: 95%
3b: 97%
3c: 91% (93% D, $d_{13.9}$)
3d: 94% (95% D, $d_{18.0}$)
3e: 87% (97% D, $d_{14.5}$)
3f: 88% (1.1% D, $d_{0.17}$)
3g: 95%
3h: 94%

Scheme 2. Activation of the fatty acids **1 a**–**1 h** with CDI (**2**). The percentages refer to isolated yields without recrystallization. The numbers in parentheses show the degree of deuteration of the aliphatic chains in the products.

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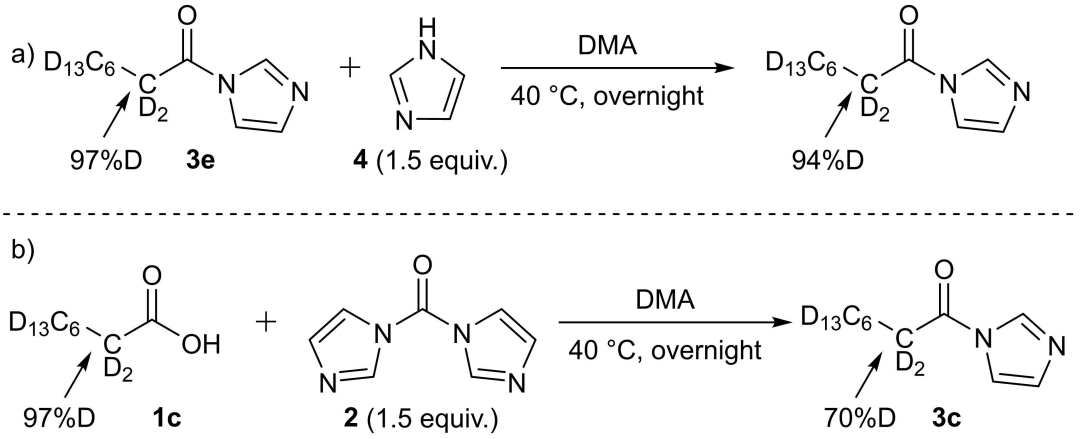
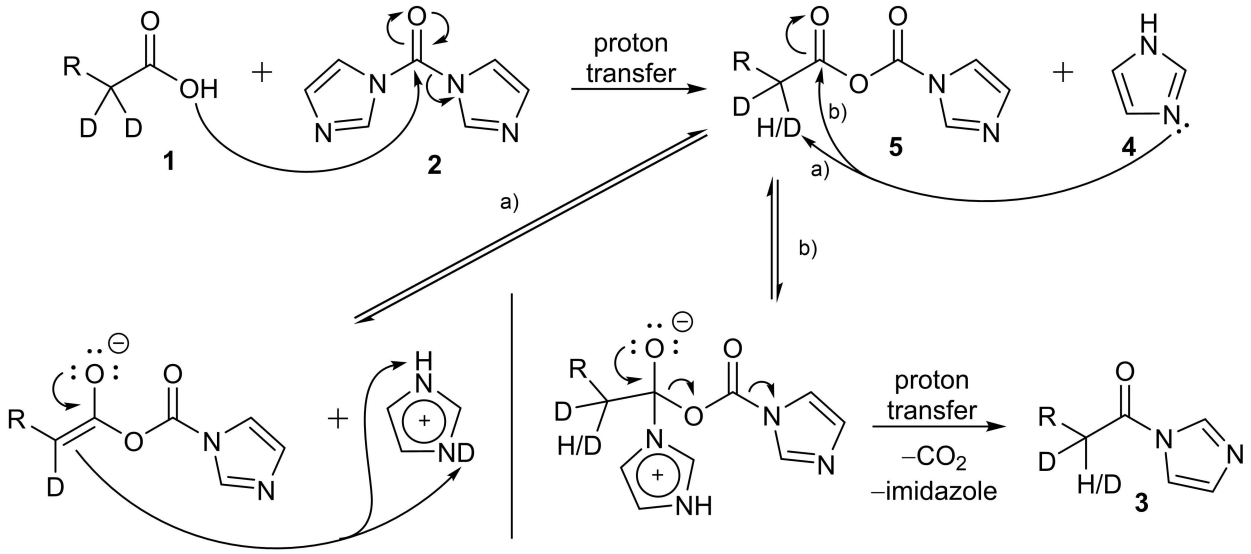
case of the protiated octanoic acid **1f** we observed an incorporation of 0.17 equivalents of deuterium in the α-position during the reaction with CDI (**3f**). Finally, the CDI-activation was also applied successfully to hexadecanoic acid (**1g**) and technical grade linoleic acid (**1h**, 62% linoleic acid, 30% oleic acid and 8% other fatty acids) to obtain the corresponding products **3g** and **3h** in 95–94% yield.

As we assumed that the imidazole, which is formed as a side product of the reaction, was responsible for the exchange, we stirred the perdeuterated compound **3e** with 1.5 molar equivalents of imidazole (**4**) in DMA at 40 °C overnight (Scheme 3a). This resulted in only a slight exchange in the α-position of approximately 3%, most likely due to the low α-acidity of amides.[12] Therefore, the much higher deuterium exchange observed during the reaction of the perdeuterated acid **1c** with CDI (Scheme 3b) could not have taken place after the completed activation. Previous studies by *Staab* and *Maleck*[13] had shown that the activation of carboxylic acids with CDI proceeds according to a stepwise mechanism via a mixed anhydride (**5**). Since the deuterons of this mixed anhydride are significantly more acidic due to the strong electron withdrawing effect of the O–C=O-group, we propose the following mechanism for the exchange (Scheme 4): The imidazole **4** that is formed in the first step can attack the mixed anhydride **5** in two different ways. It can either reversibly remove a deuteron in the α-position to form an enolate (path a) or attack the carboxylic carbon (path b). During the reversal of the enolization, the imidazolium ion can either transfer a proton or a deuteron. Our results suggest that the transfer of the proton is favoured, presumably due to an isotope effect. Finally, path b) will result in the irreversible elimination of CO_2 and imidazole to form the activated fatty acid.

This behaviour could be explained by assuming that the ND-bond in 1*D*-imidazole is significantly more stable than the NH-bond in 1*H*-imidazole. To confirm this theory, we performed density-functional theory (DFT) calculations on a model system (Scheme 5). All structures as well as their Gibbs free energies at 313 K (40 °C) were calculated at the B2PLYP/def2-TZVPP level of



Scheme 3. Mechanistical investigations regarding the proton deuterium exchange in the α-position.



Scheme 4. Proposed mechanism of the partial hydrogen exchange in the α-position.

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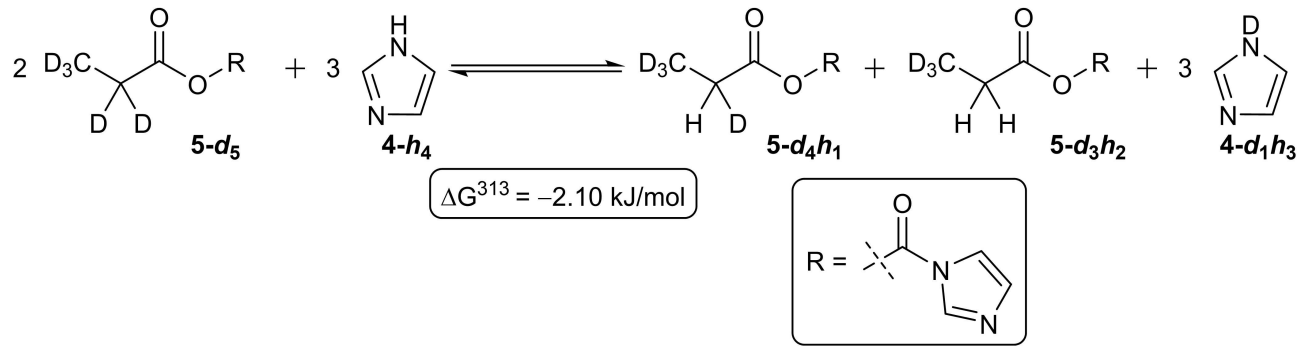
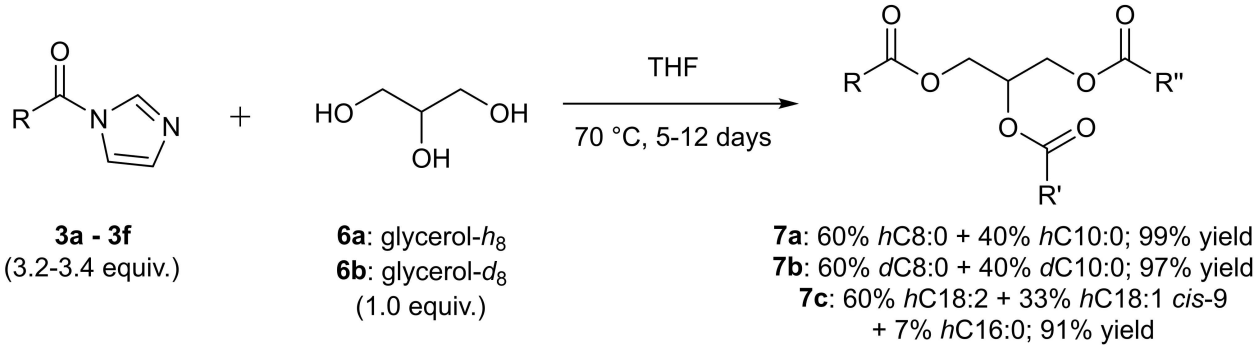


Scheme 5. Model system for the deuterium proton exchange in the α-position calculated at the B2PLYP/def2-TZVPP level of theory.



Scheme 6. Synthesis of the triacylglycerides **7a**–**7c** using the CDI-activated fatty acids **3a**–**3h**.

theory. Thus, we could confirm that the reaction of two equivalents of the fully deuterated mixed anhydride **5-d_5** with three equivalents of 1*H*-imidazole **4-h_4** to form the mono protiated anhydride **5-d_4h_1**, the double protiated anhydride **5-d_3h_2** and three equivalents of 1*D*-imidazole **4-d_1h_3** is energetically favoured by 2.10 kJ/mol. Therefore, the reason for the partial deuterium exchange in the α-position seems to be indeed the energy gained through the deuteration of imidazole.

2.2. Synthesis of Triacylglycerides

With the activated fatty acids **3a**–**3h** in hand, we proceeded to the formation of the triacylglycerides (Scheme 6). In a first step, we aimed to reproduce a commercially available protiated MCT oil containing 60% C8 and 40% C10-acid. Thus, we mixed a total of three molar equivalents of the corresponding CDI-adducts **3a** (1.80 mol. equiv.) and **3b** (1.20 mol. equiv.) with glycerol-h_8 (**6a**) in tetrahydrofuran (THF). The reaction flask was degassed, sealed with a Teflon stopper, and heated to 70 °C while monitoring the reaction via size exclusion chromatography (SEC, Figure 1). This was necessary, to avoid partial hydrolysis due to traces of humidity. After two days of reaction time the mixture contained mostly the desired triacylglyceride alongside small amounts of diacylglyceride. As the SEC showed that the activated fatty acids were almost completely used up, we added a small excess (totalling 3.2 molar equivalents) to

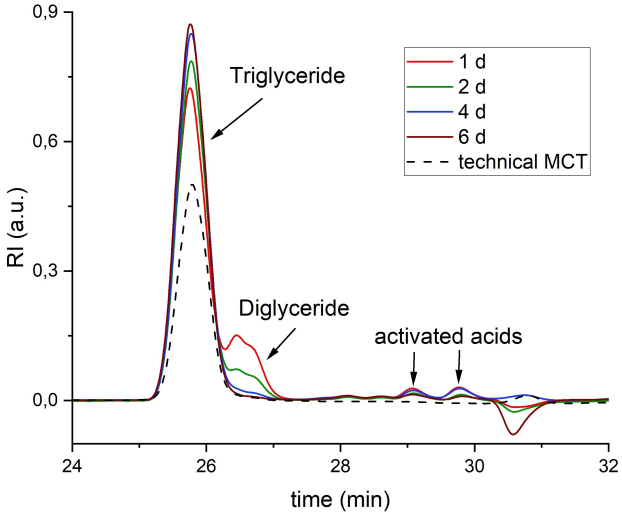


Figure 1. SEC spectra used to monitor the formation of the MCT oil **7a**.

push the reaction to completion. After an additional four days of reaction time the SEC showed complete conversion with the formed triacylglyceride perfectly matching the spectrum of the commercial product. We performed a simple aqueous work-up to remove the imidazole and remaining starting materials and obtained the desired MCT oil **7a** in 99% yield with high purity as confirmed by ^{1}H-NMR.

Encouraged by our results, we set out to synthesize a deuterated version of the MCT oil. As a deuteration degree of

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93% was deemed sufficient for masking the oil in our planned neutron experiments we decided to utilize the activated fatty acids **3c** and **3d** despite their reduced deuteration in the α-position, alongside glycerol-d_8 (**6b**). This time the reaction proceeded significantly slower and full conversion required 9 days (Figure 2). This difference in reaction speed most likely results from a kinetic isotope effect of the deuterated hydroxyl groups (see Supporting Information for additional experiments). Nevertheless, we obtained 97% of the deuterated MCT oil **7b** on a 20 g scale (84% yield based on the free acids).

Finally, we aimed to proof that our protocol can be utilized with unsaturated fatty acids. Thus, we synthesized the synthetical plant oil mimic **7c** from 0.18 molar equivalents of the activated palmitic acid **3g** and 3.22 molar equivalents of the CDI adduct of technical grade linoleic acid **3h** in 91% yield after 12 days of reaction time (Figure 3).

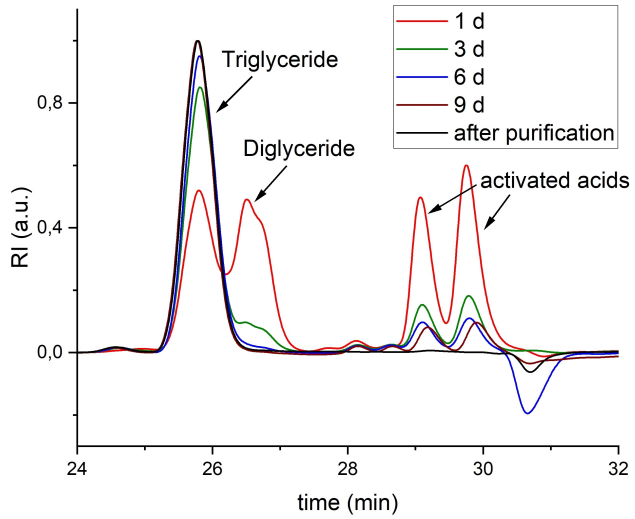

Figure 2. SEC spectra used to monitor the formation of the deuterated MCT oil **7b**.

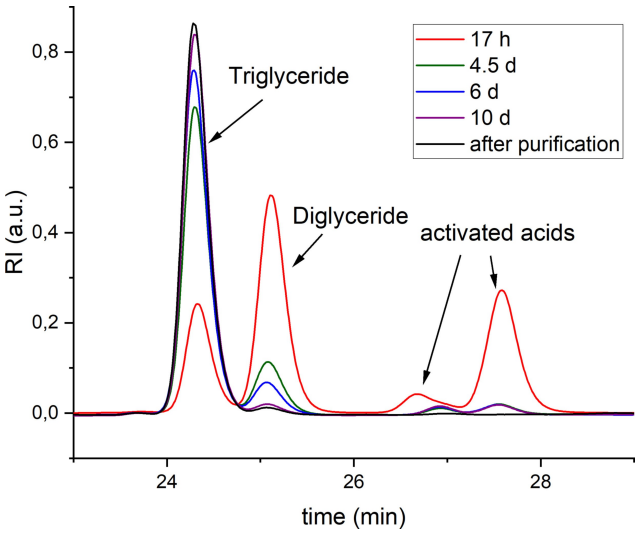

Figure 3. SEC spectra used to monitor the formation of the synthetic plant oil mimic **7c**.

3. Conclusions

In summary we have developed a practical methodology for the synthesis of triacylglycerides utilizing CDI-activated fatty acids. This method has been used to synthesize both protiated and deuterated MCT oil as well as a synthetic plant oil mimic containing unsaturated fatty acids. Compared to previously established methods our protocol produces high yields and excellent purity without needing any complex purification procedures, which is especially relevant for the synthesis of isotope labelled compounds. In addition, the use of deuterated fatty acids has allowed us to gather new insights into the mechanism of the activation of fatty acids with CDI. We proved that the nucleophilic attack of imidazole at the carboxyl group competes with a deprotonation in the α-position. These findings have been supported by DFT calculations.

Experimental Section

Materials and Methods

All chemicals were used as received. Pt/C (10 wt%), DMA (anhydrous, 99.8%), CDI (≥97%), Palmitic acid (≥98%), linoleic acid (technical grade, 62% linoleic acid and 30% oleic acid according to supplier CoA), glycerol (≥99%), THF (anhydrous, 99.9%) and Na_2CO_3 (≥99.5%) were purchased from *Sigma-Aldrich Co*, USA. HCl (≥37%) and $MgSO_4$ were purchased from *Merck KGaA*, Germany. *iso*-Hexane (HPLC grade) and $CHCl_3$ (Normapur) were purchased from *vwr International LLC*, USA. Octanoic acid (98+%) and decanoic acid (99%) were purchased from *ThermoScientific Co LLC*, USA. Glycerol-d_8 (99% D) and D_2O (99.90%) were purchased from *Eurisotop SAS, Cambridge Isotope Laboratories Inc.*

NMR spectra were collected on a *Bruker Co* (USA) Avance III 600 MHz spectrometer equipped with a Prodigy cryoprobe with a 5 mm PFG AutoX DB probe or a *Varian* INOVA 400 MHz spectrometer and analysed using MestReNova (*Mestrelab Research S.L.*). All samples were measured at 295 K. Samples were either diluted in $CDCl_3$ (for ^{1}H-NMR) or $CHCl_3$ (for ^{2}H-NMR). 1,2,4,5-Tetrabromobenzene or benzyl chloride were used as internal standards to determine the deuteration degree and purity of the products. The full spectra are part of the supporting information.

Size-exclusion chromatography (SEC) experiments were carried out using an *Agilent Technologies Inc.* (USA) 1260 Infinity SEC instrument equipped with an Optilab T-rex differential refractive index (RI) detector and three PolyPore columns at 40 °C. The solvent was a mixture of tetrahydrofuran (THF), *N,N*-dimethylacetamide (DMA), and acetic acid (84:15:1 by volume) at a flow rate of 1 mL/min. The RI signals were analysed using ASTRA software (*Wyatt*) alongside OriginPro (*OriginLab*).

All density-functional theory calculations have been performed with *Orca* Version 5.0.3.[14] Geometry optimizations and frequency calculations (NumFreq) in the gas phase have been performed using the double-hybrid B2PLYP functional[15] in combination with the def2-TZVPP basis set.[16] In order to account for the reaction temperature of 40 °C (313.15 K), the entropies obtained at 298.15 K were multiplied by 1.05 when calculating the *Gibbs* free energies. Solvent effects were initially estimated using the SMD continuum solvation model,[17] however they were found to be negligible when dealing with molecules that only differ in one hydrogen isotope and were therefore omitted in all future calculations. Detailed

results from the DFT calculations can be found in the supporting information.

Deuteration of Fatty Acids

d-Octanoic acid (**1 c**) was prepared via platinum catalysed hydrothermal deuteration with D_2O:[18] 20.0 g (139 mmol, 1.0 equiv.) octanoic acid, 5.55 g (139 mmol, 1.0 equiv.) NaOH, 2.7 g (13.9 mmol, 10 mol%) 10 wt% Pt/C and 300 mL D_2O were mixed in a pressure reactor. The reactor was sealed and heated to 220 °C for 3 cycles of 3 days each. After every cycle the H_2O/D_2O mixture was replaced with fresh D_2O. Once the deuteration was completed the catalyst was removed through filtration. The filtrate was acidified to pH 2 with hydrochloric acid and the aqueous phase was extracted with diethyl ether (3x). The combined organic phases were dried over $MgSO_4$ and concentrated *in vacuo* to give the desired deuterated fatty acid. ^{1}H-NMR (400 MHz, $CDCl_3$) $\delta = 9.70$ (br s, 1 H), 2.30 (s, 0.02 H), 1.58 (s, 0.05 H), 1.43 (s, 0.06 H), 1.24 (m, 0.21 H), 0.83 (s, 0.07 H); ^{2}H-NMR (61 MHz, $CDCl_3$) δ 2.50–2.15 (m, 2 H), 1.59 (s, 2 H), 1.41–1.11 (m, 8 H), 0.97–0.72 (m, 3 H).

d-Decanoic acid (**1 d**) was prepared analogously. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.65 (s, 1 H), 2.31 (s, 0.02 H), 1.58 (s, 0.02 H), 1.35–1.16 (m, 0.12 H), 0.83 (s, 0.04 H); ^{2}H-NMR (61 MHz, $CDCl_3$) δ 2.50–2.14 (m, 2 H), 1.74–1.47 (m, 2 H), 1.47–1.07 (m, 12 H), 0.98–0.71 (m, 3 H).

In order to exchange the carboxylic protons of *d*-octanoic acid with deuterium (**1 e**), 1.44 g (10 mmol, 1.0 equiv.) of the acid were stirred with 20 mL (1.1 mol, 110 equiv.) of D_2O for 1 hour in a closed vessel. Then the mixture was briefly centrifuged to separate the emulsion. The organic phase was carefully separated with a glass pipette and dried under high vacuum. ^{1}H-NMR analysis using dry toluene-d_8 as a solvent confirmed a deuteration degree of the carboxylic acid of $\geq$98%. The material was then stored under argon protective gas to prevent back exchange with moisture. The same procedure was also applied to protiated octanoic acid (**1 f**).

General Procedure for the Activation of Fatty Acids with CDI

The fatty acid (**1**, 1.0 equiv.) was dissolved in anhydrous DMA (1.5–2.0 M) inside an argon filled glovebox. Then CDI (**2**, 1.5 equiv.) was slowly added, which resulted in the formation of CO_2 gas. The reaction flask was fitted with a balloon and stirred inside the glovebox at 40 °C overnight. Then the reaction mixture was poured into distilled water (30 times more than the amount of DMA). If the product (**3**) precipitated as a solid it was filtered off, washed with cold water and dried under high vacuum. For liquid products the organic phase was separated. Then, the aqueous phase was extracted with $CHCl_3$ and the combined organic phases were dried over $MgSO_4$. Subsequently, the solvent was evaporated and the product dried under high vacuum. The products could be stored under argon at room temperature for several months without significant decomposition. Exposure to moisture would however result in partial hydrolysis within weeks.

1-(1H-Imidazol-1-yl)octan-1-one (3 a)

1-(1*H*-Imidazol-1-yl)octan-1-one was synthesized according to the general procedure using 14.4 g (100 mmol) octanoic acid (**1 a**) to obtain 18.4 g (94.7 mmol, 95%) crude product as a colourless powder. Recrystallization from n hexane resulted in 15.9 g of the title compound (81.8 mmol, 82%) as colourless needles. ^{1}H-NMR (600 MHz, $CDCl_3$) $\delta = 8.16$ (s, 1 H), 7.47 (s, 1 H), 7.10 (d, J = 1.7 Hz, 1 H), 2.85 (t, J = 7.4 Hz, 2 H), 1.80 (p, J = 7.4 Hz, 2 H), 1.44–1.37 (m, 2 H), 1.37–1.25 (m, 6 H), 0.89 (t, J = 6.8 Hz, 3 H). The analytics match with previously published literature information.[19]

1-(1H-Imidazol-1-yl)decan-1-one (3 b)

1-(1*H*-Imidazol-1-yl)octan-1-one was synthesized according to the general procedure using 10.0 g (58 mmol) decanoic acid (**1 b**) to obtain 12.5 g (56.2 mmol, 97%) crude product as a colourless powder. Recrystallization from n hexane resulted in 11.0 g of the title compound (49.3 mmol, 85%) as colourless needles. ^{1}H-NMR (600 MHz, $CDCl_3$) $\delta = 8.16$ (s, 1 H), 7.47 (s, 1 H), 7.09 (d, J = 0.9 Hz, 1 H), 2.85 (t, J = 7.4 Hz, 2 H), 1.79 (p, J = 7.5 Hz, 2 H), 1.44–1.37 (m, 2 H), 1.36–1.20 (m, 10 H), 0.88 (t, J = 6.9 Hz, 3 H). The analytics match with previously published literature information.[19]

1-(1H-Imidazol-1-yl)octan-1-one-$d_{13.9}$ (3 c)

1-(1*H*-Imidazol-1-yl)octan-1-one-$d_{13.9}$ was synthesized according to the general procedure using 15.0 g (94 mmol) *d*-octanoic acid (**1 c**, 97% D) to obtain 17.8 g (85.5 mmol, 91%) crude product as a slightly yellow powder. The crude product was used in the next step without any further purification. ^{1}H-NMR (400 MHz, $CDCl_3$) $\delta =$ 8.16 (s, 1 H), 7.47 (s, 1 H), 7.10 (d, J = 0.9 Hz, 1 H), 2.87–2.78 (m, 0.61 H), 1.81–1.72 (m, 0.08 H), 1.40–1.20 (m, 0.27 H), 0.87–0.80 (m, 0.06 H); ^{2}H-NMR (92 MHz, $CDCl_3$) δ 3.01–2.49 (m, 1.4 H), 1.89–1.55 (m, 2 H), 1.50–1.05 (m, 8 H), 0.99–0.67 (m, 3 H).

1-(1H-Imidazol-1-yl)decan-1-one-$d_{18.0}$ (3 d)

1-(1*H*-Imidazol-1-yl)octan-1-one-$d_{18.0}$ was synthesized according to the general procedure using 15.0 g (78 mmol) *d*-decanoic acid (**1 d**, 98% D) to obtain 17.7 g (73.6 mmol, 94%) crude product as a colourless powder. The crude product was used in the next step without any further purification. ^{1}H-NMR (400 MHz, $CDCl_3$) $\delta = 8.15$ (s, 1 H), 7.47 (s, 1 H), 7.10 (d, J = 0.9 Hz, 1 H), 2.86-2.78 (m, 0.56 H), 1.78–1.73 (m, 0.05 H), 1.38–1.17 (m, 0.12 H), 0.85–0.79 (m, 0.03 H); ^{2}H-NMR (92 MHz, $CDCl_3$) δ 3.09–2.50 (m, 1.4 H), 1.89–1.56 (m, 2 H), 1.47–1.06 (m, 12 H), 0.96–0.71 (m, 3 H).

1-(1H-Imidazol-1-yl)octan-1-one-$d_{14.5}$ (3 e)

1-(1*H*-Imidazol-1-yl)octan-1-one-$d_{14.5}$ was synthesized according to the general procedure using 1.6 g (9.7 mmol) of *d*-octanoic acid (**1 e**, 97% D, with deuterated carboxylic acid group) to obtain 1.8 g (8.4 mmol, 87%) crude product as a white powder. ^{1}H-NMR (400 MHz, $CDCl_3$) $\delta = 8.15$ (s, 1 H), 7.47 (s, 1 H), 7.09 (s, 1 H), 2.85–2.75 (m, 0.07 H), 1.79–1.69 (m, 0.08 H), 1.39–1.16 (m, 0.28 H), 0.87–0.76 (m, 0.07 H); ^{2}H-NMR (61 MHz, $CHCl_3$) $\delta = 2.74$ (br s, 2 H), 1.66 (br s, 2 H), 1.40–0.92 (m, 8 H), 0.74 (br s, 3 H).

1-(1H-Imidazol-1-yl)octan-1-one-$d_{0.17}$ (3 f)

1-(1*H*-Imidazol-1-yl)octan-1-one-$d_{0.17}$ was synthesized according to the general procedure using 1.2 g (8.3 mmol) of octanoic acid (**1 f**, with deuterated carboxylic acid group) to obtain 1.4 g (7.3 mmol, 88%) crude product as a white powder. In order to determine the amount of deuterium incorporated in the α-position a ^{2}H-NMR spectrum with bromobenzene-d_5 as internal standard was recorded. ^{1}H-NMR (400 MHz, $CDCl_3$) $\delta = 8.16$ (s, 1 H), 7.47 (s, 1 H), 7.09 (s, 1 H), 2.85 (t, J = 7.4 Hz, 1.84 H), 1.80 (p, J = 7.3 Hz, 2 H), 1.48–1.22 (m, 8 H), 0.89 (t, J = 6.4 Hz, 3 H); ^{2}H-NMR (61 MHz, $CHCl_3$) $\delta = 2.84$ (s, 0.17 H based on internal standard).

1-(1H-Imidazol-1-yl)hexadecan-1-one (3 g)

1-(1*H*-Imidazol-1-yl)hexadecan-1-one was synthesized according to the general procedure using 1.0 g (3.9 mmol) hexadecenoic acid

(**1 g**) to obtain 1.1 g (3.7 mmol, 95 %) crude product as a colourless powder. The crude product was used in the next step without any further purification. ^{1}H-NMR (400 MHz, $CDCl_3$) δ = 8.16 (s, 1 H), 7.48 (s, 1 H), 7.10 (d, J = 0.9 Hz, 1 H), 2.85 (t, J = 7.4 Hz, 2 H), 1.80 (p, J = 7.4 Hz, 2 H), 1.26 (s, 24 H), 0.92–0.83 (m, 3 H).

CDI adduct of technical grade linoleic acid (3 h)

5 g technical grade linoleic acid (**1 h**, 17.8 mmol, 62 % linoleic acid, 30 % oleic acid and 8 % other fatty acids according to CoA of the supplier) were reacted according to the general procedure. As the product did not solidify upon pouring the reaction mixture into water, the organic phase was separated, and the aqueous phase extracted with $CHCl_3$. The combined organic phases were then dried over $MgSO_4$, and the solvent evaporated. After additional drying under high vacuum the product slowly solidified at room temperature. We obtained 5.5 g (16.8 mmol, 94 %) of a slightly yellow solid. Analytics showed that the product contained 66 % linoleic acid, 31 % oleic acid and 3 % saturated fatty acids. The crude product was used in the next step without any further purification. ^{1}H-NMR (400 MHz, $CDCl_3$) δ = 8.15 (s, 1 H), 7.47 (s, 1 H), 7.09 (d, J = 0.9 Hz, 1 H), 5.44–5.25 (m, 3.25 H), 2.85 (t, J = 7.4 Hz, 2 H), 2.77 (t, J = 6.3 Hz, 1.26 H), 2.10–1.96 (m, 4.25 H), 1.80 (p, J = 7.4 Hz, 2 H), 1.46–1.20 (m, 17.13 H), 0.93–0.82 (m, 3 H).

General Procedure for the Synthesis of Triacylglycerides

Glycerol or glycerol-d_8 (**6**, 1.0 equiv.) and the desired activated fatty acids (**3**, total of 3.0 equiv.) were dissolved in anhydrous THF inside an argon filled glovebox. The flask was sealed with a Teflon stopper and removed from the glovebox. Subsequently, the reaction mixture was frozen in liquid nitrogen and the argon was removed under high vacuum. The flask was resealed under vacuum and placed into an oil-bath at 70 °C. The reaction was stirred for multiple days, and continuously monitored via SEC using an RI detector. If necessary additional activated fatty acids were added (0.1 equiv. at a time) to drive the reaction to completion. Typically, the reactions required 3.2–3.5 equiv. of fatty acids and a reaction time of 5–12 days. Once the SEC showed full conversion the THF was evaporated. The residue was dissolved in iso-hexane, and washed with aqueous HCl (1 M, 2 times), a saturated aqueous solution of Na_2CO_3 (2 times) and brine (1 time). Then, the organic phase was dried over $MgSO_4$ and the solvent was evaporated. The final product (**7**) was dried under high vacuum and analysed via NMR.

MCT-oil (7 a)

The MCT oil **7 a** was synthesized according to the general procedure using a total of 1.31 g (14.3 mmol, 1.0 equiv.) glycerol-h_8 (**6 a**), 5.33 g (27.4 mmol, 1.92 equiv.) 1-(1*H*-imidazol-1-yl)octan-1-one (**3 a**) and 4.07 g (18.3 mmol, 1.28 equiv.) 1-(1*H*-Imidazol-1-yl)decan-1-one (**3 b**). The reaction was completed after 5 days at 70 °C and yielded 7.14 g (14.2 mmol, 99 %) of the title compound as a colourless oil. ^{1}H-NMR (600 MHz, $CDCl_3$) δ = 5.26 (tt, J = 6.0, 4.3 Hz, 1 H), 4.29 (dd, J = 11.9, 4.3 Hz, 2 H), 4.14 (dd, J = 11.9, 6.0 Hz, 2 H), 2.35–2.26 (m, 6 H), 1.64–1.56 (m, 6 H), 1.28 (m, 29 H), 0.90–0.84 (m, 9 H).

Deuterated MCT-oil (7 b)

The deuterated MCT oil **7 b** was synthesized according to the general procedure using a total of 3.75 g (37.4 mmol, 1.0 equiv.) glycerol-d_8 (**6 b**), 15.0 g (71.8 mmol, 1.92 equiv.) 1-(1*H*-imidazol-1-yl)octan-1-one-$d_{13.9}$ (**3 c**) and 11.5 g (47.9 mmol, 1.28 equiv.) 1-(1*H*-imidazol-1-yl)decan-1-one-$d_{18.0}$ (**3 d**). The reaction was completed after 9 days at 70 °C and yielded 20.3 g (36.4 mmol, 97 %) of the title compound as a colourless oil. ^{1}H-NMR analysis using 1,2,4,5-tetrabromobenzene as an internal standard showed a total deuteration degree of 93 %. ^{2}H-NMR (61 MHz, $CHCl_3$) δ = 5.24 (br s, 1 H), 4.49-3.90 (m, 2 H), 4.12 (br s, 2 H), 2.27 (br s, 4.26 H), 1.55 (br s, 6 H), 1.21 (br s, 29 H), 0.82 (br s, 9 H).

Synthetic plant oil mimic (7 c)

The synthetic plant oil mimic **7 c** was synthesized according to the general procedure using a total of 378 mg (4.10 mmol, 1.0 equiv.) glycerol-h_8 (**6 a**), 227 mg (0.74 mmol, 0.18 equiv.) 1-(1*H*-Imidazol-1-yl)decan-1-one (**3 g**) and 4.37 g (13.2 mmol, 3.22 equiv.) of the CDI adduct of technical grade linoleic acid (**3 h**). The reaction was completed after 12 days at 70 °C and yielded 3.26 g (3.7 mmol, 91 %) of the title compound as a slightly yellow oil. NMR analytics showed, that the oil contained 60 % linoleic acid, 33 % oleic acid and 7 % saturated fatty acids (mostly palmitic acid). ^{1}H-NMR (400 MHz, $CDCl_3$) δ = 5.44–5.28 (m, 9.20 H), 5.28–5.23 (m, 1 H), 4.29 (dd, J = 11.9, 4.3 Hz, 2 H), 4.14 (dd, J = 11.9, 6.0 Hz, 2 H), 2.77 (t, J = 6.4 Hz, 3.61 H), 2.36–2.27 (m, 6 H), 2.09–1.96 (m, 11.19 H), 1.68–1.52 (m, 6 H), 1.41–1.21 (m, 51.88 H), 0.93–0.81 (m, 9 H).

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Conflict of Interests

The authors declare no conflict of interest.

Data Availability Statement

The data that support the findings of this study are available in the supplementary material of this article.

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