



Compact NMR spectroscopy for real-time monitoring of a biodiesel production



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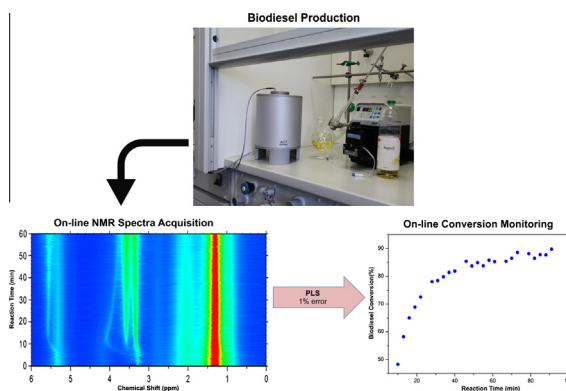
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HIGHLIGHTS

- On-line monitoring of a transesterification reaction by compact ¹H NMR spectroscopy.
- Developed PLS models from NMR-spectra recorded at 1 T to monitor biodiesel conversion.
- Real-time observation of methanol partitioning between glycerol and oleic phases.
- Resonance frequency of hydroxylic MeOH protons used to predict the conversion ratio.

GRAPHICAL ABSTRACT



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ABSTRACT

The use of biodiesel shows innumerable advantages compared to fossil fuels, since biodiesel is a biodegradable and non-toxic fuel. Nowadays, most of the biodiesel commercialized in the world is produced by the transesterification reaction of vegetable oils with methanol and basic catalysis. Understanding the reaction kinetics and controlling its optimum progress for improving the quality of the final product and to reduce production costs is of paramount importance. The present work explores compact ¹H NMR spectroscopy to follow the course of the transesterification reaction in real time. For this purpose the magnet is integrated into a flow setup which allows one to transport the neat solution from the reactor into the measurement zone and back again into the reactor. A multivariate calibration model applying Partial Least Squares regression was built to analyze the measured data and to obtain information about the biodiesel conversion ratio with errors on the order of 1%. This information is used in combination with a Lorentzian deconvolution of the spectra to estimate the relative concentrations of methanol present in the ester-rich phase in comparison with the one in the glycerol phase, the second medium involved in the reaction mixture. Finally, we demonstrate that the conversion ratio can also be monitored by measuring the chemical shift of the hydroxylic protons of methanol and glycerol present in the ester-rich phase. These results demonstrate that a compact NMR spectrometer can provide spectra with good quality and time resolution suitable for real time quality control of biodiesel production.

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1. Introduction

During the last decade the biofuel market has grown remarkably strong. The *International Energy Agency* (IEA) estimates that

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the biofuel share on a global scale in road transport will be approximately 7% in 2030, while in 2004 it was only 1% [1]. In this scenario, the biodiesel products from vegetable oils and bioethanol are the two major fuels employed as additives or substitutes to the fossil fuels originating from petroleum. The use of biodiesel shows numerous advantages compared to fossil fuels, since biodiesel is a biodegradable and non-toxic fuel, coming from a renewable energy source, and shows pollutant emission profiles lower than those of mineral diesel besides the fact that it can be directly applied to diesel engines due to its similar physical properties [2].

Nowadays, most of the biodiesel commercialized in the world is produced by the transesterification reaction of vegetable oils with methanol and basic catalysis (see Scheme 1). The transesterification reaction of a triglyceride with an alcohol to produce alkyl esters of fatty acids (biodiesel) consists of a sequence of 3 consecutive and reversible reactions, where di- and monoglycerides are produced as intermediates and glycerol as byproduct. Stoichiometrically, 1 mol of a triglyceride reacts with 3 moles of an alcohol to form 3 moles of alkyl esters and 1 mol of glycerol [3]. Nevertheless, by the fact that it is a reversible reaction, different conditions prevailing during the reaction, such as reactant concentrations or temperature can affect the final yield. Incomplete reactions result in higher amounts of mono- and diglycerides in the final product and hamper the separation process of the esters and the glycerol, which is rich in methanol and degrades the final quality of the produced fuel [4]. To control the quality of the produced fuel and to minimize the process cost, the transesterification reaction of triglycerides needs to be monitored.

Several chemical and physical analytical techniques have been applied to monitor the transesterification reaction of triglycerides in a chemistry laboratory such as IR, NIR, Raman Spectroscopy, ^1H and ^{13}C NMR, GC, HPLC and viscosimetry. However, only spectroscopic techniques such as IR and NIR have been extensively applied to tackle this goal directly in biodiesel production processes by on-line monitoring, as they are faster, cheaper and more suitable for automation on process control [5–7].

Although, high-resolution proton nuclear magnetic resonance spectroscopy (^1H NMR) was one of the first spectroscopic techniques applied to monitor the transesterification reaction of triglycerides [8], it was difficult to apply it directly in an industrial process control environment. The first and major obstacle is the high cost for acquiring the spectrometer. Furthermore, the use of superconducting magnets to generate the required magnetic field entails high maintenance costs (liquid nitrogen and helium), which curbs its wider use. This limitation, however, can be overcome with compact permanent-magnet based spectrometers instead of superconducting ones. Permanent magnets and electromagnets were used in the early days of NMR but superconducting magnets, which provide higher field strength and consequently larger chemical shift spread and sensitivity, eventually replaced them. However, recent advances in permanent magnet technology and magnetic field shimming strategies have led to small magnets suitable for compact and mobile NMR devices capable of measuring high-resolution ^1H NMR spectra of liquids at low field [9–18]

allowing the implementation of such devices into industrial environments for process monitoring [19,20].

In this context, the present work explores the application of a compact high-resolution NMR spectrometer, built with permanent magnets, for the on-line monitoring of the transesterification reaction of vegetable oils, allowing the analysis of the samples in its reacting medium without any separation process and further dilution in deuterated solvents. In order to accurately quantify the conversion of biodiesel during the reaction, a multivariate calibration model applying partial least squares (PLS) regression was developed and implemented with errors of 1% in the ultimate estimation of the conversion. Lorentzian deconvolution of the spectra was also applied to estimate the relative concentrations of methanol present in the ester-rich phase, in comparison with the ones residing in the glycerol phase, the second medium involved in the reaction mixture.

2. Experimental part

2.1. Reagents and materials

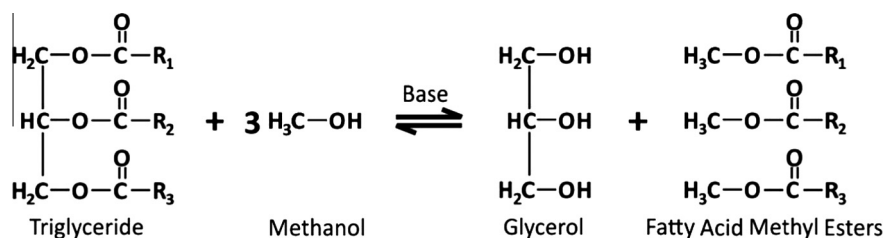
The transesterification reactions were executed with rapeseed oil (*K-Classic*, bought in a neighborhood market in Aachen, Germany), methanol and sodium hydroxide (*Roth*, analysis grade). Furthermore, acetic acid (*Roth*, analysis grade), magnesium sulfate (*Roth*, technical grade) and deuterated chloroform (*Chemotrade*, 99.8% with 1% TMS) were used in this work.

2.2. Instruments

An early prototype of a desktop magnet provided by Magritek GmbH, was employed for measurements of ^1H NMR spectra. This device is able to generate a magnetic field strength of 1 T (42 MHz Larmor frequency for ^1H) with homogeneity better than 0.04 ppm for the sample region covered by the RF coil surrounding a 5 mm glass tube as described in [18]. The magnet was controlled by a KEA² spectrometer, which was connected to a microcomputer. All of the spectra were acquired without dilution of the sample or addition of reference compounds. The high-field ^1H NMR spectra were measured on a *Bruker-400* spectrometer at a proton frequency of 400 MHz.

2.3. Transesterification reactions

All transesterification reactions were carried out in a three-necked reaction flask (reactor) of 500 mL, including a helix mechanical stirrer and a digital thermometer. Initially, 1.18 g of NaOH were dissolved in methanol and then added to 250 mL of rapeseed oil. This mixture amounts a molar ratio of 6:1 of methanol and oil and 0.5% (m/m) of NaOH related to oil. All reactions were carried out at ambient temperature of 27 ± 2 °C, keeping only the ambient laboratory temperature controlled.



Scheme 1. Scheme of a general base-catalyzed transesterification reaction of a triglyceride with methanol.

2.4. Flow setup

For the acquisition of the ^1H NMR spectra at 1 T, the reaction mixture was pumped through the magnet bore with a peristaltic pump (Ismatec, model ISM404B). The measurements were conducted with a glass flow cell 40 mm long with an outer diameter of 5 mm. Taking into account that the RF coil is approximately 5 mm long, by adjusting its position along the axis of the flow cell an optimum pre-polarization volume was set to allow the mixture to reach the Boltzmann equilibrium before entering into the RF coil region. PTFE tubes with an inner diameter of about 1 mm were used to deliver the reaction mixture from the reaction flask to the NMR cell aided by the peristaltic pump. Figure 1 shows the experimental set-up for monitoring the transesterification reaction using the compact NMR spectrometer. The spectrometer was operated in two different modes: (1) In continuous mode the reaction mixture was continuously pumped through the magnet bore at 0.9 mL/min and the ^1H NMR spectra were acquired every 10 s from the beginning until the end of the reaction. Under this condition the residence time in the sample loop was about 2 min; (2) In intermittent mode the reaction mixture was pumped at 2.0 mL/min into the measurement cell in 3 min intervals. The cell was filled with the reaction mixture in 1 min and 15 seconds and subsequently the pump was stopped. After flow stabilization during a time of 10 s a spectrum was acquired. Noteworthy is the fact that during this time, in which the solution reaches the static state, no liquid–liquid phase separation was observed. This was tested in the laboratory by optical inspection before the actual experiment. For this purpose the glass cell was placed outside the magnet bore and otherwise the same experimental protocol was followed.

Reference ^1H NMR spectra were measured with a conventional high-field spectrometer. For this purpose 3 mL of the reaction mixture were sampled from the reactor with a Pasteur pipette at appropriate intervals and straightaway transferred to a centrifuge tube of 15 mL containing 150 μL of acetic acid. The tube was quickly covered and manually shaken to stop the transesterification reaction. Afterwards, the samples were washed with brine and centrifuged at 5000 rpm. An aliquot of 50 μL of the supernatant was diluted in 1 mL of CDCl_3 present in an NMR tube for acquisition of the high-field ^1H NMR spectrum [21].

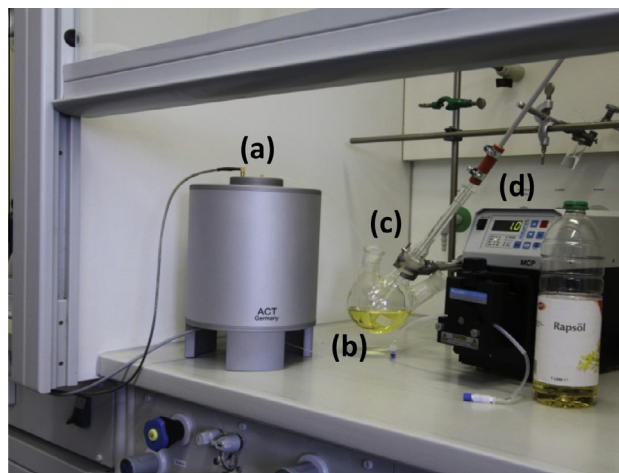


Fig. 1. Experimental set-up for monitoring the transesterification reaction. (a) Permanent magnet. (b) Reactor composed of a three-necked flask containing a mechanical stirrer. (c) Digital thermometer and Teflon tubes to transport the reaction mixture. (d) Peristaltic pump.

2.5. Data analysis

The conversion ratios (expressed in mol percentage ratios) of the triglycerides were calculated from the spectra acquired at low and high field. For the spectra measured at 1 T the conversion ratios were determined by two different methods: (1) by the ratio of the areas of the ester proton peak and olefinic proton peak from oil and ester using spectral deconvolution with Lorentz functions (see detailed description in Section 3.2.1) and (2) by the developed PLS calibration model (see detailed description in Section 2.5.1).

The data generated by both methods were compared with those obtained from the high-field spectra (400 MHz). In this case the standard procedure [8] is to compute the ratio between the areas under the ^1H NMR signals from the methoxylic protons peak (A_1) of the fatty acid esters and the α -carbonyl methylenic proton peak (A_2) from the esters and glycerides. In order to take the different number of ^1H nuclei into account contributing to each NMR signal, the factor 2/3 was included. Then the equation for the conversion ratio (C) expressed in % becomes [8]:

$$C = \left(\frac{2A_1}{3A_2} \right) \times 100. \quad (1)$$

Random experimental errors in preparing the samples are expected to be of the order of 1–2%. Then, we take this value as the error associated with the measurements of the conversion ratio measured by high-field NMR.

2.5.1. Multivariate calibration

A multivariate calibration model applying PLS regression was built to analyze the ^1H NMR data acquired with the compact device during monitoring of the transesterification reaction. PLS is currently the most widely used method applied for multivariate calibration of spectral data and its theory is well described in the literature [22–24]. The application of low-field NMR spectroscopy combined with chemometric tools, especially PLS, is emerging as a powerful tool for biofuel and edible oil applications [15–17,25], where its use is growing and competing with the more traditional techniques like MIR and NIR.

For the PLS calibration step, two transesterification reactions were studied by the low- and high-field techniques for 91 min reaction time. For both techniques, 10 spectra of each reaction were acquired at reaction times of 10, 16, 22, 31, 40, 52, 61, 70, 82 and 91 min. For the measurements at 1 T the spectra were acquired in the intermittent mode previously described. The multivariate calibration model was built using the Unscrambler software (Camo, model X 10.2). The triglyceride conversion ratios derived from the high-field ^1H NMR spectra were used for reference (vector “Y”) to build the multivariate model, and the low-field ^1H NMR spectra were applied as the data matrix “X”.

The PLS calibration model was validated with data from a third transesterification reaction, used as an external group to the calibration model previously developed. In the same way as described above, ^1H NMR spectra were acquired at low and high field. The reference values of the conversion ratios were calculated from the high-field ^1H NMR spectra whilst the matrix data values (matrix “X”) were derived from the low-field ^1H NMR spectra.

3. Results and discussion

3.1. On-line monitoring the transesterification reaction by compact NMR spectroscopy

Figure 2a shows 1 T ^1H NMR spectra of the individual compounds that coexist during the transesterification reaction [18]. It is noted that the ^1H NMR spectra for rapeseed oil (I) and the

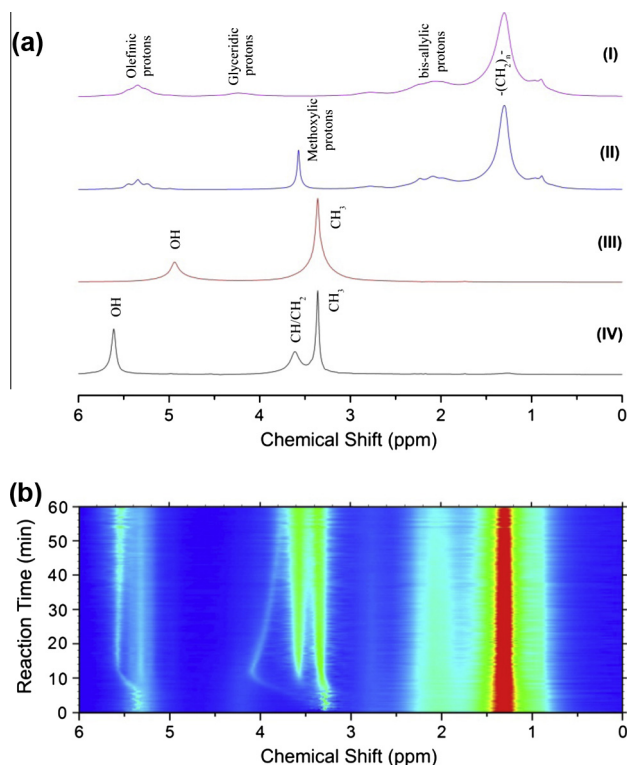


Fig. 2. (a) 1 T ^1H NMR spectra of I rapeseed oil, II rapeseed biodiesel, III pure methanol and IV the lower phase decanted after the transesterification process. (b) Stack of low-field ^1H NMR spectra acquired every 10 s for the rapeseed oil transesterification reaction (continuous mode).

rapeseed methyl esters (biodiesel, II), produced in the transesterification reaction of rapeseed oil, are very similar, with small differences in the chemical shift only for the methoxylic protons present in the methyl esters, which show a characteristic peak at 3.57 ppm and for the glyceridic protons present in the triglycerides, which show a peak at 4.24 ppm. This difference in the chemical shifts suggests using the methoxylic protons to monitor the triglyceride conversion to methyl esters during the transesterification process. Nevertheless, the carbinolic protons of glycerol (CH/CH₂ in spectrum IV), produced during the oil conversion into biodiesel, show a broad peak at 3.61 ppm, which overlaps with the methoxylic proton peak of the esters, disabling the straight forward integration of the signal from the protons for monitoring the transesterification process. This situation also applies to the high-field ^1H NMR spectra when measuring the spectrum of the reaction mixture without cleaning the sample.

During the course of the transesterification reaction the interplay among the elements described generates a continuous change of the solution medium and this is reflected in the NMR spectrum. Figure 2b depicts ^1H NMR spectra stacked along the reaction time axis covering the initial 60 min of the process. The reaction starts when methanol is added to the oil, present in the flask. At the beginning of the reaction the peaks in the spectra belong to signals corresponding to the triglycerides of the rapeseed oil (1.30 ppm, 2.00 ppm, 4.24 ppm and 5.32 ppm), CH₃ (3.30 ppm, methyl protons) and OH (3.40 ppm, hydroxylic protons) of the methyl alcohol. During the first minutes of the reaction, some distortions in the ^1H NMR spectra are noted mainly close to 3.30 ppm due to the difficulty of solubilizing methanol in the oleic phase (named ester-rich phase). After this period (5–7 min), a peak corresponding to the methyl esters (biodiesel) and the formation of glycerol appears at 3.60 ppm. This peak corresponds to the methoxylic protons of the fatty acid methyl esters and the carbinolic protons of glycerol.

Simultaneously, a peak starts to appear at 5.3 ppm and shifts up to 5.58 ppm (at 13 min). This peak corresponds to the hydroxylic protons of glycerol and methanol dissolved in the glycerol-rich phase. Another remarkable feature of the spectra is the behavior of the hydroxylic protons of methanol and glycerol present in the ester-rich phase during the course of the reaction. At the beginning of the reaction this peak is observed at 3.40 ppm and then it suffers an accentuated shift to 4.10 ppm at around 13 min of the reaction time returning finally to lower field values (3.70 ppm at the end of the reaction).

Figure 3 shows four spectra extracted from the stack plot (Fig. 2b) for four different reaction times, where an increase of the intensities of the peaks related to the fatty acid methyl esters and glycerol (4), methanol and glycerol present in the glycerol-rich phase (7) are noted. Moreover, the inset in Fig. 3 highlights the chemical shift of the hydroxylic protons of methanol and carbinolic protons of glycerol present in the ester-rich phase as a function of the reaction time.

The assignment of the peaks previously observed was validated by analyzing test samples with NMR spectroscopy at field strength of 1 T and 9 T. Test samples involving different concentrations of oil, methanol, biodiesel and glycerol, as well as different mixtures of methanol (containing also NaOH) and glycerol in different proportions of biodiesel were prepared and analyzed. The concentrations applied in the mixtures respected the observed values for each reactant and product found during the transesterification reaction.

3.2. Triglyceride conversion into fatty acid methyl esters

As can be seen in Fig. 2a IV, the lower phase decanted after the transesterification process shows negligible traces of triglycerides, making it possible to obtain the conversion of triglycerides due to the chemical reaction from the analysis of the ester-rich phase. To monitor the conversion ratios of the triglycerides into fatty acid methyl esters (FAMES) during the transesterification reaction, the peak corresponding to the methoxylic protons was used. However, the carbinolic protons of glycerol (CH/CH₂ in Fig. 2a, spectrum IV), produced during the oil conversion into biodiesel, show a broad peak at 3.61 ppm, which overlaps with the methoxylic proton peak

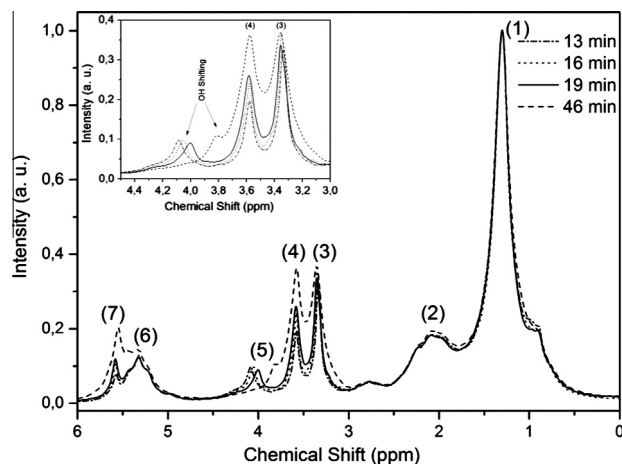


Fig. 3. Four spectra extracted from Fig. 2 at different reaction times. Peaks (1), (2) and (6) correspond to the protons common to rapeseed oil and rapeseed biodiesel, (3) methyl protons of methanol, (4) methoxylic protons in the ester and carbinolic protons of glycerol, (5) hydroxylic protons of methanol and glycerol present in the ester-rich phase and (7) hydroxylic protons of methanol and glycerol present in the glycerol-rich phase. Inset: Expanded region showing the changes of the chemical shifts of the hydroxylic protons of methanol and glycerol present in the ester-rich phase during the reaction.

of the FAMES, disabling the straight forward integration of this signal for the transesterification process monitoring.

3.2.1. Determination of the conversion ratios by deconvolution of the spectra

In a first attempt to overcome this problem a spectral deconvolution method based on Lorentz functions was developed and implemented in *Matlab*. The spectral region of interest is from 3 to 6 ppm (Fig. 4a), where most of the chemical changes during the reaction are evidenced as varying intensities and frequency shift of the different NMR signals. Since many of the relevant peaks (3, 4, 5, 6 and 7 in Fig. 4a) in this region interfere among them the method looks for their characterization by obtaining values of the individual areas, linewidths and resonance frequencies.

In order to fit the NMR signals from 3 to 6 ppm, the solution flowing through the pipe was assumed to be composed mainly of oil, biodiesel, glycerol and two methanol components, one dissolved in the ester-rich phase (methanol_{EP}) and the other in the glycerol-rich phase (methanol_{GP}). The NMR signal $f(x)$ from each of these constituents, for example methanol_{EP}, was modeled as a superposition of Lorentz functions $f_{\text{MethEP}}(x) = L_1(x) + L_2(x)$, where L_1 and L_2 identify the hydroxyl and methyl groups, respectively, of methanol_{EP}. Each Lorentz function, $L_i(x) = \frac{2 \times a_i}{\pi} \times \frac{w_i}{(4(x-x_{ic})^2 + w_i^2)}$, is characterized by three parameters: area a , width w , and frequency center x_c . In general, the areas of the Lorentz functions corresponding to different functional groups of a molecule are related to the amounts of protons under each peak, in the previous example $a_2 = 3a_1$ (1:3) and for the rest of the components the relations are shown in Fig. 4b. Except for the signal corresponding to the protons of the OH groups, the central frequencies x_c of each line rarely vary and in some cases coincide (vertical lines in Fig. 4b). Moreover, no significant changes in the values for the linewidths are observed between spectra. The signal from the olefinic protons at 5.32 ppm consists of a complex pattern given by spin-spin

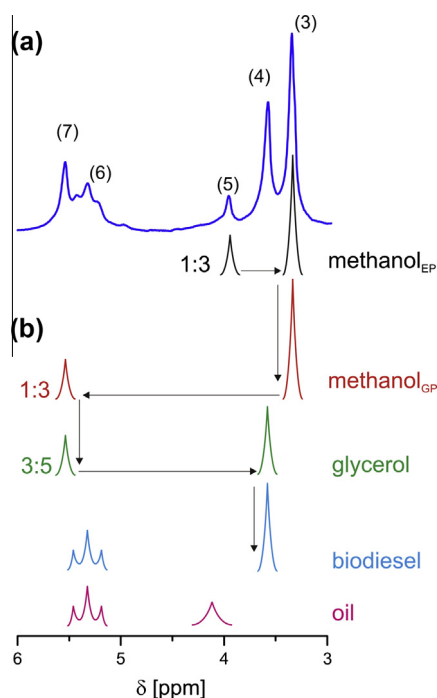


Fig. 4. (a) ^1H NMR spectral region where most of the changes are observed during the transesterification reaction. The peak labels agree with those in Fig. 3. (b) Schematic representation of the different fit functions used to deconvolve the NMR spectrum. Each schematic peak corresponds to a Lorentz function from which individual areas, widths and central frequencies are obtained (see text).

interactions relating to the double bonds corresponding to different kinds of unsaturated fatty acids present in the oil and ester [18]. For the present purpose, a good compromise was obtained by fitting it with three Lorentzians keeping the area and width relation between them constant. Under these conditions the whole fit process is quite robust (R^2 varied between 0.99 and 0.95) and fast (between 5 and 9 s).

Once the fit has been obtained, the areas corresponding to the methoxylic proton peak of the FAMES and the olefinic protons from oil and ester are used to determine the conversion ratio by using a relation similar to Eq. (1). Since the number of protons under the olefinic signal is undefined (because of the FAME profile of the oil/ester mixture) a calibration curve was created to determine the normalization factor in a similar way as the factor (2/3) is used in Eq. (1). For that, the 1 T ^1H NMR spectra of 5 different synthetic samples containing different ratios of rapeseed biodiesel and rapeseed oil, representing 18%, 38%, 60%, 82% and 100% of conversion, were acquired. The value obtained was 0.88, which was used to compute the conversion ratio during the whole transesterification reaction (Fig. 5). The average error obtained from the deconvolution procedure was of the order of 10%.

3.2.2. Determination of the triglycerides conversion ratios by multivariate calibration (PLS)

In order to improve the accuracy to determine the triglycerides conversion ratio by compact NMR spectroscopy at 1 T, a procedure based on a multivariate method using partial least squares (PLS) regression was developed and evaluated. For that, three rapeseed oil transesterification reactions were monitored by low- and high-field ^1H NMR, where the first two reactions were applied to build the calibration model and the last one to validate the model. The conversion ratios of the triglycerides, determined by high-field ^1H NMR for reaction times between 10 and 91 min were used as reference values to build the calibration model for low-field ^1H NMR.

Evaluation of several types of data processing and variable selection options for the low-field ^1H NMR spectra showed that the model with lowest errors for prediction was the one that used the spectral region between 1 and 6 ppm without apodization for the FID, followed by Fourier transformation, zero-order phase correction, and baseline correction. The values of the determination coefficients were $R^2 = 0.999$ and $R^2 = 0.979$, and the root mean

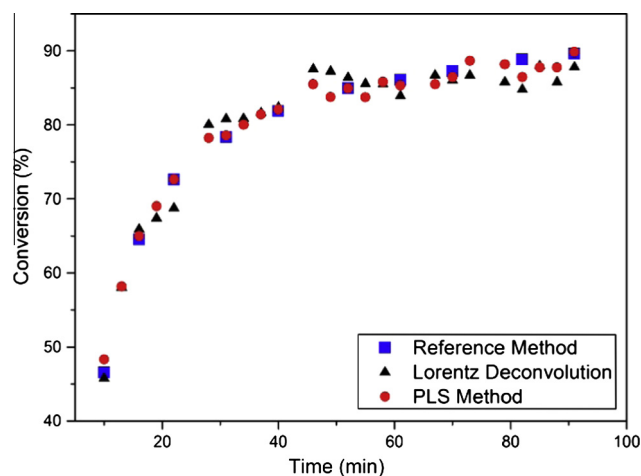


Fig. 5. Conversion ratios for a transesterification reaction monitored as function of time by high-field NMR (reference method, blue squares) and low-field NMR (intermittent mode): Lorentz deconvolution (black triangles) and PLS method (red circles). (For a color version of this figure the reader is referred to the web version of this article.)

square error of calibration (RMSEC) and cross validation (RMSECV) were 0.34% and 1.8% of conversion.

For the validation step of the developed model, the data acquired for the third transesterification reaction was used. In this case, the model showed a root mean square error of prediction (RMSEP) of 1.0% of conversion for 10 validation samples. In this way, the developed model was used to determine the conversion ratios for all the spectra acquired during monitoring of the transesterification reaction.

Figure 5 shows the conversion ratios determined from the low-field NMR measurements by Lorentz deconvolution and by the PLS model, compared with the reference method (high-field spectra) for the same transesterification reaction monitored in the intermittent mode. It can be noted from Fig. 5 that the Lorentz deconvolution method is less precise than the PLS method, which produces a kinetic curve in better agreement with the one determined by the reference method.

3.3. Shifting of the hydroxylic protons

The fact that no appreciable change in temperature neither in pH of the reaction solution was measured during the whole reaction time, suggests that the shifting of the hydroxylic protons of methanol and glycerol in the ester-rich phase observed in Fig. 2b likely originates from the continuous change in composition of the reaction medium. Initially, the reaction medium consists basically of an emulsion formed by rapeseed oil, methanol and sodium hydroxide. As the reaction proceeds, esters and glycerol are formed and oil and methanol are consumed, which changes the intersolubility of the reaction medium caused by a continuous polarity change of both phases (ester-rich and glycerol-rich phases). In particular, the concentration of alcohols in both phases is influenced by this effect as depicted in Fig. 6. This figure shows the relative concentrations of hydroxylic protons of methanol and glycerol present in the ester-rich phase in proportion to the total amount of hydroxylic protons (present in both phases). To find these values the areas of peaks 5 and 7 (Fig. 4), acquired during the continuous monitoring of the reaction, were deconvolved in each spectrum as explained in the previous subsection. Subsequently, the relative concentrations were determined using Eq. (2).

$$OH_{rel} = \left(\frac{OH_{EP}}{OH_{EP} + OH_{GP}} \right) \times 100\%, \quad (2)$$

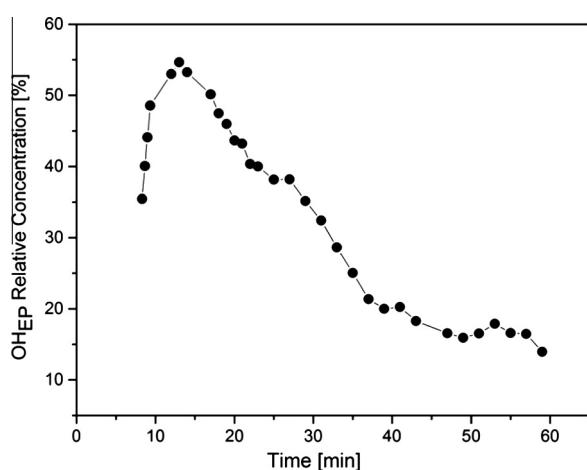


Fig. 6. Relative concentration of hydroxylic protons of methanol and glycerol present in the ester-rich phase (calculated from spectra acquired in the intermittent mode).

where OH_{rel} stands for the relative amount of hydroxylic protons present in the ester-rich phase, OH_{EP} represents the area of the peak of hydroxylic protons of methanol and glycerol present in the ester-rich phase (peak 5 in Fig. 4), and OH_{GP} is the area of the peak of the hydroxylic protons of methanol and glycerol dissolved in the glycerol-rich phase (peak 7 in Fig. 4).

During the first minutes of the reaction the concentration of the hydroxylic protons in the oleic phase grows rapidly, which can be readily seen up to approximately 13 min of the reaction time (Fig. 6). This might be related to the continuous formation of esters in this phase which generates a more polar medium, compared to oil at the beginning of the reaction, where the alcohol can be easily dissolved. During this period a shift in the resonance frequency of the hydroxylic proton peak of the methanol and glycerol to high field values is also observed in the spectra (4.10 ppm, Fig. 2b). By combining the information of both figures it is possible to obtain the chemical shift change for the hydroxylic protons during the transesterification reaction as a function of the methanol and glycerol concentration. This dependence suggests the formation of hydrogen bonds between the hydroxylic protons of methanol and glycerol and the different species present in the reaction medium. The proportional shifting to higher frequencies due to stronger bonding caused by the increase in concentration is in agreement with other studies [26].

As the reaction proceeds the glycerol concentration increases and starts to separate from the ester-rich phase. Thus, the methanol concentration begins to be partitioned between the ester-rich phase and the glycerol-rich phase. Figure 6 shows that after 13 minutes of reaction the concentration of hydroxylic protons in the ester-rich phase decreases continuously. It is important to consider that some methanol is being consumed by the transesterification reaction, which can explain this behavior. Nevertheless, the glycerol formation also occurs due to the transesterification reaction contributing to the NMR signal and that is why a sharp decrease in the concentration in this period is not expected.

3.3.1. Determination of the conversion ratio from the OH chemical shift

The continuous shift in the resonance frequency of the hydroxylic protons of methanol and glycerol in the ester-rich phase can be used to indirectly monitor the progress of the transesterification reaction in terms of the triglyceride conversion into methyl esters. For that a curve was built plotting the values of the chemical shifts of the hydroxylic proton obtained from a transesterification reaction monitored in the intermittent mode with low-field 1H NMR spectroscopy versus the conversion ratio of this reaction determined with the reference method (high-field NMR). In order to assure a single-valued function the portion of the spectra after 13 min (maximum frequency shift) of the reaction was taken. Subsequently, these data were fitted with a second-order polynomial to correlate the chemical shift values of the hydroxylic protons of methanol and glycerol in the ester-rich phase with the conversion ratio.

Figure 7 compares the results determined by polynomial regression and by the PLS model for the same set of data, acquired by monitoring a transesterification reaction by low-field NMR spectroscopy. Excellent correlation was found between the curve derived with the second order polynomial and that determined by the developed PLS model.

The results obtained with the chemometric model and with the shifting of the frequency resonance of the hydroxyl group show a lower error in the conversion ratio determination than those obtained by Lorentz deconvolution of the spectra. A possible justification for this issue might be attributed to the different spectral overlap of the signals (e.g. glycerol and biodiesel, methanol in different phases) which complicate the deconvolution of the spectral

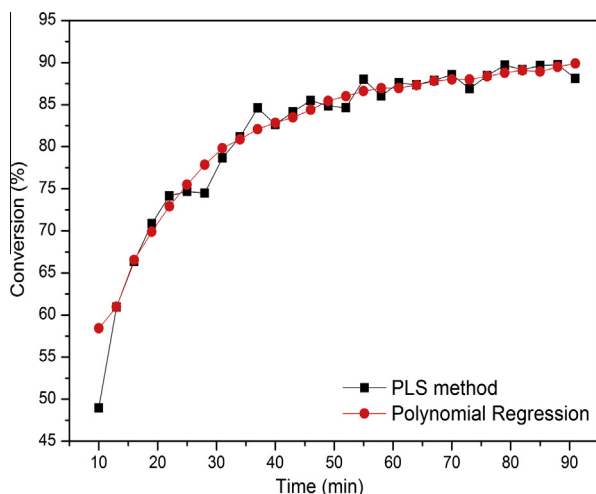


Fig. 7. Comparison of the two developed methods to determine the conversion ratio of the triglycerides by online monitoring. Black curve – PLS method. Red curve – second-order polynomial fit of the chemical shift values of the hydroxylic protons of methanol and glycerol in the ester-rich phase (both for the intermittent mode). (For a color version of this figure the reader is referred to the web version of this article.)

region with the methoxylic proton peak of biodiesel and the glyceridic proton peaks of oil and limit the accuracy of determining the oil conversion ratio. To overcome this problem, the oil conversion ratio was determined in an indirect way using the integrals of the olefinic protons of the fatty acid chains as internal reference (through the procedure described in Section 3.2.1), which is expected to accumulate higher errors. In spite of this, the three approaches implemented at 1 T show good agreement with the previously proposed method [8] to determine the conversion ratios of the triglycerides to produce biodiesel during the transesterification reaction.

4. Conclusions

This work reports the use of a compact ^1H NMR spectroscopy for on-line monitoring of a transesterification reaction to produce biodiesel. With the help of a glass flow cell directly coupled to the spectrometer it was possible to acquire spectra from the reaction medium every 10 s without any pre-treatment of the sample. The acquisition of different spectra in short time intervals enabled the observation of chemical changes in the reaction with good time resolution. This proved to be important in particular at the beginning of the reaction, where the peak related to the hydroxylic protons of methanol and glycerol and the methanol partitioning between the two phases that compose the reaction shifts significantly.

Three different methods were applied to determine the vegetable oil conversion into biodiesel in real time from the measurements performed at 1 T: Lorentz deconvolution of the spectra; multivariate calibration applying Partial Least Squares (PLS); and the correlation between conversion ratio and chemical shift of the hydroxylic protons of the alcohols.

The Lorentz deconvolution showed errors on the order of 10% for the determination of oil conversion into biodiesel, whereas the PLS method made it possible to obtain errors on the order of 1% of conversion. It must be recalled that PLS is a secondary methodology, which depends on a primary method to build a calibration model. It is also important to consider that variations of feedstock or even of the reaction variables could strongly affect the results. Nevertheless, in controlled environments, as those found in pro-

duction plants, the use of chemometric tools is completely feasible and already reality [27]. On the other hand, the deconvolution method allows one to determine the relative concentrations of methanol and glycerol in the different phases that compose the reaction medium during the transesterification reaction of the vegetable oil.

In a way similar to the PLS method, the method based on the chemical shift of the hydroxylic protons proposed in this work is very simple and suitable for automation of industrial processes. Nevertheless, this method also requires a calibration step by another analytical technique and will be more suitable for a controlled environment, similar to the PLS case.

Besides demonstrating the use of compact NMR spectroscopy for online monitoring a transesterification reaction conducted in a batch reactor in the lab (non-steady process), the results could be equally applicable to monitor or control continuous flow processes such as the ones performed in stirred tank or flux reactors within an industrial environment, where the outcome of the measurements can be analyzed in real time and used as for feedback into the production process to control the reactor conditions for improving the process efficiency. The information acquired in real time can be used to better comprehend the reaction mechanism of biodiesel production, catalyst application, and to optimize the separation techniques by which biodiesel, methanol and glycerol are recovered in a biodiesel plant [28] in order to reduce process costs. These results evidence the versatility of compact NMR, which can be a powerful tool in the biodiesel industry.

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