Determination of Carbon Chain Lengths of Fatty Acid Mixtures by Time Domain NMR

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Abstract Average carbon chain length is a key parameter that defines the quality of liquid biofuels. In this paper, a method for the determination of carbon chain lengths of fatty acid mixtures is presented. The approach is based on proton relaxation rates measured by time domain nuclear magnetic resonance. The spin–spin relaxation rates R_2 were used for the estimation of the carbon chain lengths. The method was examined for the set of samples with different mean lengths of the main linear carbon chain. Samples were prepared using four different fatty acids and mixtures of two, three or four of these fatty acids. The correlation coefficient between the known and measured values was equal to 0.994. Based on the relaxation theory, a linear-like dependence between the relaxation rate and carbon chain length was briefly shown, which endorses the experimental results. The developed methodology for determining carbon chain lengths offers robustness and rapidity, which are significant advantages when it comes to online use of the method in real industrial environments.

1 Introduction

In recent years, the increasing use of liquid biofuels and the tightening of environmental legislation have created a need for faster and more exact evaluation of fuel properties. Determination of averaged carbon chain length (CL) is important for liquid fuels, because it is one of the key parameters defining their quality and properties. CL also strongly affects other quality parameters, such as viscosity, higher heating value and cetane numbers of liquid fuels [1, 2]. The parameters of

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liquid fuels are determined based on international standards, which are time consuming, complex and difficult when applicable to online measurements.

High-resolution nuclear magnetic resonance (NMR) is one of the most informative analytical methods in chemistry. ^1H and ^{13}C NMR spectroscopy are widely used in oil and fat analysis [3]. For example, these methods have been applied for the determination of biological origin of vegetable oils [4] and to the quantitation of fatty acids profile of vegetable oils [5, 6]. The techniques based on ^1H NMR spectroscopy have been intensively developed with the aim of biodiesel composition characterization [7], monitoring the biodiesel production process [8] and evaluation of biodiesel concentration in biodiesel/diesel blends [9–11]. Furthermore, ^1H NMR spectroscopy has been presented as an alternative to gas chromatography (GC) and near-infrared (NIR) methods for routine analysis of biodiesel and gasoline [11].

However, in spite of high informativeness, accuracy and vast area of applications, high-resolution NMR spectroscopy is, in general, not applicable in industrial environments [12], because of the large size and big weight of the magnet system, the necessity to use a cryogen, the hazard of stray magnetic fields of superconducting magnets [12] and their high cost. On the contrary, small, low field or time domain NMR systems (LF-NMR or TD-NMR) equipped with permanent magnets are available for online instruments in production environments [13]. TD-NMR has become highly attractive for industrial applications due to mobility, relatively low price, easy operating, short time of analysis and a simple and non-destructive sample preparation procedure [12–14]. In particular, the robustness has made TD-NMR a standard analytical tool in quality control. The most known applications of TD-NMR, confirmed by international standards, are solid fat content determination in food and water and oil content in oilseeds [15, 16]. The international standard for hydrogen content determination in aviation fuels [17] has also been developed.

Liquid fuel analysis by TD-NMR is continuously growing. For example, the application of 1D T_2 relaxation times distribution and 2D cross-correlation T_1 – T_2 analysis to the characterization of potential biodiesel resources are highlighted in the paper [18]. The possibility of *ex situ* monitoring of the transesterification reaction by unilateral NMR is shown in the work by Cabeça et al. [19]. In most works related to TD-NMR applications to liquid fuels, the main findings are based on the correlation between T_2 relaxation time and viscosity [20, 21] and also other different fuel parameters—total acid number, refractive index, API gravity [20], cetane number and iodine value [21]. Experimentally obtained correlations of T_2 on viscosity and other physical parameters for different kinds of liquid fuel samples, oils, alkanes [20–23] were described empirically in most of the investigations. An attempt to give an interpretation of T_2 dependence on viscosity for alkanes was given in [22] using the NMR relaxation theory, considered for the shape of spherical molecules.

In this work, we have applied TD-NMR to study molecular structures of fatty acids. The correlation between relaxation rates and average carbon chain lengths of fatty acid mixtures showed the possibility of TD-NMR as a method for rapid measurement of the average CL of liquid fuels.

2 Materials and Methods

2.1 Sample Preparation

Samples with different average carbon chain lengths were prepared as different mixtures of several common fatty acids (Table 1). The fatty acids for this study were selected by their melting points, which should be lower than 25 °C to perform the measurements at room temperature. The commercially available fatty acids were used without further purification. Their properties (molecular weights, melting points and carbon chain lengths) are given in Table 1.

Average carbon chain length CL_{mix} values of the samples can be calculated knowing the molecular weights and mass parts of fatty acids using the following equations:

$$CL_{\text{mix}} = \sum_{i=1}^n a_i CL_{-i}, \quad (1)$$

where a_i is the part of molecules of i -acid in mixture, n is the number of acids in mixture and CL_{-i} is the carbon chain length of i -acid. The a_i is obtained from the equation:

$$a_i = \frac{N_i}{\sum_{i=1}^n N_i}, \quad (2)$$

where the number of molecules of i -acid, N_i , is calculated from Eq. (3):

$$N_i = \frac{m_i N_A}{M_{wi}}, \quad (3)$$

where m_i is the mass part of i -acid in the mixture (in g), M_{wi} is the molecular weight of i -acid (in g/mol) and N_A is the Avogadro constant.

Twenty samples were selected for the current study. The samples had the CL_{mix} values within the range from 4 to 18. These values are found in Table 2.

2.2 TD-NMR Measurements

The TD-NMR measurements were done using the portable NMR analyser Spin Track [24] with ^1H resonance frequency of 25.7 MHz. The diameter of the sensor hole was 10 mm. The digital receiver console provided quadrature acquisition with

Table 1 Fatty acids used for samples preparation

Common (IUPAC) name	Chemical formula	Molecular weight, g/mol	Melting point, °C	Carbon chain length CL	Producer
Butyric (butanoic) acid	$\text{C}_4\text{H}_8\text{O}_2$	88.11	−5.1	4	SIGMA-Aldrich
Caproic (hexanoic) acid	$\text{C}_6\text{H}_{12}\text{O}_2$	116.16	−3.4	6	SIGMA-Aldrich
Caprylic (octanoic) acid	$\text{C}_8\text{H}_{16}\text{O}_2$	144.21	16.7	8	SIGMA-Aldrich
Oleic (9 z -octadec-9-enoic) acid	$\text{C}_{18}\text{H}_{34}\text{O}_2$	282.46	13.0	18	MERCK

Table 2 Experimental values of carbon chain lengths CL_{mix} and spin–spin relaxation rates R_2 of fatty acid mixtures

Sample	Mass fraction				CL _{mix}	T ₂ , s	R ₂ , s ⁻¹
	CL						
	4	6	8	18			
1	0.25	0.25	0.25	0.25	7.11	0.53	1.90
2	0.3	0.3	0.4		5.86	0.66	1.52
3		0.35	0.5	0.15	7.90	0.48	2.08
4		0.2	0.5	0.3	9.15	0.41	2.43
5		0.5	0.4	0.1	7.32	0.48	2.07
6		0.15	0.15	0.7	12.62	0.25	3.98
7	1				4.00	0.93	1.07
8		1			6.00	0.66	1.50
9			1		8.00	0.47	2.13
10				1	18.00	0.17	5.79
11	0.7		0.3		4.83	0.75	1.33
12	0.5		0.5		5.52	0.67	1.49
13	0.3		0.7		6.35	0.58	1.72
14			0.7	0.3	9.80	0.35	2.88
15			0.5	0.5	11.38	0.29	3.49
16			0.3	0.7	13.44	0.25	4.06
17		0.2		0.8	13.46	0.26	3.89
18		0.5		0.5	9.50	0.40	2.51
19		0.8		0.2	7.12	0.54	1.84
20			0.2	0.8	14.71	0.22	4.60
21			0.6	0.4	10.54	0.33	3.01
22			0.8	0.2	9.13	0.39	2.56

the ringing time of around 10–12 μs . CPMG (Carr–Parcell–Meiboom–Gill) pulse sequence [25] was applied for the measurement of spin–spin relaxation rate R_2 . Echo time was equal to 6 ms; the number of 180° pulses in the sequence was 600. Relaxation delay was 2 s, and the amount of scans was equal to 8. Durations of 90° and 180° RF pulses were 7 and 14 μs , respectively. All measurements were performed at room temperature (25 °C).

The experimental relaxation rate R_2 is dependent on the composition of different fatty acid molecules in a sample [26]:

$$R_2 = \sum_{i=1}^n (P_i R_{2i}), \quad (4)$$

where P_i and R_{2i} are the proton populations and spin–spin relaxation rates of i -th fatty acid molecule; n is the number of fatty acid types in mixture.

The individual R_{2i} values corresponding to different fatty acid molecules in a mixture were not observed due to fast proton exchange [26]. It takes place as the

magnetization exchange rate k_{ex} between fatty acid molecules is much higher than the measured relaxation rate. Thus, one R_2 distinct value was obtained for each sample. It was confirmed by applying Laplace transformation to measured CPMG decays, as can be seen in Fig. 1. Therefore, all measured CPMG decays $A(t)$ were fitted by mono-exponential function defined by Eq. (5)

$$A(t) = A_0 e^{-R_2 t}, \quad (5)$$

where A_0 is the maximal signal magnitude and R_2 the spin–spin relaxation rate. A Matlab software script, written by the authors, was used for determining R_2 values.

2.3 Theory

The Bloembergen–Purcell–Pound (BPP) approach [27] is normally used for the description of spin–spin T_2 NMR relaxation in liquids. According to BPP theory [25, 27], spin–spin relaxation time T_2 or relaxation rate R_2 ($R_2 = 1/T_2$) is dependent on molecular mobility expressed as correlation time τ_c , which is the characteristic parameter of molecular mobility. For liquid samples $\omega_0 \tau_c \ll 1$ (ω_0 = the resonance frequency of TD-NMR devices) and R_2 linearly increases with the growth of correlation time as follows:

$$R_2 = 10M_2\tau_c, \quad (6)$$

where M_2 is the value of second moment, which is determined by the strength of dipole–dipole interactions between neighbouring nuclei.

The correlation time in BPP equations can be described by Stokes–Einstein–Debye equation [28, 29]:

$$\tau_c = \frac{C_r \eta V}{k_B T}, \quad (7)$$

where V is the molecule's effective volume [30], η is viscosity, T is temperature, k_B is Boltzmann constant and C_r is fitting parameter determined by experiment

Fig. 1 Laplace transformation applied to two measured CPMG decays

[28–31]. The Stokes–Einstein–Debye equation is often used in the form modified for homogeneous fluids with molecules described as spheres with hydrodynamic or Stokes radius, not molecular.

Rotational movements are the most perceptible to NMR relaxation contribution, whereas the frequency range of translational motions makes a little contribution to the NMR relaxation proton spectra, and vibrations due to their high frequency are not observed by NMR relaxation.

Correlation times for different molecular motions are the time parameters in specific correlation functions. Complex modelling is required to describe them separately. On the other hand, rotational correlation times of molecules can be calculated by known corresponding diffusion coefficients, which are described by the Stokes–Einstein equation [29]:

$$D_r = \frac{k_B T}{8\pi\eta R^3}, \quad (8)$$

where D_r is rotational diffusion coefficient, k_B is Boltzmann constant, T is temperature, η is viscosity and R is hydrodynamic (Stokes) radius of molecule.

When describing the rotational movement of molecules, it is convenient to consider that molecules have a spherical shape with hydrodynamic radius R . In the consideration that the molecule's effective volume is equal to

$$V = \frac{4\pi R^3}{3}, \quad (9)$$

the combination of the Eqs. (7–9) gives the following result [28]:

$$\tau_c \sim \frac{1}{D_r}. \quad (10)$$

Thus, it is obviously seen that the rotational correlation time is dependent on the corresponding diffusion coefficient (Eq. 10). Equation (10) is the widely used ratio for calculation of the NMR relaxation correlation time [22], as rotational molecular movements are the major contributors to NMR relaxation.

Generally molecules of different shapes can be characterised by hydrodynamic radii, thus Eq. (10) is probably applicable both to unbranched, rod-like molecules, which are mostly represented in liquid biofuels and to different types of paraffins and aromatic hydrocarbons, which are presented in petroleum fuels.

To connect the rotational diffusion coefficient with the molecular weight, the well-known ratio for self-diffusion D_{self} behaviour description for untangled polymer chains [32, 33] can be used. In case of elongated, unbranched molecules, there is direct dependence between these two parameters:

$$D_{\text{self}} \sim M_w^{-\alpha} \text{ for } M_w < M_{wc}, \quad (11)$$

where M_{wc} is the entanglement coupling molecular weight [32], α is a coefficient, equal to 1.

Combining the expressions (6), (10) and (11), it is clearly seen the linear relationship between τ_c and, therefore, of R_2 with molecular weight M_w :

$$R_2 \sim M_w \quad (12)$$

It is easy to suppose that in the case of unbranched hydrocarbons with similar chemical organisation the molecular weight is proportional to molecular size, and the latter one can be evaluated by CL. Relying on the dependence of R_2 on molecular weight M_w (Eq. 12), it is assumed the linear-like dependence of R_2 on CL:

$$R_2 \sim \text{CL}. \quad (13)$$

3 Results and Discussion

The CPMG decays for all fatty acid samples were measured and then fitted by mono-exponential function for determining spin–spin relaxation rates R_2 . The results of R_2 values are collected in Table 2. The average carbon chain lengths CL_{mix} are plotted as a function of the relaxation rates R_2 in Fig. 1 showing a linear relationship. By applying the linear fitting to the data set, the model can be calculated, as follows:

$$\text{CL}_{\text{NMR}} = a R_2 + b, \quad (14)$$

where $a = 2.914 \text{ s} \pm 0.069 \text{ s}$ and $b = 1.507 \pm 0.198$.

The values defined by Eq. (14) are in a good agreement with the CL_{mix} values, as shown in Fig. 2. The correlation coefficient between these two data sets is equal to $R = 0.994$ and standard error to 0.384. The errors can be caused by errors in R_2 determination (fitting errors) and sample preparation procedure (errors of weighting of fatty acid mass portions and mixing).

In the present study, carbon chain lengths of fatty acid mixtures have been determined by the time domain NMR, showing a good agreement with the known ones. The fatty acids were chosen by their melting points, so that it was lower than the room temperature. However, considering the method for biofuel production quality control, it should be taken into account that the fatty acid profile in real vegetable oils includes typically saturated palmitic acid (C16:0), stearic acid (C18:0), unsaturated oleic (C18:1) and linoleic acid (C18:2), [18, 34]. The specific profiles can also contain saturated and unsaturated acids of higher carbon chain lengths (more than 20) [35] besides the main components. So the carbon chain lengths of biofuels are typically longer than CLs used in the present study. As can be seen from Eq. (13) and our experimental results presented here, the relaxation rates of fatty acid molecules depend linearly on carbon chain lengths. It is quite clear that this relationship is valid in the case of longer molecules and thus also for biofuels.

In the view of better quality of biodiesel, relatively long-chain saturated and exactly monounsaturated acids are the most important because this combination will provide higher oxidation stability; the presence of double bond will decrease the melting temperature, viscosity, cloud point and pour point; and the long CL will guarantee a high enough cetane number and heat of combustion [1]. The effect of the different amount of double bonds to the NMR relaxation was not investigated in the current work. However, as the presence of double bond increases the mobility

Fig. 2 Average carbon chain lengths CL_{mix} are plotted as a function of the relaxation rates R_2 . The solid line is fitted by a linear model

(correlation time) of protons, as a result, some effect on relaxation rate R_2 is probably expected to be observed.

In this paper, a method for the determination of carbon chain lengths of fatty acid mixtures is presented. The approach is based on proton relaxation rates measured by TD-NMR. In our earlier paper [36], we have demonstrated the possibilities of TD-NMR in online analytics. The method for determining carbon chain lengths offers robustness and rapidity, which are significant advantages when it comes to online use in real industrial environments.

4 Conclusion

The results presented here illustrate that NMR relaxation can be used for the determination of average carbon chain lengths, which is one of the key parameters, for example, defining the quality properties of biofuels. The results also demonstrate the possibility of TD-NMR applying to industrial process control of liquid biofuels, and therefore, monitor their product quality, which will be still more important for the tightening environmental legislation.

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