JOURNAL OF CHEMICAL EDUCATION

pubs.acs.org/jchemeduc

Laboratory Experiment

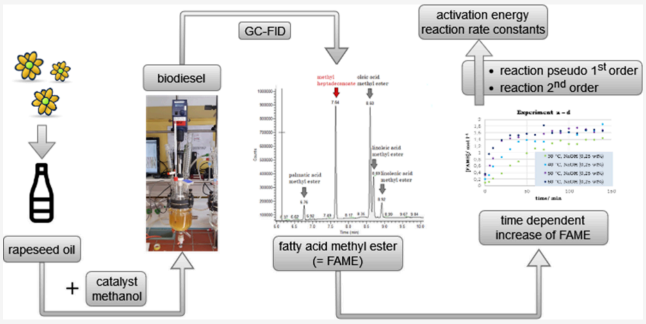
The Inexhaustible Teaching Potential of the Transesterification Reaction: The Kinetics of the Production of Biodiesel for Undergraduate Students

Sabine W. Steidl,* Horst Stangl, Ingrid Kohl, and Thomas Loerting*

Cite This: https://doi.org/10.1021/acs.jchemed.4c01335

Read Online

ACCESS | Metrics & More | Article Recommendations | Supporting Information



ABSTRACT: The replacement of conventional fuels with sustainable and renewable green fuels such as biodiesel to protect the environment is an important focus of today's research and a strong area of interest among students. Here, we develop a research-oriented experiment in the context of renewable fuels aimed at introducing fundamental concepts in physical chemistry, organic chemistry, and analytical chemistry to undergraduate chemistry students. Specifically, the students first carry out the organic chemistry task of base-catalyzed transesterification of rapeseed oil to biodiesel. Second, they execute the analytical chemistry task of observing the time dependent, temperature-controlled increase of fatty acid methyl ester concentration using gas chromatography based on flame ionization detection (GC-FID). Finally, the students carry out the physical chemistry task of extracting kinetic properties from the data, including the concepts of rate constants, reaction order, activation energies, catalysts, and mathematical modeling. These data reveal the transesterification is either a pseudo first or second order reaction, where the kinetics change a few minutes after the start of the reaction. The activation energies of 51 kJ mol^{-1} (pseudo first order reaction) and 52 kJ mol^{-1} (second order reaction) determined here are similar to literature values for similar oils and similar catalysts. Here, we have developed an inquiry-based, research-oriented experiment emphasizing deductive reasoning, which arises in the context of renewable fuels aimed at undergraduate students.

KEYWORDS: *First-Year Undergraduate/General, Physical Chemistry, Organic Chemistry, Analytical Chemistry, Inquiry-Based, Kinetics, Gas Chromatography, Green Chemistry*

■ INTRODUCTION

The idea to use vegetable oils as fuels for mobility purposes was born several years prior to the energy crisis in the 1970s and 1980s.[1] In his book "Die Entstehung des Dieselmotors" ("The Development of the Diesel Engine"), Rudolf Diesel reported the successful operation of a diesel engine with plant oil at the Paris Exposition in 1900.[1] During World War II, scientists put more effort into optimizing plant-based fuels to ensure a supply without being dependent on other countries.[2,3] The high viscosity of plant oil leads to engine deposits due to poorer atomization when injected into the combustion chamber.[2] Consequently, in 1937 a method to reduce the viscosity was established, which is still used in an optimized form.[4] This approach is not only of industrial interest, but also has a benefit to the experimental and theoretical capabilities of chemistry students.[4,5] Experiments dealing with base- and acid-catalyzed transesterification,[6−11] analytic methods, such as HPLC, ^{1}H NMR spectroscopy, ATR-FTIR, GC-MS and GC-TIC, to determine the chemical composition of biodiesel,[6,9,10,12−17] and specific topics like determining energy content of biodiesel blends[7,8] have already been successfully introduced in science education. However, besides organic and analytical chemistry, the biodiesel topic may cover one more field of chemistry, which is exploited in the present work: we here focus on introducing fundamental kinetic concepts to students that can be grasped based on the observation of the time dependent, temperature-controlled increase of FAME (fatty acid methyl ester) concentration in the course of transesterification. The analytical tool to detect FAME in our work is flame ionization detection gas chromatography (GC-FID). The subsequent mathematical modeling of the reaction kinetics allows for an understanding of reaction order, reaction rate constants, and activation energies. Even more importantly, a reflected analysis of the data allows recognizing how reaction

Received: October 29, 2024
Revised: June 18, 2025
Accepted: June 20, 2025

order and reaction rate constants may change with reaction progress. Thus, our newly developed experiment is suitable for the introduction and research-oriented deepening of kinetic concepts in an inquiry-based setting for undergraduate chemistry students.[18] Moreover, our experiment includes up-to-date and practice-oriented research in students' science education.[19−22] Comparison of the results obtained in the student study with literature studies on different oils, using different types and different amounts of catalysts, allows a deeper insight into how catalysts impact on reaction rate constants and activation energies.[19−22] This is, in fact, a vast and active area of research encompassing thousands of original experimental and theoretical studies. A suitable entry point into this literature is provided through the references cited in our manuscript. The strength of the present work lies in its integration of multiple learning concepts—ranging from analytical to bioorganic and physical chemistry—within a teacher−student instructional setting.

Finally, the socioscientific issue of this topic and the relevance to everyday life should not go unmentioned. Although, biodiesel may replace fossil fuels the large-scale agricultural land use for raw material extraction harbors the risk of the reduction of biodiversity.[23,24] Furthermore, the application of edible raw materials can lead to commodity squeeze and result in an increase of food costs.[23,24] However, biodiesel is nontoxic, free of sulfur and nearly free of polycyclic aromatic hydrocarbons.[25] The reduction of particulate matter of 75−83% compared to conventional diesel has the potential to improve air quality, especially fine dust pollution in cities.[26] By means of lively discussion about the advantages and disadvantages entailed using biodiesel the students should gain an understanding of the real-world applicability of the production of biodiesel based on the aim to replace conventional fuels with sustainable and renewable green fuels.[27,28]

■ PEDAGOGICAL BACKGROUND

The experiment was carried out with students at The Higher Technical College (HTL) Kramsach, which is a vocational higher school that offers a comprehensive, five-year education in chemical engineering for students aged 14 to 19. In total, 22 students took part in the laboratory and classroom activities for the educational case described in this manuscript. No formal ethics review was required, because the biodiesel project study was part of their regular curricular activities. All 22 students were informed about the intent of publishing the biodiesel experiment in a journal covering teaching and didactics. They were also informed that the data measured by them collectively will be published (together with more data measured by the authors of the present manuscript in preparation for the student labcourse). All 22 students provided their consent by signing a declaration of consent.

At HTL Kramsach students receive a solid theoretical and practical education both in the school's own laboratories and through internships in industry. A practical approach to teaching technical knowledge and skills prepares graduates for direct employment in the chemical industry. The training focuses on analytical chemistry, microbiology and biochemistry, process engineering, and chemical technologies. In addition to technical qualifications, students also receive a broad general education. The final school leaving examination, which includes both a subject-specific and a general part, allows graduates to obtain a general university entrance qualification. This gives them both direct entry into employment and access to universities and colleges.

The experiment is designed for high school students in the penultimate year before graduation (12th grade, ages 17 and 18 in Austria) at HTL Kramsach. Due to the chemistry and chemical engineering focus of this school and the heavy load of laboratory work (9 h a week), HTL Kramsach students acquire chemical knowledge and hands-on skills that go well beyond the typical level of high school students internationally. Consequently, this experiment is well suited for first-year undergraduate students but needs some adaptations for high-school students. In our case, we conducted the experiment over the course of several weeks in the laboratory and in the classroom. This time frame gives them the opportunity to do the actual experiment, which requires about 10 h in the lab (two blocks of 5 h). In addition, there needs to be several single hours in the classroom devoted to providing the background of the experiment and the analysis of the data. To enable implementation of the experiment within more constrained timeframes, several adaptations are possible. For example, the instructor can complete the calibration in advance, assign chromatographic peaks to the components prior to the session, and reduce the sampling duration from 150 to 100 min. These adjustments can allow the experiment to be completed within a 3 h laboratory period. Even in the absence of a laboratory, the data can be shared with students for inspection, discussion, and further analysis. In this way, important concepts in kinetics, such as reaction order or activation energy, can be taught. For this reason, the experiment can be adapted to meet the needs of undergraduate students in a global educational context. The experiment unites concepts of analytical, organic, and physical chemistry and consequently offers a broad horizon in chemistry education including environmental awareness, organic reaction mechanisms, reaction kinetics, gas chromatography, and calibration techniques. Due to the sophisticated topic of the experiment, the students should have some prior knowledge on basic experimental techniques (making dilutions, pipetting, ...), gas chromatography, and internal standard calibration to be able to successfully follow the work instructions and to focus on the understanding of the reaction kinetics associated with transesterification. Thus, the aims of the experiment can be narrowed down to following:

- The students get to know basic kinetic concepts by means of an experiment.
- The students get an insight into alternative fuels.
- The students complete the deductive path of gaining knowledge and get an insight into science research.
- The students can work independently in the laboratory and handle chemicals and equipment safely.

The experiment is guided by a research design that employs conventional scientific methods, including self-directed experimental planning, measurements, data collection, data analysis and interpretation, and critical reflection on the congruence with other research findings. Consequently, by comparing the theory of the kinetics of transesterification given in the work instruction (see Supporting Information) with the help of self-obtained experimental data and provoked reflections on observed deviations regarding the theory, the students experience a deductive way of science research. Initially, the experimental setup is planned by the students through the selection of reaction parameters such as temperature and catalyst concentration (possible ranges are provided in the students' handout) to ensure sufficient time for sample collection. Furthermore, the students are instructed (see students' handout) to compare their results with those of other groups and with the results of other research, and to explain any discrepancies using literature searches. To promote independent work in the laboratory the experiment is conducted in a partly open experimental setting. According to Bell et al. the experiment could be classified as *Structured Inquiry* or *Guided Inquiry*.[18]

The significant potential of this experiment lies in its versatility in achieving a range of learning objectives. Depending on prior knowledge and availability of time for the experiment, the focus can be narrowed. For physical chemistry classes one may focus on investigating the impact of reaction parameters on rate constants, add reaction orders, and finally add activation energies. For analytical chemistry classes, the focus could be to determine the components of biodiesel through analytical methods.

■ THEORETICAL BACKGROUND

In the following, the main and side reaction, as well as the mathematical modeling of the time-dependent reaction course using pseudo first and second order kinetics, are elaborated. Furthermore, the Arrhenius equation to determine the activation energy of the main reaction is introduced.

Transesterification and Saponification

As mentioned in the introduction, the viscosity of vegetable oil must be reduced to avoid operational problems in a Diesel motor. The viscosity of vegetable oil (approximately 27−53.6 $mm^2\ s^{-1}$) is lowered by catalyzed transesterification with methanol to fatty acid methyl ester (FAME) to approximately 3−5 $mm^2\ s^{-1}$.[29] The main reaction, transesterification, taking place in this process is given in Figure 1.[30]

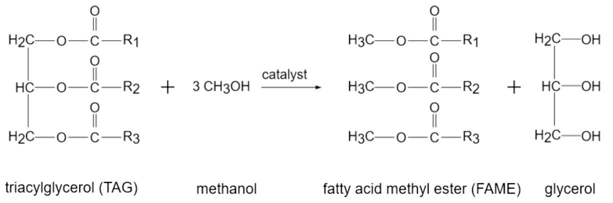


Figure 1. Main reaction, catalyzed transesterification of triacylglycerols with methanol (R_1, R_2, R_3 = long hydrocarbon chains).

Due to the presence of basic catalysts (such as sodium hydroxide or potassium hydroxide) and free fatty acids contained in the vegetable oil, the saponification (see Figure 2) can occur in addition to the transesterification.[30] The soap produced in this undesirable side reaction, leads to formation of emulsions, and thus makes further industrial processing more difficult.[30]

The reactions shown in Figures 1 and 2 suggest that vegetable oil consists mainly of triacylglycerols and free fatty acids.[31] The four main fatty acids in rapeseed oil esterified with glycerol are oleic, linoleic, linolenic, and palmitic acids. The remainder of the vegetable oil consists of mono- and diacylglycerols, water, and traces of other substances.[31]

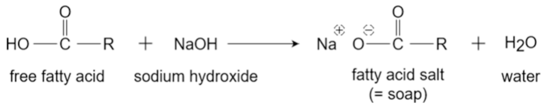


Figure 2. Side reaction, saponification (R = long hydrocarbon chains).

Kinetics of Transesterification−Pseudo First Order Reaction

eq 1 illustrates a simplified transesterification reaction. The three fatty acid residues (R_1, R_2, and R_3) of the triacylglycerol (TAG) molecule react with three methanol molecules and produce three FAME molecules and glycerol (GL) (k, k' = reaction rate constant):

$$\mathrm{TAG} + 3\mathrm{CH_3OH} \xrightarrow{k} 3\mathrm{FAME} + \mathrm{GL} \tag{1}$$

The reaction rate v of the transesterification is defined as the change of the concentration of FAME ([FAME]) over time (t):

$$v = \frac{\mathrm{d[FAME]}}{\mathrm{d}t} \tag{2}$$

It is assumed that the back reaction can be neglected due to the nonequilibrium situation. Consequently, the reaction rate depends on the concentration of the products or educts and on the rate constant k' (m, $n \in \mathbb{R}$) as

$$v = k' \cdot [\mathrm{TAG}]^n \cdot [\mathrm{Methanol}]^m \tag{3}$$

If methanol is added in excess, the concentration of methanol can be considered constant during the reaction. Moreover, the concentration of TAG can be substituted by the difference of the initial concentration of the fatty acids of the triacylclycerols ($[\mathrm{TAG_0}]$) and the concentration of FAME. This results in the following pseudo first order rate law ($k = \frac{1}{3}k'[\mathrm{Methanol}]$):

$$\frac{\mathrm{d[FAME]}}{\mathrm{d}t} = k([\mathrm{TAG_0}] - [\mathrm{FAME}]) \tag{4}$$

The solution of this differential equation can be obtained by separating the variables, integration, and defining the initial condition (initial concentration of FAME ($[\mathrm{FAME_0}]$) = 0):

$$\ln\left(1 - \frac{[\mathrm{FAME}]}{[\mathrm{TAG_0}]}\right) = k \cdot t \tag{5}$$

The integrated and linearized rate law allows determining the rate constant k, which defines the slope of the plot. Modeling the transesterification with a first order reaction is common in literature.[20,22,32,33]

Kinetics of Transesterification−2nd Order Reaction

Besides the first order reaction, the reaction kinetics of the transesterification can be modeled with a second order reaction:[33]

$$\frac{\mathrm{d[FAME]}}{\mathrm{d}t} = k' \cdot [\mathrm{TAG}] \cdot [\mathrm{Methanol}] \tag{6}$$

Substituting [TAG] with $[\mathrm{TAG_0}]$ − [FAME] and [Methanol] with $[\mathrm{Methanol_0}]$ − [FAME] ($[\mathrm{Methanol_0}]$ being the initial concentration of Methanol) yields the following rate law ($k = \frac{1}{3}k'$):

$$\frac{d[\mathrm{FAME}]}{dt} = k\cdot([\mathrm{TAG}_0] - [\mathrm{FAME}])\cdot([\mathrm{Methanol}_0] - [\mathrm{FAME}]) \qquad (7)$$

Considering the initial condition ($[\mathrm{FAME}_0] = 0$) the solution of the differential equation is as follows:

$$\frac{1}{[\mathrm{Methanol}_0] - [\mathrm{TAG}_0]} \ln\left(\frac{[\mathrm{FAME}] - [\mathrm{Methanol}_0]}{[\mathrm{FAME}] - [\mathrm{TAG}_0]}\right) - \frac{\ln\left(\frac{[\mathrm{Methanol}_0]}{[\mathrm{TAG}_0]}\right)}{[\mathrm{Methanol}_0] - [\mathrm{TAG}_0]} = k\cdot t \qquad (8)$$

If students have already been introduced into differential equations, the derivation of the integrated and linearized pseudo first order rate law is easily comprehensible for them. The derivation of the second order model demands more mathematical skills and effort. Consequently, the reaction pseudo first order is preferred in terms of didactics. Nevertheless, the modeling with a second order reaction can be a good supplement for further deepening of kinetics or to increase the level of complexity for undergraduate chemistry students.

Determination of the Activation Energy

The Arrhenius behavior, as illustrated in eq 9, of the temperature dependence of the reaction rate constants can be checked by using eq 5 or 8 to find the reaction rate constants at different temperatures. The reciprocal temperature (T^{-1}) plotted versus the natural logarithm of the reaction rate constants ($\ln k$) results in a linear regression function. The activation energy (E_A) can be determined from the slope ($-E_AR^{-1}$), and the y-axis intercept ($\ln A$) determines the pre-exponential factor A.

$$\ln k = \ln A - \frac{E_A}{R\cdot T} \qquad (9)$$

■ HAZARDS

The chemical hazard symbols, hazard, and precautionary statements are summarized for students in the working instruction (see Supporting Information). In the following paragraph, the required ingredients for the experiment and its respective hazards are shortly presented. The rapeseed oil is available in any convenient store and is not harmful. Methanol is acutely toxic, flammable and skin contact, and ingestion of this chemical should be avoided. NaOH causes burns to skin and damages eyes and, consequently, it should be handled with care and gloves. If sodium methanolate is used as alternative catalyst, students should work with gloves under a fume hood because this chemical is flammable, corrosive, and toxic. The solvent required for the determination of FAME by GC-FID is *n*-heptane. n-Heptane has effects on the environment and is flammable. It could have a negative impact on human's health, is toxic and should always be used in a well-ventilated area or hood, and away from any open flames. The internal standard and FAME are not classified as hazardous substances.

■ EXPERIMENTAL PROCEDURE

The experiment on the transesterification of triacylglycerols to produce biodiesel was executed at the Higher Institute of Technical Education HTL Kramsach (Austria) and was improved with the students as part of their industrial technology laboratory. The student's age ranges from 17 to 18 years. The experiment was done in total by about 25 students, who were split into smaller groups. All students are in their fourth year, which means they have three years of specialized chemistry education prior to this experiment. The experiment was performed in the framework of a mandatory industrial technology lab course. A group of eight to 14 students was assigned to work on the biodiesel experiment as part of their labor obligations. The work assignments of the biodiesel experiment are based on two main areas namely *biodiesel production* and *analytics*. In a joint effort, samples and data for several temperatures were collected. Thus, one group of students (four to seven students) dealt with the production of biodiesel and handed over samples to the analytics group (four to seven students) for evaluation. The latter group analyzed the samples by GC-FID, identified the individual components in the reaction mixture, and determined the total concentration of fatty acid methyl ester. Subsequently, the kinetic modeling and evaluation of these data were discussed together. In total, two groups of 8−14 students carried out these steps—so there had to be a rotation of groups in the lab. Ideal group sizes are 4−5 students per subgroup—this allows them to communicate with each other and the teacher. Two teachers would be ideal so that one can help students do the transesterification, and the other can help students do the GC analytics.

In the following section, the implementation of the experiment, possibilities of variation, and exemplary data measured in the students' laboratory are summarized.

Setup, Implementation, and Variation Possibilities of the Sampling/Experiment

For the production of biodiesel, a prefabricated apparatus (Figure 3) is used. A Huber Pilot ONE temperature control unit ensures a constant temperature in the double-walled reactor. The built-in thermometer displays the reaction mixture's temperature. The stirrer ensures homogeneous distribution of the reactants, and the reflux condenser guarantees condensation of the evaporated reactants/products.

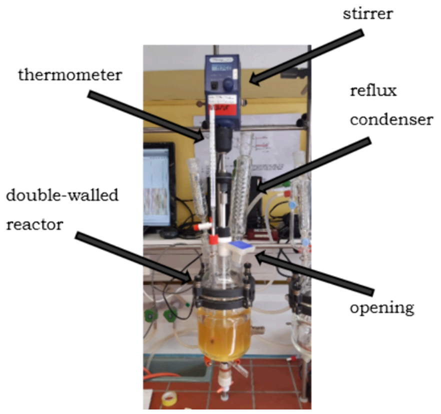


Figure 3. Prefabricated apparatus for biodiesel production.

Methanol (technically pure) and rapeseed oil are used as reactants in biodiesel production. In the biodiesel reaction, employing methanol as the short-chain alcohol results in a distinct advantage, as it prevents the formation of emulsions. Either NaOH cookies (solved in 10 mL of distilled water) or a

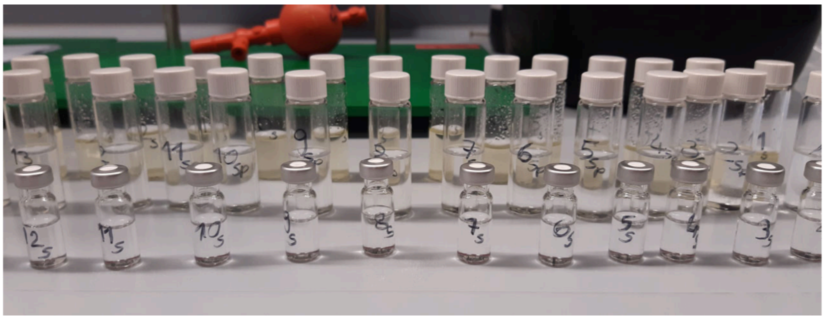


Figure 4. Sample preparation, two times centrifuged and purified sample (third row), diluted sample (second row), and sample prepared in vials for the analysis of [FAME] with GC-FID (first row).

sodium methanolate solution (25 wt % in methanol) is used as catalyst. Both catalysts lead to almost identical results since the active species is sodium methanolate in both cases.[34] To be able to follow the isothermal reaction with time, it is necessary to choose appropriate temperatures—they need to be low enough to avoid immediate reaction, but high enough to avoid reactions that are too slow for the teaching unit. The following reaction conditions were found to be most suitable to follow reaction progress in real time: 500 mL of rapeseed oil; 214 mL of technical pure methanol; 0.25 wt % catalyst (mass catalyst: mass reaction mixture); stirring at 400 rpm; temperature range 30–60 °C.

The starting point (reaction time $t = 0$ s) of the reaction is defined by the addition of the methanol and the catalyst (brought to constant temperature) to the thermostatted rapeseed oil. In order to avoid data being lost, it proved helpful for the students to prepare a spreadsheet beforehand, with prefilled sample labels and the times of sampling.

When the reaction starts, approximately 10 mL of the reaction mixture is taken at defined times and placed in centrifuge tubes, which are placed in a beaker filled with ice to slow down the reaction. The sampling has to be done in specific, predefined time intervals (especially at the beginning of the reaction), which can be stressful for students if they have not already prepared and labeled the centrifuge tubes or chosen very short time intervals. It has proven to be best to take samples after 30 s, 5 min, 10 min, and then every 10 min (for approximately two up to 3 h). The cooled samples were used without any further purification for GC-determination of FAME. In purified samples (as seen in the third row in Figure 4) we were unable to avoid unwanted reaction progress during the purification procedure.

Immediate dilution with a solution of the internal standard and *n*-heptane (see second row in Figure 4) ensures in addition to cooling on ice an immediate stop of the reaction so that the concentrations determined by GC-FID fully reflect the reaction progress in the isothermal process (see Supporting Information). In addition, the strong dilution of the sample prevents contamination of the column, so that a direct injection of the liquid sample without further purification could be carried out. Evaporating the sample prior to injection as done in the headspace technique was not necessary due to the satisfactory results. Our initial concerns about side components and overlapping peaks in chromatograms turned out be unjustified. Instead, this method has the advantage that the cooled and diluted reaction products can be injected into the GC-FID without any further processing and delay (from vials shown in the first row in Figure 4). Yet, chromatograms with baseline-separated peaks are also obtained with this method. However, heterogeneity of the composition of the liquid is still an issue. It is necessary to homogenize the liquid sample by shaking the sample using a test tube shaker at approximately 2200 rpm for approximately 1.5 min prior to taking an aliquot (see Supporting Information). This homogenization step is, in fact, much more important than adding 0.5 M H_2SO_4 to the samples for neutralizing the catalyst. We obtained essentially identical results with and without neutralization so that addition of 0.5 M H_2SO_4 can be considered unnecessary.

Sampling should be done at four different temperatures. To reduce the time required for the experiment, two groups of about 2–4 students carried out two reactions in parallel (at two different temperatures). Therefore, assuming one experiment includes preparation, sampling, dilution, and cleaning the laboratory, the sampling can be finished in two blocks of about four to 5 h.

Analytical Determination of the Amount of Methyl Ester in the Biodiesel

As mentioned above, the other group of students was responsible for the determination of the amount of FAME. A Thermo Scientific Focus GC-FID instrument with a TriPlus AS autosampler was used to determine and quantify the constituents of the biodiesel. The method used to quantitatively analyze FAME in the biodiesel samples is shown in the Supporting Information. The students had some prior knowledge of gas chromatography and calibration techniques from their school subject "analytical laboratory" in the previous year. Based on this knowledge they were able to do the identification, calibration, and evaluation of the amount of FAME in the samples with the help of a rough guide and hints from the teacher (these instructions are summarized in the Supporting Information). Figure 5 shows a chromatogram of a biodiesel sample with the peaks assigned to FAME and the internal standard. The use of an internal standard (methyl heptadecanoate) and the assignment of retention times are explained in more detail in the Supporting Information.

■ RESULTS

Rate constants at several temperatures are required to determine the activation energy of the transesterification. Consequently, the students measured FAME concentration as a function of reaction time at four different temperatures, namely 30, 40, 50, 60 °C. These results are summarized in Figure 6. The experiments are performed at different temperatures while maintaining the same reaction conditions otherwise (as summarized in Table 1). This strategy helps students to learn the concept of changing only one parameter

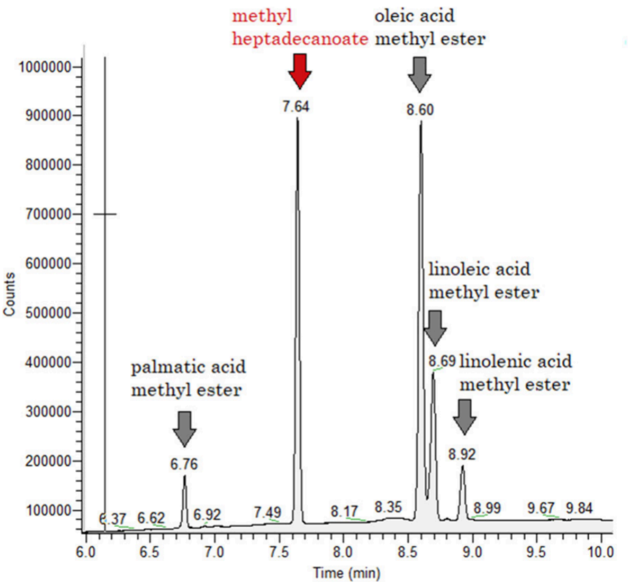


Figure 5. Chromatogram of the biodiesel sample (without any purification), palmitic acid methyl ester (retention time of 6.76 min), oleic acid methyl ester (8.6 min), linoleic acid methyl ester (8.69 min), linolenic acid methyl ester (8.92 min), and internal standard methyl heptadecanoate (7.64 min).

at a time in order to be able to grasp the impact of one specific parameter on the outcome, in this case, reaction rate constants *k*. Some minor deviations from this protocol were still done by the students, though: In contrast to experiments a and b, the samples in experiments c and d were shaken before dilution. In experiment c, two dilutions per sample were prepared and both measured in order to learn about the reproducibility of the results, where a range of approximately ±9% [FAME] was encountered. This range serves as an error bar for the concentration data. Especially at the beginning of the measurement (1st and second sampling), rather large deviations occur. That is, especially in the beginning of the reaction, large changes occur, where small differences (such as one additional second for reaction) have a large impact.

In general, the data in Figure 6 show the expected trends—the FAME concentration initially increases in a near-linear fashion. After about 40 min at 60 °C or about 90 min at 30 °C the FAME concentration levels off, where the plateau is close to a concentration of 2 mol l^{-1}. This curve shape is typical of isothermal reactions that proceed via nucleation and growth of the product phase, as, e.g., described by the Johnson−Mehl−Avrami−Kolmogorov (JMAK) equation. For the mathematical modeling of the kinetics of transesterification, the initial concentration of fatty acids of the triacylglycerols in the educt oil phase is required (see eq 5 or 8). Under the assumption that the reaction proceeds completely, i.e., all reactants react to form products), the final concentration of FAME corresponds to the initial concentration of fatty acids of the triacylglycerols.

Based on these considerations, the highest molar concentration of FAME in the previous measurements is sought. The experiment with the conditions 30 °C and NaOH (1 wt %) (see Figure 7) represents the experiment with the highest molar concentrations. The course of the molar concentration appears constant from minute 10. Consequently, $[\mathrm{TAG}_0]$ is calculated from the average of the concentrations from minute 10 to minute 90, giving a concentration of 2.2 mol l^{-1} for $[\mathrm{TAG}_0]$. This highest concentration of the fatty acids of the triacylglycerols in rapeseed oil is comparable to the literature value ($[\mathrm{TAG}_0] = 2.023$ mol l^{-1}), whereby the different values originate from different composition and production of the rapeseed oil.[35]

Moreover, Figure 7 illustrates the influence of the catalyst concentration on the reaction rate. The decrease of the catalyst concentration causes a reduction of the reaction rate and allows better observation of the start of the reaction with the aid of discontinuous sampling. Furthermore, Figure 7 shows the students that their determination of the rate constants and activation energies only applies to their reaction conditions and is only comparable to literature values with the same/similar reaction conditions.

The linearized plot of the data of experiment c according to eq 5 (reaction pseudo first order) is displayed in Figure 8. Clearly, the data do not follow the expected linear trend, except for the initial period of approximately 30 min. This indicates that the reaction mechanism changes as the reaction progresses. Typical reasons for such behavior are products inhibiting the reaction. This seems to be the case here as well—all data points beyond 30 min are below the extrapolated linear regression. The linear regression of the measurement data, shown as dotted line in Figure 8, at the beginning of the reaction is derived with the help of the software *Excel*. Similar trends are also seen for all other temperatures; see Figures S4−S11 in the Supporting Information.

Csernica and Hsu elaborated the reason for the deviation of the reaction order and strong reduction of the reaction rate later in the reaction.[36] They identified an increase of the

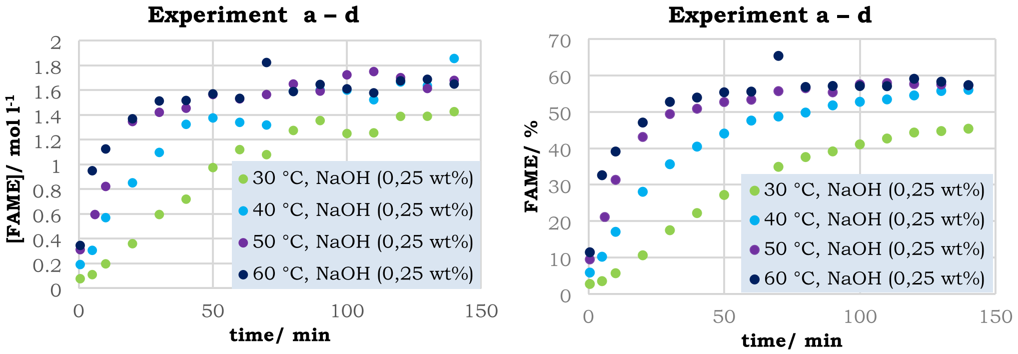


Figure 6. Measurement data: experiments a−d. 30 °C (a), 40 °C (b), 50 °C (c), 60 °C (d), NaOH (0.25 wt %), concentration (left) and yield (right).

Table 1. Reaction Conditions for Experiments a−d

Oil/mL	Methanol/mL	Catalyst/wt %[a]	Temperature/°C	Stirrer speed/rpm	Dilution factor	Sample preparation
500	214	0.25	30 (a), 40 (b), 50 (c), 60 (d)	400	43	Without purification

[a] Mass (NaOH) per mass (reaction mixture)

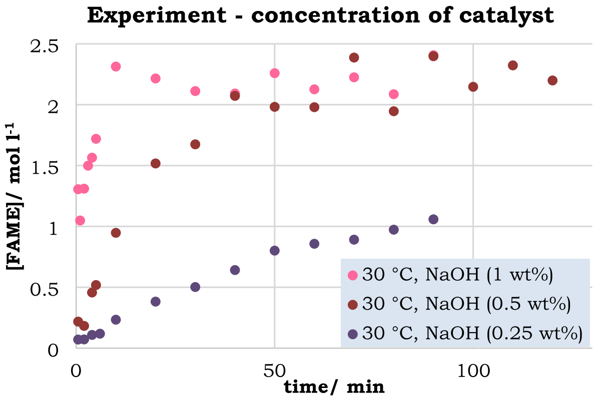


Figure 7. Measurement data: experiment concentration of catalyst. 30 °C, NaOH (1, 0.5, 0.25 wt %).

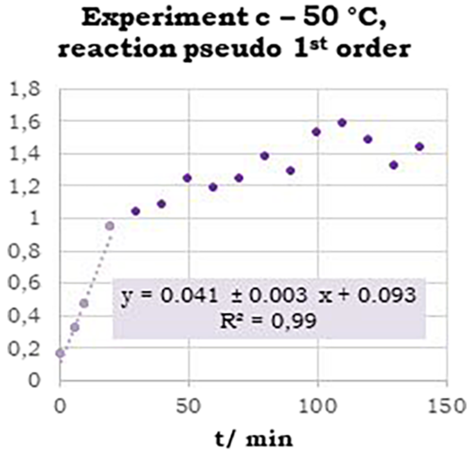


Figure 8. Experiment c. 50 °C; linear regression, reaction pseudo 1st order.

glycerol concentration and consequently reduction of mass transfer per time and solubility of triacylglycerols in the methanol phase (= glycerol inhibition). Moreover, according to Ezzati et al. the first order reaction model is suitable for the beginning of the reaction when a high initial concentration of methanol is used.[19] The aforementioned research papers could be made available to the students to provide them with insight into this research topic.

The linearized plot according to eq 8 (reaction 2nd order) of the same measurement data as shown in Figure 8 (experiment c) is displayed in Figure 9. Comparing Figure 8 and Figure 9, it cannot be determined which model better approximates the kinetics of the transesterification because both models fit the measurement data well. Therefore, as already mentioned in the chapter theoretical background, the choice falls purely for didactic reasons in the developed student experiment on the modeling of the transesterification reaction with a reaction pseudo first order.

To determine the activation energy of the transesterification, the reaction rate constants k (determined from the slope of the regression functions, as exemplarily shown in Figure 8) and associated temperatures are linearized and plotted according to eq 9. The activation energies of 50.6 kJ mol^{-1} (reaction pseudo first order) and 52.0 kJ mol^{-1} (reaction 2nd order) are determined from the slopes in Figure 10.

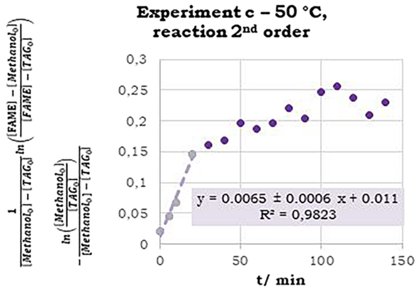


Figure 9. Experiment c. 50 °C; linear regression, reaction 2nd order.

Compared with earlier studies on the kinetics of biodiesel production, which also use basic catalysts and methanol, the activation energies are within the range of the reported literature values (25.51−57.82 kJ mol^{-1}).[20,22,32] The deviations of the activation energies in the literature are due to the different reaction conditions. The use of bentonite or higher amounts of catalysts contribute to the reduction of the activation energies.[20,22,32] We emphasize that the activation energy determined here applies to the rapeseed oil as the source material used in industry. Different values for the activation energies might be obtained for the transesterification reaction of pure FAMEs since no side reactions such as saponification affect the reaction kinetics (see Figure 2).

Assessment of Student Learning

Throughout the experiment, the students were working in individual groups and were accompanied by two teachers. Background information, next steps, and improvement of the lab procedures were constantly brought up and discussed both among students and between students and teachers. Each student was obliged to compile a laboratory report after the completion of the experiment. Such a report requires the students to introduce the topic in general and address the goals of the experiment. Students then either had to write an experimental section about the syntheses strategy and sample preparation procedures or a section on the analytical methods and procedures used. Finally, the teacher and students collaboratively analyzed the obtained data, conducted a kinetic analysis, and interpreted the results.

Whether or not "the students can work independently in the laboratory and handle chemicals and equipment safely" was constantly watched, evaluated, and, if necessary, corrected by the teachers. It can be stated that only students experienced in basic handling of laboratory equipment including instrumental analytics bring the required knowledge to successfully perform this study. The group of students who participated in the biodiesel study were well experienced in their fourth year of studies in a chemistry lab. They all had the necessary lab skills to carry out the study.

All "students got insights into alternative fuels" through an introduction by the teacher and were provided basic literature.

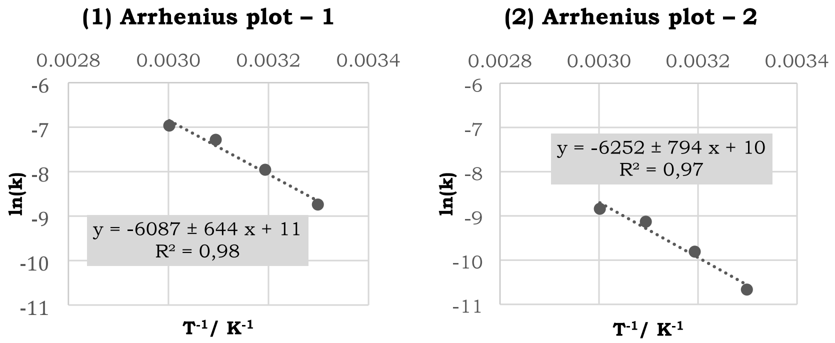


Figure 10. Arrhenius plot: reaction pseudo 1st order (1); reaction 2nd order (2).

All students were able to write a critical discussion for their laboratory report. All "students got to know basic kinetic concepts by means of an experiment". Every student was able to prepare and present a plot of the FAME concentration vs time for their laboratory report. Everybody delivered a qualitative discussion of the change of the product concentration over time depending on the conditions of the reaction. Without the help of the teacher, it was certainly difficult for the majority of the students to apply the kinetic models, learned in previous theory courses, on the data gained in order to extract the kinetic parameters, e.g., the rate constant or the activation energy.

"The students complete the deductive path of gaining knowledge and get an insight into science research." During the development of the experiment, certain obstacles had to be overcome, like finding out the TAG_0 concentration or whether a purification step of the samples prior to GC-analysis is changing the FAME concentration in the samples or not. The teams were prompted to reflect their observations and successfully changed the procedures, when necessary.

As assessed from the laboratory reports and the teacher observations in the laboratory itself, it can be stated that the students have benefitted a lot from being part of the study, especially by seeing how the experiment was improved "on the fly" as results were generated, which implied that the procedures needed to be changed (e.g., a chromatographic analysis had to be done without prior purification to not lose product in the purification step). Even more so, the students could directly see the connection between the basic kinetic concepts taught in the classroom and the kinetic data as produced in the lab. This has helped them to understand the concepts in chemical kinetics and how the concentrations, rates, etc. can be determined in practice. Nonetheless, the kinetic formalism and mathematical procedures are the largest obstacles in this experiment.

Furthermore, the results regarding the students' learning progress can be assessed in relation to the objectives outlined in the educational background section, thanks to the exchange-based guidance provided during the experiment. It was remarkable how well the students learned to handle equipment and chemicals safely over the course of the experiment. For example, using micropipettes and operating the GC-FID software posed challenges at the beginning of the experiment. However, after a few trials, the students were able to organize and conduct their work independently. The data analysis using Excel necessitated guidance from the teacher. Although the students possessed a foundational understanding of kinetics, it proved to be insufficient for conducting the analyses independently. Therefore, it is recommended that the experiment be conducted with undergraduate students who have a broader knowledge of kinetics and some experience in data analysis using Excel.

■ CONCLUSION

The production of biodiesel contains many precious didactic aspects that are important for almost all chemicals produced industrially. These aspects are of key relevance for the introduction and deepening of chemical concepts to high school and undergraduate students. The newly developed experiment here helps students to grasp concepts of importance in organic chemistry, analytical chemistry, and physical chemistry, foremost an understanding of reaction mechanisms, catalysis, and reaction kinetics. By means of optimized reaction conditions and discontinuous sampling during the basic catalyzed temperature-controlled trans-esterification of rapeseed oil with methanol to biodiesel, the students gain the opportunity to get insight into the time-dependence of isothermal reactions. Finding suitable kinetic models (pseudo first order and second order reaction) and an understanding of the deviation from the measured data, determining the rate constants and activation energy allows the students to learn the basic principles of kinetics in a deductive, inquiry-based research process.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available at https://pubs.acs.org/doi/10.1021/acs.jchemed.4c01335.

Student laboratory handout and measurement data (PDF) (DOCX)

■ AUTHOR INFORMATION

Corresponding Authors

Sabine W. Steidl – *Institute of Physical Chemistry, Innrain 52c, University of Innsbruck, A-6020 Innsbruck, Austria*; Email: sabine.steidl.97@gmail.com

Thomas Loerting – *Institute of Physical Chemistry, Innrain 52c, University of Innsbruck, A-6020 Innsbruck, Austria*; orcid.org/0000-0001-6694-3843; Email: thomas.loerting@uibk.ac.at

Authors

Horst Stangl – *HTL Kramsach, 6233 Kramsach, Austria*
Ingrid Kohl – *HTL Kramsach, 6233 Kramsach, Austria*

Complete contact information is available at: https://pubs.acs.org/10.1021/acs.jchemed.4c01335

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

The authors thank the students of the HTL Kramsach for conducting and improving this experiment. We gratefully acknowledge the University of Innsbruck for supporting open access publication.

■ REFERENCES

(1) Knothe, G.; Krahl, J.; van Gerpen, J. *The Biodiesel Handbook*; AOCS Press: Champaign, IL, 2005.
(2) Knothe, G.; Razon, L. F. Biodiesel fuels. *Prog. Energy Combust. Sci.* **2017**, *58*, 36−59.
(3) Songstad, D. D.; Lakshmanan, P.; Chen, J.; Gibbons, W.; Hughes, S.; Nelson, R. Historical perspective of biofuels: learning from the past to rediscover the future. *In Vitro Cell. Dev. Biol. Plant.* **2009**, *45* (3), 189−192.
(4) Knothe, G. Historical perspectives on vegetable oil-based diesel fuels. *Inform* **2001**, *12*, 1103−1107.
(5) Neiles, K. Y.; Bowers, G. M.; Chase, D. T.; VerMeulen, A.; Hovland, D. E.; Bresslour-Rashap, E.; Eller, L.; Koch, A. S. Teaching Collaborations and Scientific Practices through a Vertically Scaffolded Biodiesel Laboratory Experience. *J. Chem. Educ.* **2019**, *96* (9), 1988−1997.
(6) Blatti, J. L.; Burkart, M. D. Releasing Stored Solar Energy within Pond Scum: Biodiesel from Algal Lipids. *J. Chem. Educ.* **2012**, *89* (2), 239−242.
(7) La Rosa, P. D.; Azurin, K. A.; Page, M. F. Z. Soybean Oil: Powering a High School Investigation of Biodiesel. *J. Chem. Educ.* **2014**, *91* (10), 1689−1692.
(8) Knoerzer, T. A.; Hill, E. M.; Davis, T. A.; Iacono, S. T.; Johnson, J. E.; Balaich, G. J. Comparative Analysis of Fuel Composition and Physical Properties of Biodiesel, Diesel, Kerosene, and Jet Fuel. *J. Chem. Educ.* **2018**, *95* (10), 1821−1826.
(9) Yang, J.; Xu, C.; Li, B.; Ren, G.; Wang, L. Synthesis and Determination of Biodiesel: An Experiment for High School Chemistry. *J. Chem. Educ.* **2013**, *90* (10), 1362−1364.
(10) Goldstein, S. W. Biodiesel from Seeds: An Experiment for Organic Chemistry. *J. Chem. Educ.* **2014**, *91* (10), 1693−1696.
(11) Bladt, D.; Murray, S.; Gitch, B.; Trout, H.; Liberko, C. Acid-Catalyzed Preparation of Biodiesel from Waste Vegetable Oil: An Experiment for the Undergraduate Organic Chemistry Laboratory. *J. Chem. Educ.* **2011**, *88* (2), 201−203.
(12) Pierce, K. M.; Schale, S. P.; Le, T. M.; Larson, J. C. An Advanced Analytical Chemistry Experiment Using Gas Chromatography−Mass Spectrometry, MATLAB, and Chemometrics to Predict Biodiesel Blend Percent Composition. *J. Chem. Educ.* **2011**, *88* (6), 806−810.
(13) Feng, Z. V.; Buchman, J. T. Instrumental Analysis of Biodiesel Content in Commercial Diesel Blends: An Experiment for Undergraduate Analytical Chemistry. *J. Chem. Educ.* **2012**, *89* (12), 1561−1565.
(14) Habeeb, Z.; Lewis, R.; Liu, S. M.; Prosser, R. S. Qualitative and Quantitative Assessment of Biodiesel Derived from Microalgae. *J. Chem. Educ.* **2020**, *97* (10), 3791−3796.
(15) Randell, J. M.; Boon, L. M.; Nguyen, H. H.; Perez, J.; Windham, C. D.; Cermak, D. M. Biodiesel Production, Analysis, and Combustion: An Across-the-Curriculum Laboratory. *J. Chem. Educ.* **2025**, *102* (2), 765−775.
(16) Wang, X.; Chrzanowski, M.; Liu, Y. Ultrasonic-Assisted Transesterification: A Green Miniscale Organic Laboratory Experiment. *J. Chem. Educ.* **2020**, *97* (4), 1123−1127.
(17) Larraza, I.; Malko, R. Microwave-Assisted Transesterification of Cooking Oils and GC−MS Determination of Their Fatty Acid Constituents: A Research-Based Integrated Lab Experiment. *J. Chem. Educ.* **2020**, *97* (4), 1128−1133.
(18) Bell, R.; Smetana, L.; Binns, I. Simplifying Inquiry Instruction. *Sci. Teach.* **2005**, 30−34.
(19) Ezzati, R.; Ranjbar, S.; Soltanabadi, A. Kinetics models of transesterification reaction for biodiesel production: A theoretical analysis. *Renew. Energy* **2021**, *168*, 280−296.
(20) Encinar, J.; Pardal, A.; Sánchez, N.; Nogales, S. Biodiesel by Transesterification of Rapeseed Oil Using Ultrasound: A Kinetic Study of Base-Catalysed Reactions. *Energies.* **2018**, *11* (9), 2229.
(21) Pauline, J. M. N.; Sivaramakrishnan, R.; Pugazhendhi, A.; Anbarasan, T.; Achary, A. Transesterification kinetics of waste cooking oil and its diesel engine performance. *Fuel* **2021**, *285*, 119108.
(22) Pugazhendhi, A.; Alagumalai, A.; Mathimani, T.; Atabani, A. E. Optimization, kinetic and thermodynamic studies on sustainable biodiesel production from waste cooking oil: An Indian perspective. *Fuel* **2020**, *273*, 117725.
(23) Koh, L. P.; Ghazoul, J. Biofuels, biodiversity, and people: Understanding the conflicts and finding opportunities. *Biol. Conserv.* **2008**, *141* (10), 2450−2460.
(24) Tabatabaei, M.; Aghbashlo, M.; Dehhaghi, M.; Panahi, H. K. S.; Mollahosseini, A.; Hosseini, M.; Soufiyan, M. M. Reactor technologies for biodiesel production and processing: A review. *Prog. Energy Combust. Sci.* **2019**, *74*, 239−303.
(25) Atabani, A. E.; Silitonga, A. S.; Badruddin, I. A.; Mahlia, T. M. I.; Masjuki, H. H.; Mekhilef, S. A comprehensive review on biodiesel as an alternative energy resource and its characteristics. *Renewable Sustainable Energy Rev.* **2012**, *16* (4), 2070−2093.
(26) Knothe, G.; Sharp, C. A.; Ryan, T. W. Exhaust Emissions of Biodiesel, Petrodiesel, Neat Methyl Esters, and Alkanes in a New Technology. *Energy Fuels.* **2006**, *20* (1), 403−408.
(27) Eilks, I. Teaching Biodiesel: A sociocritical and problem-oriented approach to chemistry teaching and students′ first views on it. *Chem. Educ. Res. Pract.* **2002**, *3* (1), 77−85.
(28) Nida, S.; Marsuki, M. F.; Eilks, I. Palm-Oil-Based Biodiesel in Indonesia: A Case Study on a Socioscientific Issue That Engages Students to Learn Chemistry and Its Impact on Society. *J. Chem. Educ.* **2021**, *98* (8), 2536−2548.
(29) Bart, J. C. J.; Palmeri, N.; Cavallaro, S. *Biodiesel Science and Technology*; Woodhead Publishing Limited and CRC Press LLC: Cambridge, UK, New Delhi, India, Boca Raton, USA, 2010.
(30) Van Gerpen, J. Biodiesel processing and production. *Fuel Process. Technol.* **2005**, *86* (10), 1097−1107.
(31) Komers, K.; Stloukal, R.; Machek, J.; Skopal, F. Biodiesel from rapeseed oil, methanol and KOH. 3. Analysis of composition of actual reaction mixture. *Eur. J. Lipid Sci. Technol.* **2001**, *103* (6), 363−371.
(32) Wu, L.; Wei, T.; Lin, Z.; Zou, Y.; Tong, Z.; Sun, J. Bentonite-enhanced biodiesel production by NaOH-catalyzed transesterification: Process optimization and kinetics and thermodynamic analysis. *Fuel.* **2016**, *182*, 920−927.
(33) Keera, S.T.; El Sabagh, S.M.; Taman, A.R. Transesterification of vegetable oil to biodiesel fuel using alkaline catalyst. *Fuel* **2011**, *90* (1), 42−47.
(34) Mao, V.; Konar, S.; Boocock, D. G. B. The pseudo-single-phase, base-catalyzed transmethylation of soybean oil. *J. Am. Oil Chem. Soc.* **2004**, *81* (8), 803−808.
(35) Herzog, M.; Kornejew, E. Native Öle-Rohstoffquelle für Anwendungen in der Kunststoffindustrie. *Wissenschaftliche Beiträge* **2006**, *2006* (11), 44−46.
(36) Csernica, S. N.; Hsu, J. T. Inhibitory Effect of the Byproduct Glycerol Resulting from the Phase Behavior on the Transesterification. *Energy Fuels* **2013**, *27* (4), 2167−2172.

https://doi.org/10.1021/acs.jchemed.4c01335
J. Chem. Educ. XXXX, XXX, XXX−XXX