

Article

Application of Activated Carbon to Obtain Biodiesel from Vegetable Oils

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Received: 8 August 2020; Accepted: 8 September 2020; Published: 11 September 2020



Abstract: The methanolysis of refined vegetable oils (rapeseed oil, sunflower oil, corn oil and olive oil) using a heterogeneous carbonaceous catalyst was studied. Activated carbon (AC) was prepared from beech tree wood and used as the support for KOH and a lipase catalyst. The reactions were carried out for 1–4 h at 60 °C with a methanol-to-oil molar ratio of 6:1 and 0.5–1.5 wt.% KOH/AC. In contrast, the biotransformation of vegetable oils was carried out for 24 h at 40 °C with an alcohol-to-oil molar ratio of 4:1 and 5 wt.% lipase/AC. The highest methanolysis yield (99%) for the fatty acid methyl esters was obtained for lipase/AC. These data show that activated carbon is a promising supporter for KOH as well as for lipase in the transesterification reaction of vegetable oils with methanol. The use of both catalysts in the transesterification reaction may improve biodiesel production. The lipase/AC enables the reduction in methanol excess and eliminates waste formation, whereas the saponification of triglycerides is scanty when KOH/AC is used.

Keywords: biodiesel; heterogeneous catalyst; transesterification; immobilized lipase; activated carbon; bio-based catalysts

1. Introduction

Fatty acid methyl esters (FAMES) are widely used in various applications such as: biosurfactants, biolubricants, “green solvents”, hydraulic fluids, dispersing agents, as well as cosmetic and pharmaceutical products [1–5]. They are non-toxic and renewable with a high degree of biodegradability [6]. The parameters of FAMES are similar to diesel oil, and thus they can be used as a biofuel. Additionally, they have low toxicity, good fat solubility, and exquisite wetting at interfaces. This also makes them attractive components in cosmetic formulations [7].

A wide range of sources of TAGs (triacylglycerols) such as vegetable oil, animal fats, and algae oil can be used in transesterification. Fatty acid esters of alcohols obtained from triglycerides are characterized by their composition and purity [8,9]. Among the fatty acids, oleic acid, which contains only one unsaturated bond in its structure, is the most resistant to thermal oxidation. Sunflower oil contains about 40% oleic acid. However, there are hybrid varieties of sunflower oil with a high oleic acid content (over 80%). Therefore, this vegetable oil is a noteworthy source of TAGs for the synthesis of FAMES [1].

Currently, a homogeneous basic catalyst is the most commonly used industrial approach for the transesterification reaction of vegetable oils. The catalysts, such as KOH, NaOH, or CH₃ONa, are the most representative. This transesterification process has various benefits such as wide availability and low cost of catalyst, along with a high reaction rate, when gentle reaction conditions are used (atmospheric pressure and temperature below the boiling temperature of methanol or ethanol).

The conventional transesterification of TAGs prefers transesterification with short-chain alkyl alcohols, however, it has some limits. The alkaline catalyst is not effective for the conversion of oils containing high FFAs (free fatty acids) and the transesterification reaction must be carried out under anhydrous conditions [10–12]. This is because the presence of FFAs enhance the usage of the alkali catalyst. In addition, soaps are formed, complicating the separation of the ester phase from the glycerin phase, and the purification of the ester phase [13,14]. Saponification also leads to the formation of water, which promotes the generation of more FFAs and soaps during transesterification [15]. The side reactions reduce the yield of the transesterification reaction, which in turn generates additional production costs [16]. Acid homogeneous catalysts are an alternative to alkaline catalysts to convert oils containing high free fatty acids. The most common are HCl, H₂SO₄, H₂SO₃, organic sulfonic acid, and Fe₂(SO₄)₃. However, acid-catalyzed transesterification reactions are not recommended due to their comparatively slow reaction rate that requires temperatures over 100 °C and more alcohol [17–19].

Biotransformation is an alternative to the chemical TAG transesterification reaction in order to overcome the drawbacks of using basic and acidic catalysts in TAG transesterification. Lipases are hydrolytic enzymes that are well recognised as remarkable catalysts under ambient conditions, performing a myriad of chemical reactions with unparalleled enantio- and regioselectivities, making them useful in various processes. This is especially true of the pharmaceutical and cosmetics industries, and also for biodiesel production [20].

Heterogeneous catalysts have attracted considerable attention in recent years since they are less sensitive to FFA contamination during the transesterification reaction. The use of cheap catalyst alternatives would decrease the cost of the transesterification process. The advantages of heterogeneous catalysts in the production process can be seen from at least three perspectives: they can be easily separated from the products of the reaction, are reusable and have high catalytic activity, and can be reused for transesterification. Some steps of the transesterification process can be avoided, such as the purification of FAME and crude glycerol.

Of the various accessible catalyst carriers, activated carbon is a low-cost and promising solid carrier for chemical and enzymatic catalysts. Basically, activated carbon is obtained through physical or chemical activation from various carbon materials such as coal, wood, and biomass sources. Until recently, the main sources of activated carbon were anthracite and bituminous coal. Nowadays, any material with high carbon content, and also a low inorganic content, can be used for obtaining activated carbon [21–23]. Activated carbon is characterized by a wide variety of evenly spaced micropores and macropores. The activated carbon's microporosity makes it a representative material for use in catalyzing chemical reactions. Their low cost and resistance to heat and radiation make them readily used in transesterification processes of triglycerides [24,25]. The great adsorption surface area (500 m²/g to 3000 m²/g) depends on the method (physical and chemical) of activation of the biomass derived [26,27]. Activated carbons are characterized by high catalytic activity, which is due to the presence of oxides and other chemical properties on the surface, and therefore it allows them to be used as catalyst carriers [28–31].

In the transesterification reaction, activated carbon is most often used as a carrier for CaO [32,33], KOH [34–36] and lipase [37]. However, KOH/AC catalysts lead to more effective transesterification than CaO catalyst (91.3 and 87.0%, respectively) [38]. While acetate potassium was loaded on AC and used as a heterogeneous catalyst to obtain biodiesel from bitter almond oil, a yield of 93.2 wt.% was obtained. The AC surface area was 1105 m²/g. [39].

Previous studies have shown that the use of heterogeneous catalysts requires a large excess of alcohol or a large amount of catalyst in the transesterification reaction [16,18]. This study is a complement to our previous research [36], which shows the most interesting data concerning activated carbon with the oxygen groups on its surface.

The novelty of this research is to show the usage of activated carbon with a low BET (Brunauer-Emmett-Teller) surface area and a high content of oxygen groups as a carrier for catalysts (KOH and lipase) in a transesterification reaction of vegetable oils. Studies have reported cheap, efficient,

and reusable catalysts that can be performed with a small excess of alcohol and a low amount of the catalyst in the transesterification reaction of vegetable oils.

2. Results and Discussion

2.1. Characterization of the Activated Carbon

Scanning electron micrographs (SEM) of the external structure of AC, used as a catalyst carrier, are presented in Figure 1.

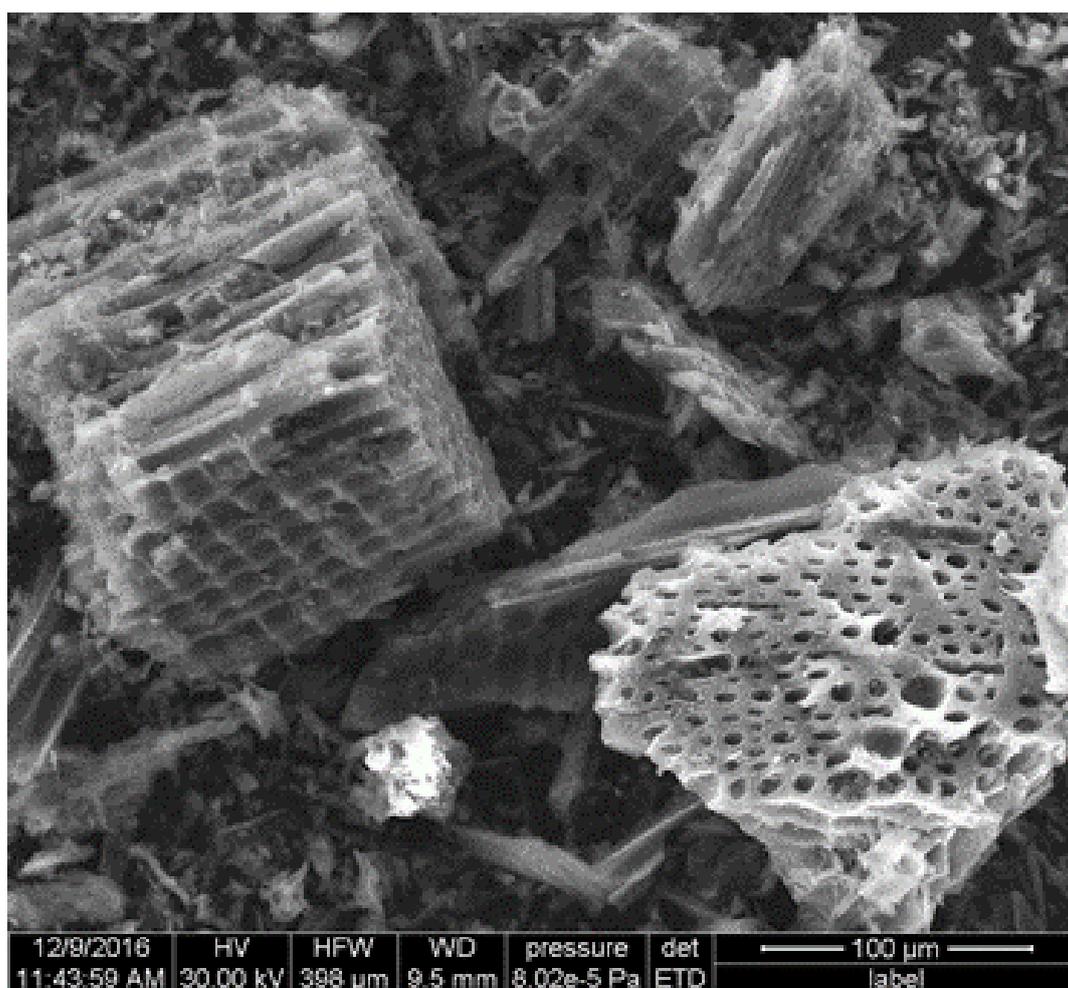


Figure 1. SEM of the powder-AC produced from beech wood.

The surface of AC, obtained from beech tree, is characterized by an irregular and porous surface, which is favorable for the adsorption process [40]. The matrices of AC present a honeycomb structure.

The characterization of activated carbon, used as a carrier of catalysts, was described in Narowska et al. [36]. The BET surface area of the activated carbons was 72 m²/g. On the adsorption properties of activated carbon, three factors have an influence, such as the surface and porous structure as well as the presence of heteroatoms on its surface, mainly oxygen [41]. The AC used in the research is composed of 84.2% carbon and has a high oxygen groups content (12.4%).

2.2. Effect of the Production Process Parameters on the Yield of Biodiesel

Homogeneous alkaline catalysts have found widespread use on an industrial scale for biodiesel production. Research currently focuses on heterogeneous catalysts, and they can offer many

perspectives. Here, a KOH catalyst was compared with KOH carried on AC. Moreover, immobilized lipase on AC was also used as a catalyst versus the chemical catalyst. The influence of the use of the KOH catalyst and reaction time on the transesterification reaction yield of RO, SO, CO, and OO are shown in Figure 2.

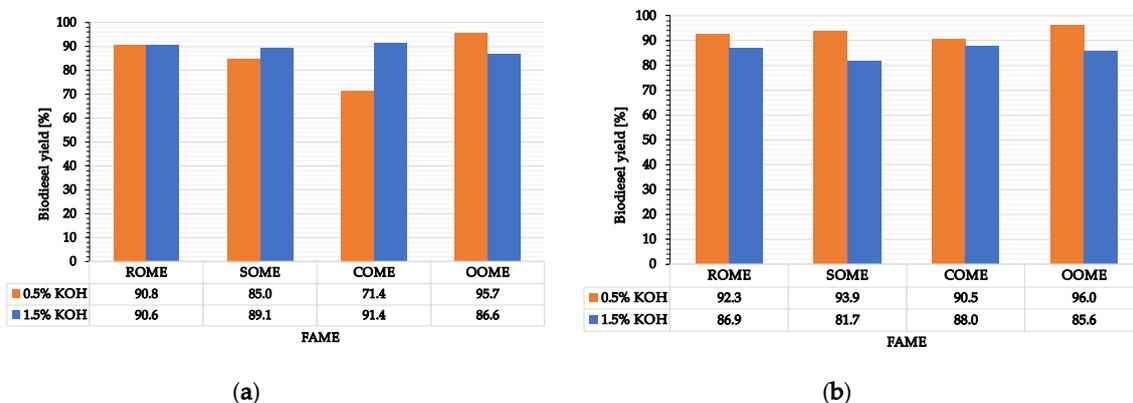


Figure 2. The yield of biodiesel obtained from vegetable oils in the presence of KOH. [Reaction time (a) 1 h and (b) 4 h].

The influence of the use of the KOH/AC catalyst and reaction time on the transesterification reaction yield of RO, SO, CO and OO are shown in Figure 3.

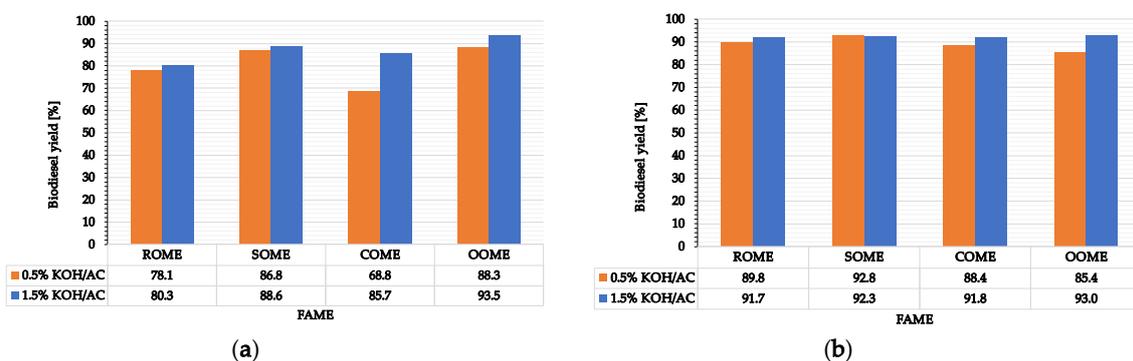


Figure 3. Yield of biodiesel obtained from vegetable oils in the presence of KOH/AC. [Reaction time (a) 1 h and (b) 4 h].

Once KOH was used in the transesterification reaction, the highest yields (>90%) for a 6:1 molar ratio were obtained for 0.5% KOH at 4 h. The highest yield was obtained when the triglyceride source was olive oil and was 96% (Figure 2), and the best properties were achieved for methyl esters of rapeseed oil (ROME) (Table 1). The products complied with the standards given in the EN-14214 [42]. A lower concentration (0.5%) of KOH resulted in a higher yield versus a higher concentration (1.5%) of KOH. Sharma et al., (2010) also found that the excess KOH amount in the transesterification reaction affected the forming of soaps (saponification) [43].

When the KOH/AC was used in the transesterification reaction, the highest yields (>91%) were achieved for 1.5% catalyst; the reaction time was 4 h. The highest transesterification reaction yield (93%) is found for olive oil in this reaction. The properties and the yield of the methyl esters do not have a significant difference (Table 2). The exception was methanolysis of olive oil where the highest yield (93.5%) was obtained for 1.5% of the catalyst with a 1 h reaction—this might be because of the unreacted TAGs, considering the properties of this reaction (Table 2). In comparison, Baroutian et al. (2011) carried out a transesterification reaction of palm oil with methanol (volume ratio; oil:methanol, 1:1), at a temperature range of 50–70 °C for 1 h in the presence of KOH loaded onto AC (obtained

from palm shell-based), characterized by a BET surface area of 1015 m²/g. The highest yield was 94% at 70 °C [44]. In turn, Buasri et al. (2012) obtained biodiesel with a yield of 86.3% when using a methanol-to-oil molar ratio of 25:1, a reaction time of 2 h, a reaction temperature of 60 °C, and waste frying oil was applied as a source of triacylglycerides. The proper amount of AC was agitated with a solution of KOH with 0.40 g/mL concentration for 24 h. The BET surface area of AC obtained from coconut shell was 898.6 m²/g, and the pore volume was 0.905 m³/g [45]. Dhawane et al. (2016) have applied AC as catalyst support for KOH in a transesterification reaction of *Hevea brasiliensis* oil. The AC was prepared from flamboyant pods (*Delonix regia*) and was characterized by the BET surface area of 820 m²/g AC. The highest yield (89.81%) was obtained for KOH loading 3.5 wt.%, with a 15:1 methanol to oil molar ratio, a reaction temperature of 55 °C, and a reaction time of 1 h [46].

In both cases (KOH and KOH/AC), the prolongation of the reaction time from 1 h to 4 h results in improved parameters of the methyl esters (Tables 1 and 2). Increasing the amount of KOH from 0.5 to 1.5% improves the parameters of biodiesel obtained from RO and CO as opposed to SO and OO; here, the increase in catalyst contribution causes a slight difference in the properties of FAMES. The most visible differences were noticed in ROME where biodiesel with the best parameters was obtained in the following reaction conditions: 1.5% KOH and 1 h. The use of KOH/AC allowed one to obtain methyl esters with slight differences in properties. An increase in the KOH/AC amount from 0.5 to 1.5% and the duration of the reaction from 1 h to 4 h improved the parameters and yield (approximately 6 to 19%). The higher yield of the heterogeneous transesterification reaction versus the homogeneous transesterification reaction can be due to the limited evaporation rate of the methanol used in the reaction. There are reports that a heterogeneous catalyst leads to an upswing of the viscosity of the reaction mixture, thereby reducing alcohol evaporation [47,48].

The use of AC to immobilize lipase is a promising prospective solution because it offers a transesterification reaction with high yield. This leads to obtaining biodiesel with good quality [49–52]. The highest yield was 99.9% for methyl esters of corn oil (COME) (Figure 4). For all oils, the second reuse of EvTr/AC (lipase Eversa Transform 2.0 immobilized onto AC) led to the highest yield. This exceeded 97% of the methyl esters yields (Figure 4). The use of EvTr/AC in the methanolysis of CO succeeded at high yield (99.9%)—this was maintained over four reaction cycles without a significant change in FAME parameters. The density, kinematic viscosity, and CFPP (cold filter plugging point) were 885 kg/m³ and 886 kg/m³, 4.7 mm²/s, and −12 to 4 °C, respectively (Table 3). However, the lowest CFPP (−13 °C) was obtained for ROME, which is even lower when using an alkaline catalyst in the transesterification of OR.

Table 1. The effect of a catalyst KOH on the properties of ROME, SOME (methyl esters of sunflower oil), COME and OOME (methyl esters of olive oil). [Molar ratio methanol-to-oil, 6:1].

FAMES	Amount of Catalyst (wt.%)	Reaction Time (h)	Density at 15 °C (kg/m ³)	Viscosity at 40 °C (mm ² /s)	CFPP (°C)
ROME	0.5	1	882	5.1	−1
	0.5	4	882	4.8	−4
	1.5	1	882	4.4	−10
	1.5	4	880	4.4	−5
SOME	0.5	1	885	4.5	3
	0.5	4	883	4.2	2
	1.5	1	883	4.2	0
	1.5	4	882	4.3	3
COME	0.5	1	885	4.2	1
	0.5	4	886	4.5	0
	1.5	1	883	4.5	−3
	1.5	4	883	4.3	−1
OOME	0.5	1	877	4.9	0
	0.5	4	877	4.6	1
	1.5	1	887	4.6	−2
	1.5	4	876	4.6	1

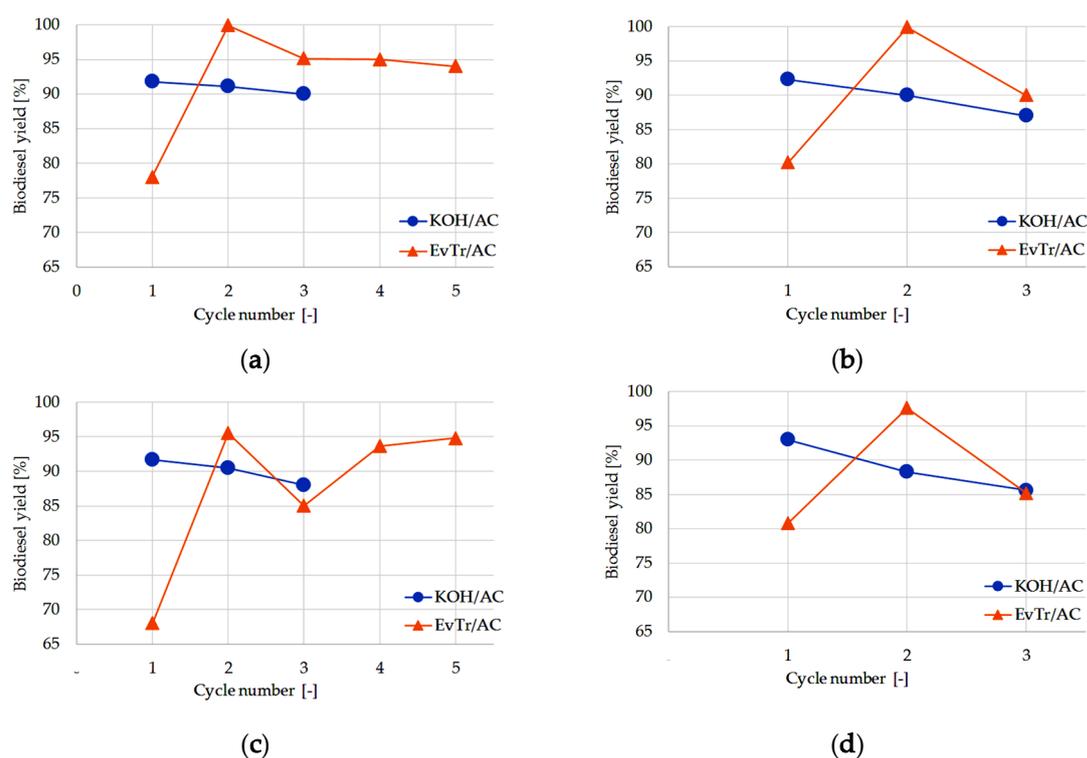


Figure 4. The yield of biodiesel obtained from vegetable oils reusing KOH/AC and EvTr/AC. [(a) biodiesel obtained from corn oil, (b) biodiesel obtained from sunflower oil, (c) biodiesel obtained from rapeseed oil, (d) biodiesel obtained from olive oil].

Dhawane et al., (2019) also carried out a transesterification reaction by using AC obtained from flamboyant pods; the BET surface area was $812.2 \text{ m}^2/\text{g}$. The highest yield (83.9%) was obtained with a catalyst loading of 3 wt.%, a molar ratio of methanol-to-oils of 6:1, temperature below $20 \text{ }^\circ\text{C}$, and the reaction time lasted for 5 h [53]. In turn, Naranjo et al. (2010) used palm oil for transesterification with six different alcohols (methanol, ethanol, 1-propanol, 1-butanol, isopropanol or isobutanol) in the presence of lipase immobilized onto two types of activated carbon, which was prepared from coconut shell. The BET surface area was $1867 \text{ m}^2/\text{g}$ and $1674 \text{ m}^2/\text{g}$ for granular activated carbon and activated carbon cloth, respectively. Biodiesel with the highest yield (100%) was obtained when isobutanol was used as alcohol for the samples with granular activated carbon, while it was 82% when activated carbon cloth was used. The other reaction conditions were: a 1:6 molar ratio of oil:alcohol and a reaction temperature of $40 \text{ }^\circ\text{C}$ [54].

Table 2. The effect of KOH/AC as catalyst on properties of ROME, SOME, COME, and OOME. [Molar ratio methanol-to-oil, 6:1].

FAME	Number of Reuse	Amount of Catalyst (wt.%)	Reaction Time (h)	Density at $15 \text{ }^\circ\text{C}$ (kg/m^3)	Viscosity at $40 \text{ }^\circ\text{C}$ (mm^2/s)	CFPP ($^\circ\text{C}$)
ROME	I	0.5	1	889	5.8	4
	II	0.5	1	907	14.5	14
	I	0.5	4	887	5.0	1
	II	0.5	4	903	11.7	15
	I	1.5	1	885	5.0	-3
	II	1.5	1	892	6.4	4
	I	1.5	4	886	4.6	-5
	II	1.5	4	890	6.4	-2

Table 2. Cont.

FAME	Number of Reuse	Amount of Catalyst (wt.%)	Reaction Time (h)	Density at 15 °C (kg/m ³)	Viscosity at 40 °C (mm ² /s)	CFPP (°C)
SOME	I	0.5	1	890	5.5	4
	II	0.5	1	916	15.8	16
	I	0.5	4	888	4.8	-5
	II	0.5	4	910	12.3	16
	I	1.5	1	886	4.6	-4
	II	1.5	1	895	6.3	5
	I	1.5	4	885	4.4	-6
	II	1.5	4	897	6.9	-1
COME	I	0.5	1	895	6.3	6
	II	0.5	1	909	14.3	15
	I	0.5	4	888	5.1	1
	II	0.5	4	905	11.7	14
	I	1.5	1	886	4.6	-5
	II	1.5	1	894	6.3	4
	I	1.5	4	886	4.3	-6
	II	1.5	4	890	6.0	-8
OOME	I	0.5	1	887	7.5	7
	II	0.5	1	915	32.3	21
	I	0.5	4	885	6.0	4
	II	0.5	4	911	29.6	20
	I	1.5	1	880	5.0	-2
	II	1.5	1	896	9.2	11
	I	1.5	4	880	4.7	-6
	II	1.5	4	890	8.6	8

Table 3. The effect of EvTr/AC as catalyst on properties of ROME, SOME, COME and OOME. [Molar ratio methanol-to-oil, 4:1; the amount of EvTr/AC was 5 wt.%].

FAME	Cycle Number	Density at 15 °C (kg/m ³)	Viscosity at 40 °C (mm ² /s)	CFPP (°C)
ROME	I	883	4.8	-13
	II	883	4.7	-13
	III	883	4.7	-12
	IV	887	5.5	-7
	V	889	6.4	-1
SOME	I	895	7.3	-6
	II	888	6.3	-8
	III	893	7.3	-7
COME	I	885	4.7	-12
	II	885	4.7	-5
	III	886	4.7	-4
	IV	891	4.1	-4
	V	893	7.3	-1
OOME	I	892	9.7	-3
	II	887	6.5	-5
	III	895	9.8	-3

2.3. The Effect of Various Sources of TAGs on Biodiesel Properties

This work studied the four most common oils used for biodiesel in Europe. Moreover, the results were compared with the previous study described in Narowska et al. [35], where waste corn oil was transesterified with methanol by the use of KOH and KOH/AC.

The characteristics of the vegetable oils used for obtaining biodiesel are shown in Table 4. The vegetable oils have different contents of free fatty acids, and waste corn oil has the highest value. In turn, sunflower oil and corn oil have the highest content of linoleic acid (approximately 55%);

rapeseed oil and olive oil have the highest content of oleic acid (two-fold more than sunflower oil and corn oil). The differences can influence the parameters of the biodiesel.

Table 4. Properties of vegetable oils used in the transesterification reaction.

No.	Property	Unit	Vegetable Oil				
			Rapeseed Oil	Sunflower Oil	Olive Oil	Corn Oil	Waste Corn Oil [36]
1	Free fatty acid content	wt. %	0.04	0.05	0.37	0.04	14.11
2	Water content	wt. %	0.08	0.05	0.04	0.04	0.07
3	Fatty acid composition	wt. %					
	Myristic; C 14:0		0.3	0.3	0.3	0.4	0.1
	Palmitic; C 16:0		4.2	7.4	14.0	10.7	10.1
	Palmitoleic; C 16:1		0.3	0.3	1.8	0.3	2.0
	Stearic; C 18:0		1.6	3.8	2.7	1.9	1.8
	Oleic; C 18:1		64.1	29.4	69.0	30.9	28.6
	Linoleic; C 18:2		18.1	56.9	10.7	54.7	55.6
4	Linolenic; C 18:3	wt. %	8.5	0.0	0.0	0.0	1.1
	Arachidic; C 20:0		0.7	0.3	0.5	0.6	0.4
	Gadoleic; C 20:1		1.3	0.1	0.3	0.2	0.3
	Behenic; C 22:0		0.4	0.9	0.1	0.2	0.0
	Erucic; C 22:1		0.2	0.2	0.0	0.0	0.0
	Lignoceric; C 24:0		0.1	0.3	0.1	0.1	0.0
	Nervonic; C 24:1		0.2	0.1	0.5	0.0	0.0
5	Density at 15 °C	kg/m ³	924	920	920	924	925
6	Viscosity at 40 °C	mm ² /s	32.0	34.0	38.2	33.7	28.8
7	CFPP	°C	21	19	20	19	20

The kinematic viscosity, density, and cold filter plugging point (CFPP) of the FAMES (Tables 1–3) significantly decreased versus vegetable oils (Table 4). The density decreased from 924 kg/m³ to 885 kg/m³ for ROME, 924 kg/m³ to 886 kg/m³ for COME, and 925 kg/m³ to 885 kg/m³ for methyl esters of waste corn oil (COMEW). The kinematic viscosity of RO, CO, and COW (waste corn oil) decreased from 32.0 mm²/s to 4.6 mm²/s, from 33.7 mm²/s to 4.3 mm²/s, and from 28.8 mm²/s to 4.1 mm²/s, respectively. The CFPP decreased from approximately 20 to −6 °C, which suggests that it can be used during a transitional period.

Enzymatic transesterification reactions of rapeseed and corn oils had density and kinematic viscosity values similar to EN 14214. According to the adopted standards, the density at 15 °C and the kinematic viscosity at 40 °C should be in the range of 860–900 kg/m³ and 3.5–5.0 mm²/s, respectively. This enzymatic transesterification also had positive effects on the CFPP of the biodiesel. That is, it reduces this value to a −3 to −13 °C range. Versus alkali transesterification, the minimum value of CFPP was −10 °C (ROME) and −8 °C (COME) for KOH and KOH/AC as a catalyst, respectively.

In the case of KOH-catalyzed methanolysis, extending the duration of the reaction time and increased catalyst concentration led to obtaining biodiesel yields with properties in accordance with EN 15214. However, these do not have a significant result on the values in the physical characteristics of FAMES. The CFPP of SOME and COME was reasonably low (0 °C and −3 °C, respectively) (Table 1) due to a greater amount of double bonds [55,56]. The chemical analysis of ROME, COME and COMEW was carried out for the esters with the best physical properties (Table 5). The biodiesel properties were very close to biodiesel standard EN-14214.

Table 5. Chemical properties of ROME, COME, and COMEw. [Molar ratio methanol-to-oil ratio of 6:1; amount of catalyst KOH/AC = 1.5 wt.%; reaction time, 4 h].

Property	Unit	FAME					EN 14214
		ROME I Use	ROME II Use	COME I Use	COME II Use	COMEw I Use	
FAME content	wt.%	96.3	83.9	91.9	82.4	98.5	≥96.5
Monoglyceride content	wt.%	0.67	0.99	0.56	1.16	0.38 ¹	<0.70
Diglyceride content	wt.%	0.55	3.62	0.30	3.03	0.24 ¹	<0.20
Triglyceride content	wt.%	1.64	8.81	0.76	7.10	0.87 ¹	<0.20
Free glycerol	wt.%	0.004	0.004	0.005	0.006	0.010 ¹	<0.02
Total glycerol	wt.%	0.422	1.691	0.271	1.476	0.230 ¹	<0.25
Acid value	mgKOH/g	0.16	0.19	0.15	0.15	0.08 ¹	<0.50
Methanol value	wt.%	0.002	0	0.001	0	0.70 ¹	<0.20

¹ data from (Narowska et al., 2019).

The content of mono-, di- and triglycerides in biodiesel is strictly defined in the EN 14214 standard, therefore the results of FAMES have been compared with it. Individual glycerides appeared in all FAMES samples, from which it could be concluded that the transesterification was incomplete. The low content of free glycerol indicates a satisfactory biodiesel purification process. Some unreacted glycerides increased the total glycerol in the samples. However, bound glycerol could be removed using vacuum distillation [57].

Biodiesel was assayed for the content of FFA, which corresponds to the acid value (AV). Its value depends on the source of TAGs used and the method of the transesterification process that is carried out. The level of FFAs in the obtained FAMES does not exceed the value recommended by the standard. Thus, the resulting biodiesels will not corrode or clog the pump due to the formation of deposits [58]. The fatty acids values agreed with the standards. Therefore, this method of obtaining biodiesel is suitable for the transesterification of both pure vegetable oils and also COW with FFAs content over 0.5%. The increased methanol content in COMEw suggests the necessity for an additional stage of biodiesel purification.

2.4. Catalyst Reusability

The reusability of the KOH/AC and EvTr/AC catalyst in the transesterification of vegetable oils with methanol was studied under optimum conditions (Figure 4). The catalysts were separated from the reaction mixture after transesterification and then used again if the physical properties of biodiesel were in accordance with EN14214 (Tables 2 and 3). The activity of heterogeneous catalysts was explained based on the obtained yields of biodiesel and their properties. The KOH/AC catalyst may be recycled without losing significant activity. The biodiesel yields were around 80–93% for both fresh and reused catalyst. After three cycles, the COME activity slightly decreased (around 2%). However, the highest losing activity was observed for OOME (around 10%). This could be due to the significant leaching of active sites of AC. In comparison, the EvTr/AC catalyst could be reused from three to five cycles with a high yield of methanolysis (68–99%), depending on the oil used. The highest yield was achieved in the second use of the catalyst (97–99%). Yields in the third cycle of EvTr/AC decreased by around 10–12% for ROME and OOME, respectively, and in the fifth cycles, yield values were without significant changes for ROME and COME. The first use of the catalyst was associated with preincubation, which progresses the methanolysis much faster in a further cycle [59]. In turn, the decrease in biodiesel yield in the third use of the catalyst might be due to the inactivated effect caused by the glycerol byproduct—the glycerol byproduct is easily adsorbed onto the surface of the immobilized lipase [37,54].

The heterogeneous catalyst KOH/WA showed a high value yield compared with other research. Baroutian et al., (2011) used KOH/AC three times with a biodiesel yield of 89.3% [44], while Buasri et al.,

(2012) used KOH/AC four times with a 80.3% yield [45]. Moreover, it was characterized by stability for the transesterification reaction with minimal loss of its catalytic activity. In turn, Dhawane et al., (2019) reused a biocatalyst (lipase immobilized onto AC) seven times and only 6–7% losses in catalytic activity were observed [53].

3. Materials and Methods

3.1. Materials

Vegetable oils (rapeseed oil, sunflower oil, corn oil and olive oil) were used as a feedstock. Refined vegetable oils were purchased from a local grocery. The powdered AC was obtained from a beech tree and the full description of carbonization with simultaneous activation is contained in [36]. Methanol, KOH and anhydrous Na₂SO₄(VI) were purchased from Chempur, Poland. All reagents were of analytical grade. The liquid lipase Eversa Transform 2.0 (lipase activity 1000 kLU g⁻¹, in an enzyme mixture containing 75 vol% of water and 25 vol% of propylene glycol) was kindly provided by Novozymes A/S (Madrid, Spain).

3.2. Methods

3.2.1. The Activated Carbon Characterization

The AC was characterized by technical analysis (according to PN-80/G-04511, PN-80/G-04512, PN-80/G-04516) and elemental analysis using a VarioEl analyzer (ELEMENTAR Analysensysteme GmbH, Langensfeld, Germany). The full description of the method is contained in Narowska et al., (2019) [36].

3.2.2. KOH/AC Catalyst Preparation

The full description of the heterogeneous base catalyst preparation is contained in Narowska et al., (2019) [36].

3.2.3. Lipase Immobilization on AC

Liquid lipase Eversa Transform was used for the immobilization on AC. The optimum conditions for EvTr/AC were defined based on modification of a previous study [54,60]. Brito et al., (2017) [60] used porcine pancreas type II Sigma and the activity of immobilized enzyme measured by hydrolysis of the olive oil emulsion method. Naranjo et al., (2010) [54] used *Candida antarctica B* lipase and the operational stability of immobilized lipase determined by the transesterification reaction of palm oil. The optimal immobilization test was found at 4 °C for 12 h in pH 7 with agitation. The lipase was mixed in 25 mM phosphate buffer and added to a vial containing AC dried at 110 °C. The proper amount of lipase and AC was calculated from the absorption capacity of 1.16 g_{EvTr}/1g_{AC}. The activity of immobilized enzyme was measured by the yield of methanolysis of vegetable oils.

3.2.4. Vegetable Oil Characterization

The Karl Fischer (Mettler, Toledo, OH, USA) titration was used as a method for water content determination. The free fatty acid was calculated according to the formula below:

$$\%FFA = AV \times f \quad (1)$$

where AV-acid value, f-factor for proper vegetable oil [61].

The AV was determined by direct titrating of the sample of TGs in the alcoholic solution against standard KOH using phenolphthalein as an indicator. The Ubbelohde type viscometer was used as an instrument to determine the kinematic viscosity. These values of vegetable oils were measured at 40 °C. The hydrometer method was applied for determining the density at 15 °C. The CFPP was determined

by measuring the lowest temperature at which the proper value of the sample still passed under a pressure of (2 kPa) through a filter with a mesh size of 45 μm within 60 s (Petrotest GmbH, Berlin, Germany). The fatty acid composition in the vegetable oil was obtained using the gas chromatograph provided with a FID (flame ionization detector) (Agilent GC 7890A automatic sampler, Santa Clara, CA, USA). The details of the parameters are described by Narowska et al. [36].

3.2.5. Biodiesel Characterization

Biodiesel yield was calculated according to the equation mentioned below:

$$\text{Biodiesel yield} = wt_{\text{biodiesel}}/wt_{\text{oil}} \times 100\% \quad (2)$$

The properties of biodiesel such as FAME content, mono-, di- and triglyceride content, free glycerol, total glycerol methanol value, acid value, density, viscosity and CFPP were measured. The details of the parameters were described by Narowska et al. [36].

3.2.6. Transesterification of Vegetable Oil with KOH as a Catalyst

The synthesis of FAME from KOH-catalyzed methanolysis of vegetable oil was performed in a 250-mL flask immersed in a water bath on the magnetic stirrer with a heating plate (neoLab, Germany, Heidelberg). The transesterification reaction was carried out at a 6:1 methanol-to-oil molar ratio and the KOH amount of 0.5% and 1.5% (based on the oil weight), at 60 °C, under the atmospheric pressure. The proper amount of KOH catalyst was dissolved in methanol and was added to the oil with the desired temperature. Then, the reaction was performed for 1 h or 4 h. When the reaction was finished, the ester phase was separated from the glycerol phase by a flask separator and washed using distilled water until the catalyst was removed. Next, the biodiesel was dried on anhydrous $\text{Na}_2\text{SO}_4(\text{VI})$.

3.2.7. Transesterification of Vegetable Oil with Lipase as a Catalyst Supported on the Activated Carbon

The transesterification reaction was performed as previously described using the lipase immobilized onto AC instead of KOH. Upon completion of the process, the reaction mixture was filtered and the EvTr/AC was reused in the subsequent transesterification reactions. Then the glycerol phase was separated from the ester phase. The esters obtained were further washed with distilled water to give a pure product. Finally, the esters were dried over anhydrous $\text{Na}_2\text{SO}_4(\text{VI})$ and filtered.

4. Conclusions

Activated carbon has potential utility as a carrier for KOH and lipase as a heterogeneous carbonaceous catalyst for the transesterification of vegetable oils. The catalyst/AC is an excellent counterpart to the homogenous catalyst and was used to obtain biodiesel from vegetable oils. This minimizes homogeneous transesterification process problems. The system has several advantages:

- The use of AC as a carrier for KOH or lipase leads to carrying out the transesterification reaction more efficiently, in comparison to the homogeneous catalyst KOH;
- The use of AC to immobilize lipase allows an increase in the stability of EvTr, which allows for reuse in subsequent cycles up to the fifth cycle. Carrying out the enzymatic transesterification of vegetable oils in the presence of EvTr/AC results in no soap formation. Therefore, the steps of FAMEs and glycerol fraction purification in the industrial process of biodiesel production could be avoided.

Author Contributions: Conceptualization, M.K. and M.L.; methodology, B.E.N., M.K. and M.L.; validation, B.E.N.; investigation, B.E.N.; writing—original draft preparation, B.E.N.; writing—review and editing, M.K. and M.L.; visualization, B.E.N.; supervision, M.K. and M.L. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by National Scientific Leadership center, grant number 327/2015/KNOW.

Acknowledgments: The authors thank Novozymes Spain S.A. for gently providing the enzyme Eversa Transform 2.0. The help and suggestions from Ramiro Martinez Gutierrez (Novozymes Spain, S.A.) are gratefully recognized. This project was partially supported by the Wroclaw Center for Biotechnology (WCB), the Leading National Research Centre (KNOW) for the years 2014–2018, and a statutory activity subsidy from the Polish Ministry of Science and Higher Education for the Faculty of Chemistry of Wroclaw University of Technology.

Conflicts of Interest: The authors declare no conflict of interest.

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