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Research article

Extraction of radish seed oil (*Raphanus sativus L.*) and evaluation of its potential in biodiesel production

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Abstract: Growing concern about replacing fossil fuels with renewable energy sources, coupled with society's concerns about environmental preservation, are the main reasons why governments have sought strategies for increased production and consumption of renewable and sustainable fuels. Radish (*Raphanus sativus L.*) belongs to the group of oilseeds of the *Brassicaceae* family, being widely cultivated in the south and central-west regions of Brazil, with physical and chemical characteristics propitious to the production of biofuels. In this context, the objective of the present work was to evaluate three different methods of oil extraction: solvent extraction, cold pressing and swelling technique, to evaluate the potential of radish oil in biodiesel production using methanol, ethanol and a mixture of both containing 60% and 40% respectively, and to evaluate the composition of fatty acids. The methodology consists firstly in the extraction of the oil, followed by transesterification reaction using 10 mL of pretreated oil and sodium hydroxide in the proportion of 1% in mass, relative to the oil. The oil samples were used in the transesterification reactions with methanol, ethanol and a mixture of methanol (60%) and ethanol (40%) in different proportions and temperatures. The results indicated that, for the oil extraction processes, the seed swelling technique presented a high extraction yield (34%). The composition of fatty acids showed presence of approximately 30% of saturated compounds and around 50% of compounds with chain up to 18 carbons, also had presence of erucic acid (up to 41%), and a high percentage of oleic acid (up to 30%). Regarding the transesterification reactions, the highest yield occurred with the use of methanol,

about 86%. Thus, in the light of the results, it can be concluded that radish oil has great potential for biodiesel production, but, other analyzes, such as acidity and flash point, should be performed to more specifically evaluate the physicochemical characteristics of biodiesel.

Keywords: radish; biodiesel; oil; transesterification; fatty acids

1. Introduction

The concept of sustainability, which has been much sought in recent years, is linked to the substitution of exhaustible sources of energy for renewable sources. In this way, sustainability its the concept of biorefinery, which aims to convert biomass into biofuels, as well as biochemicals, biomaterials and other bioproducts of commercial interest, in order to find a solution that combines economic viability with an environmentally friendly production [1,2].

Much of the energy consumed in the world is obtained through fossil fuels. However, these sources are limited and, in this way, there is a growing demand regarding the search for alternative sources of energy. Thus, vegetable oils and different biomasses appear as alternatives to substitute traditional petroleum fuels [3]. Biodiesel is an example of the use of biomass for energy production due to the great advantages over petroleum diesel, since it is non-toxic and comes from renewable sources; in addition, it has a better emission quality during the combustion process [4]. Fuels produced from vegetable oils have similar properties to fossil fuels, as well as some advantages such as sustainability, lower content of sulfuric and aromatic compounds and biodegradability [5].

In Brazil, soybean is the main raw material used for oil extraction for biodiesel production [6]. In this context, there is a need to search for other oilseeds that produce non-edible oils to produce biodiesel within the quality standards, such as radish (*Raphanus sativus*) that emerges as an excellent alternative for its use in biodiesel production, since it provides chemical and physical improvements of the soil, being used in crop rotation and has high oil content in the seeds [1,2,7].

Radish is a very vigorous plant, in 60 days covers about 70% of the soil and its planting cycle is annual, occurs between April and May and the period of duration is three months. In addition, the production of green mass is from 20 t/ha to 35 t/ha, the production of dry mass is from 4 t/ha to 8 t/ha and the grains are yellowish brown with 2 mm to 3 mm of diameter [8]. An important factor in the use of oil from the radish seed is its low viscosity, which improves engine performance and it is an excellent oil to produce biodiesel. The average oil content in the seeds is 35% by mass and the productivity is 1500 kg/ha, so when submitted to cold pressing, it provides an average of 284 L/ha of oil [7].

Seed oils are composed of a mixture of saturated and unsaturated fatty acids whose composition depends on the genetics of the cultivar, nutrient conditions, climate, soil type and the presence of diseases [9,10]. In addition, through the solvent extraction and mechanical press processes, there is generation of large amount of residual cake containing oil that is not extracted, so there is a need to obtain a new methodology to optimize and consequently increase the efficiency of the extraction process [9]. And, in relation to the production of seed cake, there is the possibility of animal feed

production due to the high amount of protein and also its use in thermochemical processes (pyrolysis) due to the characteristics of lignocellulosic biomass composed of lignin, cellulose and hemicelulose [11,12]. In the case of diesel and biodiesel products, the kinematic viscosity of radish oil is higher, and the specific mass of the products is lower, with higher superior calorific value. Moreover, a higher amount of monounsaturated fatty acids provides better physicochemical characteristics in biodiesel production such as lower levels of degradation, lower amounts of solid residues and greater oxidative stability [13,14].

The use of forage radish oil for biodiesel production generates a pie as a residue, however, according to [15], the pie produced by the oil extraction of the radish seed has a high protein content when compared to jerrycans and crambe, approximately 49.47% (dry mass), so that this residue could be classified as potential protein foods in animal diets. Another study in the literature produced biodiesel from fodder radish oil, and this biodiesel had its characteristics within the ASTM and European Union standards, showing carbon residue below 0.05 (wt.%), sulfur content below 0.005 (wt.%), viscosity of 4.87 mm^2/s at 40 ℃ and density of 877 kg/m^3 at 20 ℃ [13]. In addition, biodiesel derived from fodder radish oil presented significant amounts of erucic acid which would make radish inappropriate to the food market, making it an exclusive cultivar for the production of bioenergy.

Besides that, the energy balance of certain crops may vary, this can be explained by the different production methods and technologies involved in the process, however, according to [16,17], the energy balance of fodder radish is superior to other oilseeds such as canola, palm, castor oil and soy. The forage radish has an energy balance of 8.44, followed by soybean (5.44), palm (4.6), canola (2.19) and castor bean (1.28). According to [18], when compared to soybean and sunflower oil, the oil obtained from forage radish seeds has a higher calorific value and a higher melting point; in addition, soybean oil has lecithin and other gums in its composition, making further purification steps necessary in order to increase the cost of the biodiesel production process.

According to [14], forage radish is widely used in crop rotation, thus, its rotational cultivation with soybean would provide an improvement of the soil, as it would cause an increase of nutrients that favor plant growth. Therefore, both soybean and radish could be destined to biodiesel production, leading to the productive and quality increase of the cultivars for biofuel production processes. However, due to the difficulty of achieving high transesterification yields and greater ease in the purification process, an optimization of biodiesel production should be studied and analyzed, since the results of chemical analyzes of biodiesel showed their suitability for use in engines. Thus, as there is limited previous research on the use of forage turnip for biodiesel production, this paper aims to address this issue using the turnip forage as a potential source for use in the production of biofuels and, through the evaluation of the physico-chemical characteristics of the oil extracted from the seeds, analyze and compare with other studies the stability of the biodiesel generated, and also the best way of extracting the oil from the seeds [11,13,19,20].

Therefore, the objective of the present work is to evaluate the extraction of radish seed oil, to analyze the composition of fatty acids, to quantify biodiesel production and to evaluate the use of magnesol in the purification stage, as well as the amount of glycerin obtained in the transesterification.

2. Materials and methods

The materials and methods used in the work are described in the subtopics.

2.1. Radish seeds

The radish seeds used in the experiments were provided by the Agronomic Institute of Paran á (IAPAR), Londrina/PR, Brazil.

2.2. Oil extraction

The oil extractions of the radish were performed using three different methodologies: (I) solvent extraction, (II) cold pressing and (III) seed swelling technique followed by solvent extraction.

2.2.1. Solvent extraction

In the solvent extraction, the technique was adapted from the reference [21], where the seeds are macerated, and then 20 g of seeds are weighed for extraction of the oil using soxhlet apparatus. Thus, two types of solvents—hexane and heptane—were analyzed with experiments carried out in triplicate. The amount of solvent used was 2/3 of the contents of the round bottom flask and the extraction time was 5 hours.

2.2.2. Cold pressing extraction

Approximately 2 kg of radish seed were used for cold pressing. Model MPE-40 press, ECIRTEC brand [22].

2.2.3. Extraction by technique of seed swelling

The technique of swelling the seeds with extraction solvent consists in leaving the seeds, after maceration, immersed in the solvent so that the contact with the solvent is increased before the extraction is done using soxhlet apparatus. In this context, the seeds were immersed in solvent for 24 hours prior to extraction using soxhlet apparatus. The solvent used was hexane and the experiments were run in triplicate. It is expected that the contact of the macerated seeds with the solvent can cause a swelling of the seeds and, thus, make it possible to remove the oil contained in the innermost spaces of the samples through the facilitated diffusivity of the solvent through the pores of the vegetal matrix.

2.3. Pre-treatment of oil

Two types of pre-treatments of the oil samples were analyzed, centrifugation and vacuum filtration. Centrifugation was done for 15 minutes and 5000 rpm, and vacuum filtration was done using filter paper until the complete oil filtration [23,24].

2.4. Characterization of oil

The characterization of the oil used to produce biodiesel is extreme important, since its properties affect the quality of the biodiesel produced. In this context, the composition of fatty acids was carried out.

The tests were done on samples of oil extracted by pressing and filtrate, oil extracted by pressing and centrifuged and oil extracted by solvents (crude oil).

It was used 0.3 g of each oil sample for methanolization with 6 mL of 0.5 M NaOH solution in methanol. The reaction mixture was maintained in a 50 mL reactor with boiling pellets under reflux at about 60 ℃ for 30 min. After the elapsed time, 10 mL of methanol and 1 mL of concentrated H_2SO_4 were added and again refluxed at the same temperature for 120 min. The reaction mixture was taken out of reflux and passed into a separation funnel where it was washed three times with brine to remove salts [25].

Finally, gas chromatography was performed by the SHIMADZU Gas Chromatograph GC-14B chromatograph with methyl heptadecanoate standard at a concentration of 9.8 mg/mL and column HP-CARBOWAX 30 m × 320 μm × 0.25 μm. Thus, the samples were prepared using approximately 12 mg of product (derivatized oil), 200 μL of standard solution containing 9.8 μg of methyl heptadecanoate per 1000 μL and 800 μL of heptane (solvent). The programming of the chromatograph used was:

(1) T = 160 ℃ for 0.5 min
(2) rate of increase of temperature was 40 ℃ min^{-1} up to 190 ℃
(3) 250 ℃ for 0.5 min

Then, the chromatograms for each sample of oil used were recorded, and after that 200 μL of a standard solution of oleic acid with a concentration of 9.1 mg/mL was added to be inserted back into the chromatograph [25]. This procedure has the purpose of verifying which curve of the chromatogram belongs to oleic acid and allows the calculation of its selectivity. As the chromatograph is not coupled to a mass spectrometer, the insertion of different standards, such as oleic acid, allows to define which fatty acids are present in the oil and which curve generated by them in the chromatogram.

Finally, the selectivity calculation is performed, where the fatty acid values present in each curve are presented based on the mass of the sample used, so that 200 μL of standard were used that correspond to a mass of 1.96 mg.

The standard was eluted in the column of the chromatograph in about 7 min, so the value of the area of the standard can be obtained to be used as the basis of calculation. In addition, in around 2 min the solvent used (heptane) is evaporated and, therefore, the curve around 2 min should be

disregarded in the calculations. Each curve will represent a fatty acid and thus a different selectivity, so its calculation is done as shown in Eq 1.

$$\mathrm{Mn} = (\mathrm{An} * 1.96) / \mathrm{Ap} \quad (1)$$

Where "Mn" is the mass of each fatty acid, "An" is the area at each time and "Ap" is the area belonging to the standard.

Then, by adding all calculated masses, the total mass of fatty acids can be obtained in the samples. In addition, by relating the mass of each fatty acid to the total mass, we can obtain the selectivity of the fatty acids, that is, the percentage belonging to each one in the sample.

2.5. Transesterification

In order to calculate the amounts of oil and alcohol to be used in the transesterification, the molar mass of the radish oil was considered 903.7 g mol^{-1} and specific mass of 918 kg m^{-3} [26].

Three transesterification reactions were carried out using filtered oil, all of them being used 10 mL of oil corresponding to 0.01016 mol of oil and 1% in mass in relation to the oil of the catalyst.

In the reaction with methanol the ratio of 6 mols of methanol to 1 mol of oil was used, 2.47 mL of methanol and 0.0959 g of NaOH were added. Reaction conducted for 1 h under reflux at 65 ℃.

In the reaction with ethanol the ratio of 8 mols of ethanol to 1 mol of oil was used, 4.74 mL of ethanol and 0.0944 g NaOH were added. Reaction conducted for 1 h under reflux at 85 ℃.

In the reaction of the mixture of methanol with ethanol, the proportion of 8 mols of the mixture of 60% methanol and 40% ethanol by volume to 1 mol of oil was used, with 1.5 mL of ethanol, 2.25 mL of methanol and 0.0938 g NaOH. Reaction conducted for 1 h under reflux at 65 ℃.

In addition, two reactions were performed using centrifuged oil, one using methanol and the other using ethanol. In the reaction with methanol the proportion of 6 mols of methanol to 1 mol of oil was used, 2.47 mL of methanol and 0.0956 g of NaOH were added. Reaction conducted for 1 h under reflux at 65 ℃.

In the reaction with ethanol the proportion of 8 mols of ethanol to 1 mol of oil was used, 4.74 mL of ethanol and 0.0942 g NaOH were added. Reaction conducted for 1 h under reflux at 85 ℃. At the end of the reaction with ethanol, 1 mL of glycerin was added to the reaction mixture to facilitate separating by decantation.

2.6. Separation of biodiesel

In the reactions using methanol and the mixture of methanol and ethanol (60% and 40% respectively), the separation of the biodiesel from the glycerin was done by overnight decantation, with the biodiesel being collected using a *Pasteur* pipette to be subjected to purification. In the reaction using ethanol, the reaction mixture was left decanting overnight and later submitted to centrifugation for 15 min at 5000 rpm, the supernatant (sample with absence of glycerin) subject to the purification process with magnesol.

2.7. Purification of biodiesel

Neutralization of biodiesel impurities was made by washing with magnesol using adaptations from literature [27]. Using 1 wt.% of magnesol in relation to the mass of biodiesel at the temperature of 80 ℃ for 20 min [28]. After the reaction, hot filtration was performed to remove the magnesol from the sample.

2.8. Characterization of biodiesel

The characterization of the biodiesel produced was performed through the analysis of fatty acid composition contained in the biodiesel samples.

2.9. Gas chromatography

Gas chromatography was performed by the SHIMADZU Gas Chromatograph GC-14B chromatograph with methyl heptadecanoate standard at a concentration of 9.8 mg/mL for analysis of fatty acid composition. Samples were prepared using approximately 15 mg of product (biodiesel), 200 μL of standard solution containing 9.8 μg of methyl heptadecanoate per 1000 μL and 1 mL of heptane (solvent). The chromatograms, according to annexes, present the table containing the area (A) of each peak in the graph, with the peaks representing the fatty acids that make up the biodiesel. Therefore, adding up all the peaks will have the amount of biodiesel produced. In the chromatograms, the time peak of 2 minutes belongs to the solvent used in the analysis and its area should be disregarded for calculations. In addition, the peak around 7 min should also be disregarded because it belongs to the standard used in the preparation of samples.

After calculating the total area to be considered, the amount of standard should be found according to Eq 2, knowing that the standard concentration was 1.96 mg/mL.

$$X = (AT * \text{standard concentration}) \, / AT7 \tag{2}$$

Thus, the value of X represents the amount of fatty acid esters based on the standard used. Thus, by using the amount of sample for analysis in mg and the quantity X one can calculate, by the ratio, the yield of the transesterification.

3. Results and discussion

3.1. Oil extraction from radish seeds

Table 1 presents the extraction yields of oil from radish seeds based on each method.

Yields of extraction processes using different methods presented a high statistic difference. Extraction by swelling technique presented the highest amount of oil extracted from radish seeds, 35.13%, indicating that contact between seeds and solvent for a period of 24 h before the extraction process enabled an interaction that led to a better extraction yield. Solvent extraction showed to be more efficient when using hexane than when applying heptane, because the extraction applying

hexane as solvent enables a higher efficiency, up to 35% [29]. Thus, apolar or low polar solvents have a greater efficiency in the extraction process of radish oil, since the yield depends on the interactions between the solute and the solvent and the fatty acids present in the sample. In addition, the solvent viscosity also affects the extraction because solvents with low viscosity present high diffusivity, facilitating the diffusion within the pores of the vegetal matrix [30,31].

Table 1. Extraction processes yields including mean, standard deviation and Tukey's test significant at 5%.

Samples	Extraction yield (wt.%)	Mean	Standard deviation
Hexane extraction	17.89	20.13[a]	2.8312
	19.18		
	23.31		
Heptane extraction	14.37	13.83[b]	1.8404
	11.78		
	15.34		
Swelling technique	38.44	35.13[c]	2.9590
	34.21		
	32.74		
Cold pressed extraction	8.26	8.26[d]	-

Cold pressed extraction had a very low yield, 8.26%, when compared to literature's study which reached around 70% of extraction yield [32]. However, in that study, a double pressing technique was accomplished, which leads to a higher efficiency. Thus, it is important to highlight that the extraction process depends on grains maceration, so that bigger grains will result in lower oil extraction, and, many techniques can be applied to optimize the process and reduce costs. Therefore, it should be remembered that the extraction process is influenced by the extraction time, moisture, amount of sample, grain maceration and extraction temperature, since the temperature affects the solubility of the oil and, thus, the extraction speed [21,33,34].

3.2. Fatty acids composition

Fatty acids composition provides relevant information to evaluate biodiesel physical chemical characteristics, such as flash point, viscosity and oxidative stability. Thus, fatty acids composition of radish oil is presented on Table 2.

It is observed that in all the evaluated samples and consulted bibliography, there is a high percentage, varying between 29.1% and 34.5% of oleic acid, a monounsaturated fatty acid that enables the production of a biodiesel presenting a higher oxidative stability. Besides that, polyunsaturated fatty acids as linoleic and linolenic acid are present, but, such acids presented a high variation range in the different presented studies, so that they may be present in a small amount in the oil. The biodiesel produced from soy has around 24% monounsaturated and 60% polyunsaturated acids, there is a presence of about 15% of erucic acid, indicating that the present work can present a large amount of this acid, which is verified in the chromatographic analysis as a combination of

behenic and erucic acid. Polyunsaturated acids make biodiesel oxidation easier and this problem does not occur on biodiesel produced from radish oil due to the high presence of monounsaturated fatty acids [35]. A typical fatty acid found in radish oil is erucic acid. This substance is considered toxic, thus, when it is present in vegetable oils, it makes them inappropriate for human consumption, allowing it to be used in the production of biofuels, not interfering with the raw material of the food industry [36]. However, erucic acid still has several utilities such as good drying ability and polymerization ability that allows its use as binders, as an industrial lubricant, corrosion inhibitor and as an ingredient in the manufacture of synthetic rubber. In addition, it can be used in the production of plastic films, nylon, plasticizers, adhesives, thermal insulation and the production of cosmetics [37,38].

Table 2. Fatty acids composition of radish oil.

Fatty acid	Centrifuged oil (CO) (%)	Filtered oil (FO) (%)	Solvent extracted oil (SEO) (%)	Literature (%) [11,34]
Myristic (C14:0)	0	0	0	6
Palmitic (C16:0)	3.9	4.5	3.5	5.01–8.3
Stearic (C18:0)	0	0	0	0–3.1
Vaccenic (C18:1 cis 9)	0	0	0	0–1.4
Oleic(C18:1)	31.1	29.5	31.1	29.1–34.5
Linoleic(C18:2)	7.7	10.6	9.2	7.6–19.1
Linolenic (C18:3)	3.5	7	6	4.6–13.2
Arachidic (C20:0)	9	9.2	7.8	0.8–8.2
Gadoleic (C20:1)	0	0	0	7.9–11.2
Behenic (C22:0)	44.6	39.2	42.4	0–14.1
Erucic (C22:1)				1.2–33.3

Behenic and erucic acids were not differentiated in the chromatogram and, therefore, the data presented are a combination of the amount of the two acids, this fact may have happened due to restriction time used in the chromatographer which was 22.5 minutes, that is, maybe a larger retention time was needed to separate these two acids. Polyunsaturated acids and a decrease in saturated fatty acids make biodiesel more susceptible to oxidation, so the biodiesel from radish oil can has higher oxidative stability when compared to soy [39]. Nonetheless, long chain and/or saturated character fatty acids enable a higher cloud point, cold filter plugging point and pour point. These factors may be considered because they influence the temperature in which biodiesel can be used [40]. In the search for biodiesel of ideal composition for better engine performance, a vegetable oil with a high content of monounsaturated fatty acids (oleic and palmitoleic acid), moderate content of saturated acids and a reduction of polyunsaturated fatty acids (linoleic acid) is required since a high number of unsaturations can cause several engine drawbacks such as: oxidation, degradation and polymerization of fuels, causing the storage and transport of the fuel to be affected due to a greater formation of solid wastes [41,42].

The presence of monounsaturated fatty acids improves oxidative stability and fuel performance at low temperatures, and saturated fatty acids increase cetane number and biodiesel stability [41–44].

Thus, the presence of approximately 30% of monounsaturated (oleic and gadoleic acid), about 16% of polyunsaturated (linoleic and linolenic acid), 15% of saturates (myristic, palmitic, stearic and arachidic acid) and around 50% of compounds with a chain of up to 18 carbons make forage radish oil suitable for use in engines, in order to obtain a high combustion efficiency [35,40,41]. Recalling that it is not possible to obtain accurate counting of the saturated and monounsaturated compounds, since behenic (saturated) acid is presented in combination with erucic acid (monounsaturated acid), the selectivity of the two acids being approximately 42% based on the literature [35], we can consider the presence of erucic acid being 15% and behenic 27%. Therefore, the monounsaturated would add 45% and the saturated ones 42%, indicating a balance between the two groups of fatty acids.

3.3. Biodiesel production and purification processes

Biodiesel production presented around 10% of glycerin and 90% of biodiesel after decantation process. Amounts of biodiesel and glycerin produced, as well as amounts of magnesol used in the purification process can be seen on Table 3.

Table 3. Amounts of biodiesel and glycerin obtained through transesterification reaction and amounts of magnesol used in purification.

	Biodiesel (g)	Glycerin (g)	Magnesol (g)	% Glycerin	% Biodiesel
SMPF	6.4298	0.6853	0.0614	9.6316	90.3683
SMPC	5.8371	0.6288	0.5841	9.7248	90.2751
SEPF	10.7399	1.2233	0.1246	10.2255	89.7744
SEPC	4.6515	0.6439	0.4646	12.1596	87.8403
SSPF	4.5662	0.5831	0.0454	11.3238	88.6761

The use of magnesol enables removal of solid particles by adsorption. If not removed, these solid particles can affect biodiesel physical and chemical properties, so that, purification is required to biofuel presents a higher stability and appropriate to be used in automobiles [45–47]. Besides that, glycerin content in biodiesel production is very important, because high concentrations of glycerol can cause biodiesel storage problems because when stored together with diesel it is possible to verify the separation of the glycerin in the tanks where the fuel is stored, in addition, problems such as the formation of deposits, nozzle clogging and the emission of aldehydes are also related to the presence of glycerin in biodiesel [45,46]. Glycerin is a co-product present in the transesterification reaction of oils and fats and its determination evaluates the biodiesel purification, and the lower the glycerin presence, the better the efficiency of the transesterification process [43,45]. However, this co-product is undesirable, although it has a high commercial value, given its wide industrial application (mainly in the manufacture of cosmetics) [45,47–49]. Although glycerin is not soluble in biodiesel, it can be present in droplets, which complicates its full separation. In the United States, Europe and Brazil, accepted maximum limit of free glycerin by regulations is 0.02%, while limit of total glycerin in Brazilian regulations is 0.25%, indicating that biodiesel from radish seed oil should go through more

washings to reduce its glycerin content, so that a pure and more applicable product can be obtained [11,15].

In transesterification reaction, influence of different types of alcohols and different methods of oil extraction and purification in reaction yield were evaluated. Sodium hydroxide was the catalyst applied, because although it leads to lower yields, when compared to potassium hydroxide, it has a lower cost. Yield results of transesterification reaction were calculated based on chromatograms presented for each reaction sample (Table 4).

Table 4. Yields of biodiesel production as a function of different alcohols and pretreated oils used in transesterification reactions.

Reaction mixtures	% Yield
SMPF*	86
SMPC*	45
SEPF*	15
SEPC*	5
SSPF*	60.8

Methanol and vacuum filtration pretreated oil yielded the highest conversion of triglycerides in fatty acids methyl esters. This process reached a yield of about 86%, which is still not appropriate for biodiesel commercialization, once yield should be more than 95% [50]. Despite of that biodiesel production through methyl route is easier due to methanol reactivity, so that this route is always better than the ethanol one, both in reaction velocity and efficiency [51].

Methanol and oil pretreated through centrifugation yielded about 45% in the reaction process, so, since conditions were the same, centrifuged oil caused a huge reduction in conversion rate. This result indicates that this pretreatment was not efficient, because it reduced conversion rate in methyl esters, proving that the quality of oil used in biodiesel production is very important [3].

Regarding ethanol and filtrated oil, yield obtained was 15%, which indicates a very low conversion of triglycerides into fatty acids ethylic esters. Such fact may have happened due to the higher number of carbon in the alcohol, which affects catalyst efficiency. However, there is also the possibility of moisture presence in biodiesel, which would lead to soap formation, reducing reaction conversion. Therefore, several factors can influence triglycerides conversion in methyl or ethylic esters, such as amount of catalyst employed, molar ratio of alcohol to oil, reaction temperature, among others.

4. Conclusion

Cold pressed extraction is the most simple and quick technique evaluated, but a specific equipment is required. Besides that, extraction yield was very low (8.26%), so that, a large amount of oil remained in the cake after pressing procedure was still observed.

Solvent extraction techniques presented higher yields using hexane (20%) and lower yields for heptane (13%), so, hexane is the best solvent for extraction due to best yield results.

Seeds wetting technique presented a high extraction yield (34%) anddespite of high solvent contact time (24 h) and extraction time (5 h), it presents a high industrial applicability potential. Nonetheless, whole energetic consumption calculation needs to be accomplished to evaluate feasibility of applying this technique.

Regarding fatty acids present in radish oils samples, a large amount of oleic acid is present (up to 32%), a monounsaturated molecule that enables a better biodiesel production with a higher heating value. Thus, the presence of approximately 30% of monounsaturated (oleic and gadoleic acid), about 16% of polyunsaturated (linoleic and linolenic acid), 15% of saturates (myristic, palmitic, stearic and arachidic acid) and around 50% of compounds with a chain of up to 18 carbons make forage radish oil suitable for use in engines, in order to obtain a high combustion efficiency.

Erucic acid presence (up to 41%) contributes to radish oil application in the energetic industry, since this acid is considered toxic to food industry.

Biodiesel production resulted in about 90% biodiesel and 10% glycerin after decantation process and after purification process, it was observed an emulsification in the biodiesel produced using ethanol.

Methanol and vacuum filtration pretreated oil yielded the highest conversion rate (86%) of triglycerides in fatty acids methyl esters.

Finally, radish is not a well-known crop and it needs to be studied aiming its energetic application, not only for biodiesel, but also for bioethanol production. Its resistance to pests and diseases, its development in cold weather and insertion of nutrients on soil allow its consortium with soy, favoring an increase in biodiesel production if these cropswere cultivated together.

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Conflict of interest

All authors declare no conflicts of interest in this paper.

References

1. Santos F, Queiroz JH, Colodette J, et al. (2013) Produção de etanol celulósico a partir da cana-de-açúcar, In: Santos F, Colodette J, Queiroz JH (Eds) *Bioenergia e Biorrefinaria*: Cana-de-Açúcar e Espécies Florestais. Viçosa, 129–164.
2. Vaz JS (2011) Biorrefinarias: cenários e perspectivas. Brasília: Athalaia. Available from: http://www.bibliotecaflorestal.ufv.br/bitstream/handle/123456789/10778/EMBRAPA_Biorrefinarias-Cen%E1rios-e-Perspectivas.pdf?sequence=1.
3. Ferrari RA, Oliveira VDS, Scabio A (2005) Biodiesel de soja: taxa de conversão em ésteres etílicos, caracterização físico-químicas e consumo em geradores de energia. *Química Nova* 28: 19–23.

4. Lotero E, Liu Y, Lopez DE, et al. (2005) Synthesis of biodiesel via acid catalysis. *Ind Eng Chem Res* 44: 5353–5363.
5. EryiLmaz T, YesiLyurt MK, Cesur C, et al. (2016) Biodiesel production potential from oil seeds in Turkey. *Renew Sust Energ Rev* 58: 842–851.
6. BRASIL (2009) Ministério de minas e energia. Boletim Mensal dos Combustíveis Renováveis. Available from: http://www.mme.gov.br.
7. Wang YP, Tang JS, Chu CQ, et al. (2000) A preliminary study on the introduction and cultivation of *Crambe abyssinica* in China, an oil plant for industrial uses. *Ind Crop Prod* 12: 47–52.
8. Embrapa. Nabo forrageiro. 2018. Availble from: http://www.agencia.cnptia.embrapa.br/gestor/agroenergia/arvore/CONT000fbl23vn002wx5eo0sawqe38tspejq.html.
9. Kaymak HC (2015) Profile of (n-9) and (n-7) Isomers of Monounsaturated Fatty Acids of Radish (*Raphanus sativus* L.) seeds. *J Am Oil Chem Soc* 92: 345–351.
10. Silvestre WP, Pauletti GF, Godinho M, et al. (2017) Fodder radish seed cake pyrolysis for bio-oil production in a rotary kiln reactor. *Chem Eng Processing: Process Intensification*. Available from: https://doi.org/10.1016/j.cep.2017.12.020.
11. Ávila RNA, Sodré JR (2012) Physical–chemical properties and thermal behavior of fodder radish crude oil and biodiesel. *Ind Crop Prod* 38: 54–57.
12. Bridgwater AV (2012) Review of fast pyrolysis of biomass and product upgrading. *Biomass Bioenerg* 38: 68–94.
13. Chammoun N, Geller DP, Das KC (2013) Fuel properties, performance testing and economic feasibility of *Raphanus sativus* (oil seed radish) biodiesel. *Ind Crop Prod* 45: 155–159.
14. Santos F, Brasil LMM, Faria DJ, et al. (2018) Aproveitamento integral do nabo forrageiro (*Raphanus Sativus L.*) em processos de biorrefinaria. *Engevista* 20.
15. Souza ADV, Fávaro SP, Ítavo LCV, et al. (2010) Caracterização química de sementes e tortas de pinhão-manso, nabo-forrageiro e crambe. *Pesqui Agropecu Bras* 44: 1328–1335.
16. Reddy PP (2016) Cover/Green Manure Crops. In: *Sustainable Intensification of Crop Production*, Springer, Singapore, 55–67. Available from: https://doi.org/10.1007/978-981-10-4325_7.
17. Silva LFL (2013) Viabilidade econômica das culturas de canola, nabo forrageiro e batata doce para a produção de biocombustíveis no sul de Minas Gerais. Masters Dissertation, Postgraduate Program in Agronomy, Lavras-MG, Brazil.
18. Nicolosi G, Drago C, Liotta LF, et al. (2018) U.S. Patent No. 9873843. Washington, DC: U.S. Patent and Trademark Office.
19. Indrawan N, Thapa S, Rahman SF, et al. (2017) Palm biodiesel prospect in the Indonesian power sector. *Environ Technol Innov* 7: 110–127.
20. Soares CM, Itavo LCV, Dias AM, et al. (2010) Forage turnip, sunflower, and soybean biodiesel obtained by ethanol synthesis: Production protocols and thermal behavior. *Fuel* 89: 3725–3729.
21. Machado L, Nascimento R, Rosa G (2015) Estudo da extração de óleo essencial e de compostos bioativos das folhas de eucalipto (*Eucalyptus citriodora*). *Blucher Chem Eng Proc* 1: 5609–5616.

22. Mendonça AP (2015) Secagem e extração do óleo das sementes de andiroba (*Carapa surinamensismiq.* e *Carapa guianensisaubl.*) National Institute of Amazonian Research.
23. Berni JV, Dolfini N, Medeiros JF, et al. (2015) Análise comparativa de métodos de purificação de ésteres etílicos da blenda de sebo bovino e óleo de fritura. *Blucher Chem Eng Proc* 2: 930–937.
24. Medeiros JF, Pasa TLB, de Almeida FNC, et al. (2014) Revista brasileira de energias renováveis. *Revista Brasileira de Energias Renováveis* 3: 235–242.
25. Abreu FR, Lima DG, Hamú EH, et al. (2004) Utilization of metal complexes as catalysts in the transesterification of Brazilian vegetable oils with different alcohols. *J Mol Catal A-Chem* 209: 29–33.
26. Pagnoncelli M, Toss D, Godinho M, et al. (2014) Perfil dos ésteres obtidos pela transesterificação etanólica e metanólica do óleo de nabo forrageiro. XX Congresso Brasileiro de Engenharia Química. Florianópolis.
27. Atadashi IM, Aroua MK, Abdul AAR, et al. (2011) Refining technologies for the purification of crude biodiesel. *Appl Energ* 88: 4239–4251.
28. Faccini, Schmittcunha C, Damoraes ME, et al. (2011) Dry washing in biodiesel purification: a comparative study of adsorbents. *J Braz Chem Soc* 22: 558–563.
29. Kumar D, Singh B, Banerjee A, et al. (2018) Cement wastes as transesterification catalysts for the production of biodiesel from Karanja oil. *J Clean Prod* 183: 26–34.
30. Rezaie M, Farhoosh R, Iranshahi M, et al. (2015) Ultrasonic-assisted extraction of antioxidative compounds from Bene (Pistacia atlantica subsp. mutica) hullusing various solvents of different physicochemical properties. *Food Chem* 173: 577–583.
31. Almeida PP, Mezzomo N, Ferreira SRS (2012) Extraction of *Menthaspicata L.* volatile compounds: evaluation of process parameters and extract composition. *Food Bioprocess Tech* 5: 548–559.
32. Bressan RT (2018) Análises físico-químicas e otimização da prensagem de crambe em extrusora mecânica sob variações de temperatura e umidade relativa dos grãos. State University of Western Parana.
33. Tomishima K, Teixeira L, Carvalho T, et al. (2015) Avaliação do desempenho da extração líquido-líquido na desacidificação do óleo de coco para produção de biodiesel. *Blucher Chem Eng Proc* 1: 8843–8850.
34. Valle PW, Rezende TF, Souza RA, et al. (2009) Combination of fractional factorial and Doehlert experimental designs in biodiesel production: Ethanolysis of *Raphanus sativus L.* var. oleiferus stokes oil catalyzed by sodium ethoxide. *Energ Fuel* 23: 5219–5227.
35. Shah SN, Iha OK, Alves FCSC, et al. (2013) Potential application of turnip oil (*Raphanus sativus L.*) for biodiesel production: Physical chemical properties of neat oil, biofuels and their blends with ultra-low sulphur diesel (ULSD). *Bioenerg Res* 6: 841–850.
36. Wendlinger C, Hammann S, Vetter W (2014) Various concentrations of erucic acid in mustard oil and mustard. *Food Chem* 153: 393–397.
37. Rezende RKS, Marques RF, Masetto TE (2015) Características morfológicas e produtividade do crambe em função da adubação nitrogenada. *Agrarian* 8: 279–286.

38. Kiihl TAM, Tomm GO (2017) Banco de Germoplasma de Canola da Embrapa: conservação e multiplicação de acessos. In: *Embrapa Trigo-Artigo em anais de congresso (ALICE)*. In: SIMPÓSIO BRASILEIRO DE CANOLA, Passo Fundo. Anais... Brasília, DF: Embrapa, 2017. Available from: https://www.alice.cnptia.embrapa.br/bitstream/doc/1084148/1/CNPTID 44265.pdf.
39. Constantino AF, Lacerda JV, Santos RBD, et al. (2014) Analysis of oil content and oil quality in oil seeds by low-field nmr. *Química Nova* 37: 10–17.
40. Liu T, Jiaqiang E, Yang WM, et al. (2018) Investigation on the applicability for reaction rates adjustment of the optimized biodiesel skeletal mechanism. *Energy* 150: 1031–1038.
41. Gouveia L, Oliveira AC, Congestri R, et al. (2018) Biodiesel from microalgae. *Microalgae-Based Biofuel Bioprod*, 235–258.
42. Ashraful AM, Masjuki HH, Kalam MA, et al. (2014) Production and comparison of fuel properties, engine performance, and emission characteristics of biodiesel from various non-edible vegetable oils: A review. *Energ Convers Manage* 80: 202–228.
43. Pinzi S, Garcia IL, Lopez-Gimenez FJ, et al. (2009) The ideal vegetable oil-based biodiesel composition: a review of social, economical and technical implications. *Energ Fuel* 23: 2325–2341.
44. Ramos MJ, Fernández CM, Casas A, et al. (2009) Influence of fatty acid composition of raw materials on biodiesel properties. *Bioresource Technol* 100: 261–268.
45. Lace VO, Fraga IM, Fernandez JRC, et al. (2018) Obtenção do biodiesel metílico através da transesterificação via catálise básica do óleo de coco-da-baía (*Cocos nucifera* L). *Eclética Química J* 39: 192–199.
46. Heiden RW, Schober S, Mittelbach M (2017) Bias and imprecision in the determination of free glycerin in biodiesel: The unexpected role of limitations in solubility. *J Am Oil Chem Soc* 94: 285–299.
47. Bordignon G, Lima HJD, Martins RA, et al. (2017) Níveis de glicerina bruta na ração de suínos nas fases de crescimento e terminação. *Archivos de zootecnia* 66: 429.
48. Alves MJ, Cavalcanti IV, Resende MMD, et al. (2016) Biodiesel dry purification with sugarcane bagasse. *Ind Crop Prod* 89: 119–127.
49. Khalid A, Tamaldin N, Jaat M, et al. (2013) Impacts of biodiesel storage duration on fuel properties and emissions. *Procedia Eng* 68: 225–230.
50. Lôbo IP, Ferreira SLC, Cruz RSD (2009) Biodiesel: quality parameters and analytical methods. *Química Nova* 32: 1596–1608.
51. Joshi G, Pandey JK, Rana S, et al. (2017) Challenges and opportunities for the application of biofuel. *Renew Sust Energ Rev* 79: 850–866.