METHYLIC AND ETHYLIC BIODIESEL FUEL: METHANOLYSIS AND ETHANOLYSIS OF PULP OF PEQUI (Caryocar brasiliensis Camb) FRUIT IN NATURA CATALISED BY BASE IN A PROCESS IRRADIATED BY MICROWAVE

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METHYLIC AND ETHYLIC BIODIESEL FUEL: METHANOLYSIS AND ETHANOLYSIS OF PULP OF PEQUI (Caryocar brasiliensis Camb) FRUIT IN NATURA CATALISED BY BASE IN A PROCESS IRRADIATED BY MICROWAVE

Abstract - In this work, a synthetic route for methylic and ethylic biodiesel fuel production is reported. Biodiesel was produced by the transesterification “in natura” of pilp of pequi fruit containing vegetable oil with methanol and ethanol, using NaOH as catalyst. However, the method described here for the methylc and ethylc biodiesel fuel production, uses methanol and ethanol and employs directly the fruits, eliminating the vegetable oil chemical extraction step that usually employs in a process accelerated by irradiated by microwave (MW).

Palavras chaves: Biodiesel fuel, Microwave irradiation, Methanolysis, Ethanolysis, Pulp of pequi fruit, In natura.

1. Introduction

Pequi is a tree of the family of caryocaraceae (Caryocar brasiliensis Camb) typical of the region of the Brazilian that arrives to range up to 10 meters of height with a trunk that can arrive the 40 cm of diameter, presenting a greasy fruit with orange pulp and prickly internal wall. Recent studies had demonstrated to the yield gotten for the extration of oils and fats of the fruit of pequi, through the chemical extration (through solvent organic) of the oil of pequi (Cariocar brasiliensis Camb). The extration of the oil from mesocarpo internal (pulp of the fruit) relieved 51,9% m/m of oil of pequi, the constituent of this oil had been determined by chromatogy analysis of the FAMEs, where the following acid main ratios of the esters of fatty acid had been found: fatty acid oleic 49,7%, fatty acid palmitic 37,5% and also possesss 0,9% of esters of essences fatty acid (fatty acid ester linoleic and fatty acid ester linolenic). In a generalized manner the pulp of the fruit of pequi, possesss 56,6% in m/m of insaturates fatty acid ester ones 43,4% in m/m of saturated fatty acid esters. Already in the extration of the oil of the almond of pequi, 58,6% in m/m of oil of pequi had been gotten, where the following fatty acid esters main ratios of the greasy ones had been found: 41,1% of esters of fatty acid palmitic, 54,0% of esters of fatty acid oleic, moreover the oil of the almond of pequi possesss 7,1% of essentials esters of fatty acid (linoleic acid and acid linolenic). The oil of the almond of pequi is constituted of 58.0% in m/m of esters of insaturates fatty acid and 42.0% in m/m of esters of saturated fatty acid.

In the recent years biodiesel fuel (common name given to the alkyl esters of fatty acids) that is a part of family of biofuels and is a term used to define a fuel used in lieu of diesel fuel that is produced from vegetable oil; “bio” describes its renewability and biodegrability and “diesel” reflects its use as fuel for diesel engines), and has the following characteristics: it is virtually free of sulfur and aromatic compounds, has a cetane number equivalent to or higher than that of fossil diesel fuel, has an approximate average oxygen content of 11%, and has similar viscosity as, and a higher flash point than conventional fossil fuel, gained an important position as an environmentally friendly substitute of fossil diesel.

Biodiesel can be used pure or blended with regular diesel fuel. ASTM international defines a biodiesel bend as: “a blend of biodiesel fuel with petroleum-based diesel fuel.” Biodiesel blends are often designated with the abbreviation BXX, where XX represents the vlume (in percent) of biodiesel fuel in the blend. Thus, a blend of 70% diesel fuel
and 30% biodiesel is designated as B30, already, a fuel consisting of pure biodiesel is designated as B100.

However, it history have been started in 1893, when Rudolf Christian Carl Diesel obtained the patent for his internal combustion engine, now know as the diesel engine, the fuel initially utilized was petroleum. However, in 1900, when he introduced his internal combustion engine at the Exposition Universelle (Work Exibition) in Paris, France, he used vegetable oil (peanut oil) as fuel. Rudolf Diesel was able to demonstrate that this his engine ran perfectly well on vegetable oils. The idea behind that was to allow farmers to produce their own fuel to run their “diesel” engines, but due to economic reasons, mainly the high cost and minor availability compared to the petroleum derivatives vegetable oils were completely abandoned as fuels. The difference between fossil diesel and vegetable oil is mainly in the molecular weight of these two compounds, in the density of the vegetable oil compared to the mineral product that can cause engine operation failures and also on the viscosity of vegetable oils that is 10-20 times higher than of petroleum fuel, therefore using directly vegetable oils as a fuel can cause engine problems like injector fouling and particle agglomeration. Fossil diesel consists of hydrocarbons with an average number of 14 carbons, whereas vegetable oils consist of mixtures of triglyceride molecules; its is the esters of a tri-alcohol (glycerol) with three different fatty acids. Moreover, the molecular weight of vegetable oils is approximately three times higher than that of fossil fuel. All these problems can be overcome by the transesterification of the vegetable oils. The transesterification of vegetable oils such as soybean oil, sunflower, animal fats or other triglycerides with monohydric alcohol such as ethanol or ethanol affords a mixture of methyl or ethyl alkyl esters of the corresponding long chain fatty acids, and discharging glycerol as a co-product. These esters have a significant potential as alternative to diesel fuel since their characteristics are almost the same.

Among the techniques that have been applied for the supply of energy in the production of biodiesel, we can detach the application of ultrasonic irradiation, but the area of research of interest to us is the use of controlled microwave heating for accelerating synthetic organic transformating. Using a domestic microwave apparatus, it is possible perform reactions quickly, efficiently, and safely. While many of the publications in the literature focus on the use of sealed vessels for performing chemistry using microwave heating, it is also possible to use an open-vessel apparatus. Because the preparation methyl and ethyl biodiesel fuel from oil or fruit or seeds and methanol and ethanol is a simple transesterification, we wanted to see whether, using a comemercially available domestic microwave apparatus, it was possible to prepare biodiesel fuel rapidly and good conversations. Microwave heating is more efficient than conventional convection heating and thus would offer a resourceful way to prepare biodiesel. In addition, it may be possible to take advantage of the great rate accelerations seen in microwave-promoved chemistry to reduce the time for the biodiesel production process. The processes found in literature describe the production of biodiesel from vegetable oil and alcohol (methanol or ethanol) utilizing the irradiation for microwave as heating source, our work will be show that by using microwave irradiation the extraction of oil problems could be overcome. With this work we research group expose the possibility to use microwave irradiation for industrial manufacture of biodiesel produced from pulp of pequi fruit in natura. Since the fruit of pequi in natura used as raw material...
for production of biodiesel have snags in excess, causing problems in the extraction of the vegetable oil.

2. Experimental

2.1. Chemicals
All reagents were used as purchased; \(^1\)H and \(^13\)C NMR data were obtained in CDCl\(_3\) using internal TMS as standard, using a Advanced Brucker 400 spectrometer; mass spectroscopic analysis were performed with a Hewlett-Packard CG/MS 5988A system; gas chromatograms were obtained using a 5890 Genes II gas chromatograph and IR data obtained with a Perkin Elmer Spectrum RX spectrometer operating in the FT mode.

2.2. Production of ethylic biodiesel fuel using pulp of pequi fruit (mesocarp internal) in natura and NaOH as catalyst in a process irradiated for microwave.

All reactions were performed under atmospheric pressure in the presence of air. The mature fruits of pequi (Cariocar brasiliensis Camb) had been collected in a population of native plants proceeding from the city of Diamantina/MG in the state of Minas Gerais in the Brazil, these fruits later had been cut to the way, breaking up themselves in three distinct parts: external mesocarp, internal mesocarp (pulp of the fruit) and almond. Of these 1.0 kg of mesocarp internal was submitted to the process of drying in a microwave oven during 2 min in 340 W and 70\(^\circ\) C, after this period the dehydrated pulp was cooled until the room temperature, being after that completely worn out with the aid of a triturador of electric and stored vegetable in a bottle. In erlenmeyer (250 mL) containing 150 mL of ethanol, were added 1.0000 g NaOH, previously the mixture was agitated during 20 min the room temperature, after the a mixture was transfered to a in becker contends 100,0000 g of dehydrated and triturated pulp of pequi fruit (mesocarp internal). The reaction mixture was placed into the domestic oven of microwave of mark BRASTEMP model BMK 38ABHNA, series MD3144902 of 38 Lts and 1,7 KW. The reaction mixture was heated from room temperature to 50\(^\circ\) C using an initial microwave of 340 W (P2 or 20%) and then held at this temperature for 1 min. Upon cooling to room temperature, the reaction vessel was removed from the microwave cavity and the contents were filtered under vacuum and the pulp of pequi fruit was washed 3 times with 3 portion of 50 mL of dry EtOH, the solution was evaporated in a rotatory evaporator, in order to eliminate the excess of etanol used. The mixture was transferred to a separation funnel, where the separation of biodiesel formed and the glycerin (by-product of the reaction process) occurred for decantation. After the phases separation the traces of catalyst and ethanol were washed out with water from the esters mixture until the water layer remain completely translucent. Finally, the product was dried on anhydrous calcium. The mass of ethylic biodiesel fuel obtained was of 30.0000 (%) g and glycerin of 4.2000 g, for calculates it of the income of the process, takes in consideration the ratio of oil in mesocarpo internal (pulp of the fruit), which is of 51.9%. \(^8\) Of this form the amount of oil that contained in the 63.0000 g of pulp used in the reaction was of 30.2400 g. The reaction process was accomplished by chromatography in thin layer, where a mixture containing hexane and ethyl acetate in the ratio of 9:1 was used as eluente. The extent of the
transesterification of pulp of pequi fruit was also determined by solution $^1$H NMR spectroscopy. The relevant signals chosen for integration were those of etoxy groups in the ethylic biodiesel fuel (1.64 ppm, triplet and 2.00 ppm quartet) and those of the $\alpha$-methylene protons present in all triglyceride derivates (2.3 ppm, triplet) of pulp pequi fruit. The conversion also was calculated directly from the integrated areas of these signals.

2.3. Production of Methylic biodiesel fuel using pulp (mesocarp internal) of the pequi fruit in natura and NaOH as catalyst in a process radiated for microwave.

In a erlenmeyer (250 mL) contaning 156 mL of methanol, had been added 0.7000 g NaOH, the mixture was agitated during 20 minutes the room temperature, after that the methoxide was transfered the a becker containing 100,0000 g of pulp (mesocarp internal) of pequi fruit previously dehydrated and triturated. The reacional mixture was placed into domestic microwaves oven of mark BRASTEMP model BMK 38ABHNA, series MD3144902 of 38 Lts and 1,7 KW. The reaction mixture was heated from room temperature to 50º C using an initial microwave of 340 W (P2 or 20%) and then held at this temperature for 1 min. Upon cooling to room temperature, the reaction vessel was removed from the microwave cavity and the contents were filtered under vacuum and the pulp of pequi fruit was washed 3 times with 3 portion of 50 mL of MeOH, the solution was evaporated in a rotatory evaporator, in order to eliminate the excess of metanol used. By decantation of mixture occured the separation of the methylic biodiesel fuel (high phase) formed and the glycerin (by-product/lower phase). After the phases separation the traces of catalyst and ethanol were washed out with water from the esters mixture until the water layer remain completely translucent. Finally, the product was dried on anhydrous calcium. The mass of methylic biodiesel obtained was of 30.0000 g (%) and of glycerin 4.2000 g, the reation was accomplished by thin layer chromatography (TLC), where the mixture of acetate of etila and hexane in the ratio of 9:1 were used as eluente. The extent of the transesterification of pulp of pequi fruit was also determined by solution $^1$H NMR spectroscopy. The relevant signals chosen for integration were those of metoxy groups in the FAME (3.66 ppm, singlet) and those of the $\alpha$-methylene prótons present in all triglyceride derivate (2.3 ppm, triplet) of pulp pequi fruit. The conversion also was calculated directly from the integrated areas of these signals.

3. Results and Discussion

Considering it necessity of search for new sources of vegetal oils and fats and the importance of one politics of development supported in the inserted states in the Brazilian open pasture, we look for in this work to evaluate the potential of the fruit of pequi (Caryocar brasiliensis Camb.) as substance cousin in the production of Biodiesel. One of the main problems to use pequi (Caryocar brasiliensis Camb) as vegetable oil source is without doubts none, the great amount of very small thorns that the fruit possesss in its internal wall, what it makes it difficult in surplus its manuscript of the fruit, as much for the process of chemical oil extration (through the use of solvent organic) as, mainly for the mechanical process of extration, which would be of utmost importance for the
use of pequi as substance cousin in the production of biodiesel. Ahead of this problem, our group of research decided to use the fruit in natura for the production of biodiesel, that is, we would not extract the vegetal oil of the fruit, but yes we would direct get biodiesel of the pulp of the fruit in a process of transesterification catalyzed for alcalys and irradiated by a domestic microwaves oven. Directly to test the reaction of transesterification of the pulp of the fruit of pequi, in them we take in consideration the data found in literature, citing that in the pulp of the fruit of pequi we find around 51.9% of triacyl glycerol. Our investigations using pulp of pequi fruit as a substrate and potassium hydroxide as a base, we wanted to look at the effects of the irradiation microwave, catalyst loading, and reaction temperature and time on the transesterification of pulp of pequi fruit with ethanol to yield ethyl biodiesel fuel. Initial screening reactions were performed using a reaction process without none heating, only with mecanic agitation in room temperature. We predissolve 2.1900 g of NaOH in 31.50 mL of ethanol, the contents of the vessel were stirred either by means of a rotating magnetic for 20 min. The reaction mixture was placed into the becker (500 mL) containing 63.0000 g pulp of the fruit of pequi previously dehydrates and triturated, what theoretically it represents 32.0000 g of vegetable oil. The reaction mixture was agited by means of a rotating mecanic for 30 min and after the contents were filtered under vacuum and evaporated in a rotatory evaporator, in order to eliminate the excess of ethanol used. The reaction mixture was transferred to a separation funnel, where the separation of biodiesel formed and the glycerin (by-product of the reaction process) occurred for decantation. After the phases separation the traces of catalyst and ethanol were washed out with water from the esters mixture until the water layer remain completely translucent. Finally, the product was dried on anhydrous calcium. The mass of ethylic biodiesel fuel obtained was of 30.0000 (%) g and glycerin of 4.2000 g, for calculates it of the yield of the process, takes in consideration the ratio of oil in mesocarp internal (pulp of the fruit), which is of 51.9%. Of this form the amount of oil that contained in the 63.0000 g of pulp used in the reaction was of 30.2400 g (Equation 1).

Equation 1
The reaction process was accomplished by thin layer chromatography, where a mixture containing hexane and ethyl acetate in the ratio of 9:1 was used as eluent. The extent of the transesterification of pulp of pequi fruit was also determined by solution $^1$H NMR spectroscopy. The relevant signals chosen for integration were those of etoxy groups in the ethylic biodiesel fuel (3.66 ppm, triplet and ppm quartet) and those of the $\alpha$-methylene protons present in all triglyceride derivates (2.3 ppm, triplet) of pulp pequi fruit. The conversion also was calculated directly from the integrated areas of these signals. Analyzes it of the product (ethylic biodiesel fuel) for CG/MS demonstrated the presence of a mixture of ethylic ester, such as: oleic acid ethyl ester, ethyl ester of octadecanoic acid, ethyl ester of stearic acid and ethyl ester of linoleic acid. The specter of mass of oleic acid ethyl ester showed to the peak base in m/e = 55 and characteristic peaks of referring ester in m/e = 88 (rearrangement of Mc Lafferty) and in m/e = 101 [(CH$_3$CH$_2$COOCH$_2$CH$_3$)], as well as the specter of mass of ethyl ester of octadecanoic acid demonstrated to the characteristic peak in m/e = 101 and the peak base in m/e = 88 (rearrangement of Mc Lafferty). We wanted to look at the effects of the ethanol/pulp of the fruit ratio, catalyst loading utilized, catalyst loading and reaction temperature and time on the transeserification of vegetable oil contained in the pulp of pequi fruit with ethanol to yield ethylic biodiesel. Initial screening reactions were performed using a domestic microwave oven at 50º C using NaOH (0.6300 g) was predissolved in excess of ethanol (100 mL) and additoned 63.0000 g pulp of pequi fruit previously dehydrates and triturated, what theoretically it represents 32.0000 g of vegetable oil. The use of excess ethanol was necessary to increase the contact surface between the reagents and the pulp of pequi fruit and also for avoid lost for evaporation during the transesterification process as a consequence of the large amount of residual mass and material that do not undergoes transesterification contained in the fruits. The excess ethanol also acts as solvent for the extraction of the biodiesel and the glycerol present in the meal. This mixture was agited and placed into the domestic microwave oven and irradiated the 340 W for 1.0 min. Upon cooling to room temperature, the reaction vessel was removed from the microwave cavity and the contents were filtered under vacuum and evaporated in a rotatory evaporator, in order to eliminate the excess of ethanol used. The mixture was transfered to a separation funnel, where the separation of biodiesel formed and the glycerin (by-product of the reaction process) occurred for decantation. After the phases separation the traces of catalyst and ethanol were washed out with water from the esters mixture until the water layer remain completely translucent. Finally, the product was dried on MgSO$_4$. The mass of ethylic biodiesel fuel obtained was of 30.0000 (%) g and glycerin of 4.2000 g, for calculates it of the yield of the process, takes in consideration the ratio of oil in mesocarp internal (pulp of the fruit), which is of 51.9%. Of this form the amount of oil that contained in the 63.0000 g of pulp used in the reaction was of 30.2400 g. The reaction process was accomplished by chromatography in thin layer, where a mixture containing hexane and ethyl acetate in the ratio of 9:1 was used as eluent. The extent of the transesterification of pulp of pequi fruit was also determined by solution $^1$H NMR spectroscopy and will be of accord with the results previously obtained. The results obtained with the synthesis of ethylic biodiesel fuel using irradiation of microwaves demonstrate that the amount of base used reduced in up to 4 times in relation the amount previously used in the process of production of ethylic biodiesel fuel the room temperature, already the reaction time reduced in up to 60 times, with regard to the yield of the process the same it did not demonstrate to great variations in a process for heating using
microwaves and a process the room temperature through agitation mechanic, already with regard to amount of ethanol utilized, in the process for microwave used a bigger ratio to only prevent the loss of this solvent for evaporation. A time that in the method of irradiation for MW we use 10 times little mass of NaOH, this implies that also we use a lesser amount of water in the laudering of ethylc biodiesel fuel, in order to be able to remove the excess of used NaOH as catalyst, being a this more advantage of the use of the MW process, beyond it is clearly of the reduction in price of the total process, since not the extration of the vegetable oil, the drying of the pulp of the fruit of pequi using MW, the irradiation of 1.0 min in the MW and surely the greater easiness of separation of phases (ethylc biodiesel and glieren) after the irradiation process. Later, we decide to repeat reaction process the same using heating for microwaves, but searching to reduce the reacional time (ethylc biodiesel fuel production) in up to 6 times less. Our results are shown in Table 1.

Table 1. Screening reaction using pulp of fruit of pequi and ethanol.\(^a\)

<table>
<thead>
<tr>
<th>Entry</th>
<th>pulp of the pequi fruit/ethanol ratio</th>
<th>NaOH (% wt)</th>
<th>Time (seg)</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1:1.5</td>
<td>1</td>
<td>10</td>
<td>70</td>
</tr>
<tr>
<td>2</td>
<td>1:1.5</td>
<td>1</td>
<td>20</td>
<td>80</td>
</tr>
<tr>
<td>3</td>
<td>1:1.5</td>
<td>1</td>
<td>30</td>
<td>93</td>
</tr>
<tr>
<td>4</td>
<td>1:1.5</td>
<td>1</td>
<td>40</td>
<td>95</td>
</tr>
<tr>
<td>5</td>
<td>1:1.5</td>
<td>1</td>
<td>50</td>
<td>96</td>
</tr>
<tr>
<td>6</td>
<td>1:1.5</td>
<td>1</td>
<td>60</td>
<td>97</td>
</tr>
</tbody>
</table>

\(^{a}\)Reaction mixtures were heated to 50°C using microwave irradiation of 340 W.

Optimal reaction conditions envolved using a 1:1.5 pulp of pequi fruit/ethanol ratio, 1wt% NaOH, heating the reaction mixture to 50°C and holding at this temperature for 1.0 m (entry 3 in Table 1). This is gave a 97% conversion of pulp of pequi fruit in ethylc biodiesel fuel. A reduction in the reaction time (entries 3,4 and 5 in Table 1) practically maintained the yield same.

We next performed the reaction with methanol, for ease of operation we also predissolved the NaOH in methanol prior to the reaction. The volume of methanol used were the same volume of ethanol utilized for to produce the ethylc biodiesel fuel, however, the mass of NaOH reduced to 0.7000 g to 100.0000 g of pulp of pequi fruit, in the reaction for to produce ethylc biodiesel fuel we utilize 1.0000g of base (NaOH) to 100.0000 g of pulp of pequi fruit. Performing the reaction
using our optimum reaction conditions with 100.0000 g of pulp of pequi fruit gave a quantitative conversion 98% to methyllic biodiesel fuel.

The methyl biodiesel fuel and ethyl biodiesel fuel produced from the pulp of pequi fruit although are of different chemical composition than diesel fuel, it exhibits some similar properties. Table 2 shows a comparison of several properties of pure methyl biodiesel fuel and ethyl biodiesel fuel (B100) and pure diesel fuel.³,²¹,²²

Table 2. Comparison of some properties of biodiesel (B100), and diesel fuel.

<table>
<thead>
<tr>
<th>Fuel</th>
<th>Methylic biodiesel fuel</th>
<th>Ethylic biodiesel fuel</th>
<th>Diesel fuel</th>
</tr>
</thead>
<tbody>
<tr>
<td>Boiling point</td>
<td>&gt;200° C</td>
<td>&gt;200° C</td>
<td>160° C</td>
</tr>
<tr>
<td>Melting point</td>
<td>~4° C</td>
<td>~4° C</td>
<td>-25° C</td>
</tr>
<tr>
<td>Flash point</td>
<td>min 130° C</td>
<td>min 130° C</td>
<td>min 52° C</td>
</tr>
<tr>
<td>Specific gravity (water = 1)</td>
<td>0.88</td>
<td>0.88</td>
<td>0.85</td>
</tr>
<tr>
<td>Vapor pressure (mmHg)</td>
<td>&lt; 2</td>
<td>&lt; 2</td>
<td>0.40</td>
</tr>
<tr>
<td>Cetane number</td>
<td>min 47</td>
<td>min 47</td>
<td>min 40</td>
</tr>
</tbody>
</table>

It is important to understand that these data are approximate in nature and variations between different sources of fuel may occur. Some of these values are also minima set by Standards, such as the ASTM Standards D6751-03a for biodiesel fuels and D975-05 for diesel fuels.³,²¹ Some values such as the cetane number are similar in nature due to the fact that both liquids are used for the same function. From a fire investigation perspective, one of the most significant differences between biodiesel fuel and diesel fuel concerns the flash point. While both fuel and biodiesel fuel are combustible liquids (Flash points above 37.8° C as defined by the national fire Protection Association²³), the flash point of pure biodiesel fuel is much higher. From a boiling point perspective, diesel fuel has an initial boiling point around 185° C and an end point around 345° C. Average initial boiling point and point of methyl and ethyl biodiesel fuel produced with pulp of pequi fruit have been reported to be 300° C and 350° C, respectively. This means that biodiesel is less subject to evaporation than diesel fuel. The melting point of biodiesel is one of its main disadvantages in relation to diesel fuel, the methyl and ethyl biodiesel fuel produced with pulp of pequi fruit solidifies around 4° C, while the diesel fuel solidifies starting around -18° C.
The results presented in Table 2, had demonstrated that methylic and ethyluc biodiesel fuel produced by us from the pulp of pequi fruit in a process irradiated for microwaves, this in accordance with the specifications demanded for the ASTM for biodiesel fuel. The pequi fruit could future be classified and to be used as a source in potential of vegetable oil for the production of alternative fuel as biodiesel.

4. Conclusion
As demonstrated above in the results section, our methodology using a process for the production directly of pulp of pequi fruit “in natura” of methylic and ethylic biodiesel fuel in a process irradiated for MW and under atmospheric conditions, allows the obtantion of biodiesel fuel in excellent yields, where the reaction was complete in a matter of a few seconds and can be performed in a domestic microwave oven.

One of the advantages of this technique is to allow the one great reproducibility of the process, as well as the reduction in price of the process of production of biodiesel fuel, since the extration of the vegetable oil does not become necessary.

Acknowledgements
The authors thank the financial support from the Laboratory for the Development of Clean Technologies (LADETEL) at University of São Paulo, FAPEMIG and CNPq was greatly appreciated.

5. Referências Bibliográficas