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ORIGINAL ARTICLE

Evaluation of fuel properties for microalgae *Spirulina platensis* bio-diesel and its blends with Egyptian petro-diesel

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Abstract Biodiesel is an attractive alternative fuel for diesel engines due to its technical, environmental and strategic advantages. The most widely produced biodiesel is the esters of vegetable oils, animal fats or waste cooking oils.

In this study, the feasibility of biodiesel production from microalga *Spirulina platensis* has been investigated. The physico–chemical characteristics of the produced biodiesel were studied according to the standards methods of analysis (ASTM) and evaluated according to their fuel properties as compared to Egyptian petro-diesel. Blends of microalgae biodiesel and petro-diesel (B2, B5, B10 and B20) were prepared on a volume basis and their physico–chemical characteristics have been also studied. The obtained results showed that; with the increase of biodiesel concentration in the blends; the viscosity, density, total acid number, initial boiling point, calorific value, flash point, cetane number and diesel index increase. While the pour point, cloud point, carbon residue and sulfur, ash and water contents decrease. The observed properties of the blends were within the recommended petro-diesel standard specifications and they are in favor of better engine performance.

1. Introduction

Biofuels are becoming increasingly interesting as an alternative to fossil fuels due to increasing population, depletion of fossil fuels, global warming, and fluctuations of the crude oil prices. Biodiesel is an acceptable alternative fuel for diesel engine, due to its technical, environmental and strategic advantages. Biodiesel is well known chemically as the mono-alkyl esters of long chain fatty acids and is produced from several types of conventional and non-conventional vegetable oils and animal fats (Knothe et al., 2005). In addition to its renewability, other advantages of biodiesel include non-carcinogenic and non-mutagenic properties and its biodegradability, miscibility with petroleum diesel, lubricity, high flash point and cetane number, and absence of aromatics. The characteristics of biodiesel reduce the emissions of carbon monoxide (CO), hydrocarbons (HC) and particulate matter (PM) in the exhaust gas as compared with petroleum diesel, so is environmentally beneficial (Knothe, 2005; Lapuerta et al., 2008).

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Because of the small oil yield extracted from plant sources, however, a great deal of feedstock is needed for the commercialization of biodiesel fuel. This requires large areas of land, and can result in competition with food crops. Also, one of the most serious environmental problems today is the global warming caused primarily by the heavy use of fossil fuels. For these reasons, many scientists are looking for sources and new processing techniques which result in higher yields of fuel while requiring less land. Since the mid 70's microalgae have been extensively researched as a non-food based biodiesel feedstock due to their high oil content, rapid growth rate, high per-acre yield and ability to reduce the greenhouse gas concentration in the atmosphere. Microalgae are generally more efficient in converting solar energy and excessive amounts of CO_2 into biomass than land plants, 1 kg of algal biomass requires about 1.8 kg of CO_2 (Sudhakar et al., 2011). Algae can grow in salt water, freshwater and even contaminated water. They can grow at the sea, lakes, ponds, and on lands not suitable for food (Khan et al., 2009). Microalgae can generate as much as 40 times more oil per acre than other plants used or biofuels (Schenk et al., 2008). In addition, a variety of products may be produced from microalgae other than biodiesel, including other biofuels, animal feed, and valuable pharmaceutical and cosmetic products (Scott et al., 2010). This makes the production of biodiesel from microalgae a more cost effective process.

Spirulina platensis is plankton, filamentous, spiral-shaped, multicellular photosynthetic, blue-green microalgae. It is dominating the microflora of alkaline saline waters with pH of up to 11.0 and can exist in various types of habitats, namely soils; marches, fresh, brackish and seawaters, thermal springs and waters of industrial and domestic uses. *Spirulina* has a high phytonutrient value and pigments which have applications in healthy food, animal feed, therapeutics and diagnostics (Becker, 1994; Vonshak and Tomaselli, 2000). It is considered as an excellent food, lacking toxicity and having corrective properties against viral attacks, anemia, tumor growth and malnutrition (Hernandez-Corona et al., 2002; Mendes et al., 2003). It has been reported as animal and fish food supplement (Becker, 2004a,b).

The blend level (percentage of biodiesel in the bio–petro-diesel mixture) determines many important characteristics of the blended fuel. A lower blend level of biodiesel may reduce the expected benefits, such as fuel lubricity and tail pipe emission. A higher-than-specified level of biodiesel may exceed the engine manufacturer's recommended limitations, compromising the engine performance. Engine injection timing can be adjusted based on the blend level in order to improve the engine emission and performance (Tat and Van Gerpen, 2003). Biodiesel is usually sold at a higher price than petro-diesel fuel; therefore, the price of the fuel is dependent on the blend level.

The objectives of this work were to study the feasibility of producing biodiesel from *S. platensis* and estimate the produced by-products. It also aimed to determine the fuel characteristics of the produced biodiesel and prepare various blends of biodiesel with a typical Egyptian petroleum diesel and determine their fuel characteristics so as to select the most suitable blends which may be used as fuels for diesel engine.

2. Materials and methods

2.1. Materials

Pure sodium hydroxide as alkaline catalyst, anhydrous sodium sulfate, chloroform and methanol were purchased from Merck Co. (Germany). All other chemicals used for the preparation of cultivation medium were of analytical grade. Commercial Egyptian petro-diesel sample was obtained from a local fueling station.

2.2. S. platensis cultivation and harvest

S. platensis alga was obtained from the Microbiology Department; Soils, Water and Environment Research Institute, Agricultural Research Center (ARC), Giza, Egypt. The alga was grown and maintained in 500 mL Erlenmeyer flasks with 200 mL sterile modified Zarrouk liquid medium (Zarrouk, 1966); depleted from any nitrogen source and with a salinity of 10 g/L. The pH was adjusted to 9 prior to autoclaving at 120 °C for 20 min. The initial inoculum was adjusted to have an optical density A_{560} 0.2, dry weight 0.15 g/L and chlorophyll 8.46 mg/L. Cultures were incubated in a growth chamber under continuous shaking (150 rpm) and illumination (2000 Lux) at 32 ± 2 °C for the stationary phase.

For larger quantities of algae; *S. platensis* obtained from the above step was grown in 50 L photobioreactor (Fig. 1). Typically, alga was harvested every 48 h by centrifugation at 10,000 rpm for 15 min and washed once with distilled water, then dried using freeze drier.

2.3. Algal growth analysis

The algal growth was monitored by the change in pH, dry weight (DW), chlorophyll (Ch), electric conductivity (EC) using conductivity meter JENWAY model 4510 and optical density (A_{560}) using visible spectrophotometer JENWAY model 6300.

2.4. Phyto–chemical analysis

Dry weight of the harvested algal biomass was determined according to Vonshak (1986a,b).

Lipid extraction was done according to Bligh and Dayer (1959).

Extraction and determination of chlorophyll and carotenoids were carried out according to Holden (1965).

Extraction and determination of water soluble pigments (phycobilliprotein) *including*; allophycocyanin (APC) and C-phycocyanin (C-PC) were done according to the method described by Bryant et al. (1979).

The algal protein content was extracted and estimated by the method described by Aitken and Learmonth (1996).

***The total carbohydrate*s** were extracted as described by Chitlaru and Pick (1989) and determined by the method of Dubois et al. (1961).

The total nucleic acid concentration was estimated as described by Sambrook and Russell (2001).

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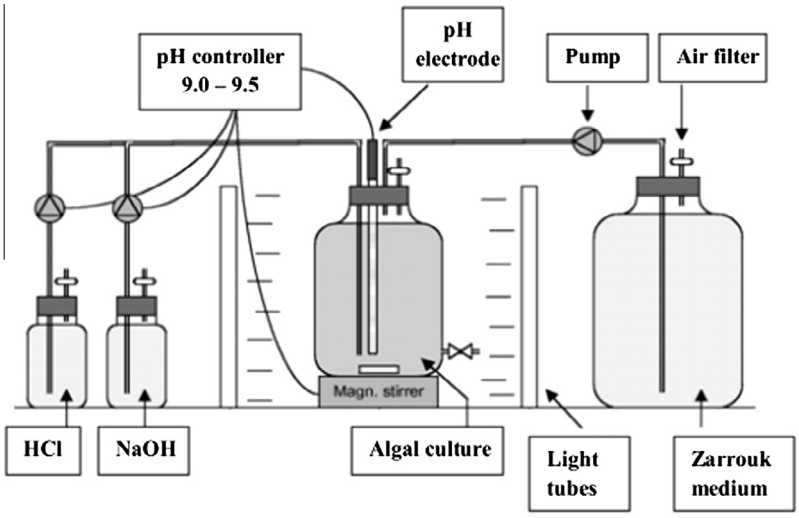

Figure 1 Schematic drawing of the photobioreactor.

2.5. Transesterification and biodiesel production

The lipid extract was mixed with methanol (1:6 w/v) and NaOH catalyst (0.6 wt%) for 3 h at 300 rpm. The solution was allowed to settle for phase separation for 16 h. The upper biodiesel layer was separated by a separating funnel, washed three times with water (5% that of ester phase) to remove any traces of methanol, NaOH or glycerol. The produced biodiesel was then dried over anhydrous sodium sulfate and its yield and that of crude glycerol were determined gravimetrically.

2.6. Gas chromatography (GC) analysis

The fatty acid methyl ester FAME composition of the produced biodiesel was analyzed using GC. The analysis was carried out by using Agilent 6890 plus, equipped with a HP-50 capillary column (0.53 mm × 30 m, 0.5 μm film) and a flame ionization detector (FID). Pure nitrogen was used as a carrier gas (4 mL/min). 250 °C injector temperature, 280 °C detector temperature, split ratio (1:50), sample size 1 μL, and the temperature program used was 80–240 °C at a fixed rate of 5 °C/min. The identification of the fatty acid esters was established by chromatography using a prepared reference mixture of fatty acid methyl esters purchased from Sigma–Aldrich.

2.7. Fourier Transform Infrared (FT-IR) Spectroscopy

Analytical FT-IR Perkin Elmer, instrument was used to identify and determine the petro- and bio-diesel structures. The spectra of all studied samples were measured in the range of 400–4000 cm^{-1} with a suitable scan resolution of 4 cm^{-1} and a scan rate of 16 cm/min.

2.8. Differential Scanning Calorimetric–Thermal Gravimetric Analysis (DSC–TGA)

The stability of biodiesel can be classified as thermal stability, oxidation stability and storage stability. However, this study focuses on thermal and oxidation stabilities. DSC–TGA analysis of the produced biodiesel was performed by Q600 SDT Simultaneous DSC–TGA. Air was purged at the rate of 100 mL/min. Temperature was programed from ambient temperature to 600 °C at the ramp rate of 10 °C/min. Oxidative stability can be measured by DSC using ASTM E 2009-02 method (2002) and was expressed as oxidative onset temperature (OOT). Heat flow out of the specimen was monitored as a function of temperature until the oxidative reaction was manifested by heat evolution on the thermal curve. OOT is defined as the temperature when a rapid increase in the rate of specimen oxidation is observed. This temperature was obtained by extrapolating the tangent drawn on the steepest slope of the reaction exothermic curve.

2.9. Physico–chemical characterization of produced biodiesel

The purified product obtained from oil esterification was tested for estimating and evaluating its fuel properties as compared to an Egyptian petro-diesel sample, using the standard methods of analysis for petroleum products, ASTM Standard Methods. The results were compared with the Egyptian standards for petro-diesel oil and European and American standards of biodiesel (EN14214 and D-6751, respectively).

Blends of Egyptian petro-diesel with different percent (2%, 5%, 10% and 20%) of the produced biodiesel were prepared on volume basis and their physico–chemical characteristics were also studied.

All the properties were analyzed in two replicates and the final results given below were obtained as the average values.

3. Results and discussion

3.1. Algal growth in photobioreactor

There was no significant change in pH at the end of incubation period, ranging between 9.24 and 9.13. This indicates the good buffering system controller. The electrical conductivity decreased from 23 dS/m to 19 dS/m. This might be attributed to the consumption of nutrients during algal growth.

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Optical density measurements for algal growth can be expressed in biomass yield, in terms of dry weight or chlorophyll (Mostafa et al., 2012). There was a great change in the optical density from 0.7 to 7.6 with a high increase in dry weight and total chlorophyll from 0.4 g/L to 4.86 g/L and from 13.65 mg/L to 108.86 mg/L, respectively.

3.2. The Phyto–chemical composition of S. Platensis

Several publications indicate a lipid content of 5–16% of dry weight in *S. platensis* by different extraction systems (Ramadan et al., 2008; Afify et al., 2010; Shalaby et al., 2010; Verma et al., 2010; Uslu et al., 2011; Mostafa et al., 2012). In this study, a lipid content of 19.55% (% wt) has been obtained. The relatively high lipid content might be due to the cultivation of *S. platensis* under stress (absence of nitrogen source and salinity 10 g/L). A similar observation was reported by Afify et al. (2010). Pigments like allophycocyanin, C-phycocyanin, carotenes and chlorophyll have many applications in food, cosmetic and pharmaceutical industries (Bhat and Madyastha, 2001; Mohammed and Mohd, 2011; Shanab et al., 2012). This study revealed yields of approximately 0.54%, 10.99%, 0.74% and 0.57% (% wt) of listed pigments, respectively. It also revealed total proteins, carbohydrates and nucleic acid contents of approximately 53.3%, 11.57% and 2.74% (% wt), respectively. All results recommend the use of the biomass after lipid extraction as animal food supplement, use pigments as natural coloring and therapeutic and/or use of carbohydrates for the production of other kinds of biofuels e.g. bioethanol. After all extraction steps were performed the waste biomass was dried, pressed and its calorific value was measured to be 39 MJ/kg. Thus it can be used as another source of bio-energy.

3.3. Algal biodiesel composition

The yield of the produced biodiesel and crude glycerol was evaluated by their weight relative to the weight of the microalgae biomass and recorded approximately 16% and 3.45% (wt%), respectively.

The FAME composition of the produced biodiesel is shown in Table 1. The FAME profile is remarkable compared to the common fatty acid profile of other biodiesels. It was characterized by a high concentration of saturated FAMEs and low concentrations of unsaturated and polyunsaturated FAMEs; approximately 90%, 8% and 2%, respectively. The high concentration of saturated FAMEs guarantees good oxidation resistance for biodiesel (Kondamudi et al., 2009). The major FAMEs were arachidic, stearic and palmitic acids methyl esters, recording approximately 32%, 28% and 15%, respectively. The presence of behenic and lignoceric acids methyl esters with concentrations of approximately 7% and 4%, respectively was also recorded. The high concentration (≈81%) of FAMEs having a carbon chain of ⩾18 carbons would impart high viscosity for biodiesel (Koberg et al., 2011).

Table 1 A summary of the identified fatty acid methyl esters (FAME).

FAME	wt%
Myristic acid methyl ester (C14:0)	3.56
Palmitic acid methyl ester (C16:0)	14.87
Palmitoleic acid methyl ester (C16:1)	0.98
Stearic acid methyl ester (C18:0)	28.11
Oleic acid methyl ester (C18:1)	5.39
Linoleic acid methyl ester (C18:2); ω6	1.12
Arachidic acid methyl ester (C20:0)	32.43
Eicosenoic acid methyl ester (C20:1)	1.94
Eicosadienoic acid methyl ester (C20:2); ω6	0.99
Behenic acid methyl ester (C22:0)	6.65
Lignoceric acid methyl ester (C24:0)	3.96
Saturated FAME	89.58
Unsaturated FAME	8.31
Polyunsaturated FAME (PUFA)	2.11

3.4. FTIR spectrophotometer analysis

FTIR spectra of petro- and bio-diesel are shown in Fig. 2. The main components of diesel are aliphatic hydrocarbons, whose chemical structures are similar to the long carbon chains of the main components of biodiesel. The characteristic absorption bands for the vibrations of C–H, around 2929 and 2850 cm^{-1} corresponding to the asymmetric and symmetric vibration modes of methyl groups, respectively, were detected. For biodiesel spectra, the FAMEs are *cis* isomers, as expected from algal oil because *trans* isomers produce a strong band at 970 cm^{-1} and a weak band at 3012 cm^{-1} while *cis* isomers give medium nearby 720 cm^{-1} and 3000 cm^{-1} bands, which are obvious in Fig. 2. FTIR spectra clearly show evident differences of both spectra. For the range from 1000 cm^{-1} to 1300 cm^{-1}, biodiesel presents many overlapping peaks, not present in diesel oil. The observation of an absorption peak around 1200 cm^{-1} may be assigned to the asymmetric axial stretching vibrations of C(C═O)–O bonds of the esters, while peaks around 1183 cm^{-1} may be assigned to asymmetric axial stretching vibrations of O–C–C bonds (Silverstein and Webster, 1998). In addition, since biodiesel is mainly mono-alkyl ester, the intense C═O stretching band of methyl ester and O–CH_3 appeared around 1739 cm^{-1} and 1172 cm^{-1}, respectively for algal biodiesel which are absent in petro-diesel spectra. In biodiesel spectra, the absence of a peak higher than 3000 cm^{-1} corresponding to –OH of carboxylic acid indicates complete transesterification. In Petro-diesel spectra, the stretch bands of aromatic hydrogen (═C–H) and aromatic (C═C) occur around 2953 cm^{-1} and 1635 cm^{-1}, respectively, which are absent in biodiesel spectra.

3.5. Thermal and oxidation stabilities

The thermal stability of the produced biodiesel was evaluated by TGA: a method used to determine relative weight loss against temperature rise of a biodiesel sample. DSC–TGA determines the temperature at which the oxidation reaction takes place. This temperature is termed as oxidation onset temperature. Fig. 3a, shows the TG/DTG curves of the produced biodiesel. The onset of one exothermic thermal event is indicated in derivative curve (DTG). The maximum rate of biodiesel weight loss occurred at approximately 372 °C, where the rate of weight loss increased to the maximum. A slower weight loss was observed at higher temperatures. The TGA curve shows 3 steps, where the first weight loss started to decrease at approximately 280 °C, and this step may be attributed to the volatilization point of the produced biodiesel. The second

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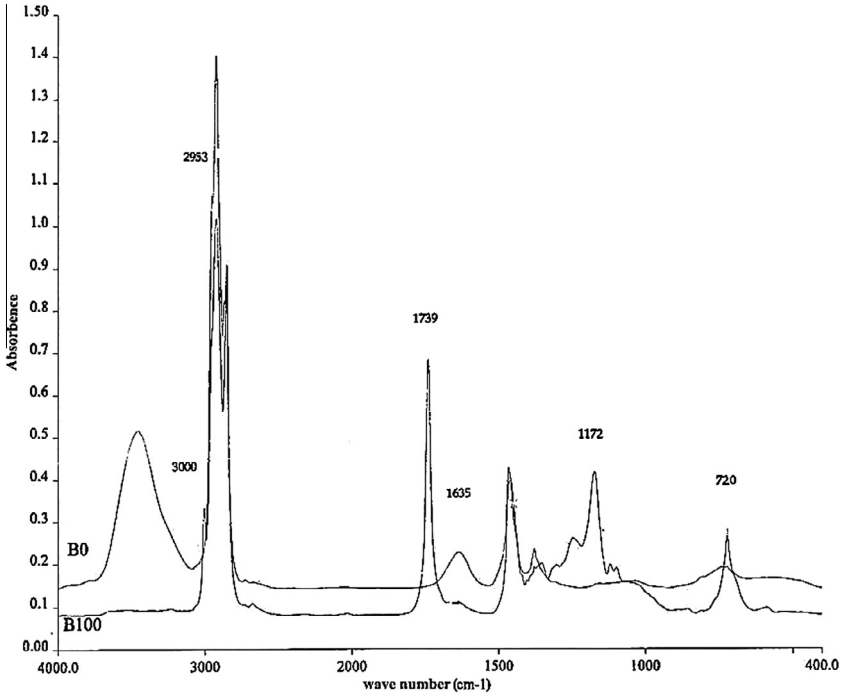

Figure 2 FTIR spectra of petro- and bio-diesel (B0 and B100, respectively).

one started to decrease at 420 °C and it may be attributed to the boiling point of esters with unsaturated bonding. The third step started to decrease at approximately 480 °C and it may be due to some algal oil that was not *trans*-esterified. The mass continues to decrease until all the biodiesel present in the sample is vaporized. Thermal stability and volatility characteristics are important in establishing the ignition quality of fuels and its lubricity properties (Sharma et al., 2009). Oxidative stability refers to the extent of fuel deterioration when exposed to air during storage. It plays a key role in the industrial applications of biodiesel. In this study, oxidative onset temperature (OOT) was used to evaluate the oxidation stability of the produced biodiesel. A high OOT value is indicative of a stable material. The DSC–TGA curves of produced biodiesel are shown in Fig. 3b. The onset of one exothermic thermal event is indicated in DSC curve with a change in enthalpy (ΔH) of 1268 J/g, confirming an oxidation reaction has been occurred. From the DSC cure, the OOT of produced biodiesel recorded was approximately 331 °C.

There is no thermal stability limit specified for biodiesel in any standard. However, practically, a biodiesel which maintains its stability up to 150 °C can be regarded as thermally stable. When the oxidation takes place, the evaporation of secondary oxidation products causes a sudden weight loss of the sample. The onset temperature of oxidation can be obtained by the intersection of the extrapolated baseline and the tangent line of the curve obtained by TGA (Fig. 3b), and recorded approximately 331 °C, which is equivalent to that obtained from DSC curve. TGA can be used for measuring the oxidation stability of produced biodiesel. The onset temperature can be used to indicate the resistance of the biodiesel sample to thermal degradation. The obtained relatively high onset temperature indicates high thermal and oxidation stability of the produced biodiesel recommending its applicability as a petroleum-based diesel substitute.

The high viscosity resulted in a lower oxygen diffusion rate, thus higher oxidation stability (Ikwuagwu et al., 2000). The degree of unsaturation influences the reactivity of the double bonds. Double bonds in polyunsaturated systems are more reactive than monounsaturated double bonds (Bajpai and Tyagi, 2006). The reason for good stability of the produced biodiesel may be due to the resistance to auto oxidation of saturated fatty acids methyl esters. As given in Table 1, the produced biodiesel has approximately 90% of saturates making it less susceptible toward oxidation.

3.6. The physico–chemical properties of the produced biodiesel

The produced biodiesel was evaluated on the basis of its distillation characteristics as well as its fuel properties compared to Egyptian petro-diesel sample and international biodiesel standards as shown in Table 2. There is a difference in viscosity compared with petro-diesel and international biodiesel standards, otherwise all the properties of the produced biodiesel are completely acceptable and meet most of the specifications. So it can be ranked as a realistic fuel and as an alternative to petro-diesel.

The iodine value for the produced biodiesel, which is a measure of un-saturation degree, recorded 102 mg I_2/100 g oil. The degree of unsaturation greatly influences fuel oxidation tendency. According to EN 14214, methyl esters used as diesel fuel must have an iodine value less than 120 g I_2/100 g oil.

The acid value measures the content of free acids in the sample, which has influence on fuel aging. The acid value of the produced biodiesel was 0.75 mg KOH/g while that of algal oil 3.05 mg KOH/g with average lowering of about 75%, indicating good transesterification process. The biodiesel TAN was higher than that of petro-diesel 0.023 mg KOH/g but within the biodiesel standards. This indicates that the biodiesel will not cause operational problems, such as corrosion and pump

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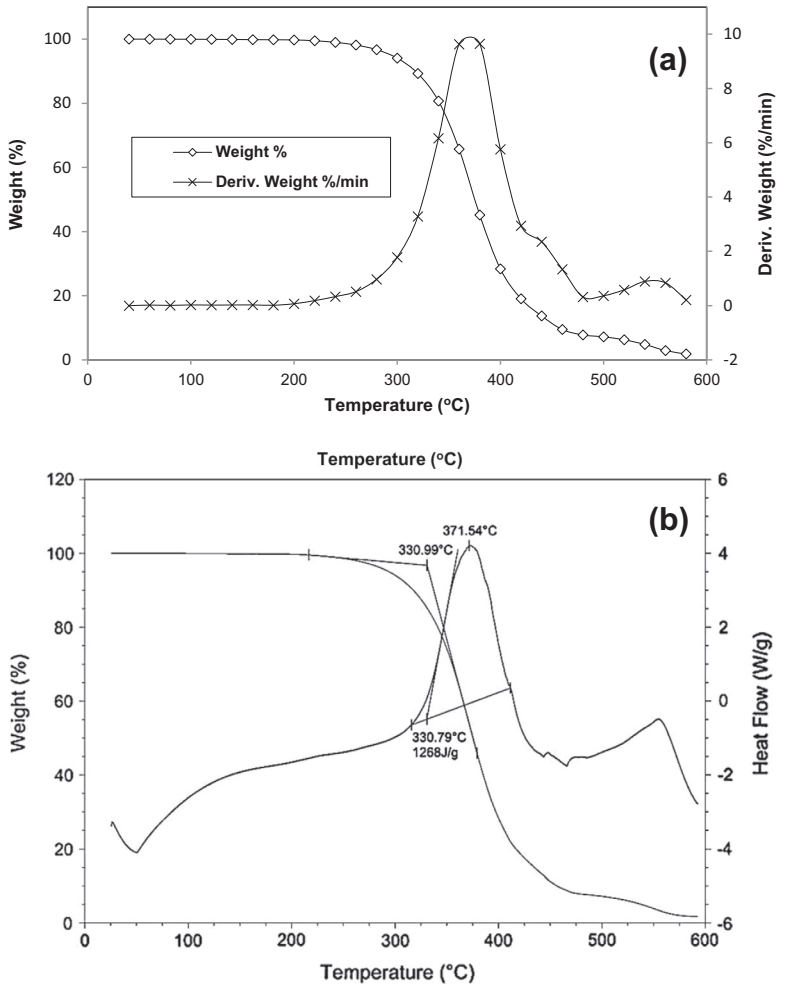

Figure 3 Typical (a) TG/DTG curves and (b) DSC/TGA thermographs for produced biodiesel.

plugging, caused by corrosion and deposit formation (Candeia et al., 2009).

The produced biodiesel has major advantages over petro-diesel. It is free of sulfur, while petro-diesel has 0.13% sulfur. So it meets the aim of petroleum industry for free sulfur diesel fuel and the biodiesel combustion will not produce sulfur oxides which lead to corrosion of the engine parts and environmental pollution. The produced biodiesel is free of ash, while that of petro-diesel was 0.009 wt%. Ash can deposit upon the engine parts causing abrasion. Biodiesel is also free of sediments. Sediments can affect the combustion of the fuel, and if abrasive, my cause excessive wear of closely fitting parts of fuel pumps and injectors. It may build up deposits in tanks and piping and may clog fuel filters which can lead to engine fuel starvation. The carbon residue in the produced biodiesel was nil. While that of petro-diesel was 0.019 wt%. This is considered as an additional advantage for biodiesel. Its water content 39 ppm was lower than that of petro-diesel 52 ppm and it was lower than the standards, 300 ppm. Water can contribute to filter blocking, cause corrosion and promote microbial growth. The density at 15 °C is an important property, mainly in airless combustion systems because it influences the efficiency of atomization of the fuel (Felizardo et al., 2006). The density of the produced biodiesel was 0.8637 g/cm^3 compared to that of petro-diesel 0.8422 g/cm^3. Fuel with high paraffinicity has a high specific gravity and a low API. The produced biodiesel was characterized by a higher specific gravity (0.8681) and a lower API value (31.5) compared to those of petro-diesel (0.843 and 36.35, respectively). Therefore volumetrically, biodiesel delivers a slightly greater amount of fuel (Sharma, 2006).

Even more than density, kinematic viscosity at 40 °C is an important property regarding fuel atomization, flow and distribution. The viscosity of the produced biodiesel was 12.4 cSt which was much higher than that of petro-diesel 3.21 cSt and it does not meet the required values for biodiesel, that must be between 1.9 cSt and 6 cSt. This inferred that the produced biodiesel would have inferior injection and atomization performance, but offer lubrication and protection for moving parts of an engine. This result also recommends preparing different blends of bio- and petro-diesel to lower the viscosity of biodiesel and improve the lubricity of the petro-diesel. Indeed, the high lubricity is particularly helpful since the trend toward ultra-low sulfur diesel fuel (ULSD) has proven problematic in this regard (Knothe, 2005).

3.6.1. Distillation temperatures DT

The DT characterize the volatility of the fuel and consequently have significant effect on the combustion characteristics of the

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Table 2 Physico–chemical properties of biodiesel compared to standard biodiesel specifications and Egyptian petro-diesel sample.

Test	Unit	Egyptian petro-diesel	Produced Biodiesel	Biodiesel (EN14214)	Biodiesel D-6751
Density @ 15.56 °C	g/cm^3	0.8422	0.8637	0.86–0.9	–
Specific gravity		0.843	0.8681	–	–
API		36.35	31.5	–	–
Kinematic Viscosity @ 40 °C	cSt	3.21	12.4	3.5–5	1.9–6
Total acid number	mg KOH/g oil	0.023	0.75	< 0.5	< 0.8
Pour point	°C	−3	−9	–	–
Cloud point	°C	6	−3	−4	–
Total S	wt%	0.13	Nil	< 0.01	< 0.05
Ash content	wt%	0.009	Nil	< 0.02	< 0.02
Carbon residue	wt%	0.019	Nil	< 0.03	< 0.05
Water content	ppm	52	39	< 500	< 300
Sediment content	wt%	Nil	Nil		< 0.05
Flash point	°C	82	189	> 101	> 130
Calorific value	MJ/kg	45.35	45.63	32.9	–
Aniline point		67	84	–	–
Cetane number		50	70	> 51	> 47
Diesel index		48	67	–	–
ASTM distillation					
IBP	°C	157	270		
50%		275	380	–	–
80%		325	403		
Copper strip corrosion @ 100 °C		1a	1a	Class 1	No. 3 Max.
Color index		3.5	1	–	–
Iodine number mg I_2/100 g oil		–	102	< 120	–

diesel engine. It is obvious from data listed in Table 2, that the produced biodiesel has high DT than those of petro-diesel. The initial boiling point IBP of the produced biodiesel was 270 °C compared to that of petro-diesel 157 °C. The mid boiling point (50%) of biodiesel was 380 °C and that of petro-diesel 275 °C. The narrow range of DT between 380 °C and 403 °C corresponding to 50% and 80% of produced biodiesel compared to wide range of petro-diesel 275–325 °C, respectively, can be attributed to the biodiesel chemical composition of FAMEs from C14 to C24. The higher DT may shorten the ignition delay of the fuel and decrease the probability of the occurrence of knocking in the diesel engine (Zheng and Hanna, 1996). The higher DT of the produced biodiesel allows to be further hydrocracked to lighter products, e.g., bio-gasoline, bio-jet fuel and biodiesel.

3.6.2. Cloud point CP and pour point PP

The cold flow properties, including CP and PP, have much lower values in the produced biodiesel (−3 and −9, respectively) than those of petro-diesel (6 and −3, respectively). This indicates better cold flow properties and increases the advantages of the produced biodiesel, as it is more suitable in cold conditions.

3.6.3. Cetane number CN, aniline point AP and diesel index DI

The CN of diesel fuel is a relative measure of the interval between the injection of the fuel into the cylinder and onset of auto ignition i.e. the ignition delay (ID) time. The higher the CN, the shorter the ID time and vice versa. The obtained biodiesel was characterized by higher CN (70) compared to that of petro-diesel (50), which would result in a higher combustion efficiency, improve engine performance, clean up emissions and can be recommended to be used in high speed engine (speeds above 800 rpm). A high cetane number diesel fuel is necessary to run such an engine; its fuel is usually specified to have a minimum CN of 52. Fuels with higher CN, which is a result of higher oxygen content will cause an engine to operate more efficiently and will tend to start more easily. Fuels with low CN will result in difficulty in starting, higher noise and exhaust smoke. The increase in saturated FAMEs content and its chain length positively enhance the cetane number of biodiesel.

AP is the lowest temperature at which the fuel is completely soluble in an equal volume of pure dry aniline. The obtained biodiesel was characterized by higher AP (84) compared to that of petro-diesel (67). A diesel fuel is a mixture of aromatics, naphthenes and paraffins, which are the three basic types of hydrocarbons found in petroleum. Aromatics have low AP therefore low DI and poor ignition quality. Paraffins, on the other hand, have relatively high AP and good ignition quality. Naphthenes are intermediate in ignition quality. Fuel with high specific gravity has high AP.

DI is more representative of the paraffincity of the fuel. The obtained biodiesel was characterized by higher DI (67) compared to that of petro-diesel (48).

3.6.4. Flash point FP and calorific value CV

The produced biodiesel has a higher FP 189 °C, compared to 82 °C for petro-diesel. So biodiesel is much less flammable fuel than petro-diesel and, hence it is much safer in handling, storage and transport.

The produced biodiesel has slightly high CV (45.63 MJ/kg) than that of petro-diesel (45.35 MJ/kg).

3.7. Physico–chemical properties of the blends

Blends of the produced biodiesel and petro-diesel were prepared, and the results of their analysis compared to the Egyptian petro-diesel standards are listed in Table 3.

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Table 3 Physico–chemical properties of prepared blends compared to standards specifications of biodiesel blends B6–B20 and Egyptian petro-diesel.

Test	Unit	Egyptian Perto-diesel standards	B2	B5	B10	B20	Biodiesel blends ASTM D-7467
Density @ 15.56 °C	g/cm^3	0.82–0.87	0.8427	0.843	0.8453	0.8462	–
Specific gravity			0.8435	0.8438	0.8461	0.8477	–
API			36.25	36.19	35.74	35.55	–
Kinematic Viscosity @ 40 °C	cSt	1.6–7	3.76	4.20	4.61	5.11	1.9–4.1
Total acid number	mg KOH/g oil	Nil	0.028	0.05	0.074	0.105	< 0.3
Pour point	°C	4.5	−3	−6	−6	−6	–
Cloud point	°C	–	3	0	0	0	–
Total S	wt%	< 1	0.11	0.09	0.033	0.021	< 0.05
Ash content	wt%	< 0.01	0.003	0.0028	0.002	0.0018	< 0.01
Carbon residue	wt%	< 0.1	0.016	0.013	0.01	0.008	< 0.35
Water content	ppm	150	49	45.5	43.8	40.9	< 500
Sediment content	wt%	< 0.15	Nil	Nil	Nil	Nil	< 0.05
Flash point	°C	> 55	84	100	108	112	> 52
Calorific value	MJ/kg	> 44.3	45.56	45.58	45.61	45.62	–
Aniline point			73	75	81	82	–
Cetane number		> 55	60	62	67	68	> 40
Diesel index		> 48	58.09	59.69	64	65	
ASTM distillation							–
IBP	°C	Min. 85% @ 350 °C	163	165	177	210	
50%			292	295	310	337	
80%			340	343	356	384	
Copper strip corrosion @ 100 °C		1a	1a	1a	1a	1a	No. 3 Max.
Color index		< 4	3.5	3	2.5	2.5	–

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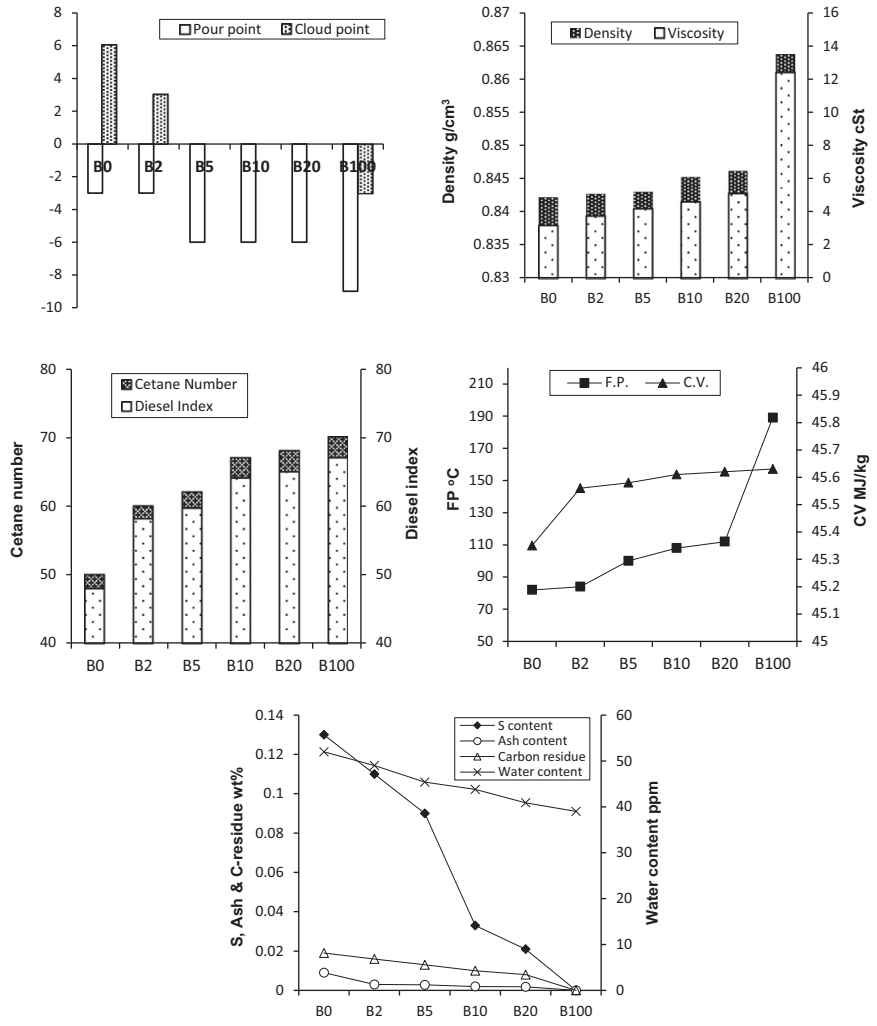

Figure 4 Some physico–chemical properties of petro-diesel (B0), biodiesel (B100) and their blends (B2, B5, B10 and B20).

Fig. 4, illustrates the changes of some physico–chemical properties after blending of petro- and bio-diesel. The data indicate that the density and viscosity values decreased with blending and increased with the increase of volume percentage of biodiesel. Their values also agree with the Egyptian petro-diesel standards and may lead to better lubricity. Although the TAN decreased with blending, its values are higher than that of Egyptian petro-diesel standards, but within the recommended blend standards. The cold flow properties, CP and PP were improved by blending. The total sulfur content decreased with increasing biodiesel percentage and led to ultra-low sulfur diesel which consequently would decrease the SOx emission and decrease environmental pollution. Ash, sediment and water contents and carbon residue in the blends decreased with the increase of volume percentage of biodiesel. This would decrease corrosion and increase the lifetime of fuel pumps, injectors and engines. The FP, CV, AP, DI, CN and DT of the prepared blends increased with increasing biodiesel percentage in the blends. This will lead to improvement in the combustion properties and ignition performance which consequently will reduce the greenhouse emissions (e.g. CO and CO_2). Also the increase in FP would lead to better handling and safety during transportation and storage. All the measured properties of the prepared blends agree with the Egyptian petro-diesel and ASTM blends standards.

4. Conclusions

The results of this study indicate that microalga *S. platensis* is a valuable candidate to be used for biodiesel production, due to its high growth rate 2.23 g/Ld, sufficient lipid content, requiring just a simple and inexpensive culture medium and producing other valuable byproducts which would decrease the overall cost of biodiesel production. It is worth mentioning that the extreme conditions of salinity and pH in which *S. platensis* can survive add to its value to be used for production of biodiesel.

The quality characteristics of biodiesel obtained in this work were in good agreement with ASTM D6751 (2007) and EN 1424 specifications. Therefore, it can be acceptable and suitable for diesel fuel. The prepared blends B5-B20 have also good comparable characteristics with the petro-diesel marketed in Egypt.

Because of the large coastal area and warm climate in Egypt, *S. platensis* has very good potential to be grown in

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Egypt, therefore it is recommended to be cultivated on a large scale production to produce non-conventional biofuels and other valuable byproducts.

Further research on non-traditional methods for lipid extraction and transesterification are undertaken now in EPRI Biotechnology lab to decrease the overall cost of biodiesel production on a large scale. Also the feasibility of producing bioethanol and biogas from the waste biomass is under study now in EPRI Lab.

Further research on hydrocracking of the produced biodiesel is recommended to introduce more flexibility of the product qualities to suit market requirements.

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