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Potentials of non-edible *Abrus precatorius* seed oil towards biodiesel production

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***Abrus precatorius* seed oil is not edible and possesses lethal toxicological properties; hence, alternative use as low cost feedstock for biodiesel production was investigated. The n-hexane extracted oil was characterized for its chemical and physical properties, and subsequently transesterified using 1% sodium hydroxide at 60°C to produce biodiesel. The biodiesel produced had acceptable quality following characterization of its fuel properties. The relative density was found to be 0.889 with kinematic viscosity of 3.34 mm^2/s within limits of Thailand biodiesel fuel standard. The acid value was 0.281 mg KOH/g, iodine value 52.43 mgI_2/g, peroxide value 3.45 mEq/kg, saponification value 227.8 mg KOH/g and 2.87% free fatty acid content. The flash point and the cetane number were 137°C and 58.3, respectively while the heat of combustion was 38.28 MJ/Kg. The low temperature operability properties of *A. precatorius* seed biodiesel determined by parameters such as cloud point, pour point and cold-filter plugging point were - 2, 1 and - 4°C, respectively. The sulfated ash value and refractive index were found to be 0.09% and 1.457, respectively. GC analysis of the fatty acid methyl esters profile revealed 50.86 and 49.1%, saturated and unsaturated fatty acid methyl esters (FAME), respectively. A percentage FAME yield of 86.1%; with higher content of methyl palmitoleate (31.94%) and a lowest value for methyldecanoate (1.27%) was obtained. Although a n-hexane oil yield was low (2.52% w/w), the results show that Abrus seed oil derived biodiesel has commercially acceptable fuel properties and may be suitable as fuel for internal combustion engine.**

Key words: Biodiesel, *Abrus precatorius*, oil, non-edible oil, fatty acid methyl ester.

INTRODUCTION

The use of vegetable oils as fuels in internal combustion engines dates back to over a century ago, when in 1912 Rudolf Diesel successfully tested peanut oil as fuel for his engine (Knothe et al., 2005). During a demonstration at

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the World's Fair, he predicted that, the use of vegetable oils for engine fuels although might seem insignificant then, but such oils may become, in the course of time, as important as petroleum and the coal tar products of the present time. His words have taken on added significance today. In the 1930s and 1940s, vegetable oils were used as diesel fuels from time to time but usually only in emergency situations. When petroleum-derived oils became available, the vegetable oils were displaced as fuels for engines (Knothe et al., 2005). The continued and increasing use of petroleum will intensify local air pollution and magnify the global warming problems caused by CO_2 emission rate above which the atmosphere can naturally contend with. Recently, the intermittent global energy crisis often prompted by the unpredictable crude oil economy and environmental issues stemming from downstream petroleum processing and burning of fossil fuels has spawned research efforts towards alternative energy. Consequently, increasing demand for bioenergy has generated a strong interest in the bioconversion of agrowastes including vegetable oils into fuels. Around one-tenth of global primary energy use is based on bioenergy sources, of which about 10% are produced from modern bioenergy in form of power, heat and fuel. Currently, biodiesel is considered a promising alternative fuel due to its renewability, better gas emission, CO_2 neutrality, non-toxic and also biodegradability (Hossain et al., 2010); excellent lubricity properties and is typically low in sulfur content, thus meeting the needs of the EPA and new generation fuels. Biodiesel (B100) is a fuel comprised of mono-alkyl esters of long-chain fatty acids derived from vegetable oils or animal fats. It is a fuel designed as a blend stock for use in blending with petroleum diesel fuel and also considered to be an alternative for petroleum-based diesel fuel. Despite possessing attractive prospects, the traditional biodiesel production still faces some bottlenecks as follows: the limited supply of lipid feedstock which generally relies on geographical and seasonal conditions, and the chemical trans-esterification which is energy consumption intensive and needs further waste treatment processes (Ibeto et al., 2011). Considerable researches have been done on vegetable oils as diesel fuel; including palm oil, soybean oil, sunflower oil, coconut oil, rapeseed oil and tung oil. Animal fats, although mentioned frequently, have not been studied to the same extent as vegetable oils (Ma and Hanna, 1999). The most common technique for producing biodiesel is trans-esterification, which refers to a catalyzed chemical reaction of triacylglycerides of vegetable oil with alcohols. However, the vegetable oil and alcohol must be substantially anhydrous and have low free fatty acid content because the presence of water or free fatty acid or both promote soap formation. Generally, trans-esterification is used to produce biodiesel from vegetable oil or animal fat containing low free fatty acid through a reaction involving alcohol and an alkaline catalyst (Ma et al., 1998; Gerpen et al., 2004; Prateepchaikul et al., 2007). When biodiesel is produced from high FFA oils by trans-esterification, the high FFA content in the oils reacts with the metallic alkoxide to produce soap (saponification) (Brown et al., 2003, Gerpen et al., 2004). In addition, if oil contains high moisture content, saponification and hydrolysis occur. These reactions cause a lower yield and washing of biodiesel very difficult. These problems can be solved via either one of four methods: enzymatic-catalyzed trans-esterification, acid-catalyzed trans-esterification, a super-critical carbon dioxide technique or a two-stage process (Ma and Hanna, 1999).

As with bioalcohol production from food based raw materials, use of edible feedstock for biodiesel production has attracted tremendous debate prompting research and development efforts to seek means of augmenting oil yield from food based feedstocks as well as identifying non-edible seed oils with potentials for biodiesel production. The production of biodiesel from edible and non-edible oil has progressively affected food uses , price, production and availability (Rashid et al., 2008). Vegetable oil seeds that do not compete with traditional food crops are needed to meet existing energy demands (Xu and Hanna, 2009). In order to achieve production cost reduction and make biodiesel more competitive with petroleum diesel, low cost feedstocks, such as non-edible oils, waste vegetable oils could be used as raw materials (Xiaohu and Geg, 2009). To reduce the impact on land resources and reserve edible feedstocks for consumption, an increase in biodiesel production will require the use of raw materials that are unconnected with food, especially non-edible oils such as the toxic *Abrus precatorious* seed oil. *A. precatorius* belongs to the family Fabaceae, and it is a creeping or climbing woody vine with pinnately compound leave (Figure 1) (Pokharkar et al., 2011). They are found in tropical climates of Nigeria, India, Thailand, Sri Lanka, the Philippine Islands, South China and West Indies (Hart, 1963).

The alkaloids of the seeds are abrine, hypaphorine, choline and precatorine. Proximate analysis of *A. precatorius* seeds revealed the presence of moisture (5.06%), oil yield (2.5%), crude protein (39.20%), crude fibre (9.08%), ash (5.38%) and total carbohydrate (42.42%) (Abu et al., 2011). *A. precatorius* seeds have been found to contain alkaloids, steroids, lectine, flavonoids and anthocyanins (Attal et al., 2010).

As part of our research and development efforts to explore edible and non-edible seed oils in Nigeria for biodiesel production, the potentials of the toxic *A. pretarious* red seed oil as feedstock for biodiesel were investigated in this study.

MATERIALS AND METHODS

Collection and processing of the samples

A. precatorius red seeds were collected and cleaned by washing with distilled water. The seeds were dried and ground into fine

Figure 1. *Abrus precatorious* plant.

coarse particles using commercial grinder.

Extraction of oil from *A. precatorius* red seeds

Dried and washed *A. precatorius* red seeds (6500 g) were crushed, using commercial grinder and fed to a Soxhlet extractor fitted with a 2-L round bottomed flask (Rashid et al., 2008). The extraction with n-hexane was executed on a water bath for 6 h. The solvent was removed under vacuum, using a rotary evaporator. The amount of oil extracted was determined as a ratio of weight of extract to the total weight of Abrus seeds used.

Determination of percentage free fatty acids (FFA) of the oil

Two grams of well-mixed *A. precatorius* sample was accurately weighed to conical flask into which 10 ml of neutralized 95% ethanol and phenolphthalein were added. This was then titrated with 0.1 M NaOH, shaking constantly until a pink colour persisted for 30 s (AOAC, 1984). The %FFA was determined using equation:

%FFA = [(Volume of NaOH)(Molarity of NaOH) / Weight of oil]*2.82

Where, 2.82 is a conversion factor for oleic acid.

Biodiesel production by two step acid-base trans-esterification

Acid pretreatment

The crude *A. precatorius* seed oil was heated to 50°C while stirring continuously using magnetic stirrer to homogenize the oil. The reaction was conducted in a 500 ml three necked round-bottomed flask attached with a reflux condenser and thermometer, and placed in a water bath with a temperature control. A concentrated sulfuric acid (2% based on oil weight) in 0.60 w/w methanol was heated to 50°C and added to the reaction flask containing pre-heated oil (Zullaikah et al., 2005; Bala, 2005). This mixture was stirred for 2 h. The reaction product mixture was then poured into a separating funnel and allowed to settle for 2 h. The top layer comprised unreacted methanol, whereas the middle layer was oil and fatty acid methylester (FAME) (small amount obtained by conversion of free fatty acids to esters), and water at the bottom layer (Zullaikah et al., 2005).

Alkaline trans-esterification

The acid pre-treated A. *precatorius* seed oil with low percentage free fatty acid was heated to 60°C in a four necked flask and stirred at 800 rpm with mechanical stirrer in a water bath. The catalyst sodium hydroxide (NaOH) 0.5% based on oil weight was dissolved in the required amount of methanol (ratio was methanol: oil = 6:1) and added to the pre-treated oil (Freedman et al., 1984). The reaction was conducted for 120 min. The resulting product was poured into a separating funnel and stood for 2 h. Two phases were distinct; biodiesel on top and the glycerol at the bottom. The two phases were separated and the excess methanol in biodiesel was recovered by using a rotary evaporator. The biodiesel was then washed thoroughly by using hot de-ionized water to wash out impurities like soap and other residues. Finally, the biodiesel was heated to 100°C for 1 h in oven to remove the moisture. Based on the initial amount of pre-treated oil, the biodiesel yield was then evaluated (Meher et al., 2006).

Analysis of physico-chemical properties of the Abrus oil extract and its biodiesel

Physicochemical properties were determined by using standard test methods. These standard values were calculated and compared with European organization (EN 14214). Kinematic viscosity (at 40°C) was determined using viscometer (Oswald U-tube) (AOAC, 1975), refractive index (at room temperature) was determined with Abbe refractometer (Alamu et al., 2008). The relative density (at 15°C) was determined using the relative density bottle (AOAC, 1975). The flash point was determined by the method of ASTMD93 using the Pensky-Martens closed cup tester. Other parameters such as ash content, cold filter plugging point, Pour point and cloud point were determined following the method described by the Association of Official Analytical Chemists (AOAC) (1984). Iodine, peroxide, acid and saponification values were determined as described by Hamilton and Hamilton (1992) while the heat of combustion was determined using bomb calorimeter. The cetane number (CN) of the biodiesel was calculated using empirical

Table 1. Percentage yield and physicochemical properties of *Abrus precatorious* seed oil.

Parameter	Amount
Oil yield (% w/w)	2.52
Relative density	0.92
Kinematic viscosity (mm^2/s)	23.4
Iodine value (mgI_2/g)	57.34
Peroxide value (mEq/Kg)	4.1
Acid value (mg KOH/g)	5.74
Saponification value (mg KOH/g)	227.8
Free fatty acid (%)	2.87

formula provided by Mohibbe et al. (2005).

Determination of fatty acid profile of biodiesel

The fatty acid composition of *A. precatorius* seed biodiesel was determined using agilent 6890 series gas chromatography (GC) equipped with flame ionization detector and capillary column (30 m×0.25 mm×0.25 mm). A quantity, 2 g of oil was weighed out in a small beaker and the exact weight was recorded. The sample was dissolved in 50 ml of chloroform and transferred into 100 ml volumetric flask and diluted to the mark. The most of the chloroform at room temperature was evaporated. One milliliter of inter-esterification reagent (20%v/v benzene and 55% v/v methanol) was added, sealed and heated at 100□C in water bath for 30 min. After inter-esterification, the extract of the methyl esters was mixed with hexane and water in a proportion of 1:1:1 and shook vigorously for 2 min. About half of the top layer (hexane layer) was transferred to the small test tube for injection. The detector temperature was programmed at 240□C with flow rate of 0.8 ml/min. The injector temperature was set at 240□C. Hydrogen was used as the carrier gas. The identification of the peaks characteristic and composition of *A. precatorius* seed biodiesel was achieved by retention times by means of comparing them with authentic standards analyzed under the same conditions (Knothe, 2005).

RESULTS

Percentage yield and physicochemical properties of *Abrus precatorious* seed oil

Table 1 shows that the yields of n-hexane oil extract of *A. precatorius* seed were very poor. The physical characterizations of the oil extract presented in Table 1 show that the oil extract has a lower relative density and lower kinematic viscosity. The Table also shows that the chemical characterization of *A. precatorius* seed oil has a lower iodine value, peroxide value, acid value with high saponification value and free fatty acid.

Biodiesel physicochemical and fuel properties

Table 2 shows the physical characterization of *A. precatorius* seed biodiesel in comparison with petro-diesel. The biodiesel from *A. precatorius* seed oil has higher density, kinematic viscosity, flash point, cloud point, pour point, cold filter plugging point, cetane number, ash contents and lower heat of combustion and refractive index than petro-diesel. *A. precatorius* seed biodiesel has lower acid value, iodine value and higher peroxide value than petro diesel.

Fatty acid profile of Abrus seed oil biodiesel

The result of fatty acid profile of *A. precatorius* seed oil biodiesel as shown in Table 3 reveals that the biodiesel contains methyl decanoate (1.27%), methyl undecanote (10.49%), methyl dodecanoate (6%), methyl eicosa-dienoate (17.16%), methyl tetradecanoate (4.59%), methyl palmitoleate (31.94%), methyl heptadecanoate (4.84%), methyl stearate (14.22%), methyl pentacosylate (3.7%) and methyl messilate (5.75%). The saturated FAME in *A. precatorius* seed oil biodiesel was 50.86% while unsaturated was 49.1%.

DISCUSSION

The oil yield content of n-hexane extract of *A. precatorius* seed was found to be 2.52%. This result is the same as the oil yield of petroleum ether extract of *A. precatorius* seed reported by Abu et al. (2011). However, the oil content suggests that *A. precatorius* seeds have low oil yield when compared to linseed (33.33%), soybean (18.35%) (Gunstone, 1999) and palm oil kernel (44.6%) (Akbar et al., 2009); although, the oil yield of *A. precatorius* seed can be improved by using other extraction methods.

The physical characterization of the oils showed that the relative density of *A. precatorius* seed oil was 0.92. The result is similar to the oil of neem (0.918) (Sekhar et al., 2009), coconut (0.91) (Alamu et al., 2010), *J. curcas* (0.901) (Belewu et al., 2010), shea butter (0.902) (Asuquo and Anusiem, 2010) and fluted pumpkin (0.908) (Ibeto et al., 2011). The relative density of *A. precatorius* seed oil is higher than the relative density specified by fuel standard. Therefore, using the oil as a biodiesel results in the delivery of a slightly greater mass of fuel which may influence engine output power since fuel injection equipment operates on a volume metering system.

The kinematic viscosity of *A. precatorius* seed oil was 23.4 mm^2/s which is above the limits of Thailand ASTM (1.9-8.0 mm^2/s) fuel standard. This value was found to be lower when compared to that of neem oil (44.00 mm^2/s) (Sekhar et al., 2009) and coconut oil (43.30 mm^2/s) (Alamu et al., 2010), but higher than that of *Jatropha curcas* oil (17.00 mm^2/s) (Wilson, 2010). Viscosity is a measure of the internal fluid friction or resistance of oil to flow, which tends to oppose any dynamic change in the fluid motion. Its value affects the atomization of fuel upon injection into the combustion chamber thus resulting to the formation of engine deposits. The high viscosities of

Table 2. Biodiesel physicochemical and fuel properties.

Parameter	Amount	Standard limits	Petrodiesel
FAME yield (%)	86.1 ± 0.01	-	-
Relative density	0.889	0.86 - 0.9[a]	0.84
Kinematic viscosity @ 40°C (mm^2/s)	3.34 ± 0.01	1.9 - 6[a], 1.9 - 8[bc]	2.98
Flash point (°C)	137 ± 0.02	Min. 130[d]	74
Heat of combustion (MJ/Kg)	38.29 ± 0.15	Min. 35[c]	42.85
Refractive index	1.457 ± 0.02	Max. 1.479[a]	1.483
Cloud point (°C)	- 2 ± 0.55	-3 to 12[d]	-16
Pour point (°C)	1 ± 0.23	-15 to 10[e]	-12
Cold filter plugging (°C)	- 4 ± 0.1	-20 to 5[f]	-18
Ash content (%)	0.088 ± 0.05	Max. 0.02[d]	0.02
Cetane number	58.3 ± 0.06	Min. 51[f]	49
Acid value (mg KOH/g)	0.281 ± 0.06	Max. 0.8[d]	0.35
Iodine value (mgI_2/g)	52.43 ± 0.2	Max. 120[f]	3.05
Peroxide value (mEq/Kg)	3.45 ± 0.17	-	0.2

Superscripts represent test methods; [a]ASTM D445, [b]Thailand ASTM, [c]EN14213, [d]ASTMD6751, [f]EN14214.

Table 3. Fatty acid profile of Abrus seed oil biodiesel.

Fatty acid methyl esters (FAME)	Amount (%) carbon molecules
Saturated FAME	50.86
Unsaturated FAME	49.1
Methyl decanoate	1.27 C_{10}
Methyl undecanoate	10.49 C_{11}
Methyl dodecanoate	6 C_{12}
Methyl eicosadienoate	17.16 $C_{20:2}$
Methyl tetradecanoate	4.59 C_{14}
Methyl palmitoleate	31.94 $C_{16:1}$
Methyl heptadecanoate	4.84 C_{17}
Methyl stearate	14.22 C_{18}
Methyl pentacosylate	3.7 C_{25}
Methyl messilate	5.75 C_{30}

vegetable oils are reduced through the process of trans-esterification (Alamu et al., 2008). Therefore, *A. precatorius* seed oil cannot be used as a fuel in engine due to high kinematic viscosity of it which would cause poor atomization of the oil in the combustion chamber thus resulting to the formation of engine deposits.

The results of the chemical characterizations of the oil extract showed that *A. precatorius* seed oil has iodine value of 57.43 mgI_2/g which classifies the oil as a non-drying oil. Non-drying oils have iodine value less than 100 (Asuquo et al., 2012). This value was lower than those of corn seed oil (103), sunflower oil (110), castor oil (83) and rubber seed oil (134.51) (Asuquo et al., 2012). Iodine value is the measure of unsaturation of fats and oil. The higher iodine value of fats and oil indicates high unsaturation and susceptibility to rancidity. This implies that the oil cannot be preserved for a long period of time. Iodine value is also used to measure the chemical stability property of oil against oxidation, and the higher the iodine value, the higher the number of double bond. *A. precatorius* seed oil has low iodine value compared to other non-drying oil such as castor oil (83) due to long chain fatty acid and higher saturated fatty acid. The low iodine value of *A. precatorius* seed oil is vital due to the fact that heating highly saturated fatty acids results in poor polymerization of glycerides which could reduce the formation of deposits in engines. It also reduces the rancidity in the oil, and makes it possible to preserve the oil and its biodiesel for long period of time.

The 4.1 mEq/kg peroxide value of *A. precatorius* seed oil showed the oxidative stabilities of the seed oil. The higher the peroxide values of oil, the greater the development of rancidity. This low peroxide value of *A. precatorius* seed oil must have resulted from proper handling of the oil during extraction, and also regulated heat treatment of the oil during extraction since heat favours oxidation of fatty acids thereby increasing the formation of peroxides (Oluba et al., 2008). The low peroxide value of *A. precatorius* seed oil showed that the oil if exposed without addition or treatment with antioxidants will be stable over a long period of time and protected against rancidity and peroxidation.

The acid value of *A. precatorius* seed oil was shown to be 5.74 mg KOH/g. This acid value was higher than those of soybean oil (2.67 mg $KOHg^{-1}$), rape seed oil (2.88 mg $KOHg^{-1}$) (Jordanov et al., 2007), palm oil (3.8 mg $KOHg^{-1}$) (Christian, 2006), but lower than that of *Cucurbita luffa* (36.47 mg KOH/g), *Brachystegia eurycoma* (27.08 mg KOH/g) (Ibeto et al., 2011). The high acid value of *A. precatorius* seed oil indicates that the oil is non-edible, but may be useful for the production of soaps, paints and biodiesel. Acid value measures the presence of corrosive free fatty acids and oxidation products. This is actually an important variable in considering the quality of oil because the lower the free fatty acid, the better the quality of oil with respect to its consumption (Balley, 1982). The percentage free fatty concentration of *A. precatorius* seed oils (2.87) is higher than the maximum limit of 2.0% (Codex Alimentairus Commission, 1993). Vegetable oils containing high free fatty acids have significant effects on the trans-esterification with methanol using alkaline catalyst. It interferes with the separation of fatty acid ester and glycerols (Ma and Hanna, 1999). The high acid value of *A. precatorius* seed oil suggests high levels of hydrolytic and lipolytic activities in the oils. Thus, this indicates that *A. precatorius* seed oil would be better converted to biodiesel using the two stage processes of esterification and trans-esterification to reduce the formation of soap.

The saponification value of *A. precatorius* seed oil was shown to be 227.8 mg KOH/g. This is higher than those of common seeds such as *J. carcus* (202.40 mg KOH/g), linseed oil (195 mg KOH/g) (Singh and Siroj, 2009) and fluted pumpkin (151.48 mg KOH/g), but lower than that of coconut (257 mg KOH/g) (Kyari, 2008). The higher saponification value of *A. precatorius* seed oil indicates the presence of high percentage of free fatty acids in the oil, and therefore implies the possible tendency to soap formation and difficulties in separation of products if utilized for biodiesel production. This would also suggest that using the oils for biodiesel production would lead to very low yields in the methyl esters.

The biodiesel yield (86.1%) of *A. precatorius* seed oil obtained is low when compared to conventional canola methyl ester (93.5%) (Leung and Guo, 2006), rape seed methyl ester (94%) (El-Diwani et al*.,* 2009), but higher when compared to *J. carcus* seed methyl ester (80.2%) (Adebayo et al., 2011) and soybean ethyl esters (66.8%) (Hossain et al*.,* 2010). The biodiesel yield of the oil was low compared to that which was specified in EN14214. This low value of *A. precatorius* seed biodiesel could be due to the formation of soap which was so prominent during the conversion process as a result of high saponification and acid values of the oil.

The properties of the triacyglycerol and the biodiesel fuel are determined by the amounts of each fatty acid that is present in the molecules. Chain length and number of double bonds determine the physical characteristics of both fatty acid alkyl ester and triacyglycerol (Mittelbach and Remschmidt, 2004). Trans-esterification does not alter the fatty acid composition of the feedstocks and this composition plays an important role in some critical parameters of the biodiesel such as cetane number and cold flow properties (Ramos et al., 2009). There are three main types of fatty acids that are present in triacyglycerols; namely, the saturated (Cn:0), monounsaturated (Cn:1) and polyunsaturated with two or three double bonds (Cn:2,3). Various vegetable oils are potential feedstocks for the production of a fatty acid methyl ester or biodiesel, but the quality of the fuel will be affected by the oil composition. Vegetable oils that are rich in polyunsaturated fatty acid such as linoleic and linolenic acids as found in soybean and sunflower oil tend to give methyl ester fuels with poor oxidation stability (Gunstone, 2004). Vegetable oil with low degree of unsaturation tends to have high freezing point. This oil has poor flow characteristic and may become solid (for example, palm oil) at low temperatures though they may perform satisfactorily in hot climates (Gunstone, 2004). The predominant fatty acid alkyl ester of *A. precatorius* seed biodiesel consists of monounsaturated fatty acid methyl ester (31.94%), polyunsaturated fatty acid methyl ester (17.16%) and saturated fatty acid methyl ester (50.86%). According to the European standard, the concentration of linolenic acid and acid containing four double bonds in FAMEs should not exceed the limit of 12 and 1%, respectively. *A. precatorius* seed biodiesel does not contain linolenic acid and fatty acid containing three or four double bonds. The long chain and higher saturated fatty acids obtained showed that *A. precatorius* seed biodiesel has higher heat of combustion, high cetane number while high concentration of methyl eicosadienoate ($C_{20:2}$,17.16%) showed that *A. precatorius* seed biodiesel has a low cold temperature properties and low viscosity. It has been reported by Rodrigues et al. (2006) that more than one unsaturation in the carbon chain lowers both the crystallization temperature and the viscosity by hindering molecular packing.

The relative density obtained for the *A. precatorius* seed biodiesel (0.889) was in agreement with the specified value reported (ASTMD445), which range from 0.860 to 0.90 for biodiesel. The trans-esterification of *A. precatorius* seed oil to biodiesel reduced the density from 0.922 to 0.889 g/cm^3. Density is a very important property of biodiesel because fuel injection equipment operates on a volume metering system; hence a higher density for biodiesel results in the delivery of a slightly greater mass of fuel. Thus, changes in the fuel density will influence engine output power due to different mass of fuel injected. The relative density of *A. preatorius* seed biodiesel is higher than palm oil methyl ester (0.878) (Jansri and Prateepchaikul, 2011), but lower than castor oil biodiesel (0.917 g/cm^3) (Encinar et al., 2010).

The kinematic viscosity of *A. precatorius* seed biodiesel (3.34mm^2/s) was found to be within the limits of U.S.A ASTM D6751 (1.9 - 6.0 mm^2/s) fuel standard, and Thailand ASTM (1.9-8.0 mm^2/s) biodiesel fuel standard. The production of biodiesel from *A. precatorius* seed oil reduced the viscosity of the oil from 23.4 to 3.34 mm^2/s. Viscosity of biodiesel depends on the structural composition of the parent or virgin oil used in biodiesel production. Viscosity increases with the number of CH_2 moieties in the fatty ester chain. For example, methyl esters of lauric, myristic, palmitic and stearic acids have kinematic viscosities of 2.43, 3.30, 4.38, and 5.85 mm^2/s, respectively (Knothe and Steidley, 2005). It also decreases with an increasing degree of unsaturation as evidenced by comparison of the methyl esters of stearic (5.85 mm^2/s), oleic (4.51 mm^2/s), linoleic (3.65 mm^2/s) and linolenic (3.14 mm^2/s) (Knothe, 2008). The higher viscosity of *A. precatorius* seed biodiesel than petro diesel is due to long chain length of fatty acid alkyl ester of *A. precatorius* seed biodiesel ($C_{16:1}$), (C_{18}), ($C_{20:2}$), (C_{30}) and higher saturated fatty acid alkyl ester (50.86%).

The flash points of *A. precatorius* seed oil biodiesel (137°C) was found to be above the minimum value (120°C) of the EN 14214 biodiesel fuel standard and the minimum value (130°C) of the ASTMD6751 biodiesel fuel standard. Flash point of a fuel is the temperature at which it ignites when exposed to a flame. Therefore, flash point is an important parameter to be considered in the handling, storage and safety of a biodiesel. The flash point of *A. precatorius* seed oil biodiesel was comparable to that of jatropha biodiesel (135°C) but lower than that of palm kernel oil biodiesel (167°C), (Alamu et al., 2008). This value was however, higher than that of neem seed oil biodiesel (120°C) and more importantly, extremely higher than that of petro diesel (74°C). The relatively higher flash point value of *A. precatorius* seed oil biodiesel showed that it does not contain methanol contaminants which would have lowered the flash point and it is of prime importance for storage and transport-tation of the fuel.

The heats of combustion of *A. precatorius* seed biodiesels (38.28 MJ/Kg) was comparable to those of soybean oil biodiesel (38.1 MJ/Kg) (Rashid et al., 2008), but greater than neem seed oil biodiesel (35.2 MJ/Kg) (Sekhar et al., 2009). The heat of combustion is important parameter for estimating fuel consumption; the greater the heat of combustion, the lesser the fuel consumption (Knothe et al., 2006). The heat of combustion or heating value is not specified in the biodiesel standards ASTM D6751 and EN14214. However, a European standard for using biodiesel as heating oil, EN 14213, specifies a minimum heating value of 35 MJ/kg. The heat of combustion increases with an increasing chain length and decreases with an increasing unsaturation. For instance, methyl stearate possesses greater energy content (40.07 MJ/kg) than methyl laurate (37.97 MJ/kg) (Knothe, 2008). Therefore, the energy content of fatty acid alkyl esters is directly proportional to chain length since longer-chain fatty acid alkyl esters contain more carbons but a similar number of oxygen atom. The heat of combustion of *Abrus* seed biodiesel was higher than the minimum value for heating biodiesel. The high heat of combustion of *A. precatorius* seed biodiesel is due to long chain length of fatty acid alkyl ester of *A. precatorius* seed biodiesel ($C_{16:1}$), (C_{18}), ($C_{20:2}$), (C_{30}) and higher saturated fatty acid alkyl ester (50.86%), underscoring the importance of Abrus seed oil-derived biodiesel as a useful petrol diesel supplement. The lower values of heats of combustion of *A. precatorius* seed biodiesel, and conventional biodiesels in general, when compared to that of petro diesel might be due to higher oxygen content and lower carbon-to-hydrogen ratio in the former than in the later.

The refractive index of *A. precatorius* seed biodiesel was 1.4567 which meets the value of ASTMD6751 of 1.479 maximum (ASTM International, 2002). It was lower than the refractive index of petro diesel (1.4831). The refractive index which is the ratio of the velocity of light in vacuum to the velocity of light in a medium is an indication of the level of saturation of the biodiesel (Oderinde et al., 2009). As chain length of fatty acid and degree of unsaturation increases, the refractive index increases. Refractive index is widely used in quality control to check for the purity and adulteration of fatty materials (Hoffmann, 1986). The lower value of refractive index of *A. precatorius* seed biodiesel than petro-diesel indicates higher purity and saturation of the biodiesel than petrodiesel.

The low temperature operability properties of *A. precatorius* seed biodiesel determined by parameters such as cloud point (CP), pour point (PP) and cold-filter plugging point (CFPP) were 2, 1 and -4°C, respectively. The cloud point of *A. precatorius* seed biodiesel is within specified range of ASTMD6751 (-3 to 12°C) biodiesel fuel standard, but higher than petro-diesel (-16°C). The pour point (PP) of *A. precatorius* seed biodiesel was within specified range of ASTMD97 (-15 to 10°C) biodiesel fuel standard, but higher than petro diesel (-12°C) while cold-filter plugging point (CFPP) is within EN 14214 (-20 to 5°C) international biodiesel fuel standards, but higher than petrodiesel (-18°C). The CP is the temperature at which the first solids become visible when cooling a diesel fuel; the PP is the temperature at which the fuel ceases to flow while cold filter plugging point is the temperature at which the diesel fuel blocks the filter device as a result of the formation of crystal agglo-merates. These parameters are related to the cold engine start, and should be sufficiently low because when the biodiesel freezes, the engine will not start (Encinar et al., 2005). An increase in chain length of saturated fatty acid methyl ester and decrease in degree of unsaturation of fatty acid result in a corresponding increase in melting point (Lee et al., 1995). For instance, compounds of similar chain length but increasing levels of unsaturation display lower melting point as evidenced by the melting point of C18:0 (methylstearate, melting point 39°C), C18:1 (methyloleate, melting point -20°C), C18:2 (methyl linoleate, melting point -35°C), and C18:3 (methyl linolenate, melting point -52°C) (Lee et al., 1995). The cloud point, pour point and cold filter plugging point of *A. precatorius* seed biodiesel (49.1% unsaturated fatty acid methyl ester and 50.86% saturated) are lower than palm oil methylester (49.9% saturated and 47.7% unsaturated) (CP = 13°C,PP=16°C) (Dunn, 2005), karanja oil methylester (68.23% unsaturated and 26.04%saturated) (CP = 3.4°C, PP = 6°C) (Bobade and Khyade, 2012), tallow oil methyl ester (55.3%unsaturated and 39.6% saturated) (CP = 17°C,PP =15°C,CFPP = 9°C) (Dunn, 2005). The lower cold temperature operability properties of *Abrus precatorius* seed biodiesel are due to high concentration of polyunsaturated fatty acid ($C_{20:2}$ =17.16%wt) of fatty acid alkyl ester of *A. precatorius* seed biodiesel. It has been reported by Rodrigues et al. (2006) that more than one unsaturation in the carbon chain lowers the cold temperature operability properties by hindering molecular packing.

The value of sulfated ash obtained for *A. precatorius* seed biodiesel 0.09% is slightly higher compared to the standard specified 0.02% max (ASTMD6751). Ash content describes the amount of inorganic contaminants, such as abrasive solids and catalyst residues and the concentration of soluble metal soaps contained in a fuel sample. It has higher ash contents than jatropha seed biodiesel [0.06%] (Adebayo et al., 2011). However, the value was higher than that obtained for diesel fuel 0.02%. The slightly higher ash contents of *A. precatorius* seed biodiesel was due to the contamination with metals from water used in purification of the biodiesel and the use of crude oil.

A. precatorius seed oil biodiesels had cetane number of 58.30 which is well above the minimum value of the ASTM D6751 (40 minimum) and EN 14214 (51 minimum) international biodiesel fuel standards. Cetane number (CN) is widely used as diesel fuel quality parameter related to the ignition delay time and combustion quality. High cetane numbers help to ensure good cold start properties and minimize the formation of white smoke. The CN of an individual compound depends upon the structure of the compound. The CN increases with an increasing chain length (that is, methyl esters of lauric (CN 67), palmitic (CN 86) and stearic (CN 101) acids), and also increases with increasing saturation of fatty acid (that is, methyl esters of stearic (101), oleic (59), linoleic (38), and linolenic (23) acids) (Knothe et al., 2006). The cetane number of *A. precatorius* seed oil biodiesel was found to be higher than some conventional biodiesels such as soybean oil biodiesel (49) (Ramos et al., 2009) and sunflower oil biodiesel (55) (Rashid et al., 2009). Thus, the higher cetane number of *A. precatorius* seed oil biodiesel when compared to petro-diesel (49) and other conventional biodiesel is due to long chain length of fatty acid alkyl ester of *A. precatorius* seed biodiesel ($C_{16:1}$), (C_{18}),($C_{20:2}$), (C_{30}) and higher saturated fatty acid alkyl ester (50.86%).

The chemical characteristics of *A. precatorius* seed biodiesel showed that the acid value of *A. precatorius* seed oil biodiesel was 0.281 mg KOH/g which is within the limits of the ASTM D6751 (0.8 mg KOH/g maximum) and EN 14214 (0.5 mg KOH/g maximum) biodiesel fuel standards. However, the acid value of *A. precatorius* seed oil biodiesel is lower than that of sunflower oil biodiesel (0.40 mg KOH/g) and *Jatropha* oil biodiesel (0.4 mg KOH/g maximum) (Wilson, 2010). The lower acid value of *A. precatorius* biodiesel when compared to other conventional biodiesels and petro-diesel (0.350 mg KOH/g) indicates that the fuel has low levels of free fatty acids in the biodiesel, which also suggests low levels of hydrolytic and lipolytic activities in the biodiesel.

A. precatorius seed biodiesel has iodine value (52.43 mgI_2/g) which is lower than the standard iodine value for biodiesel of 120 by Europe's EN 14214 specification. Iodine number is a measure of total unsaturation within a mixture of fatty acid material. The limitation of unsaturation of fatty acid is vital due to the fact that heating highly unsaturated fatty acids results in polymerization of glycerides which could lead to the formation of deposits (Mittelbach, 1994). The iodine value of *A. precatorius* seed biodiesel is lower than *Jatropha* oil biodiesel (101 mgI_2/g) (Tamalanpundi et al., 2008). The lower iodine value of *A. precatorius* seed biodiesel will allow it to be used as alternative fuel for diesel engine that leaves very small carbon deposits on the injector and in combustion chamber thus improving life of components and increasing inter service period.

The peroxide value of *A. precatorius* seed oil biodiesel was 3.45 mEq/kg. It is higher than petro-diesel (0.20 mEq/kg). The peroxide value measures the presence and amount of unstable hydroperoxide which is a portion of deteriorated biodiesel formed when oxygen from the air react with fatty esters. The peroxide value of the biodiesel can be reduced by adding of antioxidants (Frankel, 2005). The low peroxide value of *A. precatorius* seed oil biodiesels indicates little peroxidative rancidity of the biodiesels as a result of proper handling.

Conclusion

In this present studies, the physiochemical characterization of *A. precatorius* seed oil and its biodiesel properties have highlighted the potentials of the oil and its biodiesel as a very good resource. Due to high availability of the seed, ability of the seed to grow easily as well as the poisonous nature of the seed, it is considered good resource for industrial process. The oil yield of the seed which is quite small compared to other high oil yielding seeds could be modified genetically and the yield can also be improved by using other extraction methods. *A. precatorius* seed oil has high saponification value showing that it is also good for soap production. *A. precatorius* seed biodiesel is suitable when compared to conventional biodiesels and petro-diesel. Considering the percentage biodiesel yield in this study, the fatty acid methyl esters (FAME) confer on the biodiesel improved fuel qualities. Some of the fuel properties investigated had shown, to a reasonable extent, that quality biodiesel can be produced from *A. precatorius* seed. Hence, improving *A. precatorius* seed cultivation more than its present state via mechanization and genetic engineering can, therefore, facilitate its incorporation as an additional feedstock for biodiesel.

Conflict of Interests

The author(s) have not declared any conflict of interest.

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