

Article

Potential of *Chrozophora tinctoria* Seed Oil as a Biodiesel Resource

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Featured Application: Produced biodiesel through this research work can be used as a suitable fuel source instead of petroleum-derived fuels.

Abstract: Biodiesel is a renewable fuel that has been widely used in recent years. There are various resources used as biodiesel feedstocks, including animal fats, waste oils, and vegetable oils. In the present study, *Chrozophora tinctoria* seed oil is introduced as a new biodiesel feedstock. *C. tinctoria* is a weed and non-edible plant. So, the primary cost of this resource is very low, and hence it can be considered as a biodiesel source. This plant can also grow in most weather conditions. In the present study, the research team tried to produce biodiesel from *C. tinctoria* seeds through a transesterification reaction. To intensify the transesterification reaction, an ultrasonic device was used. In order to perform the transesterification reaction, potassium hydroxide was used as a catalyst. Important parameters, such as the reaction temperature, reaction time, molar ratio of methanol, and concentration of the catalyst, were adjusted. Based on the adjusted conditions, a biodiesel yield of 84% was attained. The properties of the *C. tinctoria* biodiesel was compared with the American Society for Testing and Materials (ASTM) standard. The results show the properties of a biodiesel: the density, kinematic viscosity, pour point, flash point, cloud point, and acid number are 0.868 g/cm³, 3.74 mPa, -7 °C, 169 °C, 4 °C, and 0.43 mg, respectively. The specification properties of *C. tinctoria* biodiesel can thus pass the requirement of the ASTM standard. So, *C. tinctoria* seed oil can be used as a suitable fuel source instead of petroleum-derived fuels.

Keywords: biomass; biodiesel; *Chrozophora tinctoria*; seed oil

1. Introduction

Due to an increasing global population, the use of liquid fuels, especially in the transportation sector, has grown. Reduction of fossil fuel resources, increasing greenhouse gas emissions, and increasing oil prices created the need to find sustainable, renewable, efficient, cost-effective, and low-emission alternatives [1,2]. One of the alternative fuel types is biodiesel, which was introduced as a suitable fuel instead of conventional diesel fuel for using in heating systems and specifically for diesel engines [3,4]. Chemically, biodiesel is a combination of fatty methyl ester acids that are produced by the reaction of diglycerol with alcohols in the presence of a catalyst along a process called transesterification [5,6].

Biodiesel is made from bio-oils. The number of carbons in the carbon chain of diesel fuel is similar to vegetable oils (containing 14–18 carbons). Because of the structural properties of biodiesel, this resource can play an important role as a viable alternative to conventional (diesel) energy [7]. Generally, the production price of biodiesel is high, being almost twice that of diesel's price. The price of biodiesel production consists of two main components that are the price of the raw material and price of the biodiesel production process [8,9]. Although, large amounts of oils, such as restaurant waste oil and animal fat, can be used to produce the biodiesel, the main problem with using these resources is that the transesterification reaction of some of these resources is very difficult. As a result, the cost of biodiesel production from these materials increase [10]. One of the main issues that play an important role in the final price of the biodiesel is the price of the oil that is used as the feedstock. The appropriate way to reduce the price of the produced biodiesel is the use of non-edible oils as feedstocks in the biodiesel production process, which reduces the price of finished product. So far, several researches have been done on the application of non-edible oils as the feedstock of biodiesel [11–23], such as rapeseed [24], sea mango [25], rubber seed [26], mahua [27], *Hodgsonia macrocarpa* [28], karanja [29], moringa [30], palm [31], jatropha [32], and tree of heaven [33]. So, intensive research on bioenergy resources, including crops with less food importance, or non-edible, can help to decrease the greenhouse gas emission and control climate change. The ideal non-edible resources should be water and nutrient efficient, fast growing, have a high tolerance to environmental stresses, and low invasive potential [34,35]. One of the non-edible resources that could be a novel resource for biodiesel is *Chrozophora tinctoria* seeds.

1.1. Why *Chrozophora tinctoria*?

C. tinctoria is an annual summer and warm season weed plant [36]. It can be observed under different light, soil, and moisture conditions [37,38]. Recently, some researchers suggested that *C. tinctoria* fruit could provide a new resource of oil [39,40]. The fruit of *C. tinctoria* is like a capsule in the shape of three spheres attached to a rounded-triangular structure. Fruit of *C. tinctoria* has three angular seeds [41]. *C. tinctoria* seeds are a valuable source of oil, which can be suitable for bioenergy purposes. Moreover, *C. tinctoria* is a non-edible, weed plant, so the primary cost of *C. tinctoria* seed is very low and, hence, can be introduced as a biodiesel source. This research focuses on *C. tinctoria* as a potential bioenergy crop. The biofuel production possibilities from this crop is investigated in this research paper.

1.2. *C. tinctoria* in the World and Iran

C. tinctoria is mainly found in the central/south Asia and Mediterranean region. This plant is described as native to the different countries (Figure 1), including Afghanistan, Albania, Algeria, Bulgaria, Cyprus, Egypt, France, Greece, India, Iran, Italy, Kazakhstan, Libya, Morocco, Pakistan, Palestine, Portugal, Romania, Saudi Arabia, Spain, Tunisia, Turkey, Turkmenistan, and Yemen [42,43]. By considering the weather conditions of Iran, *C. tinctoria* can be cultivated in all the regions of Iran (Figure 1). For example, different species of the *Chrozophora* genus have been found in the different provinces in Iran, such as East-Azerbaijan [37], Chaharmahal and Bakhtiari, Markazi, Qom, Qazvin, and Isfahan [36].

Based on the above literature review on the potential of *C. tinctoria* seeds as a novel non-edible resource for biodiesel production, in the present study the application of *C. tinctoria* seed oil as a novel feedstock was experimentally investigated. In the Section 2, the experimental procedure for biodiesel production from the *C. tinctoria* seed oil is presented. It is noteworthy that ultrasonic waves were also used in this research to reduce the transesterification reaction time. Finally, in the Section 3, the properties of the biodiesel are evaluated and then the properties of the produced biodiesel are compared with the ASTM standards.



Figure 1. Potential countries for *C. tinctoria* cultivation [43].

2. Materials and Methods

2.1. Raw Materials

C. tinctoria were collected from the Fars province (Southwest of Iran), in August of 2019. A Global Positioning System (GPS) was used to mark the locations (the longitude of $40^{\circ}51' - 50^{\circ}51'$ E and latitude of $20^{\circ}30' - 30^{\circ}30'$ N, and altitude of 2890 m). The sampling was done by a randomized collection in an area of about 2000 m^2 . Matured fruits were isolated manually from the aerial parts and the seeds were separated from the fruit, then the fruits were washed with sterile water, and then dried at ambient temperature for seven days. In Figure 2, the different stages of plant sampling are shown.

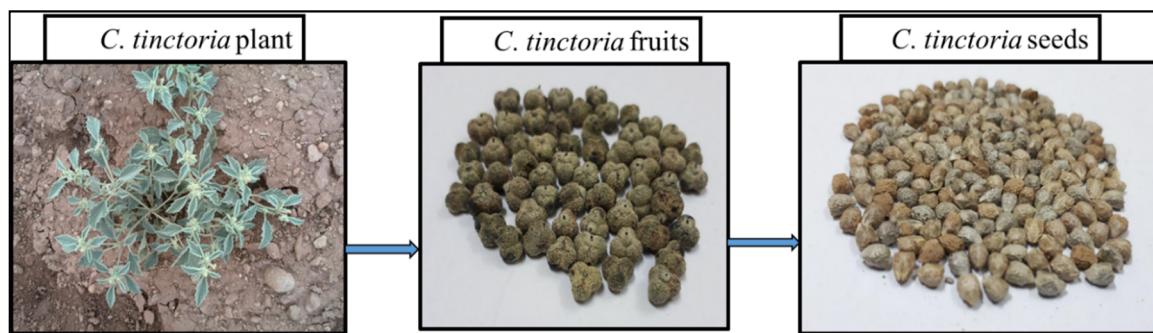


Figure 2. The plant sampling and seed separation process of *C. tinctoria*.

The seeds of *C. tinctoria* were prepared to determine their properties, and thus that of the seed oil and biodiesel. The *C. tinctoria* seeds were removed from the fruits. Seeds were powdered using a coffee grinder. Then the crushed seeds were transferred to a Soxhlet extractor to extract the oil. Oil extraction was performed using n-hexane for 8 h. The solvent was evaporated under reduced pressure at 60 °C using a rotary evaporator for 3 h.

2.2. Methodology

Fatty acids of the *C. tinctoria* seed oil were determined using Metcalfe procedure [44], and fatty acid composition were identified using a gas chromatography (Clarus580 GC) equipped with a FID detector. That equipment has been sourced from FARASOUT SANAT Eng CO, Tehran, Iran. One microliter of prepared sample was injected to the gas chromatography unit, which was equipped with a BP-30 column and FTIR detector that was kept isothermally standardized to encounter BS-EN 14103 standard. The total run time for each sample was 30 min. Fatty acid profile was determined by classifying and calculating relative peak areas.

In order to perform the transesterification reaction, an ultrasonic reactor was used. In the first step, a catalyst (e.g., potassium hydroxide (KOH)) and methanol was mixed together by a magnetic stirrer. It is noteworthy that methoxide was prepared by mixing methanol that had a molar ratio of 1:7, with 1 wt% of KOH. The prepared methoxide was added to the *C. tinctoria* seed oil. Then, the solution was transferred into the ultrasonic reactor. An ultrasonic device (Hielscher Model UP400S, USA) was applied to perform the transesterification reaction. That equipment has been sourced from FARASOUT SANAT Eng CO, Tehran, Iran. The ultrasonic device was controlled using a computer with the help of UPC software. An ultrasonic probe with a diameter of 10 mm and a height of 60 mm was used to transmit the ultrasound to the solution. The effective length of the probe that was placed inside the solution was 55 mm. The reaction time was adjusted every 5 min. The reaction temperature was adjusted to 45 ± 0.45 °C with the help of water circulating between the two reactor walls, and all experiments were performed at atmospheric pressure. After completing the reaction time, the biodiesel mixture was removed from the ultrasonic reactor. In order to stop the reaction, the biodiesel mixture was put into a cold ice bath. The mixture was stored in a separation plate and the mixture was completely separated into two phases. The lower phase was glycerin and the upper one was biodiesel. In Figure 3, the biodiesel production process is shown. After separation of the glycerin, the biodiesel should be purified. The separated part from the glycerin was washed by warm water to purify the biodiesel. In the next section, in order to remove the water from the biodiesel, the washed biodiesel was heated up for one hour in an oven (temperature was adjusted to 60 °C). Finally, a gas chromatography unit was used to identify the yield of the biodiesel. Equation (1) was used [34,45] to determine the yield of the biodiesel.

$$\text{Biodiesel yield} = \frac{\sum A - AIS}{AIS} \times \frac{MIS}{M} \quad (1)$$

where $\sum A$ is the total area under the peak of total fatty acids, MIS is the weight of the internal standard, AIS is the area under the peak for the internal standard (C19:0), and M is the weight of the sample.

After purification of the biodiesel, the most important physicochemical properties of the biodiesel, including the kinematic viscosity at 40 °C (ASTM D445), density at 15 °C (ASTM D4052), pour point (ASTM D6751), cloud point (ASTM D6751), flash point (ASTM D93), oxidation stability at 110 °C (EN 14112), acid value (ASTM D664), and cetane number (ASTM D613), were determined, based on ASTM standard [46].

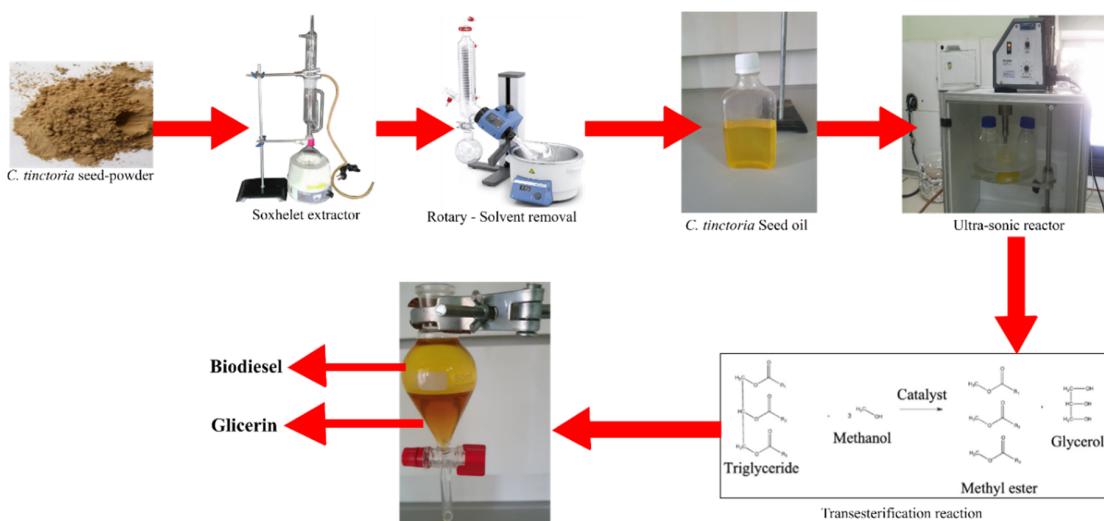


Figure 3. *C. tinctoria* biodiesel production process.

3. Results and Discussion

3.1. Seed Oil Properties

The seed and oil physico-chemical characteristics of *C. tinctoria* are shown in Table 1. The oil content was 26%. So, the oil content of *C. tinctoria* seed is appropriate, and this feedstock can thus be introduced as a feasible primary material to produce biodiesel. The kinematic viscosity of the *C. tinctoria* oil is 4.32 mPa s. The viscosity of the *C. tinctoria* oil is less than that of some other feedstocks. This property is an important characteristic of the oil as a new feedstock to produce biodiesel.

Table 1. Physico-chemical properties of the *C. tinctoria* seed oil.

Physico-Chemical Parameters	Unit	Content
Oil content	%	26
Weight of 1000 seeds	g	15
Density	g/cm ³	0.872
Kinematic viscosity	mPa s	4.32
Acid value	Mg KOH/g oil	2.5
Iodine value	g I ₂ /100 g oil	170
Water content	mg/g	50

3.2. Fatty Acid Profile

In Table 2, the fatty acid structure of the *C. tinctoria* seed oil is shown. *C. tinctoria* seed oil consists of both unsaturated and saturated fatty acids. According to Table 2, the main fatty acids of *C. tinctoria* seed oil are unsaturated fatty acids; in other words, the unsaturated fatty acid content of this oil is 91.45% of the total oil. *C. tinctoria* seed oil contains a high content of polyunsaturated fatty acids, as the main fatty acid. The main fatty acids were identified as palmitic acid (5.32%), linoleic acid (76.68%), stearic acid (3.15%), and oleic acid (13.99%).

3.3. *C. tinctoria* Biodiesel Properties

After purification of the *C. tinctoria* biodiesel, the biodiesel yield was calculated by Equation (1). The results show that the biodiesel yield from the oil is 84%. In order to investigate the properties of *C. tinctoria* biodiesel as a new resource, the physicochemical characteristics of the produced biodiesel was measured. In Table 3, the physicochemical characteristics of the *C. tinctoria* biodiesel is presented.

According to the results, it is obvious that all the physicochemical characteristics of the produced biodiesel are in agreement with the ASTM standards.

Table 2. Fatty acid composition of oil extracted from *C. tinctoria* seeds.

Fatty Acid	Content (%)
Palmitic acid (C16:0)	5.32 ± 0.047
Palmitoleic acid (C16:1)	0.11 ± 0.50
Stearic acid (C18:0)	3.15 ± 0.19
Oleic acid (C18:1)	13.99 ± 0.29
Linoleic acid (C18:2)	76.68 ± 0.16
Linolenic acid (C18:3)	0.67 ± 0.13
Σ UFA	91.45
Σ SFA	8.43
Σ MUFA	14.10
Σ PUFA	77.35
Σ UFA: Σ SFA	10.85
Σ MUFA: Σ PUFA	0.18

Note: UFA, unsaturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids.

Table 3. Properties of the biodiesel produced from *C. tinctoria*.

Parameters	Method	Limitation	Units	Content
Density at 15 °C	ASTM D 4052	0.86–0.90	g/cm ³	0.862
Kinematic Viscosity @ 40 °C	ASTMD445	1.9–6.0	mm ² /s	3.74
Flash Point, Closed Cup	D93	>130	°C	169
Cloud point	ASTM D6751	−3 to 12	°C	4
Pour point	ASTM D6751	−15 to 10	°C	−7
Acid Number	ASTMD664	0.50	mg	0.43

Viscosity is one of the important properties of biodiesel. By considering the biodiesel standard (ASTM), the viscosity of a biodiesel should be in the range of 1.9–6.0 mm²/s. The results show that the viscosity of *C. tinctoria* biodiesel is 3.74 mm²/s, which is lower than different biodiesel resources; for example, moringa (5 mm²/s) [47], karanja (6.13 mm²/s) [48], and neem (5.21 mm²/s) [47]. Another important characteristic of a biodiesel resource is its density. Density plays an important role in the fuel atomization in the combustion process of a diesel engine [49]. Based on the biodiesel standards (ASTM and EN), the range of a new biodiesel resource should be 0.86–0.9 (g/cm³). The result of the present study shows that the density of *C. tinctoria* biodiesel is 0.862 (g/cm³). By reviewing the recent studies, it is clear that the density of the *C. tinctoria* biodiesel is lower than many of the other resources, such as karanja (0.931 g/cm³) [48], moringa (0.883 g/cm³) [47], rapeseed (0.882 g/cm³) [47], and tobacco (0.888 g/cm³) [48]. Another biodiesel property that plays an important role in the corrosion of the internal parts of diesel engines, is the acid number. The ASTM standard consider a strict law for acid number. Based on the ASTM standard, the acid number of a new biodiesel should not exceed 0.5 mg KOH/g. According to the result of the present study, the acid number value of the *C. tinctoria* biodiesel is 0.41 mg KOH/g. So, this acid number is suitable for a new biodiesel resource. In this present study, other properties of the *C. tinctoria* biodiesel have been measured, including the cloud point, pour point, and flash point. The results show these properties of the *C. tinctoria* biodiesel pass the ASTM standard. It is noteworthy that the specification properties of the *C. tinctoria* biodiesel are within the ASTM standard range and that the biodiesel produced from *C. tinctoria* seed oil can be used as a suitable fuel source instead of petroleum-derived fuels.

4. Conclusions

In this research, the *C. tinctoria* seed oil, which is a weed plant, was introduced as a new resource for biodiesel production. In order to intensify the transesterification reaction, an ultrasonic device was used. Potassium hydroxide was also used as the catalyst for the transesterification reaction. Important parameters, such as the reaction temperature, reaction time, molar ratio of methanol, and concentration of the catalyst, were adjusted. Based on the adjusted condition, a biodiesel yield of 84% was obtained. The important properties of the *C. tinctoria* biodiesel were investigated. The results show that the properties of the biodiesel, namely, the density, kinematic viscosity, pour point, flash point, cloud point, and acid number, are 0.868 g/cm³, 3.74 mPa, -7 °C, 169 °C, 4 °C, and 0.43 mg, respectively. The physicochemical properties of the *C. tinctoria* biodiesel are thus in agreement with the biodiesel standard (ASTM standard). In other words, it is clear that the specifications of the physical and chemical characteristics of *C. tinctoria* biodiesel are in agreement with the ASTM standard range. So, biodiesel produced from *C. tinctoria* seed oil can be used as a suitable fuel source instead of petroleum-derived fuels.

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