

# Algae Oil Transesterification by Ultrasonic Irradiation on Sn-nano Sulfated Zirconia-Silica Catalyst

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## Authors' contributions

This work was carried out in collaboration between both authors. Author SDS designed the study, wrote the protocol, and wrote the first draft of the manuscript. Author SS managed the analyses of the study and performed the statistical analysis. Author SS managed the literature searches. Both authors read and approved the final manuscript.

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## ABSTRACT

Biodiesel production from algae oil by transesterification reaction using ultrasonic irradiation on heterogeneous catalyst was done at atmospheric condition to make the process sustainable and parsimonious. Ultrasonication reduces the reaction time as compared to the conventional methods and the solid acid catalyst gives 99.15% biodiesel yield which was obtained for oil to methanol molar ratio 1:10, catalyst 2 wt.% of oil, metal 0.4 wt.% of catalyst and 15min. reaction time. Heterogeneous catalyst and ultrasonic irradiation gives good yield and handling for biodiesel production.

**Keywords:** Transesterification; tin; solid acid catalyst; ultrasonication; biodiesel; algae oil.

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## 1. INTRODUCTION

In the new age of industrialization, fossil fuel demand is increasing rapidly for the generation of electricity and transportation. The drawback of using fossil fuels is well documented in various communications in media and scientific journals as a source of environmental problems particularly when dealing with the quality of the exhaust gases emitted from their respective combustion. Since fossil fuel reservoirs are limited so it is necessary to explore the potential for renewable, economically and environment friendly energy resources. These include solar energy, hydrogen cells and wind power. However, these technologies are still at the developing stage, and the cost of their application is very high. An alternative and renewable option is the replacement of diesel fuel by biodiesel, which consists of monoalkyl esters that are derived from organic oils, plants or animals. Biodiesel is as effective as petroleumdiesel in powering unmodified diesel engines [1] and is clean burning, non-toxic, and carbon-neutral with respect to global warming.

Presently for biodiesel production the transesterification of vegetable oil by using homogeneous acid and base catalyst such as NaOH, KOH and H<sub>2</sub>SO<sub>4</sub> were reported [2-6]. It involves high consumption of energy and the separation of the catalyst from the homogeneous reaction mixture is expensive and chemically wasteful [7]. Heterogeneous catalysts are replacing the conventional homogeneous catalysts for biodiesel production. Solid acid catalysts lead as these are easier to get separated and recycled. Additionally, they have the potential to catalyze both-transesterification of TAGs and esterification of free fatty acids. 98.53% biodiesel yield was achieved by using Na/SiO<sub>2</sub> catalyst [8]. Metal oxide of tin, magnesium, and zinc as catalysts have also been reported and leaching of metal ions was encountered in the case of basic zeolite X and ETS-10 catalysts [9-12]. Sulfated zirconia, tungstate zirconia, and sulfonated polystyrene have proved to be efficient in biodiesel production [13]. A solid acid catalyst, sometimes, faces deactivation by acidity of oil [14,15] which can be caused by partial blockage of the acid sites due to polar reagents such as glycerol. Xu et al. suggested that increase in hydrophobicity of catalyst can overcome this problem [16]. However, research activities using solid catalysts for biodiesel synthesis are limited due to low expectations about reaction rates and probable

undesired side reactions. Evidently, synthesised nano sulfated zirconia over silica core catalyst is more hydrophobic than other catalysts due to higher amount of silica [17].

Economical factor of biodiesel production from edible oil and jatropha oil is very high, so algae oil used instead of vegetable oil due to high productivity and low production cost. Under good conditions, green algae can double its biomass in less than 24 hours, additionally; green algae can have huge lipid contents, frequently over 50% [18,19]. This high yield, high density biomass is ideal for intensive agriculture and may be an excellent source for biodiesel production.

In transesterification reaction, mixing is important factor for increasing biodiesel yield. Oil and methanol are not miscible completely in biodiesel processing. Ultrasonic mixing is an effective mixing method to achieve a better mixing and enhancing liquid-liquid mass transfer [20]. Vigorous mixing increases the contact area between oil and alcohol phases with producing smaller droplets than conventional stirring [21]. Cavitation effects increase mass and heat transfer in the medium and hence increase the reaction rate and yields [22]. Ultrasonic cavitation also provides the necessary activation energy for initiating transesterification reaction. Furthermore, cavitation during sonication produces extreme local conditions and a micro environment with high temperature and high pressure, which may also create active intermediates that permit the reaction to proceed instantaneously [23].

The purpose of the present study is to utilise the self-synthesised nano sulfated zirconia over silica core [NSZS] catalyst [17] for the production of biodiesel from microalgae oil using low frequency ultrasonic irradiation. Additionally the effects of the reaction time, wave amplitude, cycle, oil to alcohol molar ratio and catalyst wt. % on the yield of biodiesel are also discussed.

## 2. EXPERIMENTAL

### 2.1 Reagents and Materials

The microalgae oil used in the transesterification reaction was non-edible and extracted from fresh water algae *Sp. neglecta*. The characteristics of algae oil are reported in Table 1. Sodium stannite trihydrate (95%) (Sigma-Aldrich). Chloroform, anhydrous sodium sulphate, methanol and standard solutions of fatty acid methyl esters (Flucka) were used.

## 2.2 Catalyst Preparation

A typical synthesis route for Nano sulfated zirconia over silica core (NSZS) was reported earlier [17]. Sn/NSZS was prepared by wet impregnation of Sn in different weight percentage on synthesised NSZS catalyst. 5 gm. catalyst extrudates was dried at 80°C for 10 hrs.in50 ml beaker and the amount of tin salt in different wt.% was calculated. For 0.4 wt.%, the amount of tin salt sodium stannite trihydrate is 0.0452 gm. Tin salt was taken in a small beaker and 3 ml double distilled water was added to it (water retention of catalyst is 0.6 ml for 1 gm NSZS catalyst). The solution so obtained was added drop wise, stirring with rod, to the catalyst extrudates. The beaker was kept for drying overnight and then for 2 hours at 120°C. The prepared catalyst was calcined at 600°C for 4 hours and designated as Sn/NSZS. Other catalysts were also prepared using this method by varying the wt. % of metal and calcination temperature.XRD of NSZS and 0.4Sn/NSZS are shown in Fig. 2.

## 2.3 Algae Oil Extraction

Oil extraction from biological materials is performed by physical method followed by chemical or a combination of the two. For oil extraction from microalgae *Sp. neglecta*, the process is usually accomplished with mechanical cell disruption followed by solvent extraction using soxhlet apparatus [24]. The mechanical cell disruption was carried out by ultrasonication. An ultrasonic probe consists of a transducer and was used to disrupt small volumes of cells by

sound waves which turn into small bubbles. The formation and cavitation of these bubbles produce shock waves that rupture the cells. The algae biomass (500 g) was mixed and homogenized with 2.5 L of methanol, 1.25 L of chloroform and 500 mL of water. The mixture was subjected to ultrasonic energy for 40 min. 1.25 L Chloroform and 1.5% w/v sodium sulfate were added to the mixture and sonicated for 20 min. The solvent extraction was done using a polar and a non-polar solvent to extract the corresponding lipid fractions from the cells. The extraction was carried out at ambient temperature (25°C). The scheme of oil extraction and physico-chemical properties of algae oil are given in Fig. 1 and Table 1 respectively.

## 2.4 Characterization

### 2.4.1 X-ray powder diffraction (XRD)

XRD of parent NSZS and Sn/NSZS catalyst after calcination at 600°C was measured by X-ray powder diffractometer (model RigakuDmax-IIIIB) Fig. 2. The measurements were conducted in a continuous  $2\theta$  scan refraction mode using Cu  $K\alpha/40$  kv/30 mA radiation ( $\lambda = 1.54056\text{\AA}$ ). The samples were scanned in  $2\theta$  range of 0–60 degree at a scanning rate of 4.0 deg./min. Crystallite size of tetragonal phase was determined from the characteristic peak ( $2\theta = 30.150$  for the (111) reflection) by using Scherrer formula with a shape factor ( $K$ ) of 0.9 as below:

$$\text{Crystallite size} = K.\lambda / W. \cos \theta$$

Where,  $W = W_b - W_s$ ;

**Table 1. Physicochemical characterization of algae biodiesel and its compression with standards specifications**

Properties	Unit specifications	Specification (biodiesel)	Specification (biodiesel)	Algae biodiesel
	ASTM	ASTM	BIS India	
Density at 15°C	kg/m <sup>3</sup>	860-900	860–900	889.3
Kinematic viscosity at 40°C	mm <sup>2</sup> /s	1.9–6.0	2.5–6.0	5.16
Acid value	mg.KOH/gm	<0.8	<0.5	0.27
Ash content	%wt.	<0.02	<0.02	0.02
Cetane number		>47	>51	54
Flash point	°C	>100	>120	148
CFPP	°C	–	–	+4
Total sulphur	ppm	<500	<500	103
Water content	ppm	<500	<500	223
Oxidation stability	IP, h	3 min.	–	5
Mono glycerides	% mass	0.8	<0.8	<0.1
Di glycerides	% mass	0.2	<0.2	<0.07
Tri glycerides	% mass	0.2	<0.2	<0.9

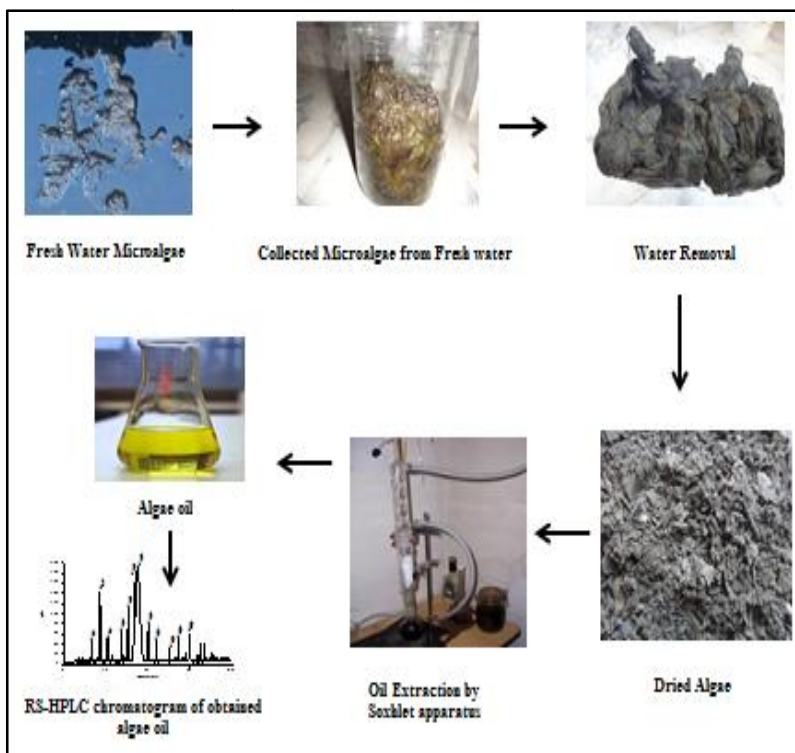


Fig. 1. Scheme of algae harvesting and oil extraction

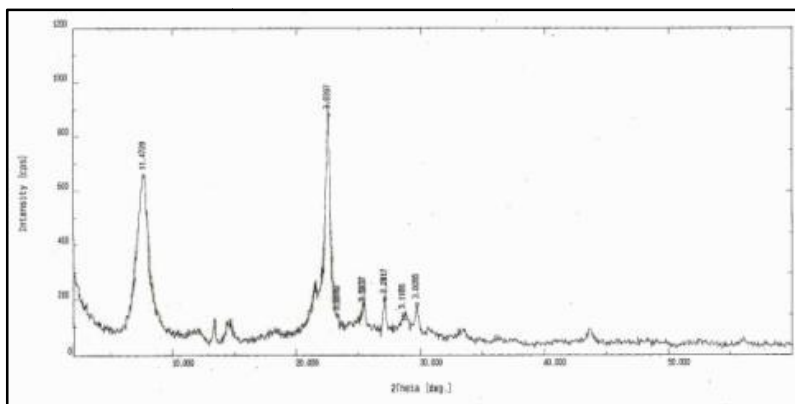


Fig. 2. XRD patterns 0.4 Sn/NSZS catalyst, calcined at 600°C

### 2.4.2 HPLC analysis

HPLC was used to analyse the purity, conversion and FAME composition of the algae oil sample. The reverse phase high performance liquid chromatography (RP-HPLC) separates different components according to their polarity. The chromatographic apparatus consisted of a model 600 pump with 600 controller, 2996 photodiode array detector and a novapak® 3.9 × 150 mm column with guard column of dimension 3.9 × 20

mm, both packed with C<sub>18</sub> particle with diameter 4 μm. (All from waters, Milford, USA). RP-HPLC method has a flow rate of 1 ml/min, an injection volume of 5 μl, a column temperature of 45°C and the UV detection is carried out at 215 nm. A 40 min gradient mobile phase (15% H<sub>2</sub>O+85% CH<sub>3</sub>OH) in 10 min, 100% CH<sub>3</sub>OH in 0 min, (60%CH<sub>3</sub>OH+15% hexane+25% propane-2-ol) in 30 min and for the last 10 min the system is back to initial state. The above method is used for the separation and determination of the compound

produced during the methanolysis of algae oil in all the experiments [25]. Fig. 3 show the HPLC chromatogram of glycerides (mono, di and tri) present in extracted algae crude oil and Fig. 4 show the methyl esters present in biodiesel produced from algae oil.

**2.4.3 Physico-chemical analysis**

Physico-chemical analysis of algae oil and algae biodiesel (Table 1, Table 2 respectively) was done by various methods. The physico-chemical parameters of algae oil such as pH, viscosity and density were determined by standard methods of analysis (AOAC, 1995). The Physico-chemical properties of algae biodiesel such as density was determined using density bottle, flash point was measured by the method of ASTM D93 using

Pensky-Martens closed cup tester, the Kinematic Viscosity at 400°C was measured by ASTM D445 method using a calibrated Viscometer with a calibration constant of 0.1057, the Sulfated Ash content was determined according to the ASTM D847 method and the carbon residue was measured using the ASTM D524 method.

**2.5 Transesterification of Algae oil [Sp. neglecta] Using Ultrasonic Irradiation**

An ultrasonic procedure (UP 200S from Hielscher ultrasonic GmbH) was used to perform the transesterification of algae oil in a 25ml Erlenmeyer type flask. The ultrasonic processes operate at 200 W and 24 kHz frequency.

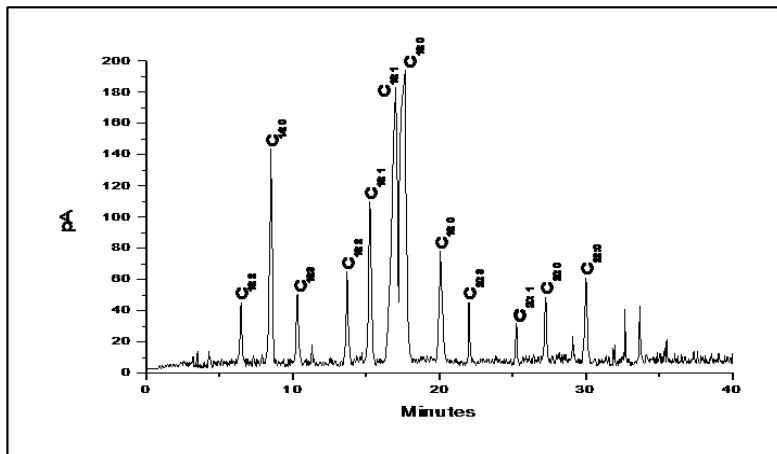


Fig. 3. RP-HPLC chromatogram of glycerides (mono, di and tri) in extracted crude algae oil

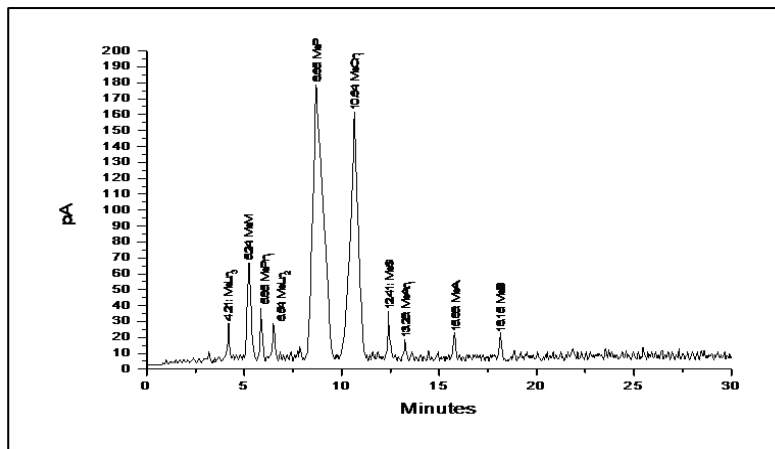


Fig. 4. RP-HPLC chromatogram of methyl esters in biodiesel

**Table 2. Physicochemical analysis of algae oil (*Sp. neglecta*)**

Properties	Unit	Value
Lipid yield	Wt.% dry mass	19.32%
pH	-	7
Density	g/cm <sup>3</sup>	0.889
Viscosity at 40°C	mm <sup>2</sup> /sec	5.67
Water content	%	0.06
Iodine value	mg/g.	62
Saponification value	mg/g.	161.4
Free fatty acid	%	1.63
Acid value	mg KOH/g	0.46 (after pre-treatment)

The amplitude and cycle were adjustable from 20% to 100% and from 0.1 to 1 cycle per ton, respectively. The ratio of algae oil to methanol at 1:10 was used in the reaction. The mixture was introduced to ultrasound waves by S7 type sonotrode submerged up to 25 mm into the solution. The ultrasonic wavecycle and its amplitude as well as the time of the reaction were adjusted by the controller at autogenously temperature. After completion of the reaction, the reaction mixture was transferred into a rota vapour flask to recover unreacted MeOH and then filter for the separation of catalyst. The filtrate was transferred into a separating funnel for phase separation. The ester mixture formed the upper layer and the glycerol form lower layer. The traces of glycerol in ester layer were washed with hot water and the biodiesel dried over the anhydrous sodium sulphate or under vacuum. At last it was analysed by reverse phase HPLC.

### 3. RESULTS

In this study an approach was to minimize the values of all the parameters which were higher in conventional methods like oil to MeOH ratio 1:15-1:20, catalyst percentage greater than 6% of oil, reaction time more than 10 hrs., reaction temperature 50-80°C and use of homogenous catalyst. Our investigations are centralized on the use of solid acid catalyst i.e. tin doped nano sulfated zirconia over silica (Sn/NSZS) for biodiesel production by the transesterification of algae oil with methanol using ultrasonic technique. The optimal conditions for this reaction are: 1:10 oil to MeOH ratio, catalyst amount 2 wt. % of oil, metal loaded 0.4 Wt.% of catalyst, ultrasonic frequency 24 KHz /200 W, wave amplitude 50%, pulse 0.7 a cycle per second and reaction time 15 minutes. Final product yield obtained was 99.15% biodiesel on these optimal conditions.

## 4. DISCUSSION

### 4.1 Effect of Metal

In the process of transesterification, ultrasonic wave produce cavitations which directly interact with catalyst at upper surface where metal atom is attached on active sites. Cavitation gives suitable atmosphere for the reaction of oil and methanol mixture on these sites. The weight % of metal loaded on catalyst is varied from 0.1% to 1.0%. However 0.4% is selected for this study as it gives maximum conversion. The extent of conversion on different metal wt. % catalyst is given in Fig. 5. It is evident that, if we increase wt. % of metal beyond 0.4%, there is no surge in conversion and it slightly decreases.

### 4.2 Effect of Catalyst Weight

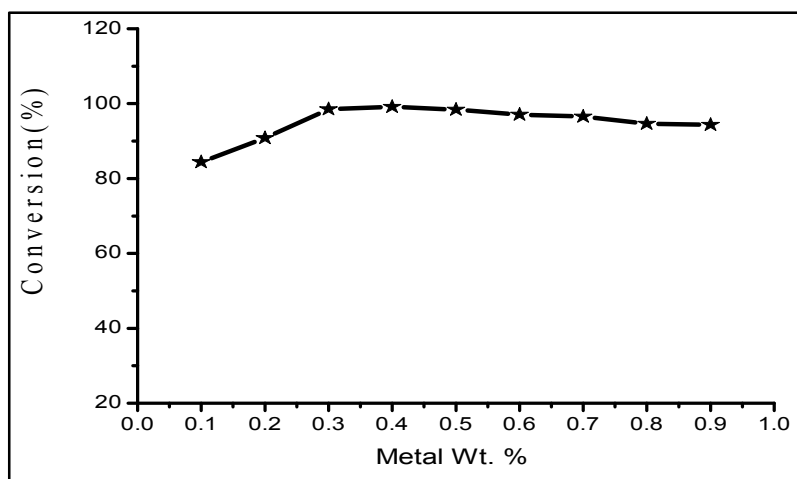
In heterogeneous catalysis, where the catalyst is in a different phase to the reactants, ultrasonic dispersion increases the surface area available to the reactants. In essence, cavitation increase the efficiency of catalyst and optimum conversion can be attained even using lower amount of catalyst. The weight % of catalyst to oil is varied from 1% to 5% (based on the weight of oil) and 2% is selected as it gives maximum conversion. If the amount of catalyst is further increased, there is no increase in the conversion of biodiesel but a slight decrease is noticed. The results are shown in Fig. 6.

### 4.3 Effect of Oil to Methanol molar Ratio

The molar ratio of oil to methanol is varied from 1:5 to 1:16. However, 1:10 molar ratio gives the maximum conversion as shows in Fig. 7. It is clear that % conversion increases with increase in the molar ratio and the maximum conversion (99.15%) is obtained at 1:10 molar ratio but beyond this the conversion % is almost constant. Thus the optimum condition for batch ultrasonic procedure to produce methyl esteris: reaction time 15 min for high ultrasonic power (50% amplitude, 0.7 s cycles each second) with 2 wt. % catalyst and oil to methanol molar ratio as 1:10. Excess methanol can, however be recovered after use.

### 4.4 Effect of Reaction Time

Ultrasonication increases the speed of the transesterification of algae oil and thus reduces the reaction time because ultrasonic cavitation mixing is an effective alternative means to achieve a better mixing in conventional processes. Ultrasonic cavitation provides the



**Fig. 5. Effect of loaded metal wt.% catalyst on algal biodiesel production over Sn/NSZS catalyst**

necessary activation energy for their action. The reaction time is varied from 0 to 30 min with an interval of 5 min. Fig. 8 shows the effect of reaction time on algae oil methyl ester content. 1:10 oil to methanol molar ratio with 2 wt.% catalyst was found to cause rapid formation of algae oil methyl ester. Within 15 min., more than 99.15% conversion was achieved but after that the conversion rate was slower and finally reached the steady state. 15 min duration is, therefore, selected for further studies. Evidently ultrasound reduces the processing time from the conventional 3–6 h batch processing to less than 15 min.

#### 4.5 Effect of Ultrasonic Amplitude

Ultrasonic amplitude affects the mixing of oil/methanol which is a key factor to get higher conversion of algae oil. The optimization of amplitude of the applied ultrasonic energy was 30%, 40%, 50%, 60%, and 70% and at this amplitude the conversion was 70%, 85%, 99.15%, 99.05%, and 98.47%, respectively (Fig. 9). As evident from the figure 50% amplitude recorded the maximum conversion of biodiesel. Further increase in the amplitude from 50% to 70% exhibits no change in conversion probably because a large number of cavitation bubbles are generated in the solution when a large amount of ultrasonic power enters the system. Many of the bubbles will coalesce forming larger, more long-lived bubbles which will certainly act as a barrier to the transfer of acoustic energy through the liquid. Another possible source of loss of efficiency in the transfer of power could be the so-called

decoupling effect. As such, 50% amplitude is selected for this study.

#### 4.6 Effect of Cycle

Ultrasound interaction to reactants, regular or irregular, affects the extent of conversion. The instrument can be used in cycle mode, with a cycle setting at 1; the reaction mixture is sonicated without interruption whereas with a cycle setting at 0.7, the mixture is sonicated for 0.7s and then the sonication stops for 0.7s. Hence in cycle mode, the ratio of sound-emission time to cyclic pause time can be adjusted continuously from 0% to 100% per second. The conversion of biodiesel was observed at the cycle 0.3, 0.5, 0.7 and 0.9s cycles each second as shown in Fig. 10. The maximum conversion 99.15% is achieved at the cycle 0.7. When the cycle was more than 0.7, there is no significant change in biodiesel production. This cycle time was, therefore, used in all the experiments.

#### 4.7 Catalyst Reusability and Comparison

Sn/NSZS catalyst has high silica content which makes it more hydrophobic and shows good stability after regeneration by calcination and washed with methanol. It was reused for the subsequent transesterification experiments. The same procedure is followed for six consecutive experiments and the results are shown in Fig. 11. It shows that the same catalyst can be used for minimum four times giving high temperature treatment after every run, to recover its activity. Sn/NSZS catalyst gives high yield in comparison with other heterogeneous catalyst with increasing time of reaction which is shown in Fig. 12.

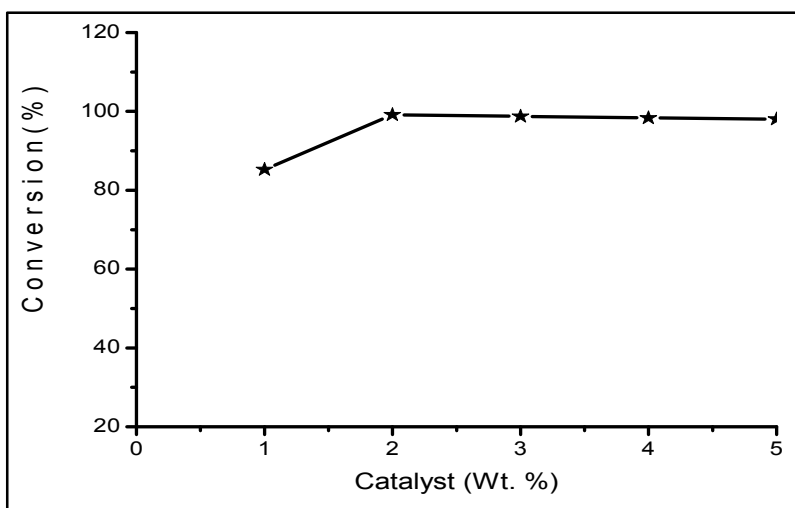


Fig. 6. Effect of loaded metal wt.% catalyst on algal biodiesel production over Sn/NSZS catalyst

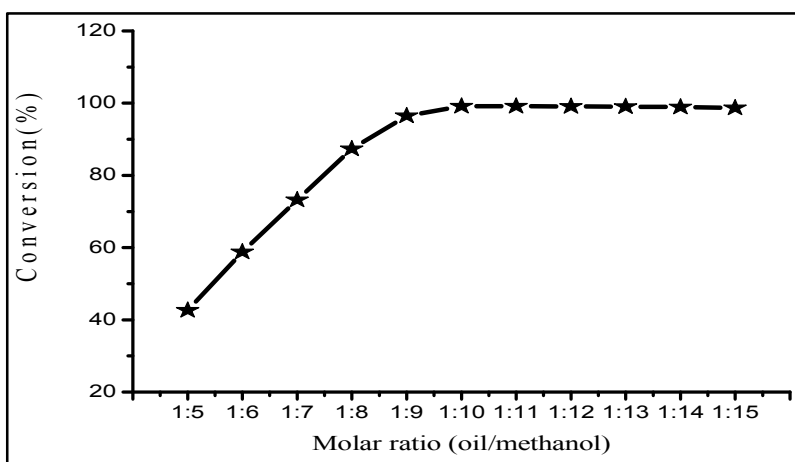


Fig. 7. Effect of oil/methanol molar ratio on algal biodiesel production over 0.4Sn/NSZS catalyst

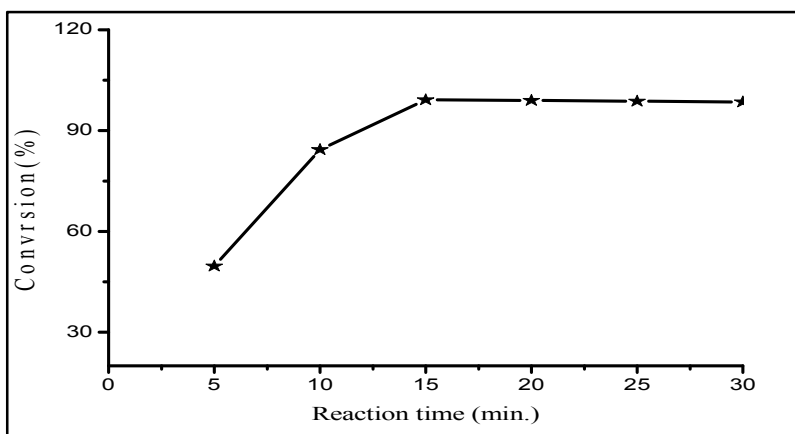


Fig. 8. Effect of reaction time on algal biodiesel production over 0.4Sn/NSZS catalyst



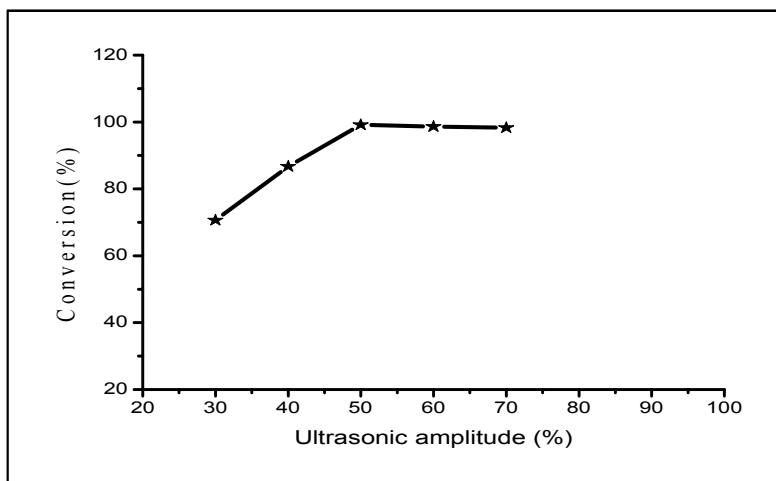


Fig. 9. Effect of ultrasonic amplitude on algal biodiesel production over 0.4Sn/NSZS catalyst

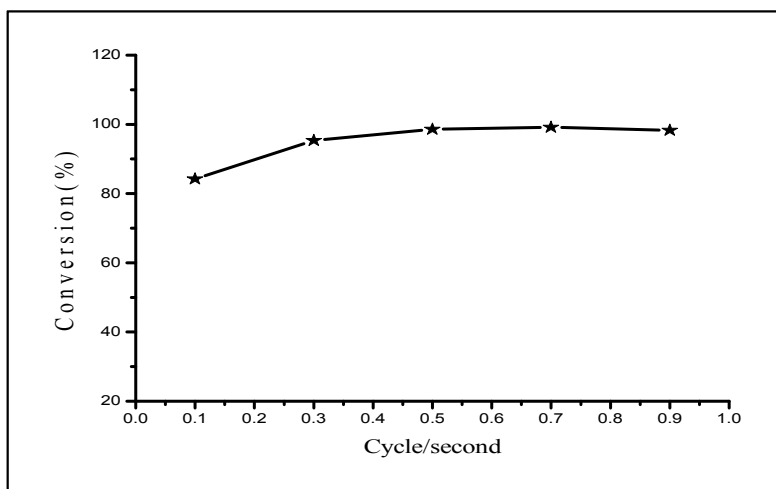


Fig. 10. Effect of ultrasonic wave cycle on algal biodiesel production over 0.4Sn/NSZS catalyst

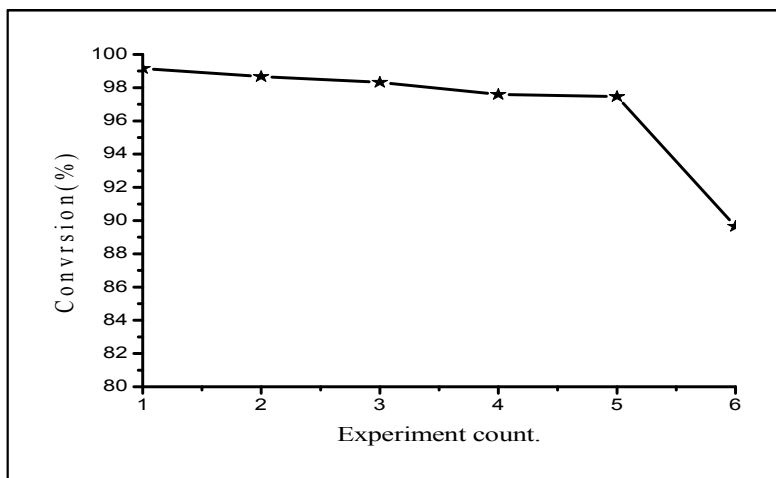


Fig. 11. Reusability of catalyst on algal biodiesel production over 0.4Sn/NSZS catalyst

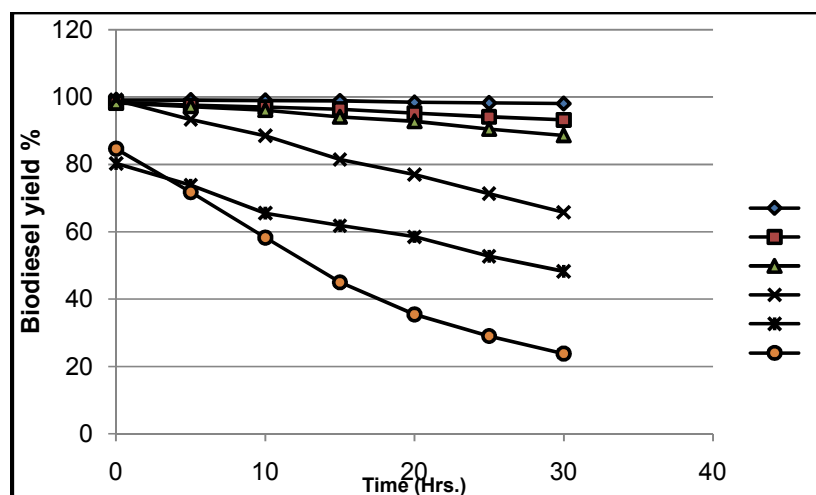


Fig. 12. Algae oil transesterification over versus reaction time for various catalysts (2 g each): (A) 0.4 Sn/ NSZS catalyst; (B) 0.4 Sn/NSZS regenerated four times by calcination; (C) 0.4 Sn/NSZS regenerated two times by calcination; (D) SZ/SiO<sub>2</sub> (Zr/Si=50:100, mol/mol); (E) zeolite Beta; (F) ZSM-5

## 5. CONCLUSION

The new solid acid metal supported core shell catalyst (Sn/NSZS) and ultrasonic assisted method for biodiesel production from algae oil by transesterification is convenient, safe and time saving. Ultrasonication provides the desired conditions to carry out the reaction under controlled atmosphere in very short reaction time and using solid acid heterogeneous catalyst (Sn/NSZS). It also gives better handling and separation. The main advantage of these studies is to prepare a suitable catalyst and a more convenient and quick procedure for the production of green biodiesel from algae oil.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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