Optimization of biodiesel production from *Chlorella Vulgaris* using CuO, NiO and MgO on zeolite 4A catalysts

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Abstract. The activities of metal oxide on zeolite as catalysts for production of fatty acid methyl ester from microalgae lipid oil were investigated by impregnation of MgO, CuO and NiO on zeolite 4A. The physical properties of the catalysts were characterized using TGA, XRD, Fe-SEM and Nitrogen Physisorption analysis. Evaluation on activity for biodiesel production from Chlorella vulgaris microalgae shows the trend of catalysts performance as CuO/Zeo>NiO/Zeo>MgO/Zeo. The amount of catalysts, the methanol to oil ratios and the length of reaction times affects the production of biodiesel with 69% of biodiesel produced using CuO/Zeolite catalysts at optimized conditions.

1. Introduction

Biodiesel or fatty acid methyl ester (FAME) is recognized as potential renewable sources of energy with similar properties as fossil fuels in terms of the chemical structure and energy content.¹,² Biodiesel is highly biodegradable and has minimal toxicity in comparison to fossil fuels, thus it can be used as alternative for fuels in various applications without major modifications.³,⁴ Production of biodiesel requires active catalyst to enhance the kinetic of the reaction and to control product selectivity.⁵ Transition metal oxide, such as Ni, Mo, W, Cu, or sulfide of group VIII and/or group VIB⁶ were often deposited on solid acid support such as aluminosilicate materials to catalyze the reaction. Zeolite has great potential as catalysts or catalysts support because of its high surface area, porosity, adsorption capacity, and easy to separate from reactants and products.⁵ The activity of zeolite is influenced by its structure, surface acidity and reaction condition. Other than the choice of the catalysts, several factors involving the temperature of the reaction, the amount of reactants relative to catalysts and the length of reaction time also plays important roles in achieving optimum yield for conversion of natural feedstock to biofuel. Transformation of natural feedstock to biofuel can be carried out using transesterification, fermentation, thermal cracking, catalytic cracking, and hydroprocessing. Transesterification is a promising method for biodiesel productions that often produced high yield of biodiesel.⁷ Finding an ideal raw feedstock for synthesis of biodiesel is important too as it will significantly affect the quality of the biodiesel and determine the viability of the process. Recently, researches have been focused on the utilization of microalgae as third generation carbon sources for biodiesel production. The efficiency
of biodiesel production from microalgae is about 15 to 300 times higher than the conventional crops when comparison is based on area basis. Apart from biodiesel, microalgae has also been investigated as sustainable carbon feedstock for production of renewable fuels for example methane, hydrogen and ethanol.

In this studies, we investigate the activities of NiO, MgO and CuO impregnated on zeolite 4A as catalysts for transesterification of lipid oils extracted from Chlorella vulgaris microalgae to produce biodiesel. Detail investigation on the effect of transesterification conditions by variations of oil-to-methanol molar ratios, the amount catalyst loading and the length of reaction time in order to obtain optimum production of biodiesel.

2. Experimental
Impregnation method was used to deposit metal oxide on zeolite support. Metal salt solutions at 15 wt % concentration were prepared by dissolution of each metal salt precursor (Mg(NO3)2.6H2O, Cu(NO3)2.3H2O, Ni(NO3)2.6H2O) in deionized water. Then, 2.05 g of zeolite was slowly added into each solution with continuous stirring. The so-called slurry was left for 4 hours at room temperature to age and followed by drying in air oven at 340 K for 16 hours. The resulting powder was calcined at different temperature from 570 K to 770 K in air furnace for 4 hours under atmospheric condition. Synthesis of biodiesel was carried out by extracting crude lipid from Chlorella vulgaris via solvent extraction method as reported by Lee et al. (Lee, Yoo et al. 2010). The Chlorella vulgaris microalgae was mixed with 400 ml of chloroform-methanol mixtures at 2:1 v/v ratio and sonicated using a sonicator (S70h Elmasonic) for 5 minutes and was continuously stirred for 3 days at room temperature. The resulting slurry was filtered and the extracted lipid was transferred into a rotary evaporator to remove the remaining solvent. 0.1 g of crude lipid was transferred in a three neck round bottom flask. Methanol was added into the mixture and stirred for 5 minutes before catalysts was added into the flask. The mixture was heated to 338 K and stirred at 300 rpm for 2 hours. Three independent variables were used which are the methanol to lipid weight ratio range from (1:12 to 1:21 w/w), variation of catalyst loading (5, 7, 9, 11 wt. %) and reaction time (0.5, 1, 2, 3, 4 hour). The calculation of biodiesel yield of FAME was performed based on peak analysis from gas chromatography. The gas chromatography (GC-7890A, Agilent) was equipped with a flame ionization detector (FID) and fitted with a HP-INNOWAX capillary column (15cm length, 0.25mm I.D., 0.25 µm film thickness) for the separation of FAME species. n-hexane was used as a solvent to dilute the samples prior to injection while methyl heptadecanoate (C18H36O2) was used as an internal standard. The average data were recorded by repeating with three injections for each sample for measurements.

2.1 Catalysts Characterization
Thermogravimetric analysis (TGA) was performed to study the thermal stabilities and to obtain optimum calcination temperature of fresh catalysts using thermo-gravimetric analyzer (TGA, Perkin Elmer Model STA 8000) at a heating rate of 283 K/min under oxygen environment from 289 K to 1273 K. The morphology was studied using field emission scanning electron (FE-SEM Gemini 500) at accelerating voltage of 5kv. Powder x-ray diffraction (XRD) patterns was analysed using Bruker Model D8 Advance Power using Cu Kα radiation at a screening speed of 2˚ min-1. Nitrogen adsorption measurements were performed at 77 K by Nova 1200 Quantachrome analyzer. The surface area was calculated using Brunauer EmmettTeller (BET) calculations. Prior to the measurement, the samples were degasses under vacum at 390 K for 5 hours.

3. Results and Discussion
3.1 Catalysts characterization.
TGA analysis were carried out on NiO/zeo, MgO/zeo and CuO/zeo catalysts following air drying at 340K. The measurement were performed to determine the structural changes of the catalysts at elevated temperatures and to obtain optimize calcination temperatures for the formation of metal oxides. The TGA profiles shown in Figure 1 revealed three weight loss regions accompanied by an exothermic DTG
peak were observed for all samples at the temperature below 600 K. The first feature observed at 470 K to 770 K is attributed to the decomposition of nitrate from salt precursor during impregnation of metal on the zeolite support. At 667 K to 1057 K, the weight loss in the TGA profiles is accompanied with a broad feature of DTG which is associated with the transformation of the hydroxide to oxide. At temperature above 1000 K, the sample is stable towards further decomposition with no discernible changes on the weight up to 1200 K. Based on the TGA/DTG profiles, the optimum temperature of 700 K was selected for the calcination of the samples to form metal oxide catalysts. We observed that the fresh Mg/Zeo and Ni/Zeo were fully oxidised at temperature above 770 K meanwhile fresh Cu/Zeo was fully oxidized at 570 K.

Analysis on the specific surface area and the total pore volume of the catalysts are important to provide information on the active surface sites for catalytic reaction. Table 2 summarized the specific surface area (SBET), total pore volume and average pore diameter of the catalysts after calcination at 700 K. There were significant differences on the surface area of the catalysts upon impregnation with metal oxide, with Zeolite 4A impregnated with copper oxide shows the highest specific surface area followed by NiO/zeo and MgO/zeo. However, the pore volume and the pore size of the catalysts were approximately similar suggesting the deposition of metal oxides occurs on the surface, but not in the pore of the zeolite. Copper oxide on zeolite following air calcination at 723 K shows high surface area ~ 45 m²/g in comparison to NiO and MgO on zeolite catalysts that may be due to the CuO is well distributed on the zeolite support relative to MgO and NiO.

### Table 2. Surface properties calcined samples of metal supported on zeolite

| Sample Notation | Specific Surface Area (SBET) | Total Pore Volume (cc/g) | Average Pore Diameter (Å) |
|-----------------|-----------------------------|--------------------------|---------------------------|
| MgO/Zeo         | 6                           | 0.0940                   | 102.00                    |
| CuO/Zeo         | 45                          | 0.1393                   | 123.80                    |
| NiO/Zeo         | 17                          | 0.0779                   | 185.50                    |
Figure 2. X-ray diffraction pattern of metal oxide on zeolite

The XRD patterns of MgO, CuO and NiO on zeolite following calcination at 700-750K are shown in Figure 2. All catalysts show peak associated with diffraction plane of Zeolite 4A at 7.27°, 10.25°, 12.49°, 16.13°, 20.51°, 21.64°, 24.16°, 27.15°. On CuO/zeolite, the diffraction peaks appeared at 2θ of 63.72°, 48.65°, 53.4°, 58.34°, 61.5°, 66.13°, 68.22° corresponded to monoclinic phase of CuO nanoparticles according to JCPDS data (80-0076). The intensity of CuO/zeolite were also significantly reduced in comparison to the diffraction patterns of zeolite 4A, MgO and NiO catalysts. In MgO/zeolite, the peaks associated to MgO were observed at 2θ of 37.9°, 43°, 62.5°, 78.63° which corresponded to the diffraction of cubic phase of MgO nanoparticles (JCPDS no. 79-0612). For NiO sample, four peaks were observed at 2θ of 44.5°, 37.4°, 43.1°, 62° which can be index to the cubic phase structure of NiO (JCPDS 44-1159).

Figure 3 demonstrates the morphology structure of zeolite 4A and metal oxide supported on zeolite obtained from FE-SEM analysis. The zeolite shows as cubic shape structure with average crystallite size of 1.4 – 2 µm. following impregnation with metal oxide, the zeolite surface shows the presence of flakes-like and granular shape crystallites with a rough surface morphology. The changes in morphology of the prepared catalysts indicating the present of metal oxide on zeolite surface as confirmed by XRD analysis.

Figure 3. FE-SEM Morphology of zeolite (a); MgO/Zeo (b); CuO/Zeo (c) and NiO/Zeo (d)

3.2 Catalytic activity

Transesterification process is an equilibrium reaction with large excess of alcohol is required to drive the reaction towards high production of biodiesel. The production of FAME is also influenced by the ratio of methanol to lipid oil presence in the system. In this study, the synthesis of biodiesel was carried out at the variation of methanol to oil ratio ratios of 1:120 up to 1:210. The transesterification was carried out at 338 K with stirring speed of 300 rpm and using 5 wt% of catalysts. The data obtained after 180 minutes of reaction are shown in Figure 4. Fig. 4a shows the production of FAME when using CuO-zeolite catalysts were significantly enhanced from 18% to 49% when the ratio of methanol to oil was
increased from 1:120 to 1:180. MgO/Zeo and Ni/Zeo catalysts showed low activities than CuO/Zeo with 7.92% and 11.94% of FAME were produced at 1:180 oil to methanol ratio. The activity of CuO/zeolite catalyst is significantly higher than MgO and NiO that can be associated with the surface area and pore volume of the catalysts. MgO and NiO catalysts have lower surface area as compared to CuO catalysts which as consequences limit the adsorption and diffusion of triglyceride within the zeolite framework to reach the active sites for transesterification process.\textsuperscript{12} When the methanol to oil ratio was further increased to 1:210, the production of FAME was significantly affected with only 25% and 20% yield in CuO/Zeo and NiO/Zeo catalysts. However, for MgO/Zeo catalysts, variation of methanol to oil ratios show negligible differences in the biodiesel yield which may be due to the lack of active sites presence in the catalysts to drive the reaction. The influence of catalyst loading was investigated by variation of the amount of metal oxide/zeolite catalysts used during catalytic transesterification of microalgae-oil from 5 - 13 wt. %. As can be seen from Figure 4(b), increasing the concentration of catalysts from 5 to 9 wt % significantly enhanced the production of FAME. The production of FAME reached optimum yield at 10 wt % of catalysts loading for all catalysts, with 19% production from MgO/Zeo, 70% from CuO/Zeo and 24% for NiO/Zeo catalysts. This indicates the efficiency of the reaction is strongly affected by the concentration of the catalyst, in which it increased the number of available sites for the reaction.\textsuperscript{13} However, when the concentration of the catalysts were further increased to 14wt % loading, the production of FAME were significantly reduced to only 10% for all catalysts.

![Figure 4](image_url)

**Figure 4.** The effect of methanol to oil ratios (a), catalysts loading (b) and reaction time (c) on the production of FAME from crude microalgae lipid when using MgO/Zeo, CuO/Zeo and NiO/Zeo catalysts. Reactions were carried out at 338 K with stirring rate of 300 rpm

This is due to the excess amount of catalysts presence in the reaction system that leads to phase separation. High concentration of catalysts also initiate and accelerated the formation of soap and therefore reduced the productivity of biodiesel.\textsuperscript{13} The effect of reaction time on transesterification reaction was investigated on MgO/Zeo, Cu/Zeo and Ni/Zeo catalysts as shown in Figure 4(c). All the catalysts reached optimum activity at different reaction times with CuO/Zeo shows optimum yield at 200 min. The reaction has reached optimum FAME yield on Mg/Zeo catalysts within 60 minutes of reaction with ~ 33% of production. Meanwhile on NiO/Zeo the reaction reached optimum biodiesel yield at 120 min. It is interesting to see that the biodiesel yields were significantly reduced when the reaction was carried out up to 250 h. extending the reaction time may have been prompted backwards reaction that leads to soap formation with similar observations were reported by Azizi et. al (2016).\textsuperscript{14}
4. Conclusion
The activities of MgO, CuO and NiO impregnated on Zeolite 4A were investigated as catalysts for transesterification of lipids oil extracted from *Chlorella vulgaris* microalgae for production of biodiesel. High production of biodiesel was obtained from CuO/Zeo catalysts followed by NiO/Zeo and MgO/Zeo. The differences on catalytic activity of the catalysts were correlated with the number of surface area and pore volume. Optimization of transesterification condition shows that under optimum conditions of 1:150 molar ratio of methanol to oil, 180 minutes of reaction and 0.09 g of catalyst loading, CuO/Zeo catalysts produced 69% of biodiesel yield.

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