

Removal of Methylene blue, *Escherichia coli* and *Pseudomonas aeruginosa* by Adsorption Process of Activated Carbon Produced from *Moringa oleifera* Bark

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Abstract- The utilization of biopolymer derived from *Moringa oleifera* bark using ZnCl₂ and H₂SO₄ as activating agents for eliminating methylene blue, *Escherichia coli* and *Pseudomonas aeruginosa* from producing wastewater. In this study, methylene blue and both bacteria were effectively adsorbed by activated carbon with lowest dosage. The activated carbon was prepared from natural-by product of *Moringa oleifera* bark by pyrolysis in a furnace at 700 °C for 1 h. The characteristics of activated carbon have been determined using Scanning Electron Microscopy (SEM), Brunauer-Emmett-Teller (BET), pHzpc (zero point charge), and FTIR spectroscopy. The obtained result were closely fitted with Freundlich isotherm model and adsorption kinetics follow the pseudo-second order model with the highest value of correlation coefficient ($R^2 \sim 1$). Adsorption quantity was dose dependent and bacteria were maximum adsorbed using 10 mg of activated carbon as well as 25 mg for methylene blue. The maximum adsorption capacity showed within 1 hour. The bacterial cells were reduced by 98 % for *E. coli*, 96 % for *P. aeruginosa* as well as methylene blue reduced 94.2 % from aqueous solution using batch adsorption methods. Adsorption process controlled by film diffusion mechanism. These results proposed that the activated carbon of *Moringa oleifera* bark can be used as a good adsorbent for the removal of Methylene blue, *E. coli* and *P. aeruginosa*.

Keywords: *Moringa oleifera* bark; Film diffusion; Dye uptake; *E. coli*; *P. aeruginosa*.

INTRODUCTION

Synthetic dye is used in many manufacturing industries for the colouring of products such as textile, paper, and paint [1]. Around 50 % of total dyes is released from textile industries [2]. Now-a-days, synthetic dye controls most of the chemical industries [3]. Methylene blue is a basic dye which is a heterocyclic aromatic compound. If consumed, it can cause irritation of mouth, throat with symptoms of jaundice in humans [4]. Methylene blue dye enter into water system can cause human cancer of breast and skin.

There are several common physical, biological and chemical methods for eliminating colouring matter from aqueous solution. These include reverse osmosis, adsorption [5], assimilation [6-8], precipitation, coagulation and ion exchange ([9], [10]). Among all methods, Adsorption is one of the major processes for dye removal from wastewater [11]. Generally, commercial activated carbon is used as adsorbent in adsorption process due to high uptake capacity, eco-friendly and non-toxic nature [12] but it is a costly process [13]. In recent times, researchers are looking into green materials for the wastewater treatment as alternative options of commercial activated carbon. There are some natural plants used as adsorbents for the removal of dye, heavy metal and bacteria from wastewater such as oil palm [13], groundnut shell [14], *Moringa oleifera* leaf [15], rice husk [16], *Moringa oleifera* seed which can be used to remove dye from industrial wastewater. Among them *Moringa oleifera* plant is a multi-purpose tree which is used for food and medicinal activities [17]. Various parts of *Moringa oleifera* such as stem, bark, and roots have antimicrobial activities [18, 19]. The main aim of this study to produce activated carbon as adsorbent from *Moringa oleifera* bark to remove methylene blue, *E. coli*, and *P. aeruginosa* from wastewater.

EXPERIMENTAL PROCEDURE

Materials

All chemicals were of analytical rating. Methylene blue solution were prepared using deionized water. *Moringa oleifera* bark was collected from outside of USIM campus and bacteria were received from the microbiological lab of USIM.

Instrumentation

Functional groups on surface of adsorbents were determined using Fourier-Transform Infrared Spectroscopy (PERKIN ELMER). The surface properties of activated carbon were identified using (SEM-HITACHI, Japan) and Brunauer-Emmett-Teller (BET: BELSORP, Japan). Centrifuge, water distillatory, pH meter, shaker, UV/Vis. Spectrophotometer (Cany 50 conc. VARIAN, Agilent Technologies, USA), analytical balance, were used in this study.

Sample collection and preparation

Moringa oleifera bark sample was collected from natural sources in-front of Akasia avenue, Nilai, Negeri Sembilan, Malaysia. Firstly, the bark was washed using water and dried under the sun for 6 hours. Then it was again washed to remove dirt from its surface and dried in an oven at 110 °C for 13 hours. The *Moringa oleifera* bark pieces were reduced into smaller particle sizes using blinder. The samples (100 g) were then chemically activated using 500 ml zinc chloride (10 % ZnCl₂) and Sulphuric acid (0.5M H₂SO₄) with ratio of 1:5 for 12 hours. Then the samples were dried in the oven at 110 °C for 12 hrs. It was grinded and sieved at 400µm. Then the sample was kept in the desiccator for further use. The precursor was placed in the furnace at 700 °C with heating rate of 10 °C/min for 60 min. During the carbonization process, purified nitrogen was flown. It was cooled in normal temperature. These activated carbon samples were washed several times with distilled water. The washed samples were dried in the oven at 110 °C for 15 hours. These were then sieved with a sieve of 400 µm. The activated carbon sample was kept in a polypropylene bottle to avoid exposure to moisture. These prepared activated carbon sample from *Moringa oleifera* bark were used for the experiments as the adsorbent in adsorption process.

Characterization of the Prepared Activated Carbon

The characterization of the *Moringa oleifera* was obtained by Fourier Transform Infrared Spectroscopy (FTIR). It was range from 400 to 4000 cm⁻¹. IR spectra were detected by disk method. The surface functional groups on the activated carbon were analysed by FTIR spectroscopy. The morphologies of the adsorbents were determined using Scanning Electron Microscope (SEM). The surface area, pore volume and pore size of activated carbon were measured by Brunauer-Emmett-Teller (BET). The system were measured by a Quanta chrome NOVA automated gas sorption system.

RESULT AND DISCUSSION

Effect of adsorbent dosage

(a) The effect of adsorbents dosage in the range of 0.005–0.05 g on the removal percent for MB dye was conducted in Fig. 1. From this figure, the removal percent increases from 25 to 100% for MB with increasing the adsorbent dose from 0.005 to 0.05 g. It was found that 94.2% from aqueous solution was removed with use of 0.025g of activated carbon. It can be said to have occurred due to available active sites present on activated carbon for the adsorption. Further increasing the adsorbent dose, the removal percentage was not significant. This may have happened due to it reaching the equilibrium state between binding adsorbate and unadsorbed adsorbate in adsorption process.

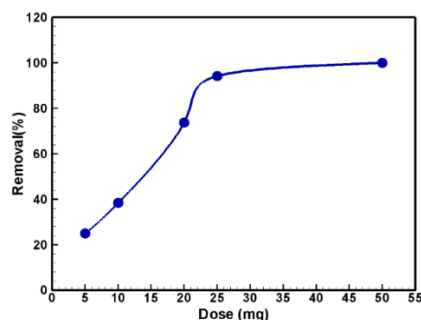


Fig. 1 Effect of dosage for methylene blue

(b) The effect of dosage of activated carbon on bacteria were studied as shown in Fig.2. The stem bark of agricultural products can show anti-bacterial activities. The number of cell for both bacteria decreased while using adsorbents. The efficacy of activated carbon on bacteria was dose dependant. The removal percentage of *E. coli* were obtained as 29.3 %, 62.7 % and 87.8 % respectively using 1, 3, 5 mg activated carbon of *Moringa oleifera* at 37 degree for 1 hour. On the other hand, the removal percentage of *Pseudomonas aeruginosa* incubated with 1, 3, 5 mg adsorbents were obtained as 59.8 %, 74.1 % and 90 % respectively. *E. coli* was decreased as 98 % and *Pseudomonas* was reduced as 96 % using 10 mg of adsorbent.

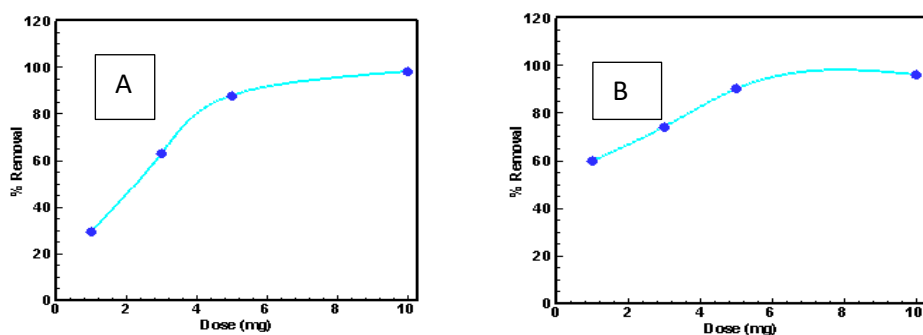


Fig. 2 Effect of adsorbent dosage for (A) *E.Coli* and (B) *Pseudomonas aeruginosa* of *Moringa oleifera*

To evaluate the number of Bacteria using Disk

About the cells (1.15×10^7) of *E. coli* and (1.73×10^7) of *Pseudomonas aeruginosa* were mixed with activated carbon (1, 3, 5, 10mg) respectively. These mixtures were incubated at 37 °C with agitation for 1 hour and after then centrifuged for 5 min. After separating, it was then placed onto nutrient agar of disks to determine the number of bacteria. All disk were placed into the incubator at 37 °C for 24 hours. After incubation, the number of bacteria were counted using magnifying glass. The results are shown in Fig 3. Summary of microbiological report are shown in Table 1. From the Table 1, the number of bacteria were counted 120, 55, 5, 0 for *E.coli* using (1 mg, 3 mg, 5 mg and 10 mg) activated carbon respectively and 165, 145, 95 and 0 for *Pseudomonas aeruginosa* bacteria using activated carbon of *Moringa oleifera* respectively.

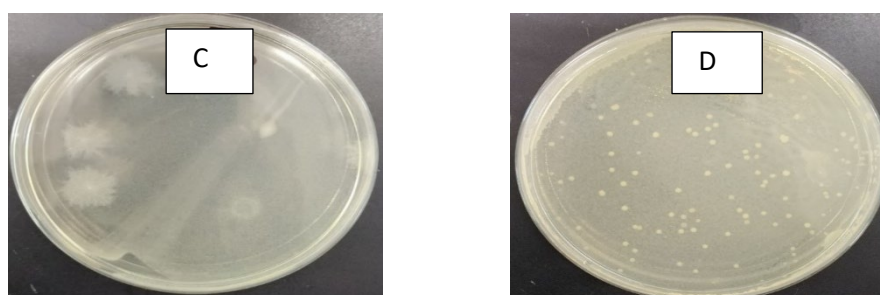


Fig. 3 Number of bacteria for (C) *E.Coli* and (D) *Pseudomonas aeruginosa* of *Moringa oleifera* by Disk methods

TABLE 1

MICROBIOLOGICAL REPORT ON *E.COLI* AND *PSEUDOMONAS AERUGINOSA* BACTERIA USING ACTIVATED CARBON OF *MORINGA OLEIFERA* BARK

Types of Bacteria	<i>Escherichia coli</i>						<i>Pseudomonas aeruginosa</i>				
	Quantity of AC	Num. of Bacteria (disk)	After 10 min. Cell removal	After 20 min. Cell removal	After 40 min. Cell removal	After 60 min. Cell removal	Num. of Bacteria (disk)	After 10 min. Cell removal	After 20 min. Cell removal	After 40 min. Cell removal	After 60 min. Cell removal
Moringa Oleifera Bark	1 mg	120				29.3%	165				59.8%
	3 mg	55				62.7%	145				74.1%
	5 mg	5	24.1%	43.4%	82.5%	87.8%	95	15.1%	35.1%	85.9%	90%
	10 mg	0				98%	0				96%

CONCLUSION

This study evaluated the use of activated carbon derived from *Moringa oleifera* as a low-cost adsorbent for the removal of Methylene blue, *E. coli* and *P. aeruginosa* in wastewater treatment. The adsorption of methylene using the activated carbon of *Moringa oleifera* bark was closely fitted by the Freundlich model. The methylene blue adsorption of activated carbon showed physical adsorption by D-R model ($E < 8$). The adsorption kinetics parameter better fit pseudo-second order model ($R^2 > 0.99$) than pseudo-first-order model and intra-particle diffusion. The increase in dose of activated carbon enhanced the bacterial adsorption capacity and at 10 mg of adsorbents, it shows maximum adsorption capacity of 98 % for *E. coli* and 96 % for *P. aeruginosa* as well as 94.2% removed methylene blue using 25 mg of activated carbon. The result suggested that the activated carbon of *Moringa oleifera* can be used as a good adsorbent to remove contaminant from wastewater.

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