Study on Activated Carbon derived from Cassava Peels as Magnetic Solid of Dispersive Solid Phase Extraction Technique for Determination of Tetracycline

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Abstract

Concern on the existence of antibiotic residues in aqueous environment has been growing in recent years since they are known to impart various severe effects on human health and environment. In general, the concentration of antibiotic residue in environment is in trace level and often below the limit of detection of analytical instruments, implying that sample preparation prior to analysis requires particular strategy. In the present work, activated carbon produced from cassava peel was converted into magnetite activated carbon followed by utilization of magnetite activated carbon produced as adsorbent for tetracycline antibiotic, in order to evaluate its potential as the material for dispersive solid-phase extraction (DSPE).Characteristics of the adsorbent were investigated using several techniques including Scanning electron microscopy (SEM) and X-ray diffraction. Several parameters influencing the adsorption by DSPE were optimized. The tetracycline adsorbed by magnetic activated carbon was analyzed using a UV-Vis spectrophotometer at a wavelength of 275 nm.

Characterizations revealed that the adsorbent has irregular surface, rough and highly porous structure with magnetic properties. Adsorption experiments indicate that equilibrium reached at pH 6, time 10 minute and adsorbate concentration 1 mg/L. By application of 20 mg adsorbent, the highest percentage of adsorption of 73 was achieved. The study indicates that magnetic activated carbon from cassava peel has promising potential as a low-cost and effective adsorbent for determination of tetracycline residues in aquatic environment.

Keywords: Adsorbent, antibiotic, cassava peel, dispersive solid-phase extraction.

Introduction

The emergence of a pandemic due to the COVID-19 virus has significantly increased the use of antibiotics, thus posing a new threat to human health in the future[7,8,24]. The lack of alternative drugs has made antibiotics as the most widely field used to prevent and treat diseases during the COVID-19 pandemic[7]. The presence of antibiotic residues is known to increase in various environments such as food, beverages, groundwater, sediments, terrestrial and rivers[1,3,5,14,20,21,28]. Antibiotics that are not absorbed by the human or animal body will be excreted through faeces and urine. Finally, the antibiotics will enter the environment and will cause various problems such as genetic changes, antibiotic resistance, damage of aquatic ecosystems and ultimately human health problems[2,10-12,18].

Therefore, monitoring of antibiotic residues in the environments is necessary to control the use and anticipate the dangers associated with antibiotics. One class of widely used antibiotics is tetracycline. Antibiotics belonging to this class are widely circulated in the markets with relatively low prices and exhibit high microbial activity so that they are widely used in human medical treatment, livestock industry and agricultural industry.

Many methods for determination of antibiotic residue have been developed using various sophisticated instruments such as HPLC, GC-MS, LC-MS/MS, CE and ELISA[13,16,27]. However, the determination of antibiotic residues in the environment is complicated due to the high complexity of the environmental matrices and the very low concentration of the residues, which in many cases lies below the detection limit of the instruments[1]. In addition, the environmental matrices my damage the equipments and instruments used.

To improve detection capability, selectivity, sensitivity and prevent equipment damage, sample preparation is very important. Traditionally, liquid-liquid extraction was widely used, however this method requires the use of large volumes of solvents and is a long-time process. To overcome these drawbacks of liquid-liquid extraction method, an alternative method that has been developed is solid phase extraction (SPE). This technique is acknowledged as green analytical chemistry (GAC) for many reasons. The SPE is considered as environmentally friendly method, since this method requires a minimum volume of solvents, therefore produces less waste than that produced by other methods. Other advantages offered by this method are simplicity in terms of sampling, relatively cheap, quick and easy application procedures, good selectivity and sensitivity and high effectiveness[5,14,25] .

Apart from the various advantages offered, the SPE method also has some limitations: the need for a large number of samples, a pressurized pump and the possibility of plugging in the SPE column. To overcome these drawbacks, a new

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technique known as dispersive solid phase extraction (DSPE) technique has been developed[14,23]. In this technique, solid sorbent is dispersed in the sample solution so that a partition equilibrium between the analyte adsorbed by the solid phase and that in the sample solution occurs. The analyte that is adsorbed on the solid phase, is then filtered or centrifuged.

The solid sorbent for DSPE is one of the important factors that determines the amount of analyte extracted. Synthetic carbon-based materials such as multi-walled carbon nanotubes (MWCNTs) and gold nanoparticles and halloysite nanotubes (Au/HNTs) have been used[22,23]. However, due to the difficulty and high production cost, the use of alternative carbon materials such as activated carbon from agricultural waste has been investigated by many workers[3,6,15]. Activated carbon is a porous material that has a large surface area and high adsorption capacity, therefore this material has been widely applied in various fields such as to remove organic pollutants[9], dye adsorbents[6] and support catalysts and supercapacitor electrodes[3].

Regardless of the advantages, some limitations of activated carbon should also be acknowledged such as difficulty in carrying out filtration because of the dispersion of fine carbon particles in the solution, increased turbidity of the solution and difficult adsorbent regeneration[9]. To overcome the problem with filtration, magnet-based separation method has been developed. With this technique, separation of the adsorbent can be accomplished faster and more efficiently leading to lower cost of investment. Magnetic-based separation is conducted using a composite of an adsorbent with a magnetic material such as black iron oxide (Fe_3O_4). The presence of magnetite iron oxide enhances chemical stability, reduces toxicity and promotes recyclability.

Various adsorbents such as biomass, zeolite, activated carbon, polymers have been magnetized by many workers[6,14,16]. Activated carbon can also be magnetized using magnetite iron oxide and the product is known as magnetite activated carbon. In addition to large surface area, magnetic property is another advantage offered by magnetic activated carbon for separation process in particular since magnetic activated carbon that has adsorbed target compound, can be separated simply by applying a bar magnet around the solution, thus eliminating the need for filtration or centrifugation.

Lampung province is the main cassava producing region in Indonesia, with total production in 2018 around 6.68 million tons with a growth rate of 1.5% per year[4]. One part of the cassava tuber that remains unutilized is cassava peel. Cassava peel is known to have high carbon content and therefore this residue is a promising raw material for production of activated carbon and activated carbon based material such as magnetite activated carbon. With respect to its potential, activated carbon derived from cassava peel was used to produce magnetite activated carbon followed by the application of the prepared magnetite as a solid phase adsorbent for extraction of tetracycline from aquatic environments.

Material and Methods

Materials: The chemicals used in this study were $ZnCl_2$, $FeCl_3{\cdot}6H_2O$, $FeSO_4{\cdot}7H_2O$, NaOH 5 M, acetone, acetonitrile, 0.1 N HCl solution, 0.1 M NaOH solution. phosphate buffer solution, acetate buffer solution and standard solution of tetracycline antibiotics. Cassava peel was kindly provided by local cassava farmers.

Instrumentations: The instruments used were Scanning Electron Microscope-Energy Dispersive X-ray (SEM-EDX) (SEM ZEISS EVO MA 10), X-Ray Diffraction (XRD) (PANalytical X'Pert3) and Ultra Violet-Visible (UV-Vis) spectrophotometer (Agilent Carry 100).

Preparation of Activated Carbon from Cassava Peel: The cassava peel was cleaned, air-dried and finally oven dried at 130 °C for 6 hours to remove the moisture. Dry cassava peels were carbonized in a furnace at 450 °C for 25 minutes. The resulting carbon was cooled and then ground with mortar and pestle and then sieved through a 106 mesh sieve. The carbon was then physically activated by heating in a furnace at 700°C for 60 minutes and then cooled.

To complete activation, the carbon was mixed with $ZnCl_2$ 30% solution and 150 mL of distilled water. The slurry formed was allowed to stand for 45 minutes and then filtered. The activated carbon obtained was washed with distilled water to a pH of 6.8 ± 0.2, dried at 115°C for 3 hours and then cooled in a desiccator until the temperature was stable and then finally sieved through a 106 mesh sieve.

Preparation of Magnetic Activated Carbon: The magnetic activated carbon was prepared with a mole ratio of activated carbon to iron salt of 2:1. A typical experiment was conducted by mixing 6.5 grams of activated carbon with 300 mL of distilled water and stirred using a magnetic stirrer at a temperature of 70^{o}C. In another container, a solution of iron salt was prepared by dissolving 7.6 grams of $FeCl_3{\cdot}6H_2O$ and 3.9 grams of $FeSO_4{\cdot}7H_2O$ in 300 mL of distilled water. The two solutions were then mixed and stirred for 30 minutes while adding 100 mL of 5 M NaOH drop wisely until a black precipitate was formed.

The resulting precipitate was filtered and washed with distilled water until the pH 6 was reached and then dried in an oven at 100°C for 3 hours to obtain magnetic activated carbon[18].

Magnetic Activated Carbon Characterization: The magnetic activated carbon prepared was characterized using SEM-EDX to evaluate the surface morphology and composition using FTIR to determine the functionality and PSA to determine the particle size and then XRD to identify the crystalline phase.

Adsorption test

Determination of optimum adsorbate concentration: Five 20 mL standard solutions of tetracycline with concentration of 0.5; 1; 1.5; 2; and 2.5 mg/L respectively were prepared. Into each solution, a mass of 10 mg of magnetic activated carbon was added and the mixture was stirred for 10 minutes at 150 rpm at room temperature. After the extraction was complete, a magnetic bar was brought closer to the beaker glass wall so that the magnetic activated carbon particles were attracted by the magnetic bar, forming the flocks so that the solution becomes clear. The solution was subjected to decantation and the filtrate was analyzed using a UV-Vis spectrophotometer, recording the absorbance of the samples at the maximum wavelength of tetracycline (275 nm) as a base to determine the optimum concentration of the adsorbate.

Determination of optimum mass of adsorbent: Five 20 mL standard solutions of tetracycline antibiotics with an optimum concentration obtained from previous experiments were prepared and 5, 10, 15, 20 and 25 mg of magnetic activated carbon were added to separate solutions. Each mixture was stirred for 10 minutes at a speed of 150 rpm at room temperature. The rest of the experiment was conducted similar to the experiment for determination of optimum adsorbate concentration and from the results, the optimum mass of the adsorbent was determined.

Determination of the optimum pH: Determination of the optimum pH was conducted using 20 mL standard solution of tetracycline with the optimum concentration and optimum mass of adsorbent at different pHs 2, 4, 6, 7 and 8. The pH of the solutions was adjusted using 0.1 M HCl solution and 0.1 M NaOH solution. Acetate buffer was used to maintain the pH of 2, 4 and 6 and phosphate buffer to maintain the pH of 7 and 8. The rest of the experiment was carried out similar to that described in the previous experiment and from this experiment, the optimum pH was obtained.

Determination of the optimum contact time: Determination of the optimum contact time was conducted using 20 mL standard solution of tetracycline with the optimum concentration, optimum mass of adsorbent and optimum pH, with varied contact times of 5, 10, 15, 20 and 25 minutes.

Results and Discussion

Preparation of magnetic activated carbon: The typical example of activated carbon preparation is shown in figure 1. After carbonization process, the carbon obtained was physically activated by heating at 700°C followed by chemical activation using $ZnCl_2$. The activated carbon is then mixed with $FeCl_3 \cdot 6H_2O$ and $FeSO_4 \cdot 7H_2O$ solution to produce magnetic activated carbon, as shown in figure 2.

Characterization using SEM-EDX: SEM micrographs showing the surface morphology of the inactivated carbon, activated carbon and magnetic activated carbon are compiled in figure 3. Figure 3a indicates the existence of inactivated carbon as a porous material. The effect of activation treatment to increase the porosity is displayed by the micrograph in figure 3b, implying that the activated carbon has surface area which also means greater absorption capacity compared to that of inactivated carbon.

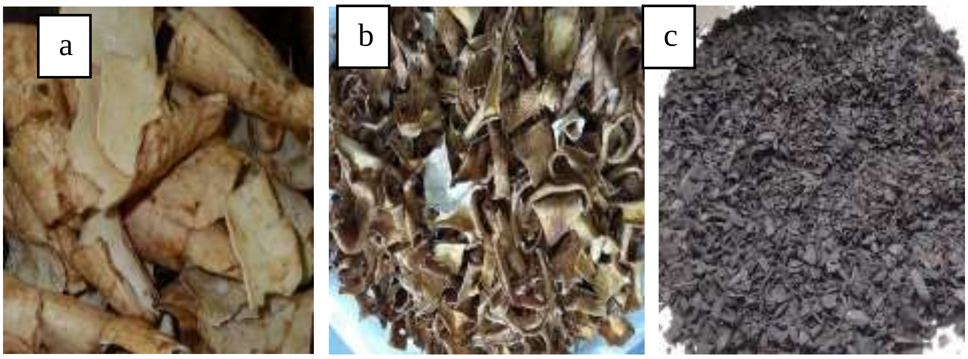

Figure 1: (a) Fresh cassava peel, (b) dried cassava peel, (c) carbonized cassava peel.

Figure 2: Magnetic activated carbon

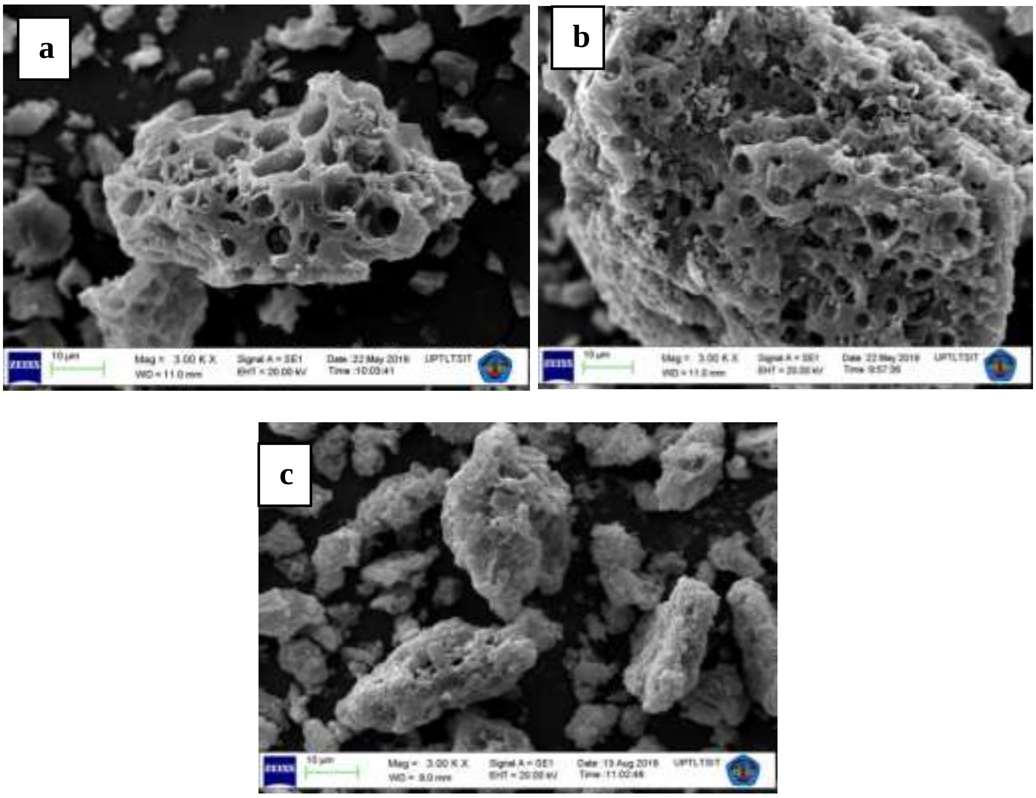

Figure 3: Results of SEM micrographs of the samples investigated: (a) inactivated carbon, (b) activated carbon and (c) magnetically activated carbon

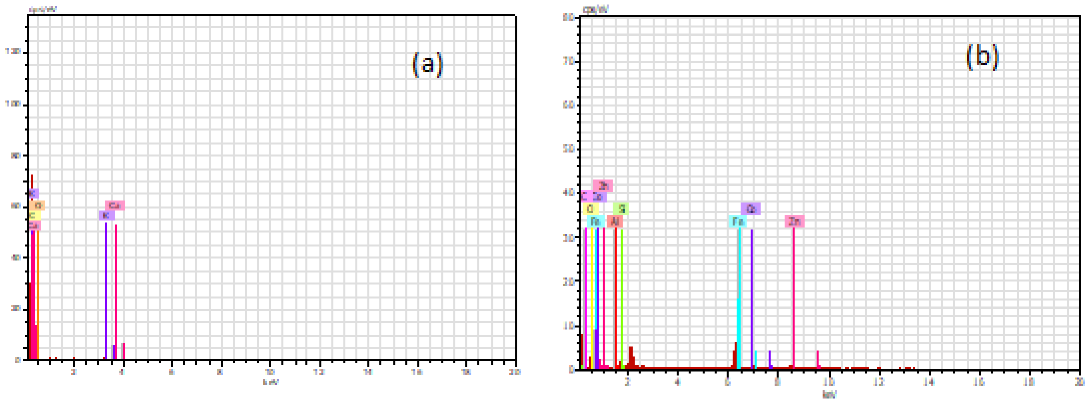

Figure 4: The results of the SEM-EDX spectrum on the adsorbent are (a) activated carbon and (b) magnetic activated carbon

The micrograph of magnetic activated carbon (Figure 3c) suggests that this material is less porous compared to the other two samples, most likely because some of the pores are filled with magnetite molecules. Regardless of this decreased porosity, magnetic activated carbon has larger molecular mass and density as a consequence. Due to this increased density, the particles of magnetic activated carbon will tend to move down in the container and therefore can be separated from the solution easier.

To confirm that the activated carbon was succesfully magnetized, the activated carbon and magnetic activated carbon were characterized using EDX. The results are presented in figure 4. As can be seen, the presence of Fe is evidently detected (Figure 4b), while in the spectrum of activated carbon (Figure 4a), this element is not detected, justifying that magnetization of the activated carbon was accomplished. Based on the figure, it can be observed that carbon has formed in the two activated carbons.

To investigate the change in structure as a result of different treatments, characterization was carried out using XRD and the results are shown in fig. 5. As can be seen in figure 5, diffractograms of inactivated and activated carbon are characterized by the presence of broad peak in the 2θ region of 20-30° which is a characteristic feature of amorphous carbon structure commonly found in activated carbon[26]. In the diffractogram of inactivated carbon (Figure 5a), several small peaks at 2θ positions of 14, 24 and 30° are observed, which are typical peaks for carbon.

In the diffractogram of activated carbon (Figure 5b), the peaks representing carbon are observed at 2θ positions of 11, 22, 24, 30 and 32°. Evidently different feature was displayed by the chromatogram of the magnetic activated carbon in which sharp peaks located at 2θ of 30,35, 43, 56 and 62° are observed. This different feature justifies the formation of magnetic activated carbon as suggested by the EDX results.

Adsorption tests: As previously described, series of adsorption experiments using magnetic activated carbon prepared were conducted to investigate the effects of various variables in order to obtain optimum conditions for adsorption of tetracycline as adsorbate. To obtain optimum concentration, the experiments with different adsorbate concentrations of 0.5; 1.0; 1.5; 2.0; and 2.5 mg/L were conducted and the results are shown in figure 6.

The graph in figure 6 clearly indicates that the highest percentage of adsorption (42.23%) was achieved using the adsorbate concentration of 1 mg/L and therefore this concentration was taken as the optimum concentration. The graph also displays decreased percentages of adsorption with increased concentrations, suggesting higher adsorbate concentrations leading to saturation of the pores of the adsorbent, therefore the adsorbent is no longer able to absorb the adsorbate.

To study the effect of adsorbent masses as a base to determine the optimum mass of adsorbent, the experiments using adsorbate with optimum concentration (1 mg/L) were conducted with different masses of the adsorbent of 5, 10, 15, 20 and 25 mg. The experimental results are presented in figure 7.

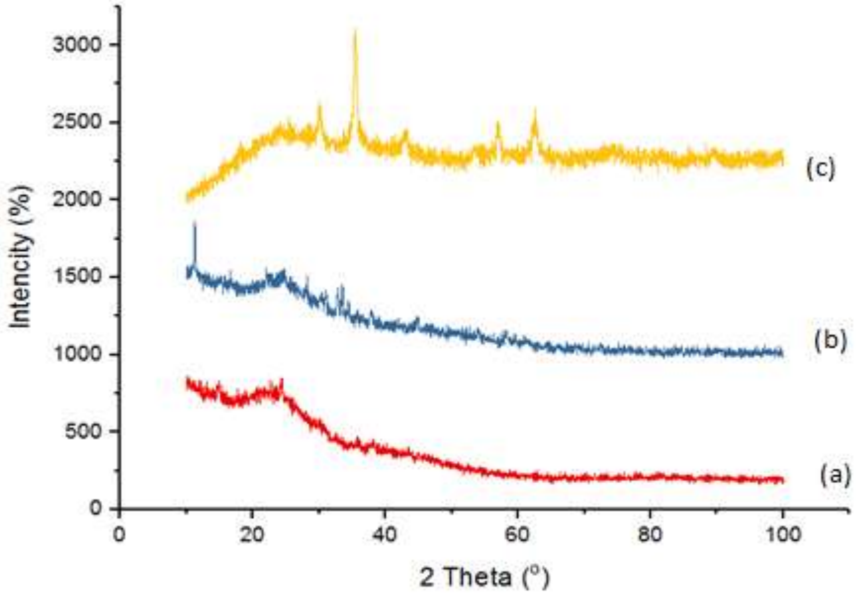

Figure 5: XRD diffractograms samples investigated: (a) inactivated carbon, (b) activated carbon and (c) magnetic activated carbon

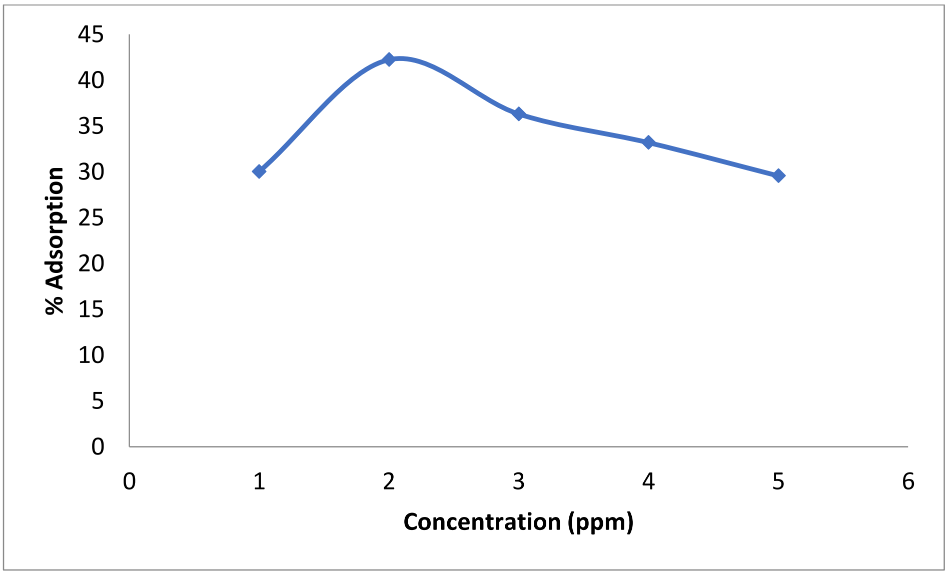

Figure 6: Effect of adsorbate concentrations on adsorption of tetracycline using magnetic activated carbon.

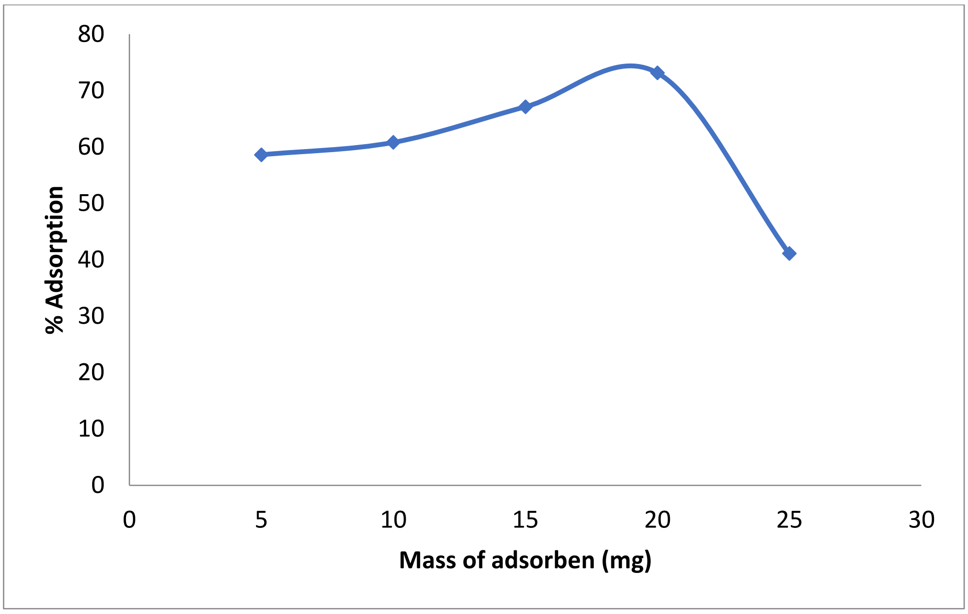

Figure 7: Effect of magnetic activated carbon masses on adsorption of tetracycline.

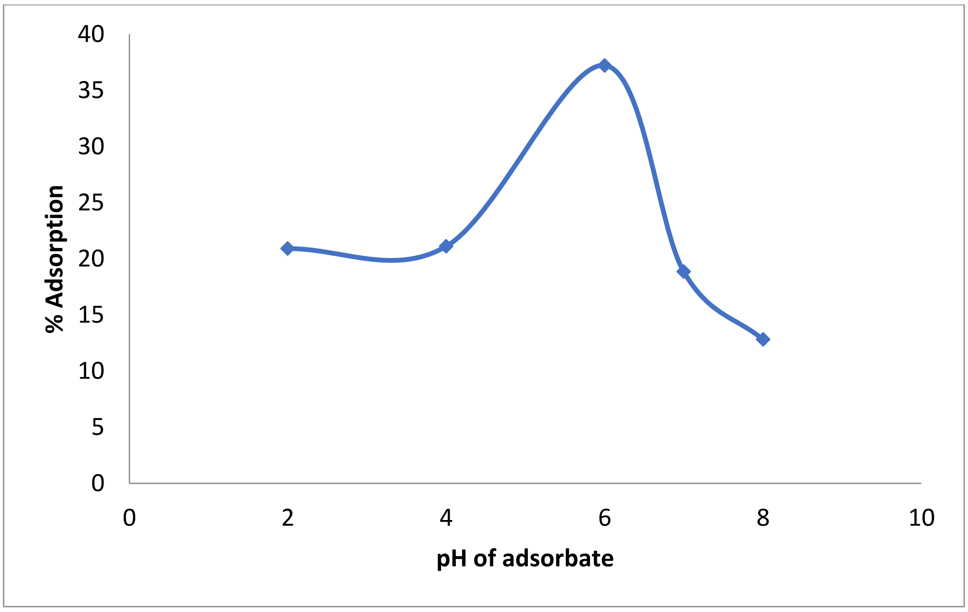

Figure 8: Effect of pH on adsorption of tetracycline using magnetic activated carbon.

As displayed by the experimental results in figure 7, the percentage of adsorption gradually increased along with the increase in the mass of the adsorbent up 20 mg where the highest percentage of adsorption of 73% was achieved, followed by sharp decrease when 25 mg adsorbent was used most likely due to saturation of the surface of the adsorbent. In this respect, it was then concluded that 20 mg is the optimum mass of the adsorbent.

To investigate the effect of pH, adsorption experiments were carried out at pH of 2, 4, 6, 7 and 8, using the adsorbate with the concentration of 1 mg/L and 20 mg adsorbent. The results obtained can be seen in figure 8. Based on Figure 8, it can be seen that the pH greatly affects the adsorption process. The optimum adsorption (37.22%) was achieved at pH = 6. This optimum adsorption is significantly higher than those achieved at more acidic conditions (pH = 2 and 4) as well as that achieved at neutral condition and more alkaline condition (pH = 8).

Tetracycline has a cation form at a pH of less than 3 and an anionic form at a pH of more than 8. While at a neutral pH, it forms zwitter ions and has a neutral charge. Based on the results obtained during the extraction process, tetracycline could be adsorbed by activated carbon, especially in the form of molecules at neutral pH. However, another study showed that pH 4 could also be selected as the optimum condition for the extraction of tetracycline using magnetically activated carbon from rice husk[17]. The experimental results showing the effect of contact times are presented in Figure 9. The experiments were carried out using the optimum concentration of adsorbate, optimum mass of adsorbent and optimum pH which were obtained from previous experiments.

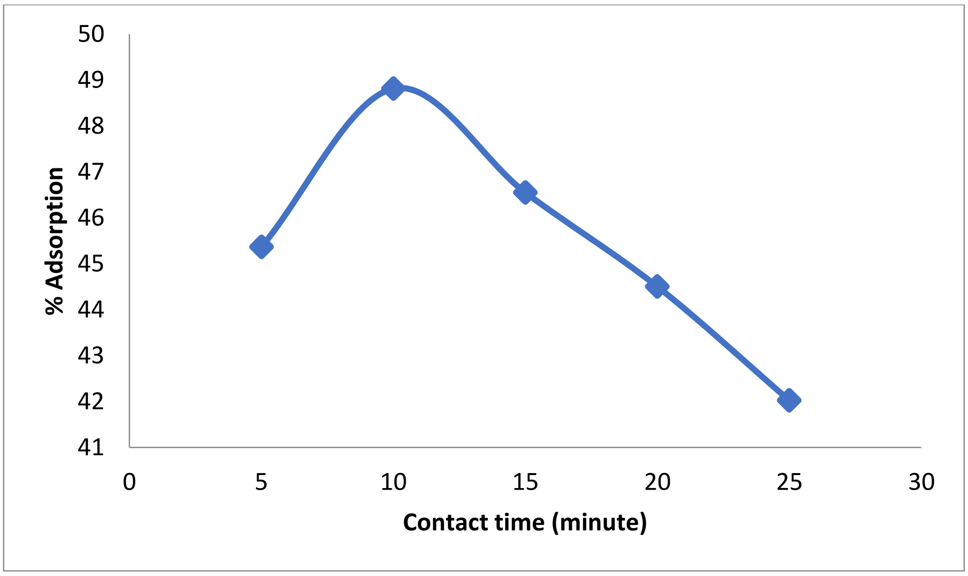

Figure 9: Adsorption test results on the effect of contact time

Based on figure 9, it can be seen that the amount of tetracycline antibiotics adsorbed increased from the first 5 minutes to the optimum time of 10 minutes with percentage of adsorption of 48.82%, while at 15 minutes to 25 minutes the adsorption decreased, most likely because prolonged time led to increased desorption of the tetracycline molecules.

Conclusion

In this study, activated carbon derived from cassava peels was successfully converted into magnetic activated carbon. The magnetic activated carbon prepared was then used as adsorbent for adsorption of tetracycline from solution. A series of experiments were conducted to investigate the effect of adsorbate concentrations, adsorbent masses, pH and contact times on percentage of adsorption. The results obtained indicate that the optimum conditions for adsorption of tetracycline were at the adsorbate concentration of 1 mg/L, the use of 20 mg adsorbent, pH 6 and contact time of 10 minutes. The highest percentage of adsorption achieved is 73 %.

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