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Suryia Manzoor

Bahauddin Zakariya University Institute of Chemical Sciences

Naseem Abbas (✉ dr.naseem.abbas@bzu.edu.pk)

Bahauddin Zakariya University Institute of Chemical Sciences

Javier Fernandez

University College London

Amir Mirza

Muhammad Nawaz Sharif University of Engineering and Technology

Ghazala Yasmin

Bahauddin Zakariya University Institute of Chemical Sciences

Muhammad Imran Khan

Xian Jiaotong University: Xi'an Jiaotong University

Zunaira Rao

Bahauddin Zakariya University Institute of Chemical Sciences

Muhammad Rizwan

Bahauddin Zakariya University Institute of Chemical Sciences

Muhammad Waqar

Bahauddin Zakariya University Institute of Chemical Sciences

Abdul Majeed

Bahauddin Zakariya University

Research

Keywords: Water pollution, Biomass, Activated Carbon, Antibiotic, Cefpodoxime, Adsorption

Posted Date: November 19th, 2020

DOI: <https://doi.org/10.21203/rs.3.rs-110525/v1>

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Reuse of Waste Materials as Precursors of Activated Carbon Synthesis for the Abatement of Antibiotic Cefpodoxime proxetil from Industrial Effluents.

Suryyia Manzoor¹, Naseem Abbas^{1*}, Javier Fernandez^{2*}, Amir Mirza³, Ghazala Yasmin¹, Muhammad Imran Khan⁴, Zunaira Rao¹, Muhammad Rizwan¹, Muhammad Waqar¹, Abdul Majeed⁵

1. Institute of Chemical Sciences, Bahauddin Zakariya University, Multan, Pakistan.
2. Department of Chemical Engineering, University College London, Torrington Place, London WC1E 7JE, UK
3. Muhammad Nawaz Sharif University of Engineering and Technology, Multan, Pakistan.
4. School of Energy and Power Engineering " Xi'an Jiaotong University, 28 West Xianning Road, Xi'an 710049, Shaanxi, PR. China.
5. Faculty of Pharmacy, Bahauddin Zakariya University, Multan, Pakistan.

*Corresponding authors details:

Email: dr.naseem.abbas@bzu.edu.pk

Institute of Chemical Sciences, Bahauddin Zakariya University, Multan, Pakistan.

Phone# 00923009109435

Email: j.fernandez.garcia@ucl.ac.uk

Department of Chemical Engineering, University College London, Torrington Place, London WC1E 7JE, UK

Abstract

The release of antibiotics to aquatic environment creates aquatic ecotoxicity and their bioaccumulation results in antibiotic resistance. Hence to lessen the risk of ecotoxicity and depletion of natural resources, it is imperative to disclose alternate precursor raw materials that can be successfully employed for the synthesis of new sorbents capable to remove antibiotics from the environment. This research reports the synthesis of activated carbon based sorbents derived from waste biomaterials like coconut shell (CNAC), walnut shell & peach stone (WNAC) and *Bombax ceiba* fruit capsule (BCAC) and their ability to remove antibiotic Cefpodoxime from the industrial effluents. Activation of carbon was done by HNO_3 and H_3PO_4 . FTIR, BET and SEM techniques were used for characterization of sorbents. Langmuir and Freundlich isotherm models were applied to study the adsorption behavior. Moreover, the experimental adsorption capacity (q_e) of the three activated carbons was found to be 32 mg/g for CNAC, 40 mg/g for WNAC and 10 mg/g for BCAC. The maximum removal efficiency of the drug was found to be 96% using the WNAC. HPLC analysis was performed to confirm the removal of Cefpodoxime from industrial effluent.

Key words: Water pollution, Biomass, Activated Carbon, Antibiotic, Cefpodoxime, Adsorption

1. Introduction

Antibiotics are being excessively utilized in pharmaceutical, veterinary and aquaculture fields. Release of these antibiotics into the environment is of great concern to the health of living beings. Their presence in groundwater, drinking water and sediments disrupts the ecosystem due to their response towards living organisms and plants even at lower concentration [1]. The exposure of pharmaceutical components on aquatic biota may cause acute persistent damage and accretion in the tissues. Reproduction abnormalities have also been observed in fish exposed to the pharmaceutical waste water effluents. Occurrence of antibiotic resistance bacteria (ARB) and antibiotic resistance genes (ARGs) have drawn much concerns recently, and they have been reported in many environmental media, such as natural rivers, hospital effluents, sewage treatment plants (STPs), harbors, and lakes [2]. The disposal of antibiotics in wastewater from manufacturing plants and the excretion of unmetabolized form by the recipients in waste water is the major source of their unnecessary exposure to the environment [3, 4]. Moreover, degradation

products of antibiotics are also being taken as a globally concerned issue. Pharmaceutical residues found in the water bodies are non-biodegradable and can easily escape the waste water treatment plants [5].

Industries have acquired a broad range of techniques and methods such as catalytic ozonation, bio-remediation, photocatalytic oxidation, coagulation and biological handlings for elimination of antibiotics from industrial wastewater treatment plants [6, 7]. Although, these advance technological processes are well serviceable for industries to complete mineralization and conversion of antibiotic molecules to simpler ones, but these are relatively high in cost and laborious to maintain on industrial scale for efficient removal [8].

Till now, researchers have tested numerous materials as drug adsorbents including activated carbon, carbon nanotubes, bentonite, zeolites, chitosan and ion exchange resins [9, 10]. The efficiency of adsorption processes is highly affected by the type of adsorbent, adsorbate properties, and the composition of precursor material [11]. ACs or modified ACs have been proved to be a potent receptive for antibiotics from the waste streams having efficiency mutable in the range of 74-100% [12]. Biomass is a sustainable resource which can be modified to produce efficient materials. One of the significant utilizations of biomass is to convert it into some valuable sorbents in which activated carbon (AC) has found its numerous applications in diverse fields. Advancement in technology has enabled the use of AC on industrial scale in the fields of catalyst support, metal recovery, electrodes, air pollution minimizer, gas storage, adsorbent for separation science field, super capacitors, food detoxification and decolorizing agent in food industry [13]. It can be used for the enrichment and separation of analytes from liquid and gaseous samples with excellent efficiency.

Performance of AC mainly depends on the experimental conditions and raw materials used for their synthesis or fabrication [14]. Granular AC can be synthesized from organic raw materials by carbonization followed by activation with different activating reagents. Corncob, coal, cotton, wood, gulf weed, fruit stones, fruit shells, etc. can be used as raw materials for AC production. Various activating reagents can be used in chemical synthesis of AC e.g. KOH, K_2CO_3 , NaOH, Na_2CO_3 , $AlCl_3$, $ZnCl_2$, $MgCl_2$, H_3PO_4 and H_2SO_4 [15, 16]. Moreover, AC derived sorbents are ecofriendly, nontoxic and hence can be easily used for removal of pollutants like pesticides, dyes and antibiotics [17]. The adsorption of antibiotics on their surface is significantly influenced by their characteristics such as surface physical morphology and functionality. Adsorptive removal

has been proved efficient, low cost, simple and versatile tool for the transportation of organic compounds from the stream of contaminants to the adsorbent's surface by using different adsorbents [18].

Cephalosporin is a class of antibiotics which includes ceftriaxone sodium, cephalexin, cefazolin, cefalotin sodium, cefpodoxime, etc. Many analytical and bioanalytical methods have been reported for the analysis of cephalosporins [19, 20]. Cephalosporins have been analysed by complexing these antibiotics with; palladium (II) chloride, copper II chelation, Zinc II complexation and parachloranilic acid complexation [21-24]. Many HPLC methods have also been introduced for the analysis of cephalosporins [25]. The use of activated carbon synthesized with controlled surface morphology and chemical characteristics for removal of antibiotics offers an impressive option to overcome the challenging situation of pharmaceutical pollution.

Our foremost objective was to study the ability of activated carbons obtained through different biomass waste for the abatement of cephalosporin, an antibiotic whose removal has not been yet investigated with activated carbon. Moreover, this work reveals the use of bombax ceiba fruit to synthesize activated carbon, which has not been explored before and is one of the common biowastes in Southeastern Asian countries. Hence, the activated carbon with defined surface functionalities was prepared from three different raw materials (mixture of walnut shell and peach stone, coconut shell and babsu) under the controlled conditions for selective removal of cefpodoxime from the industrial effluents. Cefpodoxime Proxetil (CP) is a broad spectrum, orally directed, semi-synthetic antibiotic belongs to the cephalosporin class. The chemical formula of CP is (RS)-1(isopropoxycarbonyloxy) ethyl(+) - (6R,7R) - 7 - [2 - (2 - amino - 4 - thiazolyl) - 2 - acetamido] - 3 - methoxymethyl - 8 - oxo - 5 - thia - 1 - azabicyclo [4.2.0]oct-2-ene-2-carboxylate and empirical formula of CP is $C_{21}H_{27}N_5O_9S_2$. Its molecular weight is 557.6 with pKa value is 2.5 [26] [27]. Time, pH and initial concentration have been optimized to obtain the maximum efficiency from the synthesized adsorbents.

2. Materials and methods

All the chemicals utilized in this experiment were of high analytical grade. Cefpodoxime proxetil (91.92%) was purchased from pharmaceutical company Stand pharm (Pvt.) Ltd. Highest purity (>98%) reagents were used without any purification. Methanol (99.9% Merck), potassium

dihydrogen phosphate (99.5% Merck), disodium hydrogen phosphate (99% Merck), ortho-phosphoric acid (85% Merck) was purchased and of HPLC grade. HCl (35.4% ANALAR) and NaOH (99% Merck) were used to maintain the pH of the solution. High grade KOH (85-100.5% Sigma) and HNO₃ (65% Merck) were bought from Sigma and Merck respectively. All stock and standard solutions were prepared in deionized water (MilliQ).

2.1. Synthesis of activated carbon from waste biomasses

Three different raw materials i.e. coconut shells, Bombax Ceiba fruit capsule and a mixture of walnut shells: peach stones (50:50% by mass) were selected for the synthesis of activated carbon.

The raw materials (coconut shells and a mixture of walnut shells: peach stones) were washed with deionized water, dried at 110 °C for 48 h, crushed and sieved to a size range of 1-2mm. In case of Bombax Ceiba fruit capsule, the collected raw material was washed with deionized water and dried in oven for 1 day at 110 °C. Afterwards, all of the three samples were kept in the furnace for carbonizing the raw material at 600 °C for one hour. Carbonized materials were then ground and activated with 0.1 N solution of HNO₃ and 0.1 N solution of H₃PO₄. Impregnated carbonaceous material was kept in furnace for 1 h at 650 °C.

Hence, the three types of activated carbons i.e. coconut shell based activated carbon (CSAC), walnut shells and peach stones based modified activated carbon (WPAC), Bombax Ceiba fruit capsule based activated carbon (BCAC) were thus obtained and washed with excess of deionized water until the neutral pH was achieved.

2.2. Characterization

Nature of activating reagent and precursor raw material play a vital role in creating a wide range of functional groups on AC surface. To characterize these groups, non-destructive spectroscopic FTIR instrument Shimadzu model 1800S in the range of 500-4000 cm⁻¹ was used. Frequent pores with distinct pore volume were formed by the reaction of AC with activating reagent, therefore nitrogen adsorption/desorption studies were performed to study the pore size. JEOL SEM 6360-LV microscope was used to scan the surface morphology of adsorbent.

2.3. Batch Adsorption Studies

For batch adsorption studies, 20 mL of cefpodoxime solution of various concentrations within the range of 6 to 60 mg/L was added to 0.02 g AC, shaken in a thermostatic shaker (120 rpm) for a fixed time interval, solutions were filtered and the absorbance was measured at 245 nm. Similarly, the time optimization was studied in the range of 30 to 180 minutes and pH optimization was performed within range of 1 to 7.5.

Adsorption capacity q_e (mg/g) was determined by equation 1

$$q_e = (C_i - C_e) / M \times V \quad (1)$$

Removal efficiency was calculated by

$$\%R = (C_i - C_e) / C_i \times 100 \quad (2)$$

C_i = initial concentration of sorbate (mg/L), C_e = equilibrium concentration of sorbate (mg/L),

V = volume of adsorbate solution (L), M = is the adsorbent amount (g).

Also, Langmuir and Freundlich isotherms were plotted to study the adsorption behavior of cefpodoxime on activated carbon.

2.4. Removal of Cefpodoxime proxetil from industrial effluents by using prepared adsorbents

2.4.1. Mobile phase preparation

Phosphate buffer solution (pH 4.0) was prepared by mixing 2.5g disodium hydrogen phosphate and 1.5 g potassium dihydrogen phosphate in 1 liter deionized water. Then this Phosphate buffer solution was mixed with methanol in ratio of 65:35. Prior to use, mobile phase was filtered and degassed.

2.4.2. Preparation of Sample Solution

Industrial effluents were collected from a local industry. Sample solution was spiked by adding known amount of Cefpodoxime proxetil to the stock solution of sample. To the spiked solution, activated carbon was added and under optimized conditions, samples were left for adsorption.

The activated carbon was then filtered and shaken in methanol to desorb the drug. The solution was further analyzed by HPLC.

2.5. Analysis of sample

20 mL sample solution was introduced to 0.02g of adsorbent at pH 3, shaken for 90 minutes (optimized time) and then filtered by using nylon micro filters (0.22 micron) followed by HPLC-UV (SHIMADZU LC10ATVP, C18 column) analysis of the filtrate. Analysis was followed by conditioning of the column with mobile phase for 30-40 minutes. Flow rate was maintained at 1.0 mL/min and wavelength of the detector was adjusted at 245nm.

3. Results and discussion

3.1. Characterization of adsorbent

FTIR being a conventional technique provides valuable information about the functionality of the adsorbent and hence it can be employed for the identification of functional groups present on activated carbon surface.

<<Fig. 1>>

FTIR spectra of the three activated carbons were taken to study the surface functionalization, which in turn affect the adsorption properties. In case of CNAC, a single weak and bit sharp band was observed at 3500 cm^{-1} which can be possibly attributed to the secondary amine on carbon surface. Similarly, bands at 2200 cm^{-1} , 2050 cm^{-1} , 1800 cm^{-1} and 1400 cm^{-1} were also located in the spectrum presenting the possible existence of -C-C= , -CH , C=O , -CH_2 . Similar bands were also observed on the surface of WNAC and BCAC suggesting the success of carbon surface activation, making all the three carbons viable for the adsorption.

Scanning electron microscopy (SEM) was used to observe the surface physical morphology of the activated carbon materials. Carbonization, activation and evaporation of volatile compounds resulted in widening of porous networks in AC. SEM images proved that the synthesized materials were porous with variable surface topology (figure 2). The pore volume was calculated by the amount of nitrogen gas adsorbed to the carbon surface which was found to be $0.227\text{ cm}^3/\text{g}$ for WNAC and BET surface area was calculated to be $521\text{ m}^2/\text{g}$. Similarly, CSAC presented a pore volume of $0.043\text{ cm}^3/\text{g}$ and surface area of $104\text{ m}^2/\text{g}$ and for BCAC, $0.105\text{ cm}^3/\text{g}$ and

surface area of 448 m²/g. The large pore volume can substantially enhance sorbate diffusion in activated carbons resulting in an increase in sorption capacity. WNAC presented greater pore volume and hence more sorption capacity was expected which was later confirmed by the q_e value. The q_e was the highest in case of WNAC as a sorbent.

<< Fig. 2>>

3.2. Batch adsorption studies

Three parameters (pH, time of contact, concentration of drug) were optimized to carry out batch adsorption of cefpodoxime.

Time of contact between cefpodoxime and AC was studied in the time range of 30 to 360 minutes. CNAC and WNAC required 90 minutes to attain maximum adsorption capacity (q_e) whereas in case of BCAC, 150 minutes were taken to achieve maximum q_e . Further increase in time did not affect the adsorption of the drug probably due to higher surface coverage [28]. However, other parameters were also optimized to further improve the adsorption capacity of the sorbents. For this purpose, effect of pH was studied in the next step as it can affect the degree of speciation and ionization of the adsorbate [29]. Highly basic pH was avoided as cefpodoxime decomposes above 7.5 pH [30]. Considering the figure (4a and b), drug adsorption decreased by increasing the pH of adsorbate from 3 to 4 or 5. Reduced adsorption above pH 3 explained that the drug was in its unionized form in the lower acidic region and decomposed in the strong acidic region. In figure 4c, by increasing the pH from 2 to 3 adsorptions increased rapidly (adsorption 72%) due to the fact that attraction between solute and surface functional groups was stronger than the attraction of solvent and solute molecules at this pH. On BCAC, maximum adsorption occurred at pH 4.

<< Fig. 3>>

<<Fig. 4>>

Similarly, the effect of initial concentration of Cefpodoxime was also studied. In case of CNAC and WNAC sorbents; the adsorption increased rapidly by increasing initial concentration of cefpodoxime upto 50 mg/L and maximum experimental q_e of 32 mg/g and 40 mg/g were obtained respectively. By further increasing the concentration, adsorbents showed constant q_e

value which depicted that there were no more available sites for adsorption. For BCAC, maximum experimental q_e was calculated to be 10 mg/g (figure 5).

<<Fig. 5>>

Further, Langmuir and Freundlich isotherms were applied to comprehend the adsorption phenomena of drug on modified activated carbons. Though Langmuir isotherm was primarily designed to explain the adsorption of gas molecules on homogenous surfaces, however, later it was further explored to study the behavior of solutions on solid surfaces [31].

Relationship for Langmuir isotherm:

$$1/q_e = 1/q_{max} + 1/q_{max} \cdot C_e \quad (4)$$

q_e in the Langmuir equation presents the adsorption capacity at equilibrium while the concentration at equilibrium is C_e . $1/q_{max}$ and b are constants which are obtained from regression equation where $1/q_{max}$ are intercept and b is slope [32].

In order to study the behavior of adsorbate on heterogeneous surfaces, generally Freundlich adsorption isotherm is applied and the relationship for Freundlich isotherm which was used in the following:

$$\log q_e = \log Kf + 1/n \log C_e \quad (5)$$

In equation 5, q_e is the adsorption capacity at equilibrium, C_e is the concentration at equilibrium, $1/n$ and K are constants that are calculated from regression equation where $1/n$ is slope and K is intercept. The activated carbons CNAC and WNAC predominantly followed Freundlich isotherm and the R^2 values were found to be 0.86 and 0.97 respectively. The activated carbons possess porous structure having rough surface and hence there is a possibility of multilayer adsorption. While in case of BCAC, Langmuir isotherm presented a higher R^2 value that was found to be 0.96 and for Freundlich isotherm, R^2 was 0.92. Apparently, BCAC followed Langmuir phenomenon yet it can be observed that the difference between the correlation values was quite small which may possibly suggest that none among them could predominantly define the adsorption behavior of solute on BCAC (figures 6 and 7).

<<Fig. 6>>

<<Fig. 7>>

<<Table 1>>

3.2.1. Kinetic studies

Kinetics of cefpodoxime sorption by CNAC, WNAC, and BCAC in the time range of 30 to 360 minutes was carried out at constant temperature.

The equation for the first-order kinetic model is given in equation 6.

$$\log(q_e - q_t) = \log q_e - \frac{k_1}{2.303} t \quad (6)$$

where q_e is the adsorption capacity at equilibrium and q_t represents the amount of cefpodoxime adsorbed on adsorbents (mg g^{-1}) (CNAC, WNAC, and BCAC) at time t . k_1 is the rate constant (min^{-1}). Straight line plots of $\log (q_e - q_t)$ against t were used to determine the rate constant, k_1 and correlation coefficient, R .

The second order equation may be expressed as:

$$\frac{t}{Q} = \frac{1}{k_2 Q_e^2} + \frac{t}{Q_e} \quad (7)$$

where k_2 is the rate constant of second-order adsorption (g (mg min)^{-1})[33, 34].

<<Fig. 8>>

Straight-line plots of t/q_t against t were tested to obtain rate parameters and the results suggested the applicability of this kinetic model to fit the experimental data.

The outcome of the kinetic study for all three adsorbent are given in figure 8 and table 2. Based on the correlation coefficients, the adsorptions of cefpodoxime are best described by the second order equation. The first-order equation does not fit well to the initial stages of the adsorption processes but the second-order equation better correlates to the adsorption studies.

3.3. HPLC analysis of Cefpodoxime proxetil in industrial effluents

HPLC analysis (figure 9) was performed to confirm the adsorption of respective drug on activated carbon. As WPAC presented highest sorption capacity, so it was selected for real sample analysis. The peak at 15 minutes is attributed to the cefpodoxime in spiked samples of

industrial effluents. A little shift in retention time was observed in the chromatogram of desorbed drug. This small variation in the retention time in desorption chromatogram may be due to the sample complexity (industrial effluents). The AC was able to remove maximum amount of drug from the sample which can be confirmed from the percentage removal efficiency. The percentage removal efficiency of cefpodoxime from the sample by using WNAC modified activated carbon was found to be 96%. WPAC also presented selectivity to an extent which can be clearly observed from the chromatograms in figure 9. The peak that appeared at 18 min in spiked industrial effluent before adsorption, no longer exists in the desorbed sample.

<<Fig. 9>>

Previously, C8 has been used in the cartridges for the selective adsorption of drug as a sample pre-clean up step from biological samples (blood plasma) followed by HPLC. An extraction efficiency of 95% was attained [35, 36]. However, C8 being an expensive material as compared to low cost activated carbon is impractical to be used at large scale cleanup of the drug. Hence, the waste biomass based activated carbon can be considered as a potential sorbent for removal of drug from sizeable environmental samples.

Conclusion

The study was based on the synthesis of activated carbon from different plant materials by acid activation. Characterization by FTIR, SEM and BET confirmed the porosity of activated carbon with large surface area to volume ratio. Cefpodoxime was chosen for adsorption due to its long term side effects and its accumulation in industrial waste water. WNAC showed best results for adsorption of cefpodoxime (40 mg/g) at pH 3. For real water samples, HPLC analysis was performed followed by adsorption of cefpodoxime on activated carbon. WNAC offered maximum percentage removal of 96%. HPLC analysis before adsorption and after desorption confirmed the adsorptive capability of AC towards cefpodoxime.

DECLARATIONS

Availability of data and materials

All data generated or analyzed during this study are included in this published article [and its supplementary information files].

Competing Interest

The Authors have no conflict of interest.

Funding

Not applicable

Author's Contributions Statement

This research article is the outcome of M.Phil. theses of Zunaira Rao, Muhammad Rizwan and Muhammad Waqar under the supervision of Javier Fernandez, Suryya Manzoor and Naseem Abbas. Amir Mirza wrote initial report. Ghazala Yasmin and Abdul Majeed helped to access the instruments used for the purpose of characterization as well as interpretation of the results. . Muhammad Imran Khan substantiated the analytical methods. All authors discussed the results and contributed to the final manuscript.

Acknowledgement

The authors are grateful to the Institute of Chemical Sciences, Bahauddin Zakariya University, Multan, Pakistan for providing the laboratory facilities to conduct the experimental part.

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Figures

Figure 1

FTIR of activated carbon (a) CNAC (b) WNAC and (c) BCAC.

Figure 2

SEM of activated carbon samples (a) CNAC (b) WNAC and (c) BCAC.

Figure 3

Time optimization (a) CNAC, (b) WNAC and (c) BCAC.

Figure 4

pH optimization (a) CNAC, (b) WNAC and (c) BCAC.

Figure 5

Adsorbate concentration optimization (a) CNAC, (b) WNAC and (c) BCAC.

Figure 6

Langmuir adsorption isotherms of a) BCAC, b) CNAC, c) WNAC

Figure 7

Freundlich adsorption isotherms of a) BCAC, b) CNAC, c) WNAC

Figure 8

Pseudo-second order kinetics of a. BCAC, b. CNAC, c. WNAC

Figure 9

HPLC chromatograms of industrial effluents spiked with cefpodoxime proxetil a) before adsorption of the drug b) desorbed sample

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