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Evaluation of Langmuir and Freundlich Isotherms for Removal of Cephalexin and Tetracycline Antibiotics By Sistan Sand from Water and Wastewater Samples

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Abstract
In this research, Sistan sand was used as a natural and inexpensive sorbent for removal of cephalexin and tetracycline antibiotics from water and wastewater samples. For a concentration 60.0 mg L^{-1} of cephalexin, optimum removal conditions were: pH of the sample 3.0, adsorbent amount 1.0 g, contact time 20.0 min, added amount of sodium chloride to adjust the ionic strength of the solution 7.0 g L^{-1}. Langmuir isotherm was the best fitted model for this adsorption process and adsorbent capacity was calculated to be 0.26 g g^{-1}. This adsorbent was able to remove up to 68.1% of cephalexin from wastewater. In case of tetracycline, for a 90.0 mg L^{-1} of the analyte, the optimum adsorption conditions were achieved at pH 8.0, 1.0 g of sorbent, contact time of 35.0 min and ionic strength of the solution as sodium chloride of 7.0 g L^{-1}. The isotherm was best in agreement with Freundlich model. Adsorbent capacity was 0.76 g g^{-1} and up to 76.2% of this antibiotic could be removed from wastewater.

Keywords
Cephalexin; Tetracycline; Langmuir Isotherm; Freundlich Model; Wastewater Analysis.

1. INTRODUCTION

Because of increasing consumption of pharmaceuticals, these compounds can be entered to the environment through anthropogenic excretion. Also, they cannot be removed during water treatment efficiently. Pharmaceutical could also be released to water resources from pharmaceutical manufacturers, hospitals, agriculture, livestock and aquaculture [1-3]. Although the concentrations of pharmaceuticals in environmental waters are generally at ng L^{-1} to µg L^{-1} levels, they can create cytotoxicity and ecotoxicity to living organisms that makes a global issue using increasing environmental concerns. Particularly, the occurrence and consumption of antibiotics (as major category of pharmaceuticals) has led to entrance of the antibiotic in both natural media and ecosystems that has been recognized as a major threat for public health worldwide [1, 4-6]. Cephalaxin (CEP, a type of semi-synthetic cephalosporin antibiotic) and tetracycline (TC) are two common antibiotic drugs. Because of their broad antibacterial activity, they are generally applied for treatment of infection throughout the body. TC also has uses in veterinary medicine. Their continual introduction in the environment would cause serious ecological risks [1,7-9], and therefore removal of CEP and TC from environmental water is important and serves as an attractive research case.

Adsorption to solid surfaces is a simple and effective technique to remove pollutants from water and wastewater. Non-ionic polymeric resins, ion exchange resins, oxidized celluloses, H_2O_2 oxidized activated carbons, solid phase extraction, carbon nanotubes, molecularly imprinted polymers, etc. have been proposed as sorbent for the removal of CEP and TC [7-9]. Among instruments which were applied for determination of these antibiotics, Fourier transform infrared radiation [7], high performance liquid chromatography with ultraviolet detection [8], spectrophotometry [9] and luminescence spectrometry [9] are the most common techniques. In this research, Sistan sand was applied as a readily available sorbent for removal of CEP and

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TC from water and wastewater samples. To find out the optimum removal conditions, the effective parameters on removal of CEP and TC were investigated. Analysis and detection of the target analytes were performed by spectrophotometry.

2. EXPERIMENTAL

2.1. Materials and methods

Sistan sand which was used in this study as antibiotics adsorbent was collected from Sistan area, south east of Iran. All reagents and chemicals including Cephalexin and Tetracycline hydrochloride were of analytical reagent grade (Sigma-Aldrich Co., (MO, USA)) and were used without more purification. Doubly distilled water was applied overall the research. Fresh stock standard solution of 100 mg L^{-1} of the analytes was made daily using dissolving 100 mg of the target compounds in 1000 mL of ultrapure water which was then maintained at 4 ^{0}C in refrigerator away from light. Working standard solution were obtained using proper dilution of the stock solution prior analysis.

2.2. Apparatus

Spectrophotometric analysis were performed on a Perkin-Elmer model Lambda 25 double beam spectrophotometer (USA) at a wavelength of 262 for CEP and 275 for TC. A model Easyseven Metrohm (Switzerland) pH meter was utilized for the determination of pHs. A Perkin-Elmer (Bucks, UK) Fourier transform infrared (FTIR) spectrometer was applied for qualitative spectral interpretation as well as for structural elucidation of sand adsorbent. The morphology of Sistan sand was studied with scanning electron microscope (SEM), model MIRA-3-TESCAN (Czech Republic). Powder X-ray diffraction (XRD) measurements were performed on a Bruker instrument (model D8 Advance, Germany).

2.3. Calibration curve plotting

Calibration curves for the analytes were separately plotted by changing concentrations of standard solution of them. The results showed that the calibration curve was linear over a concentration range of 0.6-60.0 mg L^{-1} and 0.6-50.0 mg L^{-1} for CEP and TC, respectively. The least square equations over the dynamic linear range were:

A (for CEP) = 0.039 C (mg L^{-1}) + 0.0365 R^2= 0.998

A (for TC) = 0.0416 C (mg L^{-1}) + 0.0509 R^2= 0.997

Where, C and A are the concentration of the antibiotic and absorbance (spectrophotometer response), respectively.

2.4. Removal procedure

To investigate extraction ability of the adsorbent, 20.0 mL of drugs solution (60.0 and 90.0 mg L^{-1}, respectively for CEP and TC) were examined. pH of the solution was adjusted to 3.0 (for CEP) and 8.0 (for TC) by drop-wise addition of either 1.0 M of HCl or 1.0 M NaOH. Next, 0.25 g of NaCl and 1.0 g of sand sorbent were added to the solution and stirred for 20 (for CEP) and 35 min (for TC). Finally, the solution filtered through filter paper and drug concentrations in solution were determined by spectrophotometry.

3. RESULT AND DISCUSSION

3.1. Absorption spectra

Since spectrophotometric method was used to determine the concentration of CEP and TC antibiotics in water samples, their absorption spectra in optimized parameters were recorded at the wavelength range of 240-320 nm against the reagent blank and are showed in Fig. 1. As it is obvious, maximum absorption wavelengths were 262 nm for CEP and 275 nm for TC.

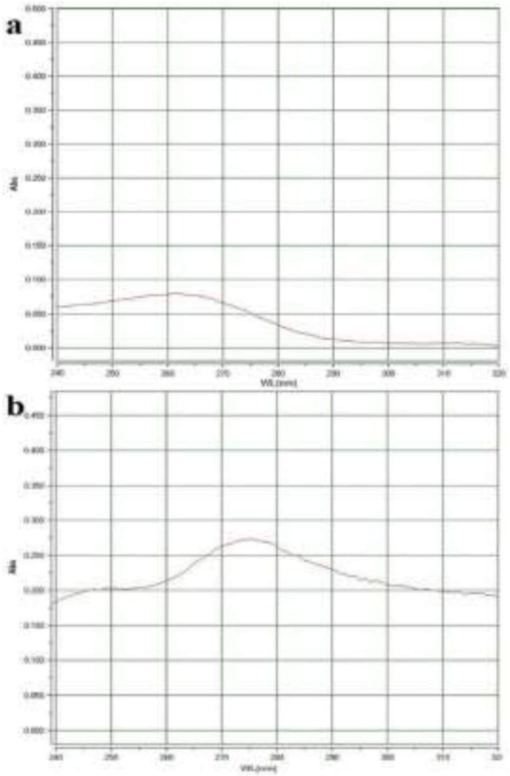

Fig. 1. Absorption spectra of 15 mg L^{-1} of CEP and TC. Conditions: sample solution, 20.0 mL; pH= 3.0 (for CEP) and 8.0 (for TC); amount of NaCl, 0.25 g; mass of sand sorbent, 1.0 g.

3.2. Preparation of sorbent

To remove contaminates from the sorbent, Sistan sand was washed 3 times by tap and then by doubly distilled water. Fig. 2 shows scanning electron microscope (SEM) images of Sistan sand after cleaning. In order to obtain uniform sand particles for better reproducibility, it was passed through laboratory sieves by different meshes (40, 60, 80, 100 and 120). Removal tests on 50.0 mg L^{-1} standard solutions of both analytes were performed with them and it was found that while smaller particles have up to 35% higher extraction efficiency, but their separation from the solution by filtration takes a long time, and finally 60 grading was used in the experiments. The average size of sand particles was around 4 μm.

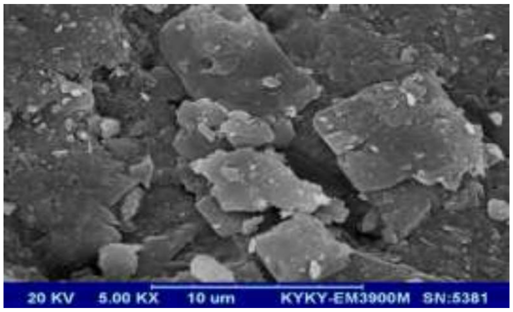

Fig. 2. SEM images of Sistan sand.

3.2. FTIR spectrum of sand

FTIR spectrum of sand was recorded and depicted in Fig. 3. The main band at 1004 cm^{-1} shows that the nature of the sand is quartz and also the presence of quartz is proved with the other bands at 776, 695, 531 and 462 cm^{-1}. The peak at 1750 cm^{-1} is related to the vibration of the carbonyl group.

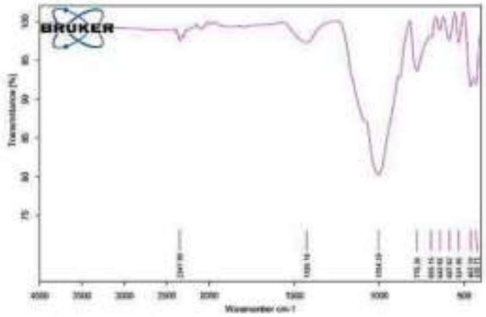

Fig. 3. FTIR spectrum of Sistan sand.

3.3. Optimization of removal protocol

To achieve maximum extraction of the antibiotics by Sistan sand, different factors which potentially could affect the removal process were checked out based on one-variable-at-a-time method. Aliquots of aqueous standard solution of each analyte at a concentration of 60.0 and 90.0 mg L^{-1}, respectively for CEP and TC were used for the following experiments. Each run was repeated at least three times.

3.3.1. Effect of pH of the solution

The effect of the pH value on the removal of CEP and TC from aqueous samples were studied in the range of 2.0 to 9.0. The percentage of CEP removal was increased from 2.0 to 3.0 and then decreased. At low pHs, H^+ absorbed with the basic group of the sorbent. So, it become positive and electrostatic attraction of CEP increases. At higher pHs, due to the presence of OH^- ions and its higher mobility in comparison with drug, CEP cannot be effectively adsorb and its removal is reduced. For TC, the removal efficiency was at the highest point at pH 8.0, which is may be due to the ionization of the antibiotics. In subsequent experiments, the pH of solutions were adjusted to 3.0 (for CEP) and 8.0 (for TC).

3.3.2. Effect of ionic strength of the solution

In this study, the influence of salt on the adsorption process was studied at the presence of sodium chloride within the concentration range of 0.05 to 10.00 g L^{-1}. It was observed that by increasing the amount of NaCl up to 7.00 g L^{-1}, the efficiency of removal of drugs increased, while after this point, the efficiency became constant. This effect can be probably due to the more ease of protonation of the drugs at a media with higher ionic strength which makes them positively charged; so they become more available to be electrostatically bond to the adsorbent.

3.3.3. Effect of the amount of the adsorbent

To obtain the highest absorption of CEP and TC, the amount of sand for suggested technique was changed from 0.1 to 1.1 g. With increasing mass of sand up to 1.0 g, the removal efficiency increased and further increase in amount of sorbent expressed no considerable change (Fig. 4). Percentage of removal increased by increasing the amount of sorbent because of increasing number of active surface sites available. After a certain point, almost all antibiotic was adsorbed and further increasing in the amount of sand had no effect on the efficiency.

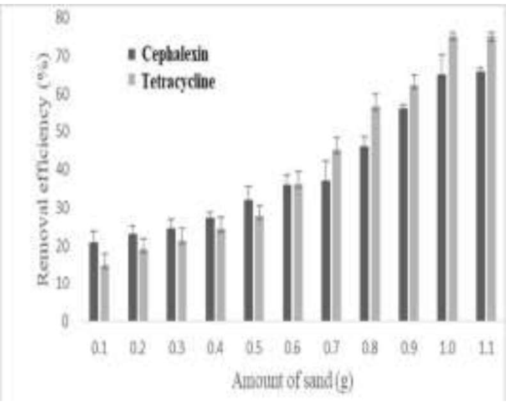

Fig. 4. Effect of amount of adsorbent. Conditions: sample solution, 20.0 mL; pH= 3.0 (for CEP) and 8.0 (for TC); 0.25 g of NaCl; concentration of the analytes, 60.0 and 90.0 mg L^{-1}, respectively for CEP and TC.

3.3.4. Effect of contact time

The influence of the contact time on the absorbance surface was also investigated. The removal efficiency was increased up to 20 min (for CEP) and 35 min (for TC) and then became constant. Accordingly, 20 (for CEP) and 35 min (for TC) were selected as optimal contact time.

3.3.5. Effect of initial concentration of antibiotics

To investigate of the drug concentration effect on removal efficiency, experiments were performed at different initial concentration of CEP (20-60 mg L^{-1}) and TC (25- 90 mg L^{-1}). The percentage of removal increased with increasing of initial concentration of CEP and TC up to 60 and 90 mg L^{-1}, respectively, and then became constant. The drugs initial concentration provides the mass transfer force. At lower concentrations, the number of activated sites of the sand sorbent are high and all analyte molecules can interact by them. At higher concentrations of drugs due to saturation of active sites on the sorbent, removal percentages became constant.

3.4. Equations related to removal efficiency of the analytes and sorbent capacity

The percentage of surface adsorption of drug was calculated from eqs. 1 and 2.

Removal efficiency (%) = $(C_0-C_e)/C_0)\times100$ (1)

C_e=(width of the origin of calibration curve– observed absorption)/(slope of calibration curve) (2)

Where C_0 and C_e are the initial and equilibrium (concentration that remains after absorption of the drug of the solution by sand sorbent) concentrations.

The amount of absorbed analytes in unit of sorbent mass was achieved from eq 3.

$q_e=((C_0-Ce)\times V)/m$ (3)

Where V, m and q_e are volume of sample (L), mass of sorbent (g) and sorbent absorbent capacity (mg g^{-1}), respectively [10].

3.5. Determining surface adsorption isotherms

Isotherms of surface adsorption show the relationship between the concentrations of adsorbed analytes in unit of sorbent mass and concentration of residual analytes in solution. By study of isotherms of surface adsorption with mathematical techniques, the properties of sorbent surface, absorption energy and its mechanism is investigated. Isotherms of surface adsorption is investigated by a defined mathematical model that has two or more unknown parameters, matched by experimental values. Finally, after finding suitable values for parameters, the desired isotherm curve was drawn and its proximity to the experimental data was examined. In the present research, Excel® software was used to perform of mathematical operations and also unknown values and curves were plotted by the same software. Two models surface adsorption isotherms, Langmuir and Freundlich isotherms, have been investigated in this study. To investigate the proximity of the achieved values of calculations of isotherms by the experimental values, the R parameter is defined (eq. 4). Smaller R means that there is only a small difference between the experimental data and models data is existed [11].

$R= (\sum(Q_i- q_i)^2/(m-2))^{½}$ (4)

Q_i: experimental value of the absorbed drug concentration with the sand sorbent (mg g^{-1})

q_i: the amount of absorbed drug concentration estimated by the isotherm equation (mg g^{-1})

m: number of obtained data of experimental method

a) Investigation of Langmuir isotherm on surface adsorption of CEP and TC

Langmuir isotherm is the most common isotherm for investigating the adsorption process. It is applied for single-layer adsorption on a surface including a limited number of similar adsorption sites. The model is based on the following three hypothesis:

1: each adsorption site holds a single absorbed molecule

2: all adsorption sites are similar

3: there is no interaction between adsorbed molecules at adjacent absorption sites.

According to the hypotheses mentioned and the assumption of equality of adsorption rate and analyte desorption on the surface at equilibrium, the Langmuir equation as eq. 5 [12]:

$Q_e=(K_L\times C_e\times q_{max})/(1+(K_L\times C_e))$ (5)

q_e: amount adsorbed on the sorbent in equilibrium (mg g^{-1})

q_{max}: maximum possible adsorption on the sorbent (mg g^{-1})

C_e: adsorbed molecule concentration in equilibrium (mg L^{-1})

K_L: associated constant by surface adsorption heat (L mg^{-1}).

b) Investigation of Freundlich isotherm on surface adsorption of CEP and TC

Freundlich isotherm is based up on the non-uniformed adsorption of analyte in occupied active sites and shows the degree of interaction between adsorbed analytes and the sorbent. The Freundlich equation is an empirical equation based on the adsorption on a heterogeneous surface and states that the absorbed molecules are absorbed as multi-layer. An optimal surface adsorption has a value of *n* between 1 and 10. Greater values of *n* are referred to stronger interaction between sorbent and absorbed analytes. When *n* is equal to 1, it means that absorbed analytes has the same energy for all absorption sites (eq. 6) [13].

$$q_e=K_f C_e^{1/n} \quad (6)$$

n: absorption intensity

KK_f: constant related to adsorption capacity in a single concentration

Under the optimal conditions achieved for removal of the drugs, solutions with different concentrations in the linear range were prepared and examined. After removal of antibiotics, the concentration of the analytes in solution were obtained and the curves of various isotherms were plotted and evaluated accordingly. The Langmuir (Figs. 5a and b) and Freundlich (Figs. 5c and d) isotherms showed to be the best matches with experimental results. Absorption capacity of sand sorbent for removal of CEP and TC (according to eq. 3) were calculated as 263 mg g^{-1} and 769 mg g^{-1} for CEP and for TC, respectively. Table 1 shows calculated parameters by matching adsorption isotherms of CEP and TC between Sistan sand sorbent and experimental data.

3.6. Real samples analysis

To evaluate the efficiency of the suggested protocol in real samples, the procedure was employed to remove CEP and TC from tap water and from an industrial wastewater released from a poultry food factory as real samples. Results showed that the analytes were not existed in them, therefore, to evaluate the effect of sample matrix on technique efficiency, samples was spiked with the analytes in various concentrations and the experiments were carried out under optimized conditions. Removal efficiency of CEP and TC with Sistan sand was calculated and depicted in Table 2. Comparison of the research by other reported methods can be found in Table 3.

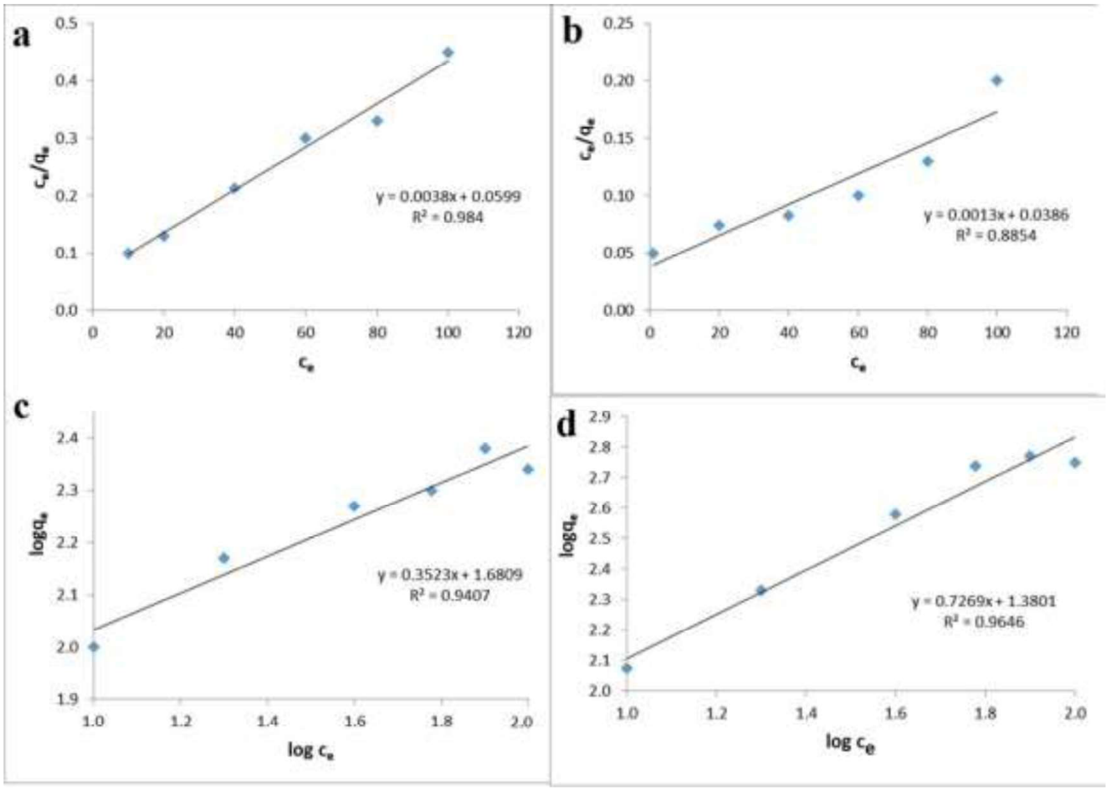

Fig. 5. Langmuir and Freundlich isotherms curve for surface absorption of CEP (a (Langmuir isotherm) and c (Freundlich isotherm)) and TC (b (Langmuir isotherms) and d (Freundlich isotherms)) by the sand sorbent (q_e (point in compared to the obtained curve of experimental data and Q_i (linear curve)).

Table 1. Calculated parameters by matching adsorption isotherms of CEP and TC with experimental data.

Analyte	Isotherm	n*	K	q_{max} (mg g^{-1})	R^2
CEP	Langmuir	-	0.063 (L mg^{-1})	263.0	0.98
	Freundlich	2.83	5.300 ($mg^{1-1/n}$ $L^{1/n}$ g^{-1})	-	0.94
TC	Langmuir	-	0.033 (L mg^{-1})	769.0	0.88
	Freundlich	1.30	3.938 ($mg^{1-1/n}$ $L^{1/n}$ g^{-1})	-	0.96

* Absorption intensity

Table 2. Results of removal efficiency for CEP and TC from industrial wastewater and tap water with Sistan sand sorbent.

Analyte	Spiked concentration (mg L^{-1})	Removal efficiency (%)		Repeatability (RSD%, n=3)
		Wastewater	Tap water	
CEP	30	68.17	66.08	3.6
	40	65.60	65.17	2.9
TC	10	76.27	74.14	2.5
	20	71.10	70.08	2.1

Table 3. Comparison of the adsorption capacity of various sorbents for CEP and TC drugs.

Analyte	Sorbent	Sample	Adsorption capacity (mg g^{-1})	Ref.
CEP	Activated carbon	Aqueous solutions	48.78	[14]
	Activated Carbon	Aqueous solutions	199.60	[15]
	Activated-carbon-impregnated magnetite composite	Aqueous solution	114.9	[16]
	Sistan sand	Wastewater	263.0	This work
TC	Iron modified zeolite	Environment water samples	476.2	[17]
	Activated carbon	Environment water samples	471.1	[18]
	Activated fiber carbon heated by microwave	Aqueous solution	339.0	[19]
	Sistan sand	Wastewater	769.0	This work

4. CONCLUSION

In this research, Sistan sand as an inexpensive and readily available adsorbent was used for the removal of two antibiotics cephalexin and tetracycline from water and wastewater samples. Optimum conditions for adsorption such as pH, amount of adsorbent, ionic strength of the solution, contact time and initial drug concentration for two antibiotics were investigated and optimized. Under optimal conditions, the Langmuir and Freundlich isotherms were demonstrated the best matches for CEP and TC, respectively. Absorption capacity of Sistan sand for removal of CEP and TC were calculated 263 mg g^{-1} (for CEP) and 769 mg g^{-1} (for TC).

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بررسی ایزوترم‌های لانگموئر و فروندلیچ برای حذف همزمان آنتی‌بیوتیک‌های سفالکسین و تتراسایکلین توسط ماسه بادی سیستان از نمونه‌های آب و فاضلاب

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چکیده

در این تحقیق، از ماسه بادی سیستان به عنوان یک جاذب طبیعی و ارزان برای حذف آنتی بیوتیک های سفالکسین و تتراسایکلین موجود در فاضلاب استفاده شد. در حذف آنتی بیوتیک سفالکسین برای غلظت ۶۰/۰ میلی گرم بر لیتر، شرایط بهینه برای جذب سطحی عبارتند از pH محلول ۳/۰، مقدار جاذب برابر یک گرم، زمان تماس ۲۵/۰ دقیقه و مقدار سدیم کلراید برای تنظیم قدرت یونی محلول ۰/۷ گرم بر لیتر. داده ها تطابق خوبی با ایزوترم لانگموئر نشان دادند و ظرفیت جاذب ۰/۲۶ گرم بر گرم محاسبه شد. جاذب توانست حداکثر ۶۸/۱ درصد از سفالکسین را در نمونه فاضلاب حذف کند. برای جذب سطحی آنتی بیوتیک برای غلظت ۸۰/۰ میلی گرم بر لیتر تتراسایکلین، در شرایط بهینه، بهترین pH برابر ۸/۰ بود و مقدار جاذب برابر ۱/۰ گرم و بهترین زمان تماس ۳۵/۰ دقیقه تعیین گردید. مقدار نمک اضافه شده برای تنظیم قدرت یونی محلول، ۰ / ۷ گرم بر لیتر می‌باشد. مطالعات ایزوترم، تطابق خوبی با مدل فرندلیچ نشان دادند و ظرفیت جاذب ۰/۷۶ گرم بر گرم گزارش شد که می‌تواند تا ۷۶/۲ درصد از این آنتی‌بیوتیک را از فاضلاب حذف نماید.

واژه‌های کلیدی

سفالکسین؛ تتراسایکلین؛ ایزوترم لانگموئر؛ ایزوترم فرندلیچ؛ تجزیهٔ فاضلاب.