



Ciprofloxacin Retention in Agricultural Soil

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Abstract In the present work, the adsorption and desorption process of the antibiotic Ciprofloxacin (CIPRO) in an agricultural soil was studied through batch experiments. Freundlich's model satisfactorily described all the experimental data. A high KF value was obtained, $8.52 \text{ L}^{1/n} \text{ mg g}^{-1}$, indicating a high affinity of CIPRO for the soil, with adsorption percentages of 63% in all cases. On the other hand, the desorption was very low, assuming in all cases less than 7% with respect to the amount previously adsorbed. Based on these results, we can affirm that CIPRO presents a low risk of transfer from the soil to other environmental compartments.

Keywords Ciprofloxacin, retention, agricultural soils, antibiotics

Introduction

In recent years, there has been a growing concern about the presence of antibiotic residues in the environment, because they can generate different negative impacts on human and ecological health [1,2]. Ciprofloxacin (CIPRO) is a broad-spectrum antibiotic, belonging to the fluoroquinolone class, which are widely used in both human and animal medicine, and which reach the soil mainly through the application of animal excretion and sewage sludge on agricultural fields [3-5]. Once in the soil, the adsorption-desorption process will condition its transport to other environmental compartments (surface and groundwater and crops), its degradation and its bioavailability towards soil microorganisms [2,6], for what is of special interest its study. Therefore, the main objective of this work is to study the adsorption and desorption behavior of the CIPRO antibiotic in an agricultural soil through batch experiments. The results of this study will contribute to a better understanding of the environmental fate of this pollutant, as well as its possible risks to human and ecological health.



Materials and methods

Antibiotic

CIP (HPLC grade, $\geq 98\%$ purity) was purchased from Sigma-Aldrich. CIPRO is an amphoteric molecule, with two pKa values (5.90 – 8.89), so it can appear as a dipolar ion, cation and/or anion, depending on the pH of the medium.

Soil

To carry out the study, a soil dedicated to the cultivation of vegetables from the town of Santa Ana, Province of Corrientes (SA) was used. The soil is acidic, pH 5.12 (Table 1), there is a predominance of negative charges in the soil. The soil presented a CIC of 3.22. The organic matter content was also 0.90 and 0.63% and N 0.05. The texture of the soil is sandy (91.6% sand, 2.5% clay and 5.9% silt).

Table 1. Main physicochemical properties of the selected soil.

suelo	pH	MO %	CO %	Ω dS/m	N %	P mg/L	K mg/L	Ca meq/100g	Mg meq/100g	CIC meq/100g
SA	5.12	0.90	0.52	0.09	0.05	32.8	0.15	1.92	0.76	3.22

Equilibrium adsorption-desorption experiments

The equilibrium adsorption and desorption process was studied using batch experiments. Batch experiments followed the OECD guidelines for pesticides [7], with a pre-test performed to define the solid: solution ratio (1 g: 30 mL), the equilibrium period (24 h), the stability of compounds and sorption from the tube walls. In all cases, the composite losses were less than 5% (data not shown).

For this, 1 g of soil was put in contact with 30 mL of CIPRO solution containing 0.01 M CaCl₂ as ionic background. Four (4) different initial concentrations (4 – 10 mg L⁻¹) were used. These suspensions were shaken for 24 h, centrifuged, filtered and analyzed by spectrophotometry. Next, for the desorption study, 30 mL of 0.01 M CaCl₂ were added to the soil samples resulting from the adsorption process and they were subjected to the same treatment as that carried out for the adsorption process. The experimental data obtained in the adsorption process were described using the Freundlich and Dubinin- Radusevich model. All experiments, including the controls and blanks, were performed in triplicate.

Sorption models

Adsorption is a very important process because it can provide very valuable information about the mechanism, the properties of the surface and the preferences of each adsorbent for a specific sorbate. Equilibrium data were analyzed using linear (L), Freundlich (F), and Dubinin-Radushkevich (D-R) isotherms

Linear sorption isotherm is expressed as:

$$q_e = K_d \cdot C_e \quad (1)$$

The distribution coefficient, K_d , in a soil/water system is defined as the ratio between the equilibrium concentration of a compound in the sorbent and in solution [8].

$$K_d = \frac{q_e}{C_e} = \frac{(C_i - C_e)}{C_e} \times \frac{V}{m} [L/g] \quad (2)$$

where q_e is the amount of CIPRO adsorbed at equilibrium (mg/g); C_i is the initial concentration of CIPRO (mg/L); C_e is the equilibrium concentration of CIPRO (mg/L); m is the adsorbent (soil) mass (g); and V is the volume of the aqueous solution (L).

The adsorption percentage (adsorption, %) was calculated as:

$$q_d \% = \frac{(C_i - C_e)}{C_i} \times 100 \quad (3)$$

The K_d parameter of the linear model is very important to estimate the adsorption potential of dissolved contaminants in contact with soils as a natural adsorbent. K_d describes the extent and favorability of the association between CIPRO and the soil. High values of K_d indicate a strong association, while low values indicate a weak association. The degree of adsorption of an organic contaminant in the soil determined by K_d is strongly related to the properties of the soil and the contaminant.



The distribution coefficient, K_d , is equal to the slope of the linear sorption isotherm (C_e vs. q_e) [9]. Except the linear isotherm model, two-parameter isotherm models were applied in this study to describe the equilibrium sorption data of CIPRO onto soil: Freundlich [10] and D–R [11].

Freundlich isotherm is the oldest model to describe the sorption process. This model talking about adsorption on heterogeneous surfaces with uniform distribution of energy where the energy adsorption exponentially decreases by the end of the adsorption centre adsorbent [10]. The model can be described as:

$$q_e = K_F \cdot C_e^{1/n} \quad (4)$$

Where: q_e (mg g⁻¹) is the amount of CIPRO adsorbed to the soil; C_e is the concentration of CIPRO in the solution at equilibrium; K_F (L^{1/n} mg^{1-1/n} g⁻¹) is the Freundlich affinity coefficient and $1/n$ (dimensionless) is the Freundlich linearity index. The desorption results were expressed as the amount of CIPRO desorbed for each of the initial concentrations used. The Freundlich constants K_F and $1/n$ were calculated from the intercept and slope of the linear plot of $\log q_e$ versus $\log C_e$.

Dubinin–Radushkevish isotherm is used to estimate the apparent free adsorption energy. D–R model can make the difference between physical and chemical sorption [11]. D–R model can be described as follows:

$$q_e = q_m \cdot \exp(-\beta \cdot \varepsilon^2) \quad (5)$$

where q_e is the equilibrium concentration in the sorbent (mg/ g), q_m is the theoretical capacity of saturation (mg/g), β is constant of the sorption energy (mol² /kJ²) and ε is the Polanyi potential.

$$\varepsilon = RT \cdot \ln \left(1 + \frac{1}{C_e} \right) \quad (6)$$

The slope of the plot of $\ln q_e$ versus ε^2 gives β and the intercept yields the adsorption capacity, q_m . Linear regression analyses were used to determine slopes and intercepts of the linear plots and for statistical analyses of the data. By knowing the value of the constant, β can get information on the amount of free energy E (kJ mol⁻¹) per mole of sorbate and information on the type of sorbent mechanism through the equation [12, 13]:

$$E = \frac{1}{\sqrt{-2\beta}} \quad (7)$$

Validity of sorption isotherm

As linear and nonlinear analyzes are used in this study, different errors have been used to test the validity of the isotherm models: the R^2 regression coefficient of the linear analysis and the nonlinear residual mean square error (RMSE) [14], the error sum of squares (ERRSQ) [12,14], and a composite fractional error function (HYBRID) [12,15] of nonlinear analysis.

The equations are expressed as follows:

$$RMSE = \sqrt{\frac{1}{n-2} \sum_{i=1}^N (q_{e,exp} - q_{e,cal})^2} \quad (8)$$

$$ERRSQ = \sum_{i=1}^N (q_{e,exp} - q_{e,cal})^2 \quad (9)$$

$$HYBRID = \sum_{i=1}^N \left[\frac{(q_{e,exp} - q_{e,cal})^2}{q_{e,exp}} \right] \quad (10)$$

Where $q_{e,exp}$ and $q_{e,cal}$ show the experimental and calculated values and N is the number of observations in the experimental data

Linear regression analyses were used to determine the slope and intercept of the linear plots and for statistical analysis of the data. While for nonlinear analysis of two parameters models (Freundlich and Dubinin– Radushkevich isotherms), GRAPH v 4.4.2. software was use.

Results and Discussion

The calculated values of adsorption coefficients K_d as well as parameters of F and D–R model are shown in Table 2. Each data was given as a mean of three consecutive measurements The relative standard deviation was less than 10%. In most cases, the sorption coefficient obtained by the linear model is slightly higher than that obtained by the F model, while the lowest values were obtained using the D–R model.



Figure 1 shows the adsorption curves of CIPRO in the soil, which is of type L, indicating that the relationship between the amount of antibiotic in solution and the amount adsorbed to the soil decreases as the amount of CIPRO increases added.

This thus gives rise to non-linear and concave curves, suggesting a progressive saturation of the surfaces of the different soil constituents.

This type of curve also indicates that, at low concentrations of the solute, in this case CIPRO, it has a very high affinity for the soil, adsorbing almost all of the compound.

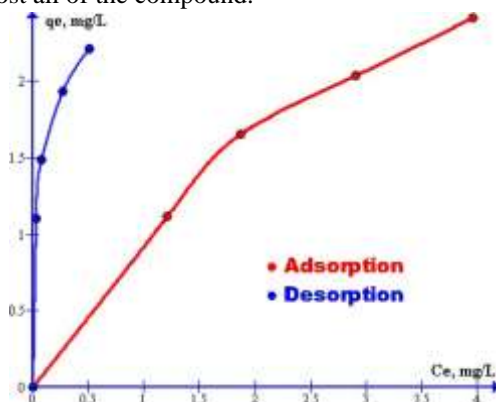


Figure 1: CIPRO adsorption–desorption curve in soil

The adsorption was very high, with adsorption percentages of 69%, which are similar to those obtained by Riaz *et al.* (2019).

The calculated values of the adsorption coefficients K_d , as well as the Freundlich and DR model parameters are shown in Table 2. Each data was given as the mean of three consecutive measurements.

The relative standard deviation was less than 10%. The adsorption coefficient obtained by the linear model is higher than that obtained by the Freundlich model, while the lowest values were obtained using the D–R model. The value of the parameter E gives us information about the type of adsorption processes: the process is characterized by physical nature if $E < 8$ kJ/mol, the process is controlled by ion exchange if the values of E are between 8 and 16 kJ/mol, and the process is chemical in nature if $E > 16$ kJ/mol [13,16]. The estimated value of E was found to be > 16 kJ/mol for all experiments (Table 2), implying chemical adsorption of CIPRO in soil.

Table 2: The distribution coefficient (K_d), Freundlich and Dubinin–Radushkevich sorption isotherm parameters obtained using the linear method ($K_F ((\text{mg/g})(\text{L/mg})^{1/n})$, β (mol^2/kJ^2), q_m (mg/g)), E (kJ/mol)

Isotherm adsorption						
Freundlich						
n	K_F	R^2	RMSE	ERRS	HIBRID	
1.515	0.989	0.9980	0.0299	0.0053	0,0025	
Dubinin–Radushkevich						
$\beta \times 10^{-7}$	q_m	E	R^2	RMSE	ERRS	HIBRID
3.845	2.536	1140.25	0.9552	0.1342	0.1081	0.0566
Isotherm desorption						
Freundlich						
n	K_F	R^2	RMSE	ERRS	HIBRID	
4.177	2.648	0.9903	0.0014	0.0114	0,0077	
Dubinin–Radushkevich						
$\beta \times 10^{-8}$	q_m	E	R^2	RMSE	ERRS	HIBRID
9.500	2.2923	2290.54	0.99905	0.0026	0.0210	0.0105

The parameters and correlation coefficients obtained from the Freundlich ($\log q_e$ vs. $\log C_e$) and D–R ($\ln q_e$ vs. ϵ^2) plots (numbers not shown) are listed in Table 2. The value of the exponent n can give a premise about the favorability of adsorption. The common opinion is that when $n < 1$ adsorption is poor, if n is between 1 and 2, adsorption is moderately difficult, while if n is between 2 and 10, adsorption is quite good [17]. According to the value of n obtained 1.515, the adsorption of CIPRO in the soil can be classified as moderately difficult, also indicating that the number of sites available for the adsorption of CIPRO decreases with the increase of CIPRO added.

The Freundlich affinity coefficient (K_F) was $2.648 \text{ L}^{1/n} \text{ mg}^{1-n} \text{ kg}^{-1}$, a value that is lower than those obtained by other authors, both for CIPRO and for other fluoroquinolones [18,19]. This discrepancy is due to the different experimental conditions used, mainly to the different range of initial antibiotic concentrations.

The K_d value is 753 mL g^{-1} (at 0.01 M CaCl_2) indicating a very high adsorption of CIPRO in the investigated soil that corresponds to the ciprofloxacin adsorption data [20,21] and fluoroquinolones in soil samples in general [18]. The composition of soil determines the degree and strength of adsorption.

Desorption was very low, 3%, in agreement with what was observed by other authors [22]. Therefore, we can affirm that the CIPRO retention process in agricultural soils is practically irreversible (Figure 1). Table 2 shows the corresponding F and DR parameters. The desorption K_F value was higher than that observed for the adsorption process, indicating that the desorption capacity is less favorable.

Adsorption-desorption hysteresis is due to the irreversible adsorption of a fraction of a chemical. It is known that several experimental factors can also contribute to hysteresis [23]. It was shown that the adsorption of CIPRO is irreversible, an adsorption-desorption hysteresis was observed in the soil. In this desorption study, the amount of CIPRO desorbed was observed to be less than the total amount of CIPRO adsorbed (Figure 1), indicating positive hysteresis and suggesting that a significant amount of CIPRO was strongly bound to soil particles and is not easily desorbed.

Mechanism

There are studies that have determined that the pharmaceutical adsorption capacity and their mobility does not depend only on the physicochemical properties of the adsorbent, such as the pH value, the content of organic matter, the cation exchange capacity and the ionic strength, but also on the properties of the adsorbent. physicochemical properties of the pharmaceutical products investigated [20,23-27] as well as the temperature of the processes of adsorption [28].

The clay content in the sediments studied and its impact on adsorption mechanisms are negligible. Furthermore, the adsorption of CIPRO in soil samples is a consequence of an expansion of the montmorillonite spacing due to interlayer adsorption [27]. Some authors explain the adsorption of fluoroquinolones on montmorillonite as a result of cation exchange in such a way that adsorption is not completely limited to the number of available exchangeable adsorbent sites, but to the number of fluoroquinolone cation species [18]. This fact is closely related to the results obtained. In fact, taking into account the physical and chemical properties of the studied soil (Table 1), it is evident that in the soil the main mechanism is by cation exchange. This is justified by the high percentage of ionization at the studied pH.

Ciprofloxacin is an amphoteric molecule that has two ionizable functional groups, the acidic 3-carboxyl group ($pK_{a1} = 6.08$) and the alkaline N-4 of the piperazine substituent ($pK_{a2} = 8.41$). The pH value of the soil is 5.12, and according to this pH the predominant form is the cationic one. The content of each ionic species of the CIPRO can be calculated based on the pH value and the pK_a value [29]. The calculated values are 90.1% cationic and 9.89% neutral. During adsorption in soil, most CIPRO molecules exist as cations at pH 5.12 and can be electrostatically attracted by negatively charged soil particles [18].

Error analysis

The Figure 2 shows the comparisons of the D–R and Freundlich isotherms for CIPRO sorption obtained by the nonlinear regression method at a temperature of $25 \text{ }^\circ\text{C}$. Table 2 shows the sum of squared errors (ERRSQ), residual



root mean squared error (RMSE) and hybrid fractional error function (HYBRID) and were calculated using nonlinear methods. Based on these results, the Freundlich model is more appropriate than the D–R model. The different isotherms obtained were analyzed by non-linear curve fitting with GRAPH v 4.4.2 software.

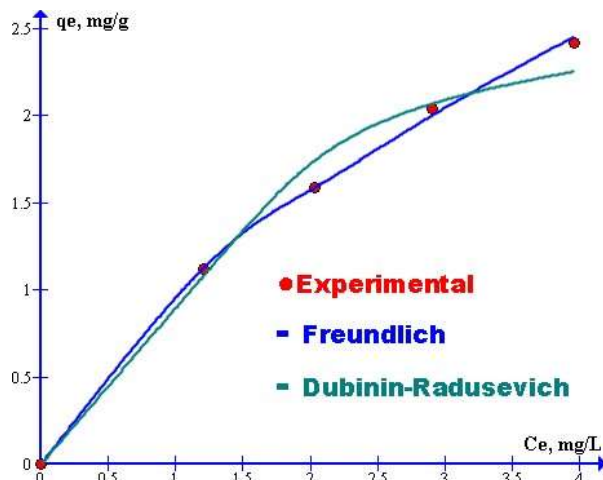


Figure 2: Comparison the D–R and Freundlich isotherms for CIPRO adsorption obtained by non-linear regression method at a temperature of 25 °C

Conclusion

The adsorption of ciprofloxacin in soil depends on the properties of the soil and the adsorption conditions. CIPRO adsorption was found to increase with an increase in the initial CIPRO concentration and the cation exchange capacity of the soil. In addition, the adsorption of CIPRO strongly depends on the pH value of the soil (values not shown).

Ciprofloxacin showed a very high affinity for soils, with adsorption percentages higher than 73% in all cases, and desorption percentages lower than 7%. Based on these results, it is expected that this contaminant does not present a high risk of leaching and runoff into surface and groundwater. At the same time, its bioavailability will also be limited, thus not presenting a significant risk to human health and the environment, compared to other contaminants that have a lower affinity for soils.

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