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Effects of Two Cations on Gastrointestinal Absorption of Ofloxacin

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A study was performed to establish the effect of Al^{3+} and Fe^{2+} cations on the absorption of ofloxacin when it is administered orally at a dose of 200 mg. The study was carried out with nine volunteers, who each received three treatments (A [200 mg of ofloxacin], B [200 mg of ofloxacin plus 11 g of colloidal aluminum phosphate], and C [200 mg of ofloxacin plus 1,050 mg of $FeSO_4$]) according to a Latin square design; the washout period was 1 week. The analytical technique was a microbiological diffusion method. The pharmacokinetic parameters were calculated from the cumulative urinary excretion data and from a sigma-minus plot. The total amount of ofloxacin excreted in urine had a mean value of 163.59 ± 22.13 mg when ofloxacin was administered alone, 152.41 ± 18.76 mg when it was administered with Al^{3+}, and 146.49 ± 14.85 mg when it was administered with Fe^{2+}. No statistically significant differences were found in the *F* values (fractions of dose absorbed) obtained with ofloxacin alone and ofloxacin plus Al^{3+} ($P = 0.341$). When ofloxacin alone was compared with joint administration with Fe^{2+} the value of *F* decreased 10.85%; this difference is statistically significant ($P = 2.623 \times 10^{-2}$).

Ofloxacin is included in the fluoroquinolone group together with ciprofloxacin, norfloxacin, pefloxacin, and lomefloxacin, among other drugs. From the qualitative point of view, these compounds have similar kinetic characteristics, although they exhibit important quantitative differences that affect mainly their absorption and metabolism (4, 8–10, 14).

It has been reported that the combined administration of agents containing Al^{3+}, Mg^{2+}, or Fe^{2+} considerably reduces the degree of absorption of some of the quinolones, such as ciprofloxacin (5, 6, 15), norfloxacin (13, 20), and pefloxacin (22).

The aim of the present work was to establish whether this kind of interaction occurs in the combination of ofloxacin and Al^{3+} or Fe^{2+}, since there are few data regarding this in the literature and the published data are not comparable. Hoffken et al. (7) have shown that a sizable decrease in the absorption of ofloxacin occurs when it is administered simultaneously with antacids; more recently, Flor et al. (4) have shown that a slight decrease in the absorption of ofloxacin occurs when it is administered 2 h after the administration of a dose of antacid.

MATERIALS AND METHODS

This study was carried out at two levels: in in vitro and in vivo experiments.

In vitro assay. To study the passage of ofloxacin across intestinal membranes, we employed the method described by Wilson and Wiserman (25) with slight modifications. In particular, we injected unreversed intestinal sacs, 5 cm in length, from Wistar rats with 0.5 ml of a solution containing ofloxacin (1 mg/ml) in the presence and in the absence of the Al^{3+} cation. The intestinal sac, always made with the fragment of gut corresponding to the first partition after the choledoch, was placed into a glass Erlenmeyer flask containing 25 ml of Krebs-Ringer bicarbonate solution (21) maintained at 37°C.

Each day, the experiment was carried out with two fragments of intestine under identical experimental conditions; one fragment was used to study the passage of ofloxacin in the absence of Al^{3+}, and the other was used to study the passage of the drug in the presence of the cation. Samples were collected from the interior and exterior of the intestinal sacs at previously scheduled times (10, 20, 30, and 45 min), and the value of the in vitro absorption constant (K_i), together with the total amount absorbed after 45 min (Q_i), was calculated. The experiment was repeated with 10 intestinal fragments for each situation (absence or presence of Al^{3+}). The source of Al^{3+} was $KAl(SO_4)_2 \cdot 12H_2O$ (Fluka S.A.), and the concentration of Al^{3+} was 0.43 mg/ml, for a molar concentration ratio of ofloxacin to Al^{3+} of 3:1.

In vivo assay. The in vivo assay was carried out with nine healthy volunteers of both sexes, with ages between 22 and 32 years and a mean weight (± standard deviation) of 61 ± 12 kg. None of the volunteers had received any medication for at least 1 week prior to the experiments, and all gave consent to participate after being informed of the purposes and nature of the study.

All volunteers received treatment A, which consisted of the oral administration of 200 mg of ofloxacin; treatment B, which comprised the same dose of the quinolone plus 11 g of colloidal aluminum phosphate (Fosfalumina; Sehering-Plough); and treatment C, which comprised the same dose of ofloxacin plus 1,050 mg of $FeSO_4$ (two tablets of Fero-Gradumet; Abbott). The washout period was 1 week. The subjects had fasted for 12 h with water ad libitum before receiving medication and did not eat until 2 h had elapsed since administration. The study was carried out according to a Latin square design (24). Urine samples were collected over the 33 h after administration of ofloxacin, and the drug concentrations in the urine samples were determined.

Analytical technique. In all cases (samples from the in vitro assay and urine samples), a microbiological plate diffusion method was used (16), with *Bacillus subtilis* as the assay organism for the determination of ofloxacin levels. Standard solutions were prepared with urine (from healthy volunteers taken before ofloxacin administration) for analysis of the urine samples and with Krebs-Ringer bicarbonate solution (21) for the samples from the in vitro part of the study. The

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concentration ranges were 100 to 5 and 10 to 0.5 μg/ml, respectively.

This microbiological technique was studied and contrasted with a high-performance liquid chromatography technique that was developed in our laboratory for the measurement of ofloxacin in biological fluids. Statistical comparison of the methods showed that there were no significant differences between the concentrations determined by the two methods (17).

Kinetic analysis. Linear and nonlinear regression analysis was carried out with the data for ofloxacin levels obtained from the in vitro experiments. These data fit a monoexponential kinetic process defined by the equation $Q_r = Q_o \cdot e^{-K_i t}$, where Q_r is the amount of drug remaining inside the intestinal sac at time t, Q_o is the initial amount of ofloxacin in the intestinal sac, and K_i is the constant of passage of ofloxacin through the membrane. The values of K_i in the presence and absence of Al^{3+} were obtained by linear regression from the relationship between log Q_r and t.

The cumulative urinary excretion curves were constructed by graphing the total amounts of ofloxacin excreted in urine against time. The total amount excreted (U_∞) and the fraction of dose absorbed (F) were calculated. The F value was calculated from the U_∞/D ratio, since ofloxacin is sparingly metabolized and almost completely eliminated in an unaltered form through the kidneys (4). By using the sigma-minus plot (23), the elimination rate constant was calculated.

Statistical analysis. After the normal distribution of the data was checked by the Shapiro-Wilks test (19), a two-way analysis of variance was performed for statistical comparison of the results. To calculate the statistical power of the assay, the equation $Z_\beta = (\Delta/\sigma)\sqrt{n} - Z\alpha$ was used (1), where Δ is the difference considered to be significant ($\Delta = 20\%$), σ is the standard deviation of the parameters compared, α is the level of significance ($\alpha = 0.05$), and n is number of individuals ($n = 9$).

RESULTS

The passage of ofloxacin through the membrane of the intestinal sacs occurs through a first-order process whose constant has a mean value (± standard deviation) of (3.22 ± 1.19) × 10^{-3} and (3.74 ± 1.66) × 10^{-3} min^{-1} in the absence and presence of Al^{3+}, respectively (Fig. 1). The total amount absorbed at 45 min after the beginning of the in vitro experiments was 124.57 ± 58.1 μg when the solution contained ofloxacin alone and 111.85 ± 57.79 μg when it included Al^{3+}. There were no significant differences between the values of K_i obtained under the experimental conditions employed ($P > 0.4$), nor were there any significant differences between the values of Q_i ($P > 0.5$).

The results obtained in the experiments involving the healthy volunteers are shown in Fig. 2 and Table 1. Figure 2 shows the mean curves of urinary excretion of ofloxacin for treatments A, B, and C, and Table 1 shows the mean pharmacokinetic parameters obtained with these treatments.

The total amount of ofloxacin excreted in urine (U_∞) had a mean value (± standard deviation) of 163.59 ± 22.13 mg when administered alone, 152.41 ± 18.76 mg when accompanied by Al^{3+}, and 146.49 ± 14.85 mg when administered with Fe^{2+}, representing fractions of administered dose (F) of 81.78% ± 11.06%, 76.17% ± 9.35%, and 72.91% ± 7.44%, respectively. No statistically significant differences between the first two values of F were found ($P = 0.341$); the value of F corresponding to the administration of ofloxacin together with $FeSO_4$ was reduced by 10.85%. The statistical power of the assay, calculated as described in Materials and Methods, was about 90%.

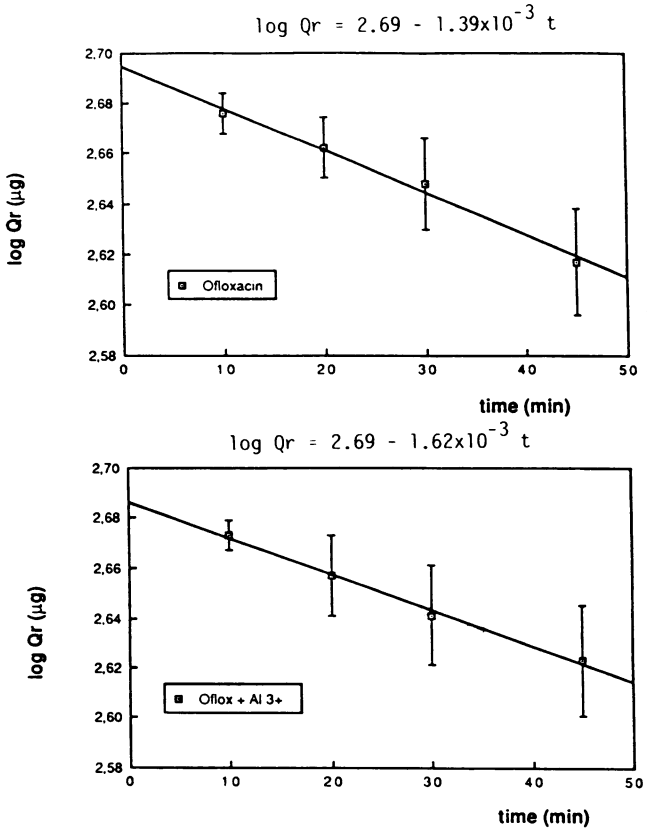

FIG. 1. Linear relationship established between log Q_r and time (t). Oflox, ofloxacin. Error bars show standard deviations.

DISCUSSION

As mentioned above, there is a large body of information in the literature about the effect that certain divalent or trivalent cations may exert on the absorption of some quinolones, such as ciprofloxacin, norfloxacin, or pefloxacin; however, very little information, most of it nonhomogeneous, on the interaction of this kind of quinolone with cations has been published.

This was the background that prompted us to carry out the present study, which involved both in vitro and in vivo experiments. The results obtained point to some differences in the kinetic behavior of ofloxacin and those of other quinolones when the drugs are administered jointly with a nonsystemic antacid agent containing Al^{3+} or a ferrous salt.

The in vitro assay shows that the presence of Al^{3+} does not modify the capacity for passage of ofloxacin across the intestinal membranes; this suggests that no modifications in the bioavailability of this quinolone would occur if it were administered jointly with antacid agents containing the cation. Indeed, the results of the experiments carried out with healthy volunteers confirm the hypothesis postulated from the results of the in vitro assay. No statistically significant differences between the mean pharmacokinetic parameters of ofloxacin administered jointly with the antacid agent used and ofloxacin administered alone ($P = 0.341$) were found. Moreover, comparison of the three treatments, A, B, and C,

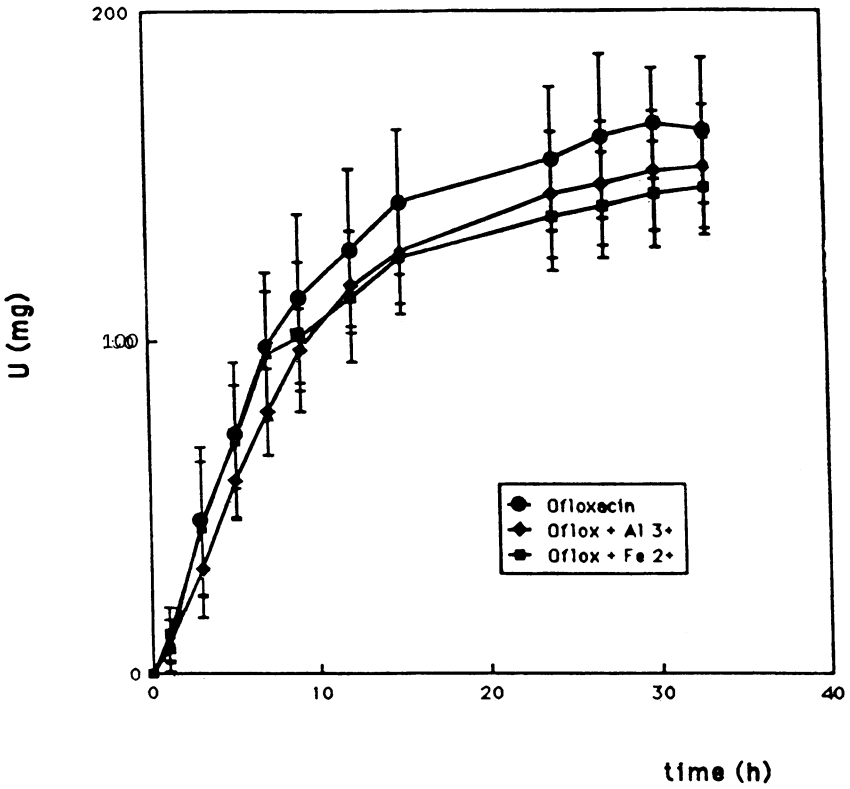

FIG. 2. Mean curves of urinary excretion of ofloxacin (Oflox). Error bars show standard deviations.

together reveals no statistical differences in the values of F or U_∞ ($P = 0.138$ and 0.153, respectively), which means that these differences are not relevant. When ofloxacin is administered with Fe^{2+}, the amount absorbed decreases slightly and the value of U_∞ is reduced by 10.85%. Statistical comparison of F values for treatments A and C reveals significant differences ($P = 2.623 \times 10^{-2}$); nevertheless, the magnitude of the reduction in the percentage of absorption is not very important from the clinical point of view.

These results are conclusive with regard to this hypothesis and clearly show that the oral absorption of ofloxacin is not modified as a result of the simultaneous administration of 11 g of colloidal aluminum phosphate. With 1,050 mg of ferrous sulfate, absorption decreases slightly.

These results should be interpreted with caution; one cannot affirm that ofloxacin does not interact with antacids in any conditions of administration, but it is clear that its absorption is not modified by the administration of a single dose of antacid containing Al^{3+}. In any case, it is clear that its behavior differs significantly from those of other quinolones such as ciprofloxacin, whose absorption is seriously decreased by the coadministration of Al^{3+}, Mg^{2+}, or Fe^{2+}.

TABLE 1. Mean pharmacokinetic parameters of ofloxacin (± standard deviation)

Treatment	F(%)	U_∞(mg)	k_{el} (h^{-1})[a]	$t_{1/2}$ (h)[b]
Ofloxacin	81.78 ± 11.06	163.59 ± 22.13	0.15 ± 0.03	4.76 ± 0.99
Ofloxacin plus Al^{3+}	76.17 ± 9.35	152.4 ± 18.76	0.14 ± 0.02	4.97 ± 0.81
Ofloxacin plus Fe^{2+}	72.91 ± 7.44	146.49 ± 14.85	0.13 ± 0.02	5.57 ± 1.03

[a] k_{el}, elimination rate constant.
[b] $t_{1/2}$, half-life.

Differences among the few published reports about ofloxacin interaction can be found; Hoffken et al. (7) affirm that ofloxacin oral absorption decreases by 69% when the drug is administered in combination with an antacid containing Al^{3+} and Mg^{2+} (10 oral doses), whereas Flor et al. (4) affirm that ofloxacin oral absorption decreases slightly (about 20%) when the drug is administered 2 h after a dose of an antacid containing Al^{3+} and Mg^{2+}. These results are very similar to ours, considering that Flor et al. found high standard deviations in the relative bioavailability and that they used the area under the concentration-time curve from 0 to 12 h (AUC_{0-12}) instead of the $AUC_{0-\infty}$ to calculate the bioavailability. Taking into account that a delay in the absorption of ofloxacin is apparent from their results (time to maximum concentration of drug in serum increases to 2 h), the use of the AUC_{0-12} instead of the $AUC_{0-\infty}$ could be the reason for the differences between their and our results. Another reason could be that the antacid they administered contained Al^{3+} and Mg^{2+} and the one we administered contained only Al^{3+}.

Although in some publications (2, 18) the formation of nonabsorbable chelates has been suggested as the possible mechanism responsible for the decrease in the degree of absorption of some quinolones in the presence of divalent or trivalent cations, this mechanism remains to be elucidated. The structural differences between ofloxacin and the rest of the quinolones could account for the different behavior of the former in the presence of Al^{3+}. However, it would be of great interest to carry out studies designed to establish the

true mechanism of the interaction and to explain the differences found for ofloxacin.

The clinical importance of any interaction that modifies the absorption processes of drugs is evident, since a decrease or blockage in absorption prevents the access of the drugs to target sites at the desired concentrations. In the case of antibiotics and chemotherapeutic agents, the risk this involves is double. On one hand, it could lead to therapeutic failure because concentrations could become subtherapeutic. On the other hand, because one is dealing with an antibacterial agent, this kind of situation may favor the selection of resistant strains.

Accordingly, it is essential to consider the interaction between the quinolone agents and antacid agents containing cations when setting up therapy with these drugs. Taking into account the fact that the antibacterial spectra of the quinolones currently used in clinical practice are similar, a possible alternative would be to use ofloxacin when the patient is simultaneously receiving a nonsystemic antacid agent or ferrous salts. If the etiology of the infective process indicates that another quinolone with which interaction occurs should be used, a dose regimen in which at least 2 h elapse between the administration of the quinolone and the administration of the antacid agent must be followed, as suggested by other authors (11, 12).

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