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Structure-Absorption Relationships of a Series of 6-Fluoroquinolones

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The physicochemical constants and some structural parameters (topological, steric, and electronic) of eight third-generation monofluorate quinolones (six uncommercialized and two used clinically [ciprofloxacin and enrofloxacin]) were determined: pK_a, intrinsic solubility (S_0), chromatographic capacity factor, partition coefficient (P), valency molecular connectivity, molecular volume, molecular surface area, dipolar moment, and charges associated with each atom of the molecule. The apparent intestinal absorption rate constants (K_{abs}) in rat (in vivo perfusion) and the MICs at which 90% of the isolates are inhibited (MIC_{90}s) against 100 *Escherichia coli* strains were also determined. We sought to establish simple nonlinear and multiple linear correlations between K_{abs}, on the one hand, and lipophilic parameters and other physicochemical and structural parameters estimated. Of the nonlinear functions examined, the hyperbolic had the best correlation between K_{abs} and P, which was in accordance with the Wagner-Sedman (J. G. Wagner and A. J. Sedman, J. Pharmacokinet. Biopharm. 1:23–50, 1973) equation, whereas, after application of the stepwise multiple linear regression method, a multiple linear correlation with some predictive value could be established only between K_{abs} as a dependent variable and log P and log S_0 as independent variables. In conclusion, the K_{abs} and MIC_{90} of the quinolone CNV 8902 suggest that it is a sufficiently interesting compound to warrant the investigation of its potential therapeutic use orally.

The 1962 synthesis of nalidixic acid by Lesher and Froelich (10) started a new line of research into chemotherapeutic agents: quinolones. Today quinolones are a widely used family of antibacterial agents in clinical practice. At first, they were indicated only for treatment of infections of the urinary tract, but later their major contribution to the treatment of several systemic pathologies was confirmed, arousing great interest in the field of chemotherapy.

This growing interest was reflected in three different chronological stages, clearly defined by the advances in developing new molecules: first-generation quinolones, which include oxolinic acid, piromidic acid, piperamic acid, cinoxacin, rosoxacin, and the ones characterized by the 4-quinolone nucleus; second-generation quinolones, which became available in 1975, when Gerster (7) synthesized flumequine, and whose components are characterized by the introduction of fluor into position C-6 on the molecule; and third-generation quinolones, whose development started with synthesis, by Koga et al. in 1980 (8), of a compound with a fluor atom in position C-6 and norfloxacin, with a substitution for piperazine, at C-7, and which included compounds such as ciprofloxacin, ofloxacin, pefloxacin, and enoxacin. These structural modifications brought about a broadening of the antibacterial spectrum in that the resultant compounds are active even against *Staphylococcus aureus* and *Pseudomonas aeruginosa* and possess superior pharmacokinetic characteristics, such as good bioavailability when taken orally, greater tissue penetration, and a long half-life. Since then, a large number of new molecules, many of them polifluorate derivatives, have been synthesized.

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In daily clinical practice, administration of fluoroquinolones is mainly oral. Therefore, once the antimicrobial activity of new substances has been verified, it is a logical next step to study the process of their absorption into the gastrointestinal tract and to try to establish correlations between their rate constants and some physicochemical parameters.

The aims of this study were, first, to determine the following physicochemical constants for eight second-generation monofluorate quinolones: pK_a, intrinsic solubility (S_0), chromatographic capacity factor (k'), and partition coefficient (P). Then, we wanted to calculate some structural parameters, topological parameters (valency molecular connectivity, dipolar moment, and molecular volume), and electronic ones (charges connected with each atom of the molecule), to determine the apparent intestinal absorption rate constants (K_{abs}) by using in vivo perfusion with rats; and to try to establish simple nonlinear correlations or, alternatively, multiple linear correlations between lipophilic parameters or other physicochemical and structural parameters and K_{abs}.

MATERIALS AND METHODS

Chemicals and reagents. Eight fluoroquinolones were examined in this study: two in clinical use, ciprofloxacin and enrofloxacin, supplied by Chemo Ibérica, and six uncommercialized ones provided by Cenavisa S.A. Laboratories (Reus, Spain) (3). Four of the compounds have the piperazine heterocycle at position 7 and constitute a homologous subseries with a methylene group at position 4 of this ring. The remaining compounds are the homologous morpholine, thiomorpholine, and 4-hydroxypiperidine, all in relation to the piperazine group at position 7 of the common nucleus. Chemical structures are shown in Fig. 1.

Disodium hydrogen phosphate, potassium dihydrogen phosphate, potassium dichromate, sodium tetraborate, sodium chloride, sodium hydroxide, 85% phosphoric acid, sodium heptansulfonate, and high-performance liquid chromatography (HPLC) grade methanol and acetonitrile were purchased from E. Merck (Darmstadt, Germany). Ethylurethane was provided by Fluka Chemie A. G. (Buchs, Switzerland).

One hundred strains of *Escherichia coli* (American Type Culture Collection)

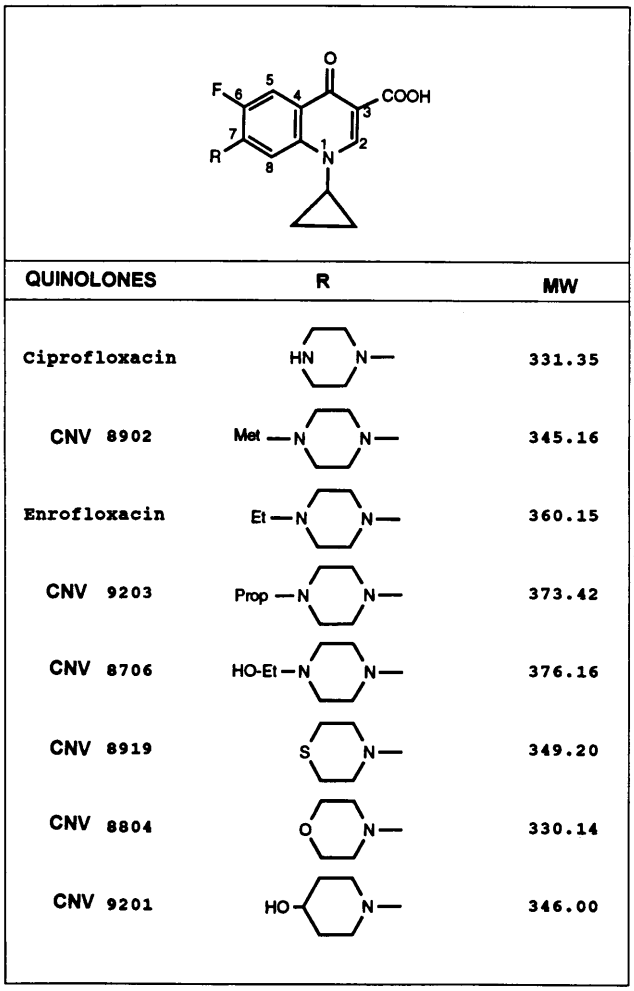

FIG. 1. Chemical structures of compounds studied. MW, molecular weight.

were supplied by the Department de Microbiologia at the Facultat de Medicina, Universitat Autònoma de Barcelona, Barcelona Spain.

Animals. Fasted albino male rats (Sprague-Dawley), weighing between 180 and 210 g, were used in the in situ study to calculate the intestinal absorption rate constant. Animals were supplied by the Facultat de Farmàcia, Universitat de Barcelona (Barcelona, Spain). A minimum of five animals per compound were used.

HPLC. For calculation of physicochemical parameters (pK_a, S_0, P, and k') and K_{abs} HPLC was used, with UV detection at 280 nm, and validated within and between days. The chromatograph (HPLC system 400; Kontron Ins., Zurich, Switzerland) was equipped with two pumps (model 420), a variable 432 UV detector set at 280 nm, a 460 autosampler, and a 450 data system. The analyses were carried out at room temperature with a Lichrocart column (125 by 4.5 mm). The mobile phase, consisting of methanol–acetonitrile–10 mM phosphate buffer with 0.0028% sodium heptansulfonate (pH 2.5), was pumped at a flow rate of 1 ml/min and a 20-μl sample injection volume. The organic and aqueous proportions of the mobile phase for each quinolone analyzed are shown in Table 1.

pK_a and S_0. The pK_a values of the eight fluoroquinolones were determined at 37°C by the solubility method. For this, nine buffered solutions with different pH values (covering 0.5 U between values) were prepared. The pH values were 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5, and 9.0. All the solutions were prepared with a constant ionic strength 0.15 (16). Solutions of KH_2PO_4 and Na_2HPO_4 at 0.066 M were used for pH between 5 and 7.5, and solutions of $Na_2B_4O_7$ at 0.025 M were used for pH between 8 and 9. If necessary, the ionic strength was adjusted to 0.15 with NaCl. The fluoroquinolones were added in excess to the appropriate buffer and stirred for 24 h in a shaking water bath at 37°C. The pHs of the suspensions were measured at 24 h and adjusted if necessary. If the pH was adjusted, the sample was agitated for an additional 24 h. The solutions were then filtered (pore size, 0.45 μm), diluted appropriately, and analyzed by HPLC. All experiments were conducted in sextuplicate.

In the case of the fluoroquinolones with a single value of pK_a—CNV 8919, CNV 8804, and CNV 9201—this value was calculated by simple nonlinear regression, fitting the following function to the experimental results:

$$S = S_0 \cdot 10^{(pH - pK_a)} + S_0$$

where S is the solubility at the pH value of the assay. In the case of fluoroquinolones with two values of pK_a, whose solubility, as they are amphoteric substances, corresponds to that of the two substances ionized (AH_2^+ and A^-) and to the amphoteric substance (AH), the calculation of the two values of pK_a was performed by using a program proposed by Asuero (1), which also provided the S_0 value and the corresponding standard deviations (SD) for the three parameters.

***P* and k′.** To determine P, *n*-octanol and phosphate buffer solution (pH 7.0) phases were presaturated with each other for at least 24 h before the experiment. Solutions of each quinolone in phosphate buffer were prepared and agitated for 24 h with a variable volume of the organic phase depending on the compound assayed. After centrifugation at 2,000 × *g* for 15 min, the concentration in buffer phase was analyzed by HPLC; the concentration in octanol was calculated from the difference between the initial and final concentrations in the buffer phase. Six replicates were used for the calculation of P.

k' was obtained by reversed-phase HPLC. It is defined as $(t_r - t_0)/t_0$, where t_0 is the retention time of the control substance which theoretically is not retained in the column and t_r is the retention time of the substance analyzed. In order to obtain the value of t_0, a 0.3 mM solution of potassium dichromate was used as a control.

Calculation of structural and electronic parameters. (i) Topological parameters. The molecular connectivity index of order i was calculated with the INDICES program (4), and the molecular connectivity index of valency was calculated with the CONIND program (4), both running on a PC 486.

(ii) Steric parameters. Molecular surface area and volume were calculated with a PC version of the MOLSV program (14), adapted by K. J. Tupper at the University of Indiana from the program developed by Graham H. Smith. The program was run on a PC 486. The Cartesian coordinates introduced in the program corresponded to structures with the most effective possible geometry, based on the data obtained from calculations with the AM1 semiempirical method.

(iii) Electronic parameters. The net charge associated with each atom of the molecule, the dipolar moment, and the energies of the HOMO and LUMO molecular orbitals were calculated by the AM1 semiempirical method (5), once the molecular geometry had been made as effective as possible. These calculations were performed on an IBM 3090-170 in the Area de Fisocoquímica at the Facultat de Química de Tarragona (Tarragona, Spain).

Rat in situ intestinal absorption. Intestinal absorption studies were carried out by using a recirculation system shown in Fig. 2. The system consisted of a reservoir containing 40 ml of a fluoroquinolone solution (pH 7.0), to be perfused while being stirred, and a peristaltic pump (Minipuls 3; Gilson, Middleton, Wis.) producing a flow of 1 ml/min.

The rats were anesthetized with 1 g of ethylurethane/kg of body weight intraperitoneally. Only two small incisions were made to expose the beginning of the small intestine and the cecum. In order to prevent enterohepatic recycling, the bile duct was ligated. Glass cannulae were inserted at both ends of the intestine. The small intestine was washed gently with an isotonic phosphate buffer solution (pH 7.0). Then, the 40 ml of the perfusion solution was recirculated for a total of 2 h. Samples of 0.5 ml were taken from the reservoir every 15 min. Variations in volume of the perfusion solution and in pH were checked in turn. As a perfusion solution, an isotonic phosphate buffer solution (pH 7.0), at 0.04, 0.15, and 0.4 mM concentrations for each quinolone, was used. After the compound was dissolved, the pH of the solution was tested and adjusted to 7.0 if necessary. K_{abs} was calculated from the drug remaining in the reservoir of the recirculation system, with absorption taken to be a first-order process; that is to say, K_{abs} was calculated by linear least-squares regression analysis of the natural logarithms of the remaining fluoroquinolone concentrations (C) versus time (t): $\ln C = -K_{abs} \cdot t + \ln C_0$, where C_0 is the initial concentration of each quinolone. The data used to calculate the regression line are concentrations estimated between 15 and 120 min.

TABLE 1. Organic and aqueous proportions of the mobile phase for HPLC

Compound	O/A[a]
Ciprofloxacin	40:60
CNV 8902	40:60
Enrofloxacin	40:60
CNV 9203	40:60
CNV 8706	40:60
CNV 8919	65:35
CNV 8804	50:50
CNV 9201	40:60

[a] O/A, organic/aqueous.

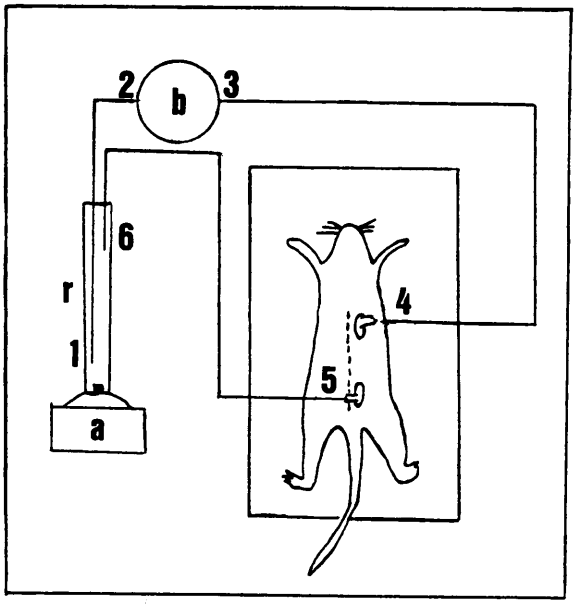

FIG. 2. Schematic diagram of the perfusion method for the rat duodenal segment. a, magnetic stirrer; r, reservoir; b, peristaltic pump. Lengths of numbered segments: 1 plus 2, 60 cm; 3 plus 4, 33 cm; 5 plus 6, 30 cm.

Assay of antibacterial activity (MIC_{90}). The minimal concentration that inhibits 90% of the bacterial strains analyzed (MIC_{90}) was used as a descriptive parameter of antibacterial activity. One hundred strains of *E. coli* were used to determine MIC_{90}s (6). Antibacterial activity was determined by serial dilutions in agar, according to the specifications of the National Committee for Clinical Laboratory Standards (12). The inoculum was applied, with a Steers replicator, to Müller-Hinton agar plates containing serial quantities of quinolone (17). The plates were incubated at 37°C for 20 h and inspected immediately.

Statistical analysis. To determine the linearity of the analytical method used, analysis of variance (ANOVA) was used to compare the mean response/concentration ratios. Five straight lines with six points each were used for each fluoroquinolone.

To study the potential linearity of the absorption process of some of the fluoroquinolones assayed, the K_{abs} values of ciprofloxacin at three concentrations (0.04, 0.15 and 0.4 mM) were subjected to a one-way ANOVA, and those of enrofloxacin and CNV 8902 at concentrations of 0.04 and 0.4 mM were analyzed by Student's t test. The significance level adopted was 0.05.

The K_{abs} values for the eight fluoroquinolones at 0.04 mM were compared by using the Kruskal-Wallis Z test. Multiple comparisons were performed by means of the Bonferroni test at $\alpha = 0.05$.

Fitting of models to data. In order to establish the possible correlations between K_{abs} as a dependent variable and 49 theoretical parameters used as potential independent variables, the simple nonlinear and multiple linear correlations were fitted to the data by least-squares regression.

For the simple nonlinear least-squares fit the program MULTIABS, based on the MULTI program of Yamaoka et al. (21), was used, and for the multiple linear fit the SPSS statistical package (11) was used. In the series of fits, the dependent variable (y) was K_{abs} or log K_{abs} and the independent variables (x) was composed of two sets of data: the physicochemical and the theoretical (topological, steric, and electronic) parameters. The fit functions were as follows. The *hyperbolic* function was $y = K_m \cdot x^n/(Q + x^n)$ where K_m is the constant for the maximum rate of transference across the lipophilic membrane and n and Q are constants for a determined system of partition. The *double linear logarithmic* function was $\log y = a + b \cdot \log x$, where a is the intercept and b is the slope. The *Multiple linear* function was $y(\log y) = a + bx_1 + cx_2 + dx_3$, where x_1, x_2, and x_3 are independent variables, a is the intercept, and b, c, and d are the coefficients of the parameters.

The correlations for all the eight fluoroquinolones studied, as well as for the four elements of the homologous subseries, were determined.

RESULTS

The analytical method was linear in all cases within the range of the concentrations under study and was sufficiently precise (coefficients of variation, between 0.75 and 9.35%) and accurate (relative errors, between −7.39 and +8.68%). The limit of detection ranged between 8 and 10 ng/ml.

TABLE 2. pK_a

Compound	Mean ± SD	
	pK_{a1}	pK_{a2}
Ciprofloxacin	6.20 ± 0.10	8.59 ± 0.10
CNV 8902	5.83 ± 0.05	7.57 ± 0.05
Enrofloxacin	6.26 ± 0.03	7.81 ± 0.03
CNV 9203	5.99 ± 0.13	8.46 ± 0.13
CNV 8706	6.02 ± 0.01	7.27 ± 0.01
CNV 8919	6.37 ± 0.17	
CNV 8804	6.37 ± 0.06	
CNV 9201	7.72 ± 0.22	

pK_a and S_0 means with SD are shown in Tables 2 and 3, respectively. Table 4 lists k' and P means ± SD, Table 5 lists K_{abs} means ± SD, and Table 6 lists the MIC_{90}s, used as a descriptive parameter of antibacterial activity.

Among the simple nonlinear functions assayed, the best correlation was found with the hyperbolic one, in agreement with the Wagner-Sedman equation (20). After the application of the simplex method for K_{abs}) and P and the subsequent computer application of the Marquardt algorithm, the following values were obtained: $K_m = 1.6482 \pm 4.5952$, $n = 0.5895 \pm 0.700$, and $Q = 5.6020 \pm 17.4733$. As can be observed, the parameters are not very reliable and lead to errors in predicted K_{abs} values between −0.32 and +18.52%.

A double logarithmic simple linear correlation was also obtained between K_{abs} and P, but the correlation coefficient obtained ($r = 0.886$) reflected the low predictive value of the fit. Considering only the four components of the homologous subseries, a relatively acceptable fit was obtained ($r = 0.993$), although it still contained errors between the experimental values of K_{abs} and the predicted values, which ranged from −21 to +10%.

After the exhaustive application of the stepwise multiple linear regression method, only one multiple linear correlation with predictive value ($r = 0.952$) could be established, between K_{abs} as a dependent variable and log P and log S_0 as independent variables: $K_{abs} = 0.2173 + 0.2618 \cdot \log P - 0.1339 \cdot \log S_0$ (standard error of the calculation, = 0.1073; fit coefficient, = 0.8702, $r^2 = 0.907$, $r = 0.952$).

DISCUSSION

The pK_a values calculated for fluoroquinolones with amphipathic structure (ciprofloxacin, enrofloxacin, CNV 8902, CNV 9203, and CNV 8706) are on the order of 6 to 7 for pK_{a1} and between 7 and 8 for pK_{a2}. These data agree with the literature (15, 16); values for CNV 8919, CNV 8804, and CNV 9201, with

TABLE 3. S_0 at 37°C

Compound	Mean S_0 ± SD (mg/ml)
Ciprofloxacin	0.1320 ± 0.0050
CNV 8902	0.1510 ± 0.0080
Enrofloxacin	0.4160 ± 0.0200
CNV 9203	0.3680 ± 0.0170
CNV 8706	1.3590 ± 0.2230
CNV 8919	0.0022 ± 0.0006
CNV 8804	0.0079 ± 0.0008
CNV 9201	0.1680 ± 0.0419

TABLE 4. k' and P in octanol–pH 7.0 buffered solution

Compound	k'	Mean $P \pm$ SD
Ciprofloxacin	0.080	0.035 ± 0.0005
CNV 8902	0.096	2.602 ± 0.172
Enrofloxacin	0.112	2.370 ± 0.421
CNV 9203	0.136	9.592 ± 0.250
CNV 8706	0.072	0.470 ± 0.045
CNV 8919	3.016	16.170 ± 2.710
CNV 8804	1.056	1.595 ± 0.062
CNV 9201	3.288	1.642 ± 0.082

TABLE 6. In vitro antibacterial activities

Compound	MIC_{90} (μg/ml)
Ciprofloxacin	0.025
CNV 8902	0.025
Enrofloxacin	0.050
CNV 9203	0.100
CNV 8706	0.200
CNV 8919	0.400
CNV 8804	0.200
CNV 9201	0.800

only one pK_a value, are between 6 and 7. Given the similarity of the pK_a values of the eight fluoroquinolones, it is to be expected that the potential changes in pH which could occur at the absorption sites will almost all behave similarly in terms of their ionization.

Of the eight fluoroquinolones, the one with greatest S_0 is CNV 8706 (1.359 ± 0.223 mg/ml), whereas the one with the least S_0 (0.0022 ± 0.0006 mg/ml) is CNV 8919.

The values of k' range between 0.08 (ciprofloxacin) and 3.28 (CNV 9201). The range of values of the lipophilic parameter (P) for the eight fluoroquinolones assayed spans some three orders of magnitude: CNV 8919's is the highest (16.17 ± 2.71), and ciprofloxacin's is the lowest (0.035 ± 0.0005).

K_{abs} values displayed no important variations in the volumes of the solutions perfused throughout the experiment, which were always below 6%; the variations between the pH at the end of the experiment and that at the beginning (pH 7.0) were irrelevant for all the cases, below 0.6 U. As a result, no correction for volume or pH was made in the K_{abs} calculations.

TABLE 5. K_{abs} in situ in the rat

Compound (mM)	Mean $K_{abs} \pm$ SD (h^{-1})	No. of replicates
Ciprofloxacin		
0.04	0.035 ± 0.035	6
0.15	0.041 ± 0.020	5
0.40	0.045 ± 0.020	5
CNV 8902		
0.04	0.332 ± 0.085[a]	5
0.40	0.350 ± 0.141	5
Enrofloxacin		
0.04	0.440 ± 0.040[a]	7
0.40	0.500 ± 0.100	7
CNV 9203 (0.04)	0.668 ± 0.013[a,b,c]	5
CNV 8706 (0.04)	0.032 ± 0.022[b,c,d]	6
CNV 8919 (0.04)	0.885 ± 0.223[a,b,c,d,e]	5
CNV 8804 (0.04)	0.556 ± 0.101[a,b,e,f]	6
CNV 9201 (0.04)	0.277 ± 0.052[a,d,e,f,g]	5

[a] Different from the value for 0.04 mM ciprofloxacin (the Kruskal-Wallis Z test with Bonferroni's correction [α = 0.05] was used for all comparisons).
[b] Different from the value for 0.04 mM CNV 8902.
[c] Different from the value for 0.04 mM enrofloxacin.
[d] Different from the value for 0.04 mM CNV 9203.
[e] Different from the value for 0.04 mM CNV 8706.
[f] Different from the value for 0.04 mM CNV 8919.
[g] Different from the value for 0.04 mM CNV 8804.

In parallel, a study about the potential linearity of the absorption process for some of the fluoroquinolones assayed was carried out. Three concentrations of ciprofloxacin, 0.04, 0.15, and 0.4 mM, subjected to a one-way ANOVA, showed the absence of statistically significant differences ($P = 0.842$) among the average K_{abs} values. The study was conducted with enrofloxacin and CNV 8902. It was not possible to do the same with the other five fluoroquinolones, as their solubility in the perfusion liquid was below the minimum required. The same extreme concentrations (0.04 and 0.4 mM) were assayed for enrofloxacin and CNV 8902, but the intermediate concentration wasn't, as it was assumed that it would not diverge appreciably from linearity, as had been the case with ciprofloxacin. Student's t test, for unpaired data, made clear the absence in both cases of statistically significant differences between the K_{abs} values for the two concentrations. In principle, the absorption process for at least these three fluoroquinolones is linear under these experimental conditions, which agrees with the results obtained with other fluoroquinolones (2, 9, 18).

The least value calculated for K_{abs} was for CNV 8706 (0.032 ± 0.022 h^{-1}). However, this value was not significantly different from that found for ciprofloxacin (0.035 ± 0.035 h^{-1}). The other fluoroquinolones had higher values: CNV 8919 and CNV 8804 had K_{abs} values of 0.885 ± 0.223 h^{-1} and 0.556 ± 0.101 h^{-1}, respectively, which were above the value calculated for enrofloxacin (0.440 ± 0.042 h^{-1}). As to the effect that nitrogenated heterocyclic substitutes at position C-7 of the base molecule could have on absorption, it should be noted that the thiomorpholinic substitution in CNV 8919 may have been responsible for the higher K_{abs} obtained while the piperazinic one in ciprofloxacin may have been responsible for a lower value. The sequential introduction of methylene groups at the position 4 nitrogen atom of the heterocycle of position 7 of the base molecule appeared to be the cause of the increase in K_{abs}, as the highest K_{abs} (0.668 ± 0.013 h^{-1}) was obtained in this homologous subseries for the element displaying the longest aliphatic chain (CNV 9203). The replacement of a hydrogen atom in the aliphatic chain of one of the compounds of the homologous subseries (enrofloxacin) by a hydroxyl group (CNV 8706) caused a significant decrease in K_{abs}, from 0.440 ± 0.040 to 0.032 ± 0.022 h^{-1}.

As Tables 3 to 5 show, the decrease in S_0 seems to favor intestinal absorption, whereas the fluoroquinolone with the highest P has the highest K_{abs} among these heterologous compounds.

After the Kruskal-Wallis Z test was applied, statistically significant differences among the average K_{abs} values of the eight fluoroquinolones emerged; Bonferroni's test allowed the existing differences between pairs of values to be determined. The lowest K_{abs} was for CNV 8706, and the highest was for CNV 8919; the substitution in position 7 probably affected the K_{abs}. CNV 8706 displayed, as a differential structure, a hydroxyl group in the aliphatic chain of the nitrogen in position 4 of the

piperazine substitute, which seemed to have a negative effect on the absorption process. Overall, these results agreed with other data from the literature (19, 22).

With the aim of establishing structure-absorption correlations, at first the simple nonlinear hyperbolic correlation between K_{abs} and P was determined, as this was the most suitable function for understanding the absorption process which also had predictive potential (13). With a solely predictive aim, other parameters were also assayed; these included lipophilic (k'), physicochemical (S_0 and pK_{a1}), and structural (topological, steric, and electronic) parameters. In all these cases the correlation observed was unsatisfactory for prediction, and therefore the results obtained have been omitted here.

Analysis of the homologous subseries, composed of the four fluoroquinolones differentiated by the number of -CH_2- groups, found an acceptable simple hyperbolic correlation between K_{abs} and P, with relatively reliable parameters and an acceptably good fit, especially considering that the first element in the series may behave atypically, as it lacks the common repetitive group (first-term effect). In light of these results and the subsequent statistical treatment, the Wagner-Sedman (20) hyperbolic model seems to explain, to some degree and with the necessary reservations for such a small series, the intestinal absorption process for the four compounds in the subseries. For perfectly homologous compounds and the hydroxypiperazinic derivative (CNV 8706), the fit deteriorates rapidly and lacks any predictive value.

The evaluation of a high number of theoretical parameters (topological, steric, and electronic; see Materials and Methods) undertaken, together with the calculation of other physicochemical parameters, obtained experimentally, aimed to maximize not only the possibility of establishing simple nonlinear correlations but also multiple linear correlations between two or more independent variables and K_{abs} or its logarithm. However, due to the relatively low number of fluoroquinolones, it was not possible to establish statistically standard multiple correlations, with more than two or three independent variables. Bearing in mind the correlation matrix of 51 variables analyzed (49 potential independent variables and 2 potential dependent variables [K_{abs} and its logarithm]), a multiple linear correlation with certain predictive value ($r = 0.952$) could be established only after the application of the stepwise multiple linear regression method, between K_{abs} as a dependent variable and log P and log S_0 as independent variables.

It seems that the heterologous character of the eight fluoroquinolones assayed is probably the reason why simple (hyperbolic or bihyperbolic) nonlinear correlations cannot be established between K_{abs} and either the physicochemical parameters obtained experimentally or the structural (topological, steric, and electronic) parameters. However, it can be concluded that for the eight fluoroquinolones studied here, an increase in the lipophilic parameter aids intestinal absorption, whereas the opposite occurs with S_0, whose increase affects absorption negatively.

Finally, it should be noted that the K_{abs} for CNV 8902 ($0.332 \pm 0.085\ h^{-1}$) was significantly higher than that for ciprofloxacin ($0.035 \pm 0.035\ h^{-1}$) (Table 5), although their intrinsic activities, expressed as MIC_{90}s against 100 *E. coli* strains (0.025 mg/liter), were practically equivalent (Table 6). These intrinsic activities were, in turn, from 2 to 32 times higher than those of the other fluoroquinolones. Therefore, CNV 8902 should be considered a sufficiently interesting compound to warrant the verification of its possible therapeutic value after oral administration.

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