Photochemistry of the Phototoxic Drug Lomefloxacin: Paths Observed in the Presence of Amines or NaOH and from the Methyl Ester

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The photochemistry of the fluoroquinolone drug lomefloxacin has been examined in aqueous solution in the presence of aliphatic amines (0.01 and 0.2 M) as well as 0.01 M NaOH; a fairly efficient decomposition (Φ = 0.2 to 0.4) takes place. Under the above conditions the product distribution is the same and differs from that previously observed in neutral and acidic aqueous solution. Five products have been characterized, all of them resulting from reductive elimination of the fluorine in the 8-position and some alteration of the piperazine side-chain. All of the products are rationalized as resulting from the heterolytic cleavage of the C^8–F bond in triplet lomefloxacin to give the corresponding triplet aryl cation. Due to the multiplicity, the C^8 site has a carbene rather than a localized cation character. Under these conditions — the amino group in the 4′-position is free — the main process is formal hydride transfer from the 3′-position and the minor one attack of the 4′-amino group at the C^6–F bond. In accordance with this rationalization, the methyl ester of lomefloxacin exhibits a closely analogous photochemistry under neutral conditions. The cationic intermediate may be involved in the observed phototoxicity of the drug, via an oxygen-independent path.

Introduction

Probably because of the frequent exposure to sunlight typical of today's lifestyle, photoinduced adverse side-effects caused by drugs that absorb significantly natural or artificial light are a matter of growing concern.[1−3] As a consequence, the photochemical properties of drug molecules are investigated in order to provide a rationale for the observed effects. As an example, antibiotic fluoroquinolones (FQ) are successfully employed in the treatment of a variety of infections, but exert significant phototoxic and skin photosensitizing effects upon exposure to UV-A irradiation, which is efficiently absorbed by these drugs.[4−11] Furthermore, some of these drugs appear to increase the inherent photogenotoxic and photocarcinogenenic potential of UV light.[12−17] FQs therefore deserve a particular attention in this respect, both because they are widely used and because most of the known derivatives are phototoxic to some degree. It is increasingly apparent that there is no single photochemical mechanism behind the toxic effect. Some of these drugs, such as ofloxacin and rufloxacin, are rather photostable ($\Phi_{dec} << 0.1$)[18−24] and their action is possibly due to oxygen sensitization [Equation (1)],[25−33] which is thought to be the most common mechanism involved in phototoxicity.

$$FQ^* + O_2 \rightarrow FQ + {}^1O_2 \text{ or } O_2^{\cdot -} \rightarrow \text{toxicity} \quad (1)$$

On the other hand, many fluoroquinolones are quite photoreactive in water (Φ = 0.1 to 0.5 and above) and, interestingly, undergo heterolytic C−F bond fragmentation,[18,22,33] an unexpected (and little precedented) reaction due to the strength of the bond (ca. 120 kcal/mol). Apparently, the charge delocalization in the heteroaryl cation makes ionic cleavage possible, at least in a protic solvent.[34] The fluoroquinolones of this group, which is larger than the previous one, are modest oxygen sensitizers,[31] clearly due to the short lifetime of the excited states, which makes Equation (1) unimportant, although they are even more phototoxic than the previous group. As an example, lomefloxacin (LOM-H), a widely used drug of this family, is known to be photogenotoxic; indeed, it has been used as a model for determining the photogenotoxic potential of drugs.[12−17,35,36] This molecule is characterized not by oxygen sensitization but by efficient defluorination from the 8-position (Φ = 0.5). Several pieces of evidence suggest that fluoride is liberated in a unimolecular step and that the aryl cation **2** is formed [see Scheme 1 and Equation (2)].[18,37,38]

$$Ar{-}F^* \rightarrow Ar^+ + F^- \quad (2)$$

This point has a bearing on the rationalization of the phototoxicity, which may involve the cation. This would be an example of the rarely invoked, oxygen-independent mechanism shown in Equation (3), where **2** plays the role of the active intermediate X.

$$FQ^* \rightarrow X \rightarrow \text{toxicity} \quad (3)$$

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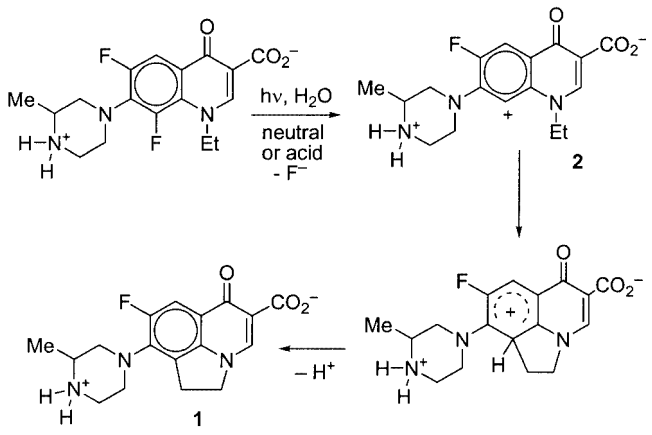
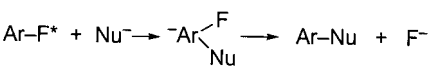


Scheme 1

On the other hand, the role of **2** has a chemical interest, since very little is known about the behavior of aryl cations due to the difficult access to such species.[39] Recent results support a spin-dependent chemistry for these intermediates. In the singlet state, aryl cations are unselective electrophiles, for which addition to the solvent is generally the main process. Triplet aryl cations, on the other hand, have a carbenoid character and either react with π nucleophiles such as alkenes and aromatics (although *not* with n nucleophiles such as alcohols) or abstract hydrogen from H donors (alcohols behave as hydrogen donors, not as nucleophiles).[40] With electron-donor-substituted aryl cations, such as the amino-substituted quinolones we are presently considering, the triplet is the lowest-lying state and is expected to be the reactive state. In the case of LOM-H, defluorination is accompanied by insertion into the neighboring *N*-ethyl group, virtually the only process occurring in aqueous solution, which yields pyrroloquinoxaline **1** (Scheme 1).[22,37] This has been considered a typical example of a "carbenoid" intramolecular reaction, of which there is another example in the (low yield) cyclization of the *o*-propylphenyl cation.[41]

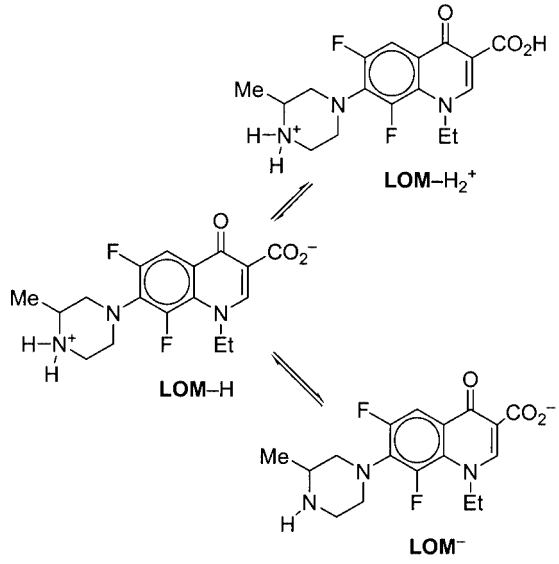
Furthermore, photochemical defluorination of FQs may follow different paths. As mentioned above, unimolecular heterolysis has been proposed for LOM-H, a 6,8-difluoro derivative, but with some 6-monofluoro analogs it has been demonstrated that defluorination occurs via an S_NAr2-type mechanism [Equation (4)], which involves addition of water (NuH) to the excited state to give the corresponding 6-hydroxyquinolone.[22,23]



(4)

The above considerations raise two questions. The first one is whether the use of a stronger nucleophile, such as an amine or OH^-, rather than water, may make a S_NAr2 path operative also with lomefloxacin. The second one, should unimolecular fragmentation occur also in the presence of such nucleophiles, is whether these may interfere with the chemistry of the cation.

Obviously, it must be taken into account that FQs are amino acids and the addition of nucleophiles/bases changes the ionic forms present (Scheme 2).[33,42,43] Photochemical conversion of lomefloxacin into compound **1** occurs with maximal quantum yield under neutral conditions when it is present as the zwitterion LOM-H, and remains the predominant reaction also under acidic conditions (where the cation LOM-H_2^+ is present), although with a lower efficiency.[38] Preliminary experiments have shown that fluoride is also liberated under basic conditions from the anion LOM^-, although the product distribution was not investigated.[38]



Scheme 2

We report here the results obtained in aqueous solutions in the presence of amines and mineral bases. In order to test the role of ionic forms, the study was extended also to the carboxylic ester LOM-Me, since in this case the piperazine nitrogen is non-protonated also under neutral conditions.

Results

Irradiation of lomefloxacin (5×10^{-4} M) in aqueous solution containing 0.01 M diethylamine gave a mixture of five main products, which did not include pyrroloquinolone **1**, by far the main product in the absence of the amine, as shown by HPLC. The same product distribution was obtained in the presence of 0.01 M triethylamine. The irradiated solutions were stirred with 1% ethyl chloroformate in chloroform and the organic layer was treated with ethereal diazomethane to transform the photoproducts into non-ionic derivatives that could be separated by silica-gel chromatography. The structure of the derivatized compounds obtained from such treatment and separation was

determined from their analytic and spectroscopic properties, thus allowing the characterization of the products initially formed.

The main products are two isomers that have lost the fluorine atom at the 8-position and undergone a modification of the piperazine group. The structures of these compounds were assigned as enediamines **3** and **4**, and they were obtained as the *N*(4′)-ethoxycarbonylated methyl esters **3′** (23%) and **4′** (18%; see Table 1 and Scheme 3). The signals of the enediamine system are particularly diagnostic in the ^{1}H and ^{13}C NMR spectra (see Exp. Sect.). Furthermore, a smaller amount of another pair of 8-defluorinated quinolones was obtained, namely products **6** and **7**, in which the piperazine chain has lost two carbons and has been degraded to an aminoethylamino group; they were isolated again as the *N*(4′)-ethoxycarbonylated methyl esters **6′** and **7′** (3% each), which have been previously characterized.[44] Finally, a product resulting from a more deep-seated transformation was obtained. In this case, both fluorine atoms have been lost from the 6- and 8-positions. The NMR spectroscopic data suggested that position 6 had been amino-substituted and, further, that an *N*-methyl and an *N*-formyl group were present. These pieces of evidence, and others presented in the Exp. Sect. (in particular NOE experiments supporting the location of the *N*-methyl group close to CH-8), allowed us to assign the unexpected structure of pyrazinoquinolone **5** (isolated as derivative **5′**, 10%) to this compound.

Table 1. Photoproducts from the irradiation of lomefloxacin and lomefloxacin methyl ester after treatment with ethyl chloroformate and isolation by chromatography

Conditions	Products (% yield)
$NHEt_2$, 0.01 M[a]	**3′** (23), **4′** (18), **5′** (10), **6′** (3), **7′** (3)
$NHEt_2$, 0.2 M[a]	**3′** (20), **4′** (18), **5′** (10), **6′** (10), **7′** (8)
NaOH, 0.01 M	**3′** (21), **4′** (20), **5′** (9), **6′** (10), **7′** (10)
NaOH, 0.01 M[b]	**5′** (10), **6′** (14), **7′** (12)
H_2O[c]	**3′** (18), **4′** (14)

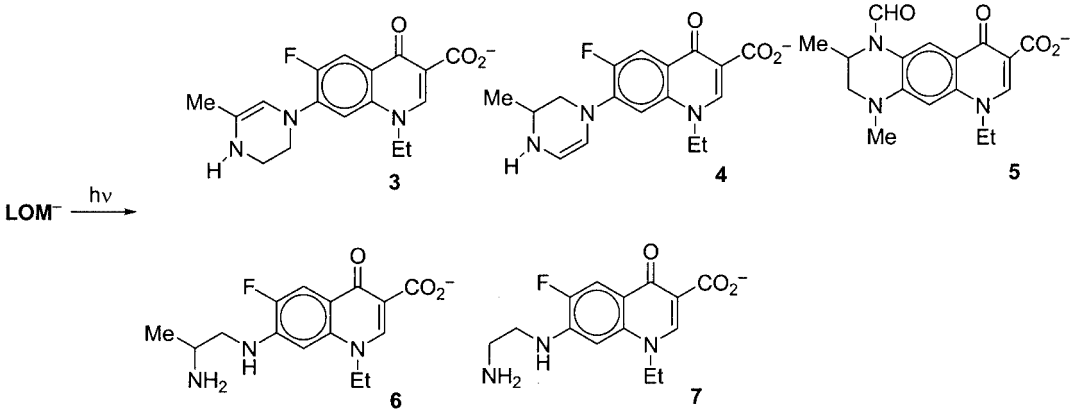
[a] Virtually identical results when using the same concentration of NEt_3. [b] The irradiated solution was kept for 2 hours at 35 °C before treatment with ethyl chloroformate and chromatography. [c] Results for lomefloxacin methyl ester.

Examination of the irradiated solution by HPLC/MS showed four peaks, with masses corresponding to products **5** (t_R = 5.3 min), **7** (5.5), **6** (6.2), and **3**+**4** (7.3). The ratio of the peaks fitted well with the ratio of the derivatized products **3′**−**7′** isolated above, and this gave us confidence that the photochemistry is well depicted by Scheme 3, although the yields reported above may underestimate the initial yields somewhat due to the isolation procedure. Some checks at partial (ca. 25 and 50%) conversion showed the same peaks in essentially the same ratio.

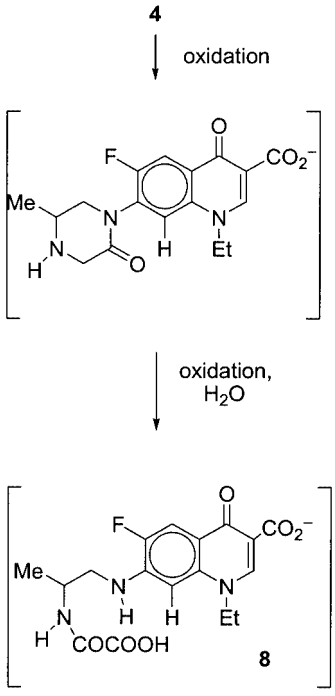
The photochemistry was then examined in the presence of 0.2 M amine (diethylamine and triethylamine). The product distribution was the same as in the previous case, although with somewhat larger amount of products **6** and **7** (see Table 1).

Lomefloxacin was then irradiated in the presence of 0.01 M NaOH, giving again a mixture of products **3**−**7** with a ratio quite similar to that from the 0.2 M amine solution.

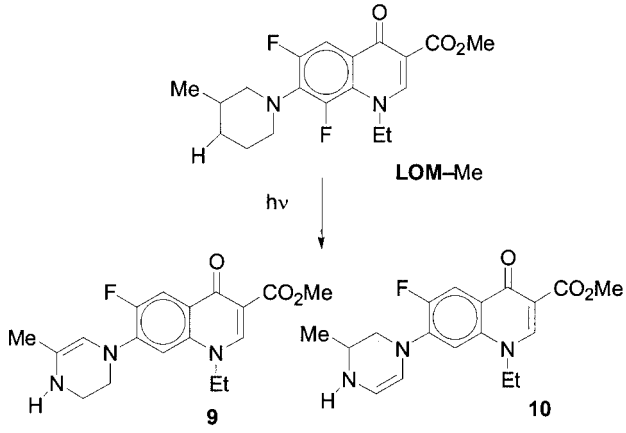
The above results refer to experiments where the solution was derivatized and extracted immediately after the end of the irradiation. However, when the irradiated 0.01 M NaOH solution was left untreated the HPLC trace changed. The peak at t_R = 7.3 min (**3**+**4**) decreased and a new peak at t_R = 5.8 min, initially quite minor, increased. After 2 h at 35 °C, the first peak disappeared and the latter one became by far the main one. The peaks corresponding to products **5**, **6** and **7** changed little, with a small increase for the latter two compounds. Derivatization and chromatography at this point gave only products **5**−**7** (see Table 1). Thus, no evidence for the structure of the t_R = 5.8 min compound was obtained, apart from the mass spectrum, which suggested a formula of $C_{17}H_{20}N_3FO_7$ or $C_{17}H_{18}N_3FO_7$. On this basis, the structure of monooxalamide **8** was tentatively suggested, indicating that oxidation and hydrolysis of compounds **3** and **4** had occurred (see Scheme 4). The same phenomenon occurred with the solutions containing 0.2 M amine and, to a lesser degree, 0.01 M amine.



Scheme 3



Scheme 4



Scheme 5

The quantum yield of the photoreaction was measured at about 20% conversion, as reported in Table 2. The values are lower than under neutral conditions (ca. 37%) and show no difference between amine- and NaOH-containing solutions. The quantum yield of liberated fluoride was in every case virtually coincident with the decomposition quantum yield.

Table 2. Photodecomposition quantum yield of lomefloxacin (LOM-H) and its methyl ester (LOM-Me) in aqueous solution

		Φ_{dec}
LOM-H	H_2O[a]	0.55
	H_2O, Et_2NH 0.01 M	0.20
	H_2O, Et_2NH 0.2 M	0.21
	H_2O, NaOH 0.01 M	0.20
LOM-Me	H_2O[a]	0.40
	H_2O, Et_2NH 0.02	0.33
	H_2O, NaOH 0.01 M	0.30

[a] Buffered at pH 7 by addition of $NaHCO_3$.

Finally, the methyl ester LOM-Me was irradiated in aqueous solution. Extraction and chromatography of the organic layer demonstrated that two products were formed under these conditions and that these corresponded to the methyl esters of the enediamines obtained from the free acid (structures **9** and **10**, Scheme 5 and Table 1). The quantum yield of the reaction (see Table 2) was slightly lower than that of LOM-H. The photoreaction of the ester was also tested in the presence of diethylamine and NaOH; the main photoproducts were again **9** and **10**, with a somewhat lower quantum yield (Table 2).

Discussion

The present study extends the range of conditions for the photochemistry of lomefloxacin. These data, in conjunction with previous results under neutral and acidic conditions, show a moderate dependence of the photoreaction efficiency on the medium. The quantum yield for decomposition is maximal under neutral conditions, and changes by a factor of up to 3 when varying the pH of the solution from 12 to 2. On the other hand the quantum yield and product distribution are the same in the presence of secondary and tertiary amines as well as in the presence of a mineral base and show a limited dependence on the amine concentration. We conclude that amines do not interact with the reactive excited state (with all FQs this is the triplet, see below)[33] either as nucleophiles or as electron donors, despite the fact that electron transfer is thermodynamically allowed;[45] they merely act by increasing the pH of the solution. This fits with the previous finding that other donors, such as the sulfite and phosphate anions, only moderately affect the photochemistry of lomefloxacin, while they completely divert the photochemistry of monofluoroquinolones,[22,44] and, as in that case, this can be attributed to the much shorter lifetime of this FQ (no strong transient detected on the nanosecond timescale),[23] which limits quenching.

Examination of the chemistry occurring supports this notion. In fact, we did not observe the reduction [via radical anion, Equation (5)] that replaces the S_NAr2 substitution for monofluoro derivatives.[22,44]

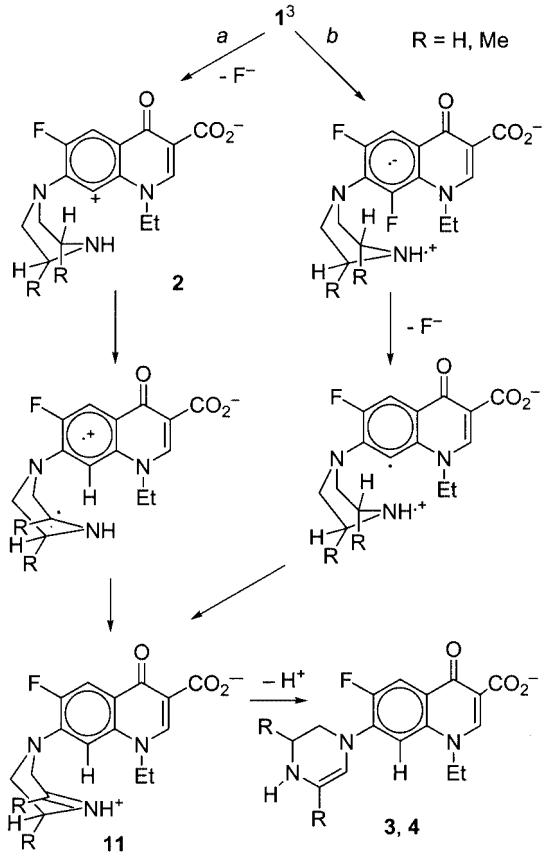
$$Ar-F^* + D \rightarrow Ar-F^{-\bullet} \rightarrow Ar^{\bullet} \rightarrow Ar\text{-}H \quad (5)$$

Rather, reduction at the 8-fluoro position, accompanied by dehydrogenation of the piperazine ring, occurs both from lomefloxacin in the presence of amines/bases and from the ester.

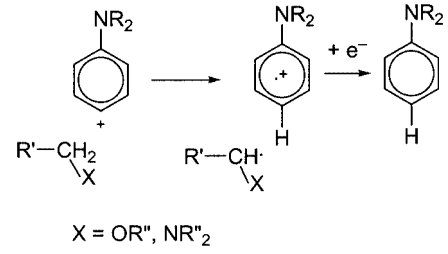
The most economic rationalization is that the primary photoprocess remains unimolecular C^8-F bond cleavage, even if the final products are different from those obtained from lomefloxacin under neutral conditions. In all of the present experiments (0.01 and 0.2 M amine, 0.01 M NaOH), more than 99% of the lomefloxacin ($pK_a = 8.78$)[46] is present as the anionic form LOM^-. Our hypothesis is that the reactive quinolone chromophore is relatively unaffected in the three ionic forms LOM-H, LOM-H_2^+, and LOM^-. The exchange of a CO_2^- for a CO_2H group at the 3-position in the latter form may affect the efficiency, but not the mode of photoreaction of the heteroaromatic moiety, while the protonation of the 4′-nitrogen in the piperazino side-chain in the two first forms has no direct bearing on the photoactive chromophore. Consistent with this, the quantum yield of reaction for lomefloxacin, while decreasing at a basic pH, remains rather high (≥0.2, see Table 2). Values that high were found in the FQ series only when a unimolecular fragmentation was involved. The S_NAr2 reactions of 6-monofluoro FQs have Φ values of about 0.1 or below. Likewise, changing to a CO_2Me group, as in LOM-Me, leads again to photodecomposition with a similar quantum yield, which decreases only slightly at pH 12, and the reaction course is similar to that observed with LOM^- under both neutral and basic conditions.

We suggest that, rather than the primary photodissociation, the pH of the medium affects the evolution of cation **2** and this is the origin of the difference with respect to the results under neutral or acidic conditions. When the piperazino nitrogen in the 4′-position is protonated, cation **2**, which, as mentioned in the introduction, is formed in the low-lying triplet state and exhibits a carbenoid chemistry, inserts into the geometrically accessible C−H bond at the β-position of the *N*-ethyl group to give **1** (Scheme 1). The C−H bond in the 3′-position of the piperazine ring would likewise be accessible and, being weaker (α to an amino group), may be expected to compete. However, protonation of the 4′-amino group in the zwitterionic form (LOM-H, present under neutral conditions) and in the cationic form (LOM-H_2^+, present under acidic conditions) inhibits the approach of this group to the positively charged heterocyclic moiety in **2**.

The situation is reversed when the 4′-piperazino group is free, as under basic conditions, when LOM^- is present, or when ester LOM-Me is irradiated under neutral or basic conditions. In this case the favored attack on 3′-H occurs via the folded conformation in Scheme 6 (path a). Insertion into the C−H bond would lead to a highly strained compound in this case, and thus hydrogen transfer takes place instead. Indeed, in the case of the 4-aminophenyl cation it has been demonstrated that alcohols and amines act as H donors, giving a radical cation that is then reduced to the final compound[34,40,47] (Scheme 7). In this case the second step would occur intramolecularly (overall a formal hydride



Scheme 6



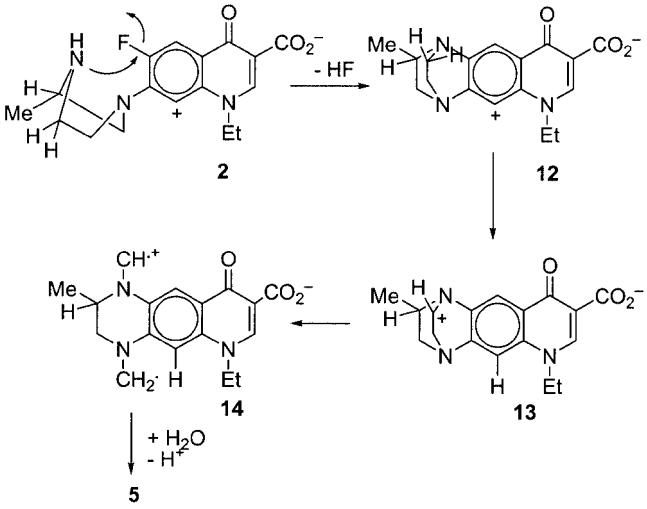
Scheme 7

transfer) to give cation **11**. As one may expect, this is followed by deprotonation from the 2′-position to yield enediamines **3** and **4** (or the corresponding esters **9** and **10** from LOM-Me), a type of photoreaction not previously found among FQs, although degradation of the piperazine moiety, probably likewise initiated by intramolecular H abstraction, has been previously observed with structurally related orbifloxacin.[48]

An alternative rationalization that can be considered is intramolecular electron transfer in the excited state, followed by fluoride loss and hydrogen transfer, finally giving again cation **11** (Scheme 6, path b). This would lead to the same result and is difficult to distinguish. Since, as mentioned above, the triplet lifetime is very short, we feel that the present data do not require that such a diversion from the usual unimolecular heterolysis is likely.

In the case of products **6** and **7**, defluorination is accompanied by oxidative hydrolysis of the piperazine side-chain. Since a rather extensive degradation takes place, it is likely that this is initiated by the photochemical step, but then involves thermal steps starting, for example, from cation **11**. Low conversion experiments showed no precursors and the detailed mechanism of this transformation is not known, although it probably involves stepwise oxidation by dissolved oxygen and is favored by a base, since the amount of **6** and **7** is lower under less-basic conditions (0.01 M amine) and the corresponding esters have not been obtained from the photolysis of LOM-Me in neutral solution. A related degradation has been previously observed for the morpholino side-chain of another drug, linezolid, which also undergoes photodefluorination.[49] Enediamines **3** and **4** are sufficiently stable and can be transformed into urethanes when treated immediately. Otherwise, they also undergo thermal oxidation to give a non-isolated product, for which formula **8** has been suggested.

The formation of minor product **5** is not obvious. Since both fluorine atoms have been eliminated in this case, we initially thought that two different (photochemical) steps were involved. However, when monitoring the reaction by HPLC we found no indication that this product results either from over-irradiation or upon standing in basic solution, the proportion of the corresponding peak not changing appreciably in such cases. Thus, we tentatively suggest a path involving, again, a reaction directly from cation **2** (Scheme 8). This involves attack by the 4′-amino group to the positively charged aromatic ring, hydrogen fluoride loss (to yield cation **12**), and hydrogen transfer to restore aromaticity. The resulting bicyclic cation **13** undergoes homolytic C−C cleavage and adds water to give the observed *N*-methyl-*N*′-formyl derivative **5**. This process appears to be promoted by bases, since it is not observed with the ester LOM-Me under neutral conditions.



Scheme 8

Conclusion

The main finding from this work is that lomefloxacin, and reasonably other 8-halo-substituted FQs, consistently undergoes efficient C−F heterolytic fragmentation under a large range of conditions, probably yielding in all cases triplet cation **2**. The triplet multiplicity gives a carbene character at C^8 to this species, rather than the reactivity of a localized carbocation, so that no solvent addition takes place in aqueous solutions, even under basic conditions or in the presence of a nucleophile such as an amine. On the contrary, this intermediate undergoes three different intramolecular reactions (insertion into the C−H bond in the β-position of the *N*-ethyl group, formal hydride transfer from the 3′-position of the piperazine side-chain, and attack by the 4′-amino group onto the C−F bond in position 6). The last two reactions give enediamines **3** and **4** (and presumably products **6** and **7**, where oxidative hydrolysis of the piperazine also takes place) and tetracyclic **5**, respectively.

This suggests that the smoothly photochemically generated (hetero)aryl cations maintain the peculiar non-reactivity with n-nucleophiles even under these "forced" conditions, and it is possible that the chemistry of such a highly reactive intermediate can be modulated by the choice of the medium.

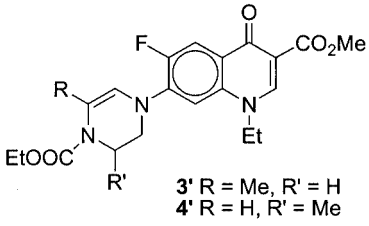
Considering that lomefloxacin is a widely used drug, one should expect that exposure of this molecule to light, for example in the skin after consumption, not only destroys the pharmaceutically active component, but also generates an aggressive reagent such as cation **2**, which may react intramolecularly as shown here, but possibly also with closely related biomolecules such as DNA bases, since the related 4-aminophenyl cation has been demonstrated to add to π-nucleophiles.[34] Such reactions would explain the oxygen-independent phototoxicity of this drug [Equation (2)]. Investigations aimed to document intermolecular reaction paths are ongoing.

Experimental Section

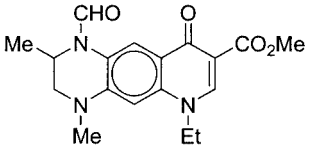
Materials: Lomefloxacin [1-ethyl-6,8-difluoro-1,4-dihydro-7-(3-methyl-1-piperazinyl)-4-oxo-3-quinolinecarboxylic acid] hydrochloride was purchased from Sigma Aldrich, St. Louis, MO. The free base (LOM-H) was prepared as previously reported.[22] The methyl ester (LOM-Me) was prepared by boiling and stirring a suspension of 1 g of LOM-H in methanol (150 mL) containing 96% H_2SO_4 (20 mL) for 2 h. The solution obtained was neutralized with NaOH, the solvent evaporated and the residue taken up with chloroform and recrystallized.

Methyl 1-Ethyl-6,8-difluoro-1,4-dihydro-7-(3-methyl-1-piperazinyl)-4-oxo-3-quinolinecarboxylate (LOM-Me): Colorless crystals, m.p. 124−127 °C. IR (KBr): $\tilde{\nu}$ = 3430, 1725, 1615, 1480, 1255 cm^{-1}. ^1H NMR ($CDCl_3$): δ = 1.2 (br. s, 3 H, piperazine CH_3), 1.55 (t, ^2J = 7 Hz, 3 H, Et CH_3), 3.1−3.3 (m, 4 H, Et CH_2 and piperazine CH_2), 3.3 (m, 2 H, piperazine CH_2), 3.45 (br. q, 1 H, piperazine CH), 3.95 (s, 3 H, OCH_3), 4.3 (m, 2 H, piperazine CH_2), 7.95 (d, $^2J_{H,F}$ = 11 Hz, 1 H, H-5), 8.3 (s, 1 H, H-2) ppm.

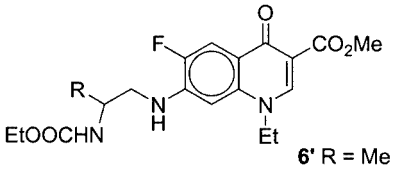
Irradiation of LOM-H: A solution of LOM-H (140 mg, 5×10^{-4} M) in doubly distilled water (800 mL) containing the appropriate amount of amine or NaOH was placed in an immersion-well apparatus, stirred and flushed with nitrogen for 30 min, and then irradiated with a 125 W high-pressure mercury arc through Pyrex while maintaining the nitrogen flux. Monitoring by HPLC (Hypersil ODS2, 250 × 4.6 mm, 5 μm, pH 3 phosphate buffer/MeCN (8:2) as eluent, flux 0.6 mL/min, λ = 275 nm) showed that the starting material had been consumed after 60 min. The solution was stirred twice for 2 h with 400 mL of 1% ethyl chloroformate in chloroform and the organic layer was separated each time. The organic layers were reunited, dried, concentrated and treated with ethereal diazomethane. The solution was evaporated and the residue chromatographed on silica gel eluting with chloroform/methanol (98:2 to 95:5 mixtures). This gave four fractions: a light-yellow oil, identified as a mixture of products **3′** and **4′**, and minor amounts of three colorless solids, two of which were identical to previously characterized samples (compounds **6′** and **7′**)[44] and the last of which was identified as compound **5′**. The key spectroscopic characteristics are reported below.



3′ R = Me, R′ = H
4′ R = H, R′ = Me



5′



6′ R = Me
7′ R = H

Mixture of 3′ and 4′: Light-yellow oil that solidifies on standing. $C_{21}H_{24}FN_3O_5$ (417.43): calcd. C 60.42, H 5.80, N 10.07; found C 60.0, H 6.1, N 9.9. IR (neat): $\tilde{\nu}$ = 1703, 1618, 1440, 1315, 1220 cm^{-1}. The NMR spectra were registered at 45 °C, since the piperazine signals were blurred at lower temperatures due to hindered rotation of the carbamate moiety.

Methyl 7-(4-Ethoxycarbonyl-2,3-dihydro-5-methylpyrazinyl)-1-ethyl-6-fluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylate (3′): ^{1}H NMR ($CDCl_3$): δ = 1.31 (t, ^2J = 7 Hz, 3 H, carbamate CH_3), 1.53 (t, ^2J = 7 Hz, 3 H, Et CH_3), 2.13 (br. s, 3 H, pyrazine CH_3), 3.65 (m, 2 H, pyrazine CH_2), 3.82 (m, 2 H, pyrazine CH_2), 3.88 (s, 3 H, OCH_3), 4.15−4.2 (m, 4 H, OCH_2 and Et CH_2), 5.88 (br. s, 1 H, pyrazine CH), 6.78 (d, $^3J_{H,F}$ = 6.5 Hz, 1 H, H-8), 8.01 (d, $^3J_{H,F}$ = 13 Hz, 1 H, H-5), 8.41 (s, 1 H, H-2) ppm; NOE effect on H-8 (δ = 6.78 ppm) upon irradiation at δ = 5.88 ppm, on CH_2 (δ = 3.65 ppm) upon irradiation at δ = 6.78 ppm, and on CH_3 (δ = 2.05 ppm) upon irradiation at δ = 5.88 ppm. ^{13}C NMR ($CDCl_3$): δ = 14.3 (CH_3), 14.4 (CH_3), 19.6 (CH_3), 42.2 (CH_2N), 47.6 (CH_2N), 48.9 (Et CH_2), 51.9 (OCH_3), 61.9 (OCH_2), 105.1 (CH-8), 109.8 (C-3), 114.1 (d, $^2J_{C,F}$ = 22 Hz, CH-5), 114.8 (d, $^4J_{C,F}$ = 5 Hz, pyrazine CH), 116.9 (pyrazine C), 123.2, 136.1 (C-4a, C-8a), 139.8 (d, $^2J_{C,F}$ = 10 Hz, C-7), 148.2 (CH-2), 150.4 (d, $^1J_{C,F}$ = 250 Hz, C-6), 153.7 (OCON), 166.3 (CO_2CH_3), 172.6 (C-4) ppm.

Methyl 7-(4-Ethoxycarbonyl-2,3-dihydro-3-methylpyrazinyl)-1-ethyl-6-fluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylate (4′): ^{1}H NMR ($CDCl_3$): δ = 1.15 (d, J = 7 Hz, 3 H, pyrazine CH_3), 1.31 (t, J = 7 Hz, 3 H, carbamate CH_3), 1.52 (t, J = 7 Hz, 3 H, Et CH_3), 3.61 (m, 2 H, pyrazine CH_2), 3.71 (m, 2 H, pyrazine CH_2), 3.88 (s, 3 H, OCH_3), 4.15 (m, 2 H, Et CH_2), 4.20 (q, J = 7 Hz, 2 H, OCH_2), 4.55 (m, 1 H, pyrazine CH), 5.81 (br. d, J = 6 Hz, 1 H, pyrazine CH=), 6.32 (br. d, J = 6 Hz, 1 H, pyrazine CH=), 6.81 (d, $^4J_{H,F}$ = 7 Hz, 1 H, H-8), 8.04 (d, $^3J_{H,F}$ = 14 Hz, 1 H, H-5), 8.42 (s, 1 H, H-2) ppm; NOE effect on H-8 (δ = 6.81 ppm) upon irradiation at δ = 5.81 ppm, on CH_2 (δ = 3.50 ppm) upon irradiation at δ = 6.81 ppm, and on CH_2 (δ = 3.71 ppm) upon irradiation at δ = 6.81 ppm. ^{13}C NMR ($CDCl_3$): δ = 14.2 (CH_3), 14.5 (CH_3), 16.1 (CH_3), 47.3 (pyrazine CH), 48.9 (Et CH_2), 51.0 (pyrazine CH_2), 51.9 (OCH_3), 61.9 (OCH_2), 104.5 (CH-8), 107.5 (pyrazine CH=), 109.8 (C-3), 111.8 (pyrazine CH=), 114.1 (d, ^2J = 22 Hz, CH-5), 123.2, 136.2 (C-4a, C-8a), 138.9 (d, ^2J = 10 Hz, C-7), 148.2 (CH-2), 150.8 (OCON), 151.7 (d, ^1J = 250 Hz, C-6), 166.2 (CO_2CH_3), 172.6 (C-4) ppm.

Methyl 1-Ethyl-6-formyl-1,4,6,7,8,9-hexahydro-7,9-dimethyl-4-oxo-3-pyrazino[2,3-*g*]quinolinecarboxylate (5′): Creamy crystals, m.p. 140−42 °C. $C_{18}H_{21}N_3O_4$ (343.38): calcd. C 62.96, H 6.16, N 12.24; found C 62.5, H 6.3, N 12.0. IR: $\tilde{\nu}$ = 1717, 1663, 1618, 1494, 1310, 1091, 1044, 1018 cm^{-1}. ^{1}H NMR (CD_3CN): δ = 1.1 (d, ^2J = 7, 3 H, pyrazine CH_3), 1.44 (t, ^2J = 7 Hz, 3 H, Et CH_3), 3.1 (s, 3 H, NCH_3), 3.25 (m, 2 H, ring H_2-8), 3.78 (s, 3 H, OCH_3), 4.25 (m, 2 H, Et CH_2), 4.9 (m, 1 H, H-7), 6.5 (s, 1 H, H-10), 7.55 (s, 1 H, H-5), 8.4 (s, 1 H, H-2), 8.7 (s, 1 H, CHO) ppm; NOE effect on H-10 (δ = 6.50 ppm) upon irradiation of the *N*-Me signal at δ = 3.10 ppm, which indicated the position of the methyl group and, by inference, of the formyl group. ^{13}C NMR (CD_3CN): δ = 13.5 (pyrazine CH_3), 15.9 (Et CH_3), 38.7 (NCH_3), 40.5 (CH-7), 48.8 (Et CH_2), 50.8 (OCH_3), 53.6 (CH_2-8), 95.5 (CH-5), 109.0 (C_q), 114.3 (CH-10), 121.0 (C_q), 121.8 (C_q), 138.0 (C_q), 142.0 (C_q), 148.1 (CH-2), 155.5 (CHO), 165.7 (CO_2Me), 172.4 (C-4) ppm.

Thermal Transformation of the Photoproducts: When the irradiated solution was kept at 35 °C for 2 h, the peak corresponding to products **3** and **4** (t_R = 7.8 min) disappeared while those corresponding to products **6** and **7** (t_R = 5.5 and 6.2 min) increased somewhat and a further peak at 5.8 min developed and became the main peak. Treatment as above and column chromatography of the solution gave only previously isolated compounds (**5′**, **6′** and **7′**). However, HPLC/MS experiments showed that the peak at 5.8 min had peaks in the mass spectrum at *m/z* = 396 (20%) [$M^+ + H_2O - 1$], 395 (100%) [$M^+ + H_2O - 2$] and 379 (18%) [M^+]; structure **8** was tentatively attributed to this product (mass 379).

Irradiation of LOM-Me: A solution of LOM-Me (150 mg, 5×10^{-4} M) in doubly distilled water (800 mL) was irradiated as above (45 min). Treatment with ethyl chloroformate in chloroform and chromatographic separation gave a single product fraction containing products **3′** and **4′**.

Quantum Yields: Quantum yields of the reaction were measured on 2-mL samples in 1-cm optical path spectrophotometric cuvettes on

an optical bench fitted with a 150-W high-pressure mercury arc and an interference filter (313 nm). The amount of the starting compound reacted (<20% in order to minimize secondary photoreactions) was assessed by HPLC as above and the fluoride anion liberated was determined by means of an ion-selective electrode. The light flux was measured by ferrioxalate actinometry.

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