

**UNIVERSITY OF MEDICINE AND PHARMACY, TÂRGU MURES
SCHOOL OF DOCTORAL STUDIES**

**Study On Metal And Inclusion Complexes Of
Fluoroquinolone Derivatives**

PHD THESIS - ABSTRACT

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Fluoroquinolones are synthetic antibacterial agents with a broad activity spectrum. The second generation fluoroquinolones, which are the focus of this thesis have a 6-fluoro substituent on the quinolone ring structure, responsible for their broad gram-negative activity and a 7-piperazinyl substituent, the last providing them greater activity against gram-positive bacteria and a longer half-life.

The appearance of clinical resistance and the withdrawal from the market of the last generation quinolones due to serious adverse effects and safety concerns led to the idea of transforming the older derivatives, but with reduced side effects of second generation in compounds with enhanced properties (increased antibacterial activity and photostability, avoidance of clinical resistance, decreased local irritation). A way of their conversion is the formation of complexes. Complexation might have advantages in various analytical procedures of determination and dosage used in the pharmaceutical industry.

Fluoroquinolones, through their C-4 keto group and C-3 carboxylic acid group are potential ligands to form **chelate complexes** with bivalent and trivalent cations and due to their hydrophobe groups and molecular dimension they can also act as guest molecules in **inclusion complexes**.

The aim of the present work was to provide spectrophotometrical methods of detection and characterization of metal complexes, to form inclusion complexes of the three second generation quinolones (norfloxacin, ofloxacin and ciprofloxacin), the elucidation of their structure and concomitant characterization by combining traditional spectrophotometric methods with modern separation technique and new computational methods. We proposed to study the electrophoretic behaviour of the optical isomers of ofloxacin in the presence of neutral cyclodextrins, as well.

After building the three-dimensional models of **ciprofloxacin** with **iron (II, III)** ions we have concluded that the most stable structures are those formed under neutral or alkaline conditions, most likely at each metal ion two bidentate ligands are bound and the remaining coordination sites are occupied by two water molecules. Fe (III) ions form more stable complexes than Fe (II).

The **UV spectrophotometric** measurements confirm the occurrence of interactions between the two components, ciprofloxacin and Fe(III), i.e. the occupancy of four coordination sites of six, namely the formation of complexes of ciprofloxacin-Fe³⁺ in a 2:1 ratio, with stability constants of order 10³ and the formation of polynuclear chains or polydentate structures besides the expected composition.

Studying the ionization of ciprofloxacin molecule in function of pH, its acidity constant was found around pH 6, which corresponds to the first stage of ionization of the molecule.

The formation of **inclusion complexes** of the three quinolone derivatives with four neutral cyclodextrins: β -CD, γ -CD, HP- β -CD and RAMEB .has been investigated.

It was established by means of **molecular modeling** that cyclodextrins act as complexing agents toward the three fluoroquinolones, the sum of the energy of the components beeing greater than the one of the complex formed, the difference indicating its stability.

The conclusions drawn from the analysis of the **IR spectra** of the complexes prepared by the method of mixing and grinding in comparison with those of the components are consistent with the data generated by computational methods.

Having photoprotective nature, cyclodextrins are masking characteristic peaks of groups which are included in their cavity. The presence of the characteristic frequency of carboxyl group in the spectra of complexes indicates that this moiety has not been embedded in the CD cavity. Some peaks assigned to C-O bond of oxazine ring of ofloxacin disappear in complexes formed with β -CD and RAMEB. Based on these results we presume that only the piperazinyl group of ciprofloxacin, respectively norfloxacin was included in the CD cavity, while in the case of ofloxacin besides the piperazinyl group a part of the oxazine ring was also encapsulated.

The small structural differences and the amphoteric character of fluoroquinolones studied is reflected also in their behavior during **capillary electrophoresis (CE)** studies.

After addition of the selectors β -CD, γ -CD, HP- β -CD and RAMEB in concentrations of 20-40 mM to the background electrolyte (25 mM borate buffer) there was observed a separation of quinolone derivatives, which indicates the formation of inclusion complexes between the three active substances and cyclodextrins.

Chiral separation of ofloxacin was performed in 0.05 M phosphate buffer at pH 3.1, by using the chiral selector RAMEB added to the buffer solution at a concentration of 40 mM. In these experimental conditions, separation occurred in 6 minutes.

Based on literature data it can be assumed that the migration of the two configurations took place in the following order: S (5.162 min) and R (5.451 min). The difference in their affinity to the host molecule allowed the separation of the enantiomers, thus CE has proved to be an eligible method for solving this challenge.

Keywords: fluoroquinolone, cyclodextrin, metal complex, inclusion complex, chiral separation, IR, UV spectrophotometry, capillary electrophoresis