

ORIGINAL ARTICLE

Determination of ofloxacin and dexamethasone in Dexaflox eye drops through different ratio spectra manipulating methods

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Abstract Different sensitive and selective spectrophotometric methods for the determination of ofloxacin and dexamethasone in their binary mixture were presented. Ofloxacin was determined simply by zero order at its λ_{\max} 293.4 nm in a linear range of 1.5–12 $\mu\text{g mL}^{-1}$ with mean percentage recovery of $100.07 \pm 0.66\%$ without any interference of dexamethasone even in low or high concentrations. Dexamethasone was determined by first derivative of ratio spectra ¹DD at 266.5 nm, ratio subtraction and mean centering at 243 nm with methods in a linear range of 2.5–27.5 $\mu\text{g mL}^{-1}$ with mean percentage recoveries of $100.09 \pm 0.70\%$, $100.00 \pm 0.72\%$ and 99.92 ± 0.62 , respectively. These methods were applied to the analysis of pharmaceutical dosage form and bulk powder where good recoveries were obtained. The proposed methods were validated according to USP guidelines.

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1. Introduction

Ofloxacin (Oflox., Fig. 1) (\pm)-9-fluoro-2,3-dihydro-3-methyl-10-(4-methyl-L-piperazinyl)-7-oxo-7H-pyrido[1,2,3-de]-1,4-benzoxazine-6-carboxylic acid¹ is a fluoroquinolone

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antibacterial agent, which is highly active against both Gram-positive and Gram-negative bacteria. It is also active against mycoplasma, chlamydia and legionella.²

The mechanism of the bactericidal effect of Oflox. is based on the inhibition of the DNA gyrase of the bacteria, the enzyme that produces a negative supercoil in DNA and thus permits transcription and replication.³

Several methods are available for analysis of Oflox. such as high-performance liquid chromatography,^{4–8} capillary electrophoresis,⁹ chemiluminescence¹⁰ and chemometry.¹¹

Dexamethasone (Dexa. Fig. 2) [9-Fluoro-11 β ,17,21-trihydroxy-16 α -methylpregna-1,4-diene-3,20-dione]¹, a potent synthetic corticosteroid with anti-inflammatory and immunosuppressive properties, is frequently used as an

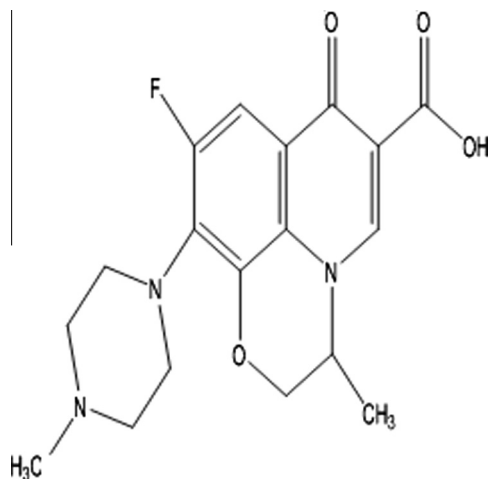


Figure 1 Structural formula for ofloxacin (MW = 361.38).

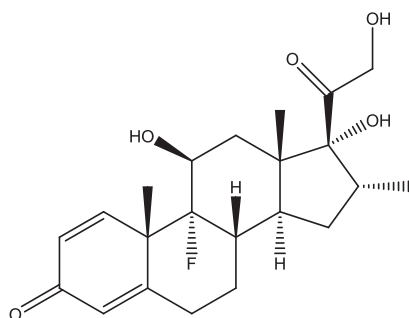


Figure 2 Structural formula for dexamethasone (MW = 392.47).

anti-inflammatory agent.¹² It has been widely used to treat inflammation, allergy and diseases related to adrenal cortex insufficiency.

Several methods had been reported in the literature describing the analysis of Dexa. including LC,^{13–17} spectrophotometry,¹⁸ TLC chromatography¹⁹ and voltametry.²⁰

Dexa. was determined with Oflox. in their binary mixture by RF HPLC²¹ or in their ternary mixture with ephedrine by microemulsion LC.²²

The aim of this study was to develop and validate spectrophotometric methods for the determination of Oflox. and Dexa. The methods were validated by using the guidelines of USP.¹

2. Experimental

2.1. Instrumentation

Spectrophotometric measurements were carried out using a double beam UV visible spectrophotometer, Shimadzu Japan, model 1601 PC, connected to IBM compatible computer and HP 800 ink Jet printer, the bundle software was a UV PC personal spectroscopy software version 3.7. The spectral band width was 0.2 nm and wavelength scanning speed was 2800 nm min⁻¹.

2.2. Chemicals and reagents

Oflox. standard material was kindly supplied by EL Nile company, Egypt. Its purity was checked in our laboratory according to the USP HPLC method¹ and was found to be 99.98 ± 0.73%. Dexa. standard material was kindly supplied by EIPICO Company, Egypt. Its purity was checked in our laboratory according to USP HPLC method¹ and was found to be 100.30 ± 1.38%. Market sample of Dexaflax eye drops was manufactured by Gamgom pharma, Saudi Arabia, each 1 ml claimed to contain 3 mg Oflox. and 1 mg Dexa., batch number KC 082 was purchased from the local market.

All reagents and solvents used were of analytical grade. 3% of 0.1 N HCl in methanol was used as a reagent for the preparation of standard solutions.

2.3. Standard stock and working solutions

Oflox. standard stock solution: 0.25 mg mL⁻¹ in 3% of 0.1 N HCl/methanol reagent.

Oflox. standard working solution: 0.1 mg mL⁻¹ in 3% of 0.1 N HCl/methanol reagent.

Dexa. standard stock solution: 0.25 mg mL⁻¹ in 3% of 0.1 N HCl/methanol reagent.

Dexa. standard working solution: 0.1 mg mL⁻¹ in 3% of 0.1 N HCl/methanol reagent.

2.4. Procedures

2.4.1. Spectral characteristics of ofloxacin and dexamethasone

The zero order absorption spectra of 9, 12 µg of Oflox. and 2.5, 27.5 µg of Dexa. solutions were recorded against 3% of 0.1 N HCl/methanol reagent as a blank over the range of 200–400 nm.

2.4.2. Construction of calibration curves

Aliquots equivalent to 15–120 µg mL⁻¹ Oflox. and 25–275 µg mL⁻¹ Dexa. are accurately transferred from their standard working solutions (0.1 mg/mL) into two separate series of 10-ml volumetric flasks then completed to volume with 3% of 0.1 N HCl/methanol reagent. The spectra of the prepared standard solutions are scanned from 200 to 400 nm and stored in the computer.

2.4.2.1. For zero order method. The content of Oflox. was determined from the measurements of its absorbance of zero order spectra at λ_{max} 293.4 nm and plotted against their corresponding concentrations. The same procedure was used to determine the content of Oflox. in the laboratory prepared mixtures and in pharmaceutical formulations.

2.4.2.2. For first derivative of ratio spectrophotometric method.

The computer stored spectra of Dexa. were divided by the previously stored spectrum of Oflox. of concentration 12 µg mL⁻¹ and the first derivative of the ratio spectra were obtained with Δλ = 4 nm and a scaling factor of ten. The content of Dexa. was calculated from the measurements of the amplitude of the first derivative peak of (Dexa/Oflox) at 266.5 nm and plotted against their corresponding concentrations. The same procedure was used to determine the content of Dexa. in the

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