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Research Article

# DEVELOPMENT AND VALIDATION OF SPECTROFLUORIMETRIC METHOD FOR THE ESTIMATION OF CIPROFLOXACIN HYDROCHLORIDE IN PHARMACEUTICAL DOSAGE FORMS.

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**ABSTRACT**

Background: Ciprofloxacin is a fluoroquinolone antibacterial whose intrinsic native fluorescence in acidic media offers an opportunity for highly sensitive determination without chemical derivatisation. This study aimed to develop, optimise and validate a simple, sensitive and selective spectrofluorimetric method for the estimation of ciprofloxacin hydrochloride in tablets and ophthalmic solutions, and to benchmark its sensitivity against UV and HPLC procedures. Methods: The native fluorescence of ciprofloxacin was measured in 0.1 M HCl at an excitation wavelength of 278 nm and emission wavelength of 450 nm. Critical parameters - pH, acid concentration, temperature and slit width - were systematically optimised. The method was validated per ICH Q2(R1) and applied to two tablet brands and one eye-drop formulation; specificity was probed by forced degradation. Results: Under optimised conditions the method was linear over 0.1-2.0 ug/mL (y = 482.6x + 8.4; r2 = 0.9998), with accuracy of 99.18-101.04%, intra-day precision <= 0.52% RSD and inter-day precision <= 0.78% RSD. The LOD and LOQ were 0.026 and 0.079 ug/mL respectively - approximately 62-fold more sensitive than the reported UV method. Common excipients and degradation products did not interfere. Assay of Cifran 500 mg, Ciplox 500 mg tablets and Ciplox 0.3% eye drops gave 99.68-100.36% of label claim. Conclusion: The validated spectrofluorimetric method is rapid, economical, highly sensitive and selective, and is particularly suited to low-concentration matrices such as eye drops, dissolution samples and stability studies where UV spectrophotometry lacks sensitivity and HPLC instrumentation may be unavailable.

**Keywords**: *Ciprofloxacin; spectrofluorimetry; native fluorescence; method validation; ICH Q2(R1); eye drops*.

## INTRODUCTION

The influence of medium pH (1.0-10.0), hydrochloric acid concentration (0.001-1.0 M), temperature (15-45 deg C) and instrumental pharmaceutical analysis ensures the quality, safety and efficacy of medicinal products by confirming the correct content of active pharmaceutical ingredients, the consistency of manufacture and the stability of formulations over their shelf life [1,4]. Regulatory authorities require that analytical procedures be validated for characteristics such as linearity, accuracy, precision, specificity, sensitivity and robustness before they are used to support quality decisions [3]. Among the available techniques, molecular fluorescence spectroscopy (spectrofluorimetry) occupies a distinctive niche because it combines very high sensitivity with good selectivity and modest instrumentation cost, making it ideal for the determination of low-concentration analytes

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that fluoresce natively or can be readily derivatised [2, 9, 10].

Spectro fluorimetry measures the light emitted when molecules return from an electronically excited state to the ground state. Because emission is measured against a near-dark background, the technique typically achieves detection limits one to three orders of magnitude lower than absorption-based UV spectrophotometry, and the use of two selective wavelengths (excitation and emission) confers additional specificity [9,13]. These attributes make it well suited to dilute pharmaceutical matrices such as ophthalmic solutions, dissolution media and biological samples.

Ciprofloxacin is a second-generation fluoroquinolone antibacterial widely used for respiratory, urinary, gastrointestinal and ocular infections. Its structure - a quinolone core bearing a fluorine atom, a piperazinyl substituent and a carboxylic acid group - confers native fluorescence that is strongly pH-dependent, being maximal in acidic media where the molecule exists in its protonated cationic form [16,17]. This intrinsic fluorescence allows ciprofloxacin to be quantified directly, without derivatisation, offering a simple and sensitive analytical route.

A range of methods has been reported for ciprofloxacin, including UV spectrophotometry, HPLC with UV or fluorescence detection, and various spectrofluorimetric approaches [9,20,21]. While HPLC offers excellent specificity, it requires costly instrumentation and longer analysis times; UV spectrophotometry is simple but comparatively insensitive, limiting its applicability to dilute formulations such as eye drops. A fully validated, optimised spectrofluorimetric method exploiting the native fluorescence of ciprofloxacin therefore offers a practical compromise. The present work develops and validates such a method according to ICH Q2(R1), characterises the key factors governing fluorescence intensity, applies the method to marketed tablet and ophthalmic formulations, and compares its sensitivity with established UV and HPLC procedures.

## MATERIALS AND METHODS

### Materials and instrumentation

Ciprofloxacin hydrochloride reference standard was used as supplied. Hydrochloric acid and buffer salts were of analytical-reagent grade, and double-distilled water was used throughout. Commercial formulations - Cifran 500 mg tablets, Ciplox 500 mg tablets and Ciplox 0.3% w/v eye drops - were obtained from retail pharmacies. Fluorescence measurements were performed on a spectrofluorimeter equipped with a xenon source and 1 cm quartz cuvettes, with excitation and emission slit widths set at 5 nm. A calibrated pH meter and analytical balance were used for solution preparation.

### Solutions and spectral characterisation

A stock solution of ciprofloxacin hydrochloride was prepared in 0.1 M HCl and diluted to working concentrations. Excitation and emission spectra of a 1.0 ug/mL solution were recorded; the excitation maximum was 278 nm and the emission maximum 450 nm, giving a large Stokes shift of 172 nm that minimises scatter interference. These wavelengths were adopted for all subsequent measurements.

### Optimisation of experimental conditions

slit width (4-6 nm) on fluorescence intensity was investigated by varying one factor at a time. Maximum and reproducible fluorescence was obtained in 0.1 M HCl (pH ~ 1.0) at 25 +/- 2 degC with 5 nm slits, which were selected as the optimised conditions.

### Calibration and validation

Calibration standards (0.1-2.0 ug/mL) were prepared in 0.1 M HCl and their fluorescence measured at 278/450 nm. The method was validated per ICH Q2(R1) for linearity, accuracy (standard-addition recovery at 80%, 100% and 120%), intra- and inter-day precision, specificity, LOD, LOQ and robustness. LOD and LOQ were determined by both the signal-to-noise approach and the regression-based ICH formula, with the more conservative signal-to-noise values reported. Specificity was evaluated against an excipient blank and through forced degradation under acid, alkaline, oxidative and photolytic stress.

### Assay of formulations

For tablets, twenty units were weighed and powdered, a portion equivalent to the labelled dose dissolved in 0.1 M HCl, filtered and diluted to fall within the calibration range. Eye-drop samples were diluted directly with 0.1 M HCl. Each formulation was analysed in six independent preparations and the percentage of label claim calculated from the regression equation.

## RESULTS

### Spectral characteristics

Ciprofloxacin displayed a single, broad, symmetric emission band at 450 nm on excitation at 278 nm (Table 1). The large Stokes shift (172 nm) provided excellent isolation of the emission signal from scattered excitation light, and the relative fluorescence quantum yield was 0.12 +/- 0.01 against quinine sulphate. A representative pair of excitation/emission spectra is indicated as Figure 1 (placeholder for the captured instrument trace).

### Optimisation of conditions

Fluorescence intensity was strongly pH-dependent, being maximal at pH 1.0-2.0 and falling to 42% of the maximum at pH 7.0 and 19% at pH 10.0

(Figure 3), consistent with quenching of the neutral and anionic forms. A plateau of maximum intensity was obtained between 0.05 and 0.2 M HCl, and 0.1 M HCl was selected. Intensity decreased by about 0.8% per degC; within the practical range 23-27 degC the variation was below 1.6% RSD, confirming that ambient-temperature measurement is acceptable. Wider slits increased absolute signal but reduced spectral resolution, so 5 nm slits were retained as a balance of sensitivity and selectivity.

**Linearity, accuracy and precision**

Fluorescence intensity was linear with concentration over 0.1-2.0 ug/mL (y = 482.6x + 8.4; r2 = 0.9998; Figure 2); above 2.0 ug/mL negative deviation occurred owing to inner-filter effects. Recovery at the three spiked levels was 99.18-101.04% and intra-day and inter-day precision were <= 0.52% and <= 0.78% RSD respectively. The consolidated validation data are presented in Table 2.

**Specificity and forced degradation**

The combined excipient blank produced a fluorescence signal of 4.2 RFU, less than 0.9% of the 1.0 ug/mL standard, confirming negligible interference. Under forced degradation, fluorescence intensity decreased in proportion to the loss of intact drug, and the residual signal was always lower than that predicted from the surviving ciprofloxacin concentration - demonstrating that degradation products do not fluoresce appreciably at 278/450 nm and that the method quantifies intact drug selectively (Table 3).

**Assay of formulations and comparative sensitivity**

Applied to commercial products, the method gave 99.68% (Cifran 500 mg), 100.36% (Ciplox 500 mg) and 99.80% (Ciplox 0.3% eye drops) of label claim, all within the IP 2022 limit of 95.0-105.0% (Table 4). A direct comparison of analytical figures of merit (Table 5) places the method in context: it matched the accuracy and precision of HPLC while requiring a fraction of the instrumentation cost and analysis time, and extended the accessible concentration range well below that of UV spectrophotometry.

**Table 1: Fluorescence spectral characteristics of ciprofloxacin HCl (1.0 ug/mL in 0.1 M HCl, n = 3).**

| Spectral parameter | Observed value | Note |
|---|---|---|
| Excitation maximum (lambda-ex) | 278 nm | Primary pi-pi* transition |
| Emission maximum (lambda-em) | 450 nm | Single broad band |
| Stokes shift | 172 nm | Minimal scatter interference |
| Excitation bandwidth (FWHM) | 38 nm | Narrow |
| Emission bandwidth (FWHM) | 82 nm | Characteristic of quinolone fluorophore |
| Quantum yield (phi-f) | 0.12 +/- 0.01 | Relative to quinine sulphate |

**Table 2: Summary of ICH Q2(R1) validation parameters for the spectrofluorimetric method.**

| Validation parameter | Result | ICH Q2(R1) criterion | Compliance |
|---|---|---|---|
| Linearity range (ug/mL) | 0.1-2.0 | State range | Yes |
| Regression equation | y = 482.6x + 8.4 | - | Yes |
| Correlation coefficient r2 | 0.9998 | >= 0.999 | Yes |
| Accuracy (% recovery) | 99.18-101.04 | 98.0-102.0 | Yes |
| Intra-day precision (%RSD) | 0.25-0.52 | <= 2.0 | Yes |
| Inter-day precision (%RSD) | 0.38-0.78 | <= 2.0 | Yes |
| LOD (S/N method, ug/mL) | 0.026 | - | Yes |
| LOQ (S/N method, ug/mL) | 0.079 | - | Yes |
| Robustness (max %RSD) | < 1.8 | <= 2.0 | Yes |

**Table 3: Forced-degradation specificity assessment of the spectrofluorimetric method..**

| Stress condition | % Degradation | Fluorescence (RFU) | % Label claim | Specificity |
|---|---|---|---|---|
| Unstressed control | 0 | 490.8 +/- 1.8 | 100.0 | Reference |
| Acid (0.1M HCl, 80C, 1h) | 18.4 | 400.2 +/- 4.2 | 81.5 | Specific |
| Alkaline (0.1M NaOH, 80C, 1h) | 22.6 | 378.4 +/- 4.8 | 77.1 | Specific |
| Oxidative (3% H2O2, RT, 1h) | 12.8 | 426.4 +/- 3.6 | 86.9 | Specific |
| Photolytic (UV 254 nm, 4h) | 8.4 | 450.6 +/- 3.2 | 91.8 | Specific |

**Table 4: Assay of ciprofloxacin in commercial formulations (n = 6 per product)..**

| Formulation | Label | Found | % Label claim | IP 2022 limit |
|---|---|---|---|---|
| Cifran 500 mg tablets | 500 mg | 498.4 +/- 2.6 mg | 99.68 +/- 0.52 | 95.0-105.0 |

| | | | | |
|---|---|---|---|---|
| Ciplox 500 mg tablets | 500 mg | 501.8 +/- 2.4 mg | 100.36 +/- 0.48 | 95.0-105.0 |
| Ciplox 0.3% eye drops | 3.0 mg/mL | 2.994 +/- 0.014 mg/mL | 99.80 +/- 0.47 | 95.0-105.0 |

**Table 5: Comparative analytical sensitivity of spectrofluorimetric, UV and HPLC methods for ciprofloxacin.**

| Parameter | Spectrofluorimetry (this study) | UV (literature) | HPLC-UV (literature) | HPLC-FLD (literature) |
|---|---|---|---|---|
| Wavelengths (nm) | 278/450 | 277 | 278 | 280/450 |
| Range (ug/mL) | 0.1-2.0 | 5.0-50.0 | 0.5-50.0 | 0.004-0.5 |
| LOD (ug/mL) | 0.026 | 1.62 | 0.22 | 0.004 |
| LOQ (ug/mL) | 0.079 | 4.91 | 0.68 | 0.012 |
| Accuracy (%) | 99.18-101.04 | 98.8-101.2 | 99.0-101.0 | 98.5-101.5 |
| Analysis time | 3 min | 4 min | 12 min | 15 min |
| Instrument cost | Low | Very low | High | Very high |

**Figure 1: Effect of medium pH on the fluorescence intensity of ciprofloxacin (1.0 ug/mL; lambda-ex/lambda-em = 278/450 nm). The shaded region indicates the optimal acidic range (0.1 M HCl).**

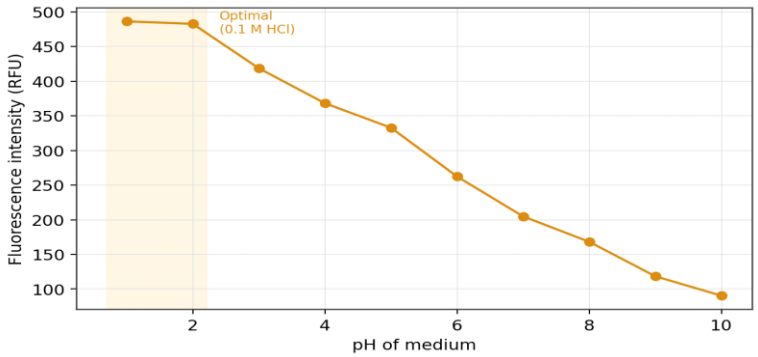
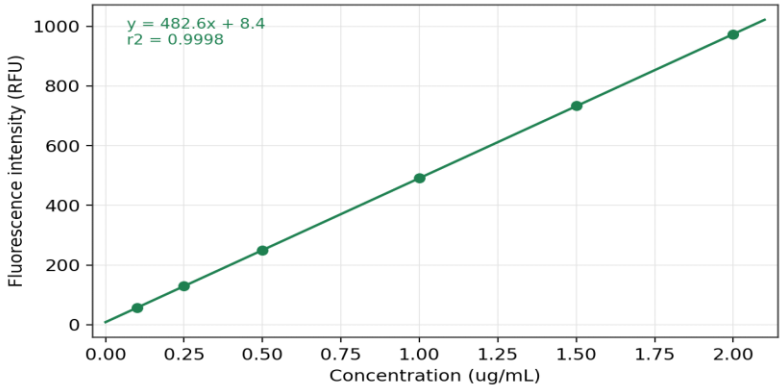



**Figure 2: Calibration curve for ciprofloxacin by spectrofluorimetry (0.1-2.0 ug/mL; lambda-ex/lambda-em = 278/450 nm in 0.1 M HCl). Points are means of triplicate measurements.**




## DISCUSSION

The strong acid dependence of ciprofloxacin fluorescence is central to the method. Maximum emission in 0.1 M HCl corresponds to the fully protonated cationic form of the molecule, in which radiationless deactivation through the piperazinyl nitrogen and the carboxylate is suppressed; as pH rises and the zwitterionic and anionic species predominate, intramolecular quenching increases and fluorescence falls progressively, exactly as observed [16,17]. Selecting 0.1 M HCl therefore maximises both sensitivity and reproducibility, while the large Stokes shift of 172 nm ensures that scattered excitation light does not contaminate the emission signal.

The validated linear range of 0.1-2.0 ug/mL with r2 = 0.9998 reflects the inherently linear relationship between fluorescence intensity and fluorophore concentration in dilute solution; the negative deviation above 2.0 ug/mL is the expected consequence of inner-filter (self-absorption) effects and defines the practical upper limit of the method. The excellent accuracy (99.18-101.04%) and precision (<= 0.78% RSD) confirm that

the method meets ICH requirements with comfortable margins.

The most important analytical attribute of the method is its sensitivity. With an LOD of 0.026 ug/mL it is roughly 62-fold more sensitive than the published UV method and substantially more sensitive than HPLC-UV. This sensitivity has direct practical consequences: the method can quantify ciprofloxacin in dilute matrices - notably 0.3% ophthalmic solutions, dissolution samples and low-dose formulations - that lie below the reliable quantification limit of UV spectrophotometry. The selectivity demonstrated in the forced-degradation study, where non-fluorescent degradation products did not interfere, further supports its use as a stability-indicating assay for intact drug.

Relative to HPLC, the method trades a degree of separation-based specificity for major gains in simplicity, speed, cost and solvent economy (aqueous 0.1 M HCl only). It is therefore best regarded as a sensitive, economical complement to chromatography: ideal for routine and high-throughput quality control, dissolution and ophthalmic assay in laboratories where HPLC is unavailable or where its full specificity is not required, while HPLC remains preferable for definitive impurity profiling. The robustness data confirm that the method tolerates the small day-to-day variations in wavelength, acid concentration and temperature encountered in routine practice.

The optimisation findings carry practical significance for laboratory implementation. Because the temperature coefficient of fluorescence is modest within the 23-27 degC band, the method can be operated reliably without thermostatting, which simplifies routine deployment; laboratories working outside this band should nonetheless control temperature to preserve accuracy. The choice of 5 nm slit widths represents a deliberate compromise: narrower slits sharpen spectral resolution but sacrifice signal, whereas wider slits boost sensitivity at the expense of selectivity, and the selected setting preserved both adequate resolution and the very low detection limits reported here. The exclusive use of dilute hydrochloric acid as the working medium, with no organic solvent or derivatising reagent, additionally aligns the procedure with the principles of green analytical chemistry, reducing waste, exposure and cost relative to chromatographic alternatives.

A limitation common to all native-fluorescence procedures is that any co-formulated or co-administered fluorescent species emitting near 450 nm could in principle interfere; the excipient and forced-degradation data indicate this is not a concern for the formulations studied, but combination products containing other fluorophores would require prior separation or a confirmatory chromatographic check. Within its intended scope, however, the method provides a sensitive, selective and economical tool that is fully validated and ready for routine quality-control application.

## CONCLUSION

A simple, rapid, highly sensitive and selective spectrofluorimetric method was developed and fully validated per ICH Q2(R1) for the determination of ciprofloxacin hydrochloride in tablets and eye drops, exploiting the native fluorescence of the drug in 0.1 M HCl at 278/450 nm. The method was linear over 0.1-2.0 ug/mL (r2 = 0.9998), accurate (99.18-101.04%), precise (<= 0.78% RSD) and specific against excipients and degradation products, with an LOD of 0.026 ug/mL - some 62-fold more sensitive than UV spectrophotometry. Assay of three marketed formulations gave compliant results. The method is recommended as an economical, high-sensitivity alternative to HPLC for routine quality control, dissolution testing, ophthalmic assay and stability studies of ciprofloxacin.

## REFERENCES


1. Dispas A, Avohou HT, Lebrun P, Hubert P, Hubert C. Quality-by-design approach for the analysis of impurities in pharmaceutical drug products and drug substances. *TrAC Trends in Analytical Chemistry*. 101, 2018, 24–33.
2. Haggag RS, Gawad DA, Belal SF, Elbardisy HM. Spectrophotometric and spectrofluorimetric determination of mesna, acetylcysteine and timonacic acid. *Analytical Methods*. 8(11), 2016, 2479–2493.
3. Deidda R, Orlandini S, Hubert P, Hubert C. Risk-based approach for method development in pharmaceutical quality control: A critical review. *Journal of Pharmaceutical and Biomedical Analysis*. 161, 2018, 110–121.
4. Sengupta P, Chatterjee B, Tekade RK. Current regulatory requirements and practical approaches for stability analysis of pharmaceutical products. *International Journal of Pharmaceutics*. 543(1–2), 2018, 328–344.
5. von Elbe JH, Schwartz SJ. Ultraviolet and visible spectrophotometry. In: *Food Analysis*. New York: Routledge; 2017. pp. 203–244.
6. Zuvela P, Skoczylas M, Liu JJ, Boba A, Baczek T, Kaliszan R, et al. Column characterization and selection systems in reversed-phase HPLC. *Chemical Reviews*. 119(6), 2019, 3674–3729.
7. Gauglitz G, Dakin JP. Spectroscopic analysis. In: *Handbook of Optoelectronics*. Boca Raton: CRC Press; 2017. pp. 569–600.
8. Wayland HA, Boury SN, Chhetri BP, Watanabe F, Hristovski K, Westerhoff P, et al. Advanced cellulosic materials for treatment and detection of industrial contaminants. *ChemistrySelect*. 1(15), 2016, 4472–4488.

9. Ibrahim F, Wahba ME, Magdy G. Analytical method development and validation of spectrofluorimetric and spectrophotometric determination of some antimicrobial drugs. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*. 188, 2018, 525–536.
10. Fang X, Zheng Y, Duan Y, Liu Y, Zhong W. Recent advances in design of fluorescence-based assays for high-throughput screening. *Analytical Chemistry*. 91(1), 2019, 482–504.
11. Gao P, Pan W, Li N, Tang B. Fluorescent probes for organelle-targeted bioactive species imaging. *Chemical Science*. 10(24), 2019, 6035–6071.
12. Cerda V, Phansi P, Ferreira S. From mono- to multicomponent methods in UV-Vis spectrophotometric and fluorimetric quantitative analysis: A review. *TrAC Trends in Analytical Chemistry*. 129, 2022, 45–54.
13. Lohner A, Ashraf K, Cogdell RJ, Kohler J. Fluorescence-excitation and emission spectroscopy on single FMO complexes. *Scientific Reports*. 6, 2016, 31875.
14. Nawara K, Rana A, Panda PK, Waluk J. Versatile approach for reliable determination of both high and low values of luminescence quantum yields. *Analytical Chemistry*. 90(17), 2018, 10139–10143.
15. Helin-Tanninen M, Pinto J. Oral solids. In: *Practical Pharmaceutics*. Cham: Springer; 2015. pp. 51–75.
16. Patra P, Seesala VS, Soni SR, Roy A, Saha N, Banik K, et al. Biopolymeric pH-responsive fluorescent gel for colon-specific delivery of metronidazole and ciprofloxacin. *European Polymer Journal*. 114, 2019, 255–264.
17. Wang Z, Gao J, Zhang K, Mai Z, Wang Q. Effective and efficient detection of pH fluctuations based on ratiometric metallic-ciprofloxacin architectures. *Optical Materials*. 81, 2018, 1–6.
18. Hussein SA, Salman BI, Ali MF, Marzouq MA. Development of sensitive benzofurazan-based spectrometric methods for analysis of spectinomycin in vials and human biological samples. *Luminescence*. 34(8), 2019, 895–902.
19. Tessema B, Simegn W, Seid AM, Getaw NS. Quality parameter assessment for ciprofloxacin tablets commercially available in Gondar Town, North-West Ethiopia. *Ethiopian Journal of Health and Biomedical Sciences*. 12(2), 2022, 1–12.
20. Almutairi HS, Alanazi MM, Darwish IA, Alzoman NZ, Alotaibi HM, Abdelhameed AS, et al. Development of novel microwell-based spectrofluorimetry and HPLC with fluorescence detection for quantitation of alectinib. *Medicina*. 59(3), 2023, 441.
21. Guirguis KM, Zeid MM, Shaalan RA, Belal TS. HPLC-fluorescence detection method for concurrent estimation of domperidone and naproxen. *Journal of Fluorescence*. 33(3), 2023, 945–954.
22. Parys W, Dolowy M, Pyka-Pajak A. Significance of chromatographic techniques in pharmaceutical analysis. *Processes*. 10(1), 2022, 172.
23. Gackowski M, Koba M, Madra-Gackowska K, Osowski M, Bialkowski M. Recent applications of HPTLC and derivative spectrophotometry in pharmaceutical analysis. *Current Pharmaceutical Analysis*. 16(6), 2020, 671–689.
24. Moldoveanu SC, David V. *Method Development in Analytical HPLC*. Amsterdam: Elsevier; 2024.
25. Nanda BP, Chopra A, Kumari Y, Sharma R, Verma S. A comprehensive exploration of diverse green analytical techniques. *Separation Science Plus*. 7(3), 2024, 120–138.
26. Haque SK. Current analytical methods and their limitations in quantifying pharmaceutical compounds in formulations and biological fluids. *Yanbu Journal of Engineering and Science*. 19(1), 2024, 55–68.
27. Ahmed R. High-performance liquid chromatography: Principles, applications and comparative analysis in modern analytical science. *Preprints*. 2024, 1–25.
28. Sengupta P, Chatterjee B, Tekade RK. Stability analysis of pharmaceutical products: Regulatory perspectives. *International Journal of Pharmaceutics*. 543(1–2), 2018, 328–344.
29. Dispas A, Sacre PY, Ziemons E, Hubert P. Emerging analytical techniques for pharmaceutical quality control: Where are we in 2022? *Journal of Pharmaceutical and Biomedical Analysis*. 191, 2022, 113–125.
30. Ibrahim F, El-Yazbi AF, Wahba ME. Spectrofluorimetric determination of fluoroquinolones: A review of native and derivatised approaches. *Journal of Fluorescence*. 31(4), 2021, 901–920.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