



## ULTRA-FAST LIQUID CHROMATOGRAPHIC DETERMINATION OF ATOMOXETINE, DULOXETINE, PAROXETINE, FLUOXETINE AND SERTRALINE IN TAP WATER, URINE, AND PHARMACEUTICAL FORMULATIONS USING 7,7,8,8-TETRACYANOQUINODIMETHANE AS A DERIVATIZATION REAGENT

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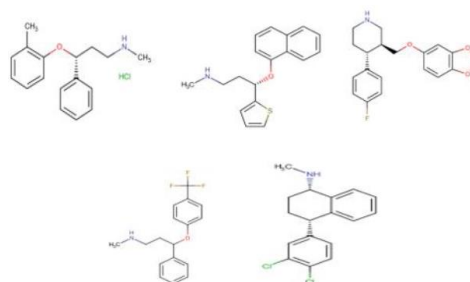
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Received May 1, 2025

The widespread consumption of pharmaceuticals, including both human and veterinary drugs, has resulted in the accumulation of these substances in aquatic ecosystems, presenting significant ecological threats. Psychoactive drugs, particularly antidepressants, raise particular concern due to their direct effects on brain chemistry. The growing contamination of global water systems, especially in urban environments, underscores the increasing presence of these substances in nature. A novel analytical method using UFLC-UV was developed and validated for the quantification of Atomoxetine (ATM), Duloxetine (DLX), Paroxetine (PRX), Fluoxetine (FLX) and Sertraline (SRT) in tap water, urine, and pharmaceutical samples. This technique relies on creating a purple chromogen through a displacement reaction with TCNQ in acetonitrile, with heating at 80 °C for 20 minutes. The processed sample was then injected into a C18 column. Separation was performed using acetonitrile–water (85:15) as mobile phase at 1.0 mL/min flow rate. Detection was carried out at 567 nm. The method showed linearity in the range of 10–100 ng/mL for all compounds. The validated procedure was successfully implemented to identify these antidepressants in tap water, urine, and pharmaceutical formulations. Recovery values were found between 95.41 to 100.85%. The LOD values ranged from 1.02 to 1.98 ng/mL in all of the sample matrices.



### INTRODUCTION

Psychiatric drugs are organic compounds designed to treat mental health disorders. It has been found that surface water and groundwater, which constitute the main sources of drinking water, contain

psychoactive substances.<sup>1–2</sup> As a result, tap water has become a significant route of exposure to psychoactive substances, even for individuals who do not use prescription psychiatric drugs or illegal drugs. The accumulation of drugs in the environment has become an increasingly concerning issue

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worldwide. Due to the slow degradation of these substances in nature, they persist in water sources

such as rivers, oceans, and groundwater, posing risks to ecosystems and human health.<sup>3-6</sup>

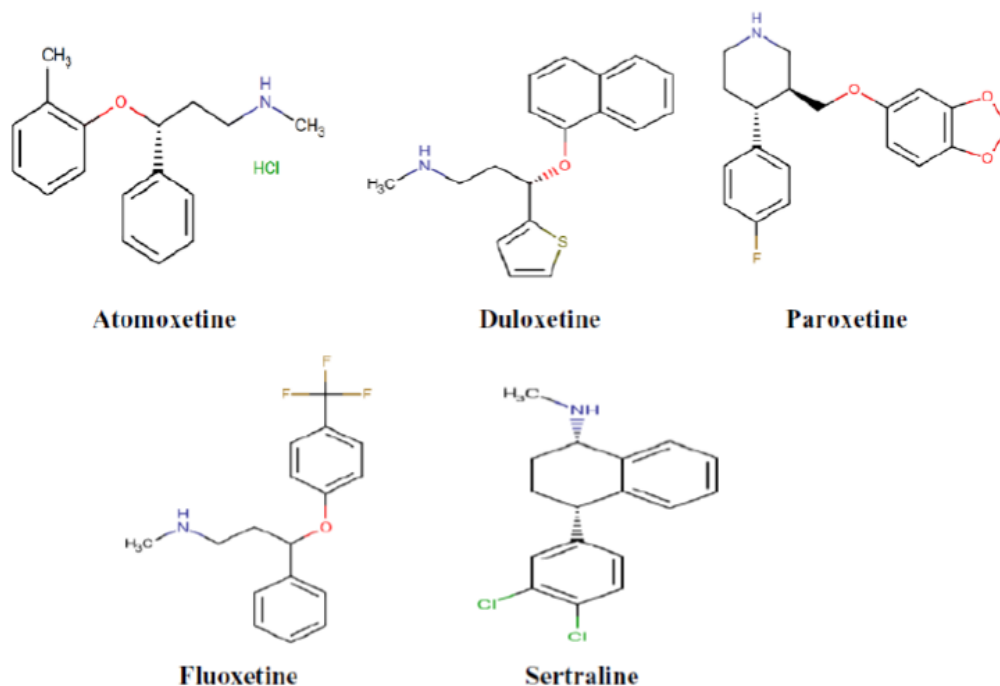


Fig. 1 – Chemical structures of antidepressants.

Multiple analytical methods have been employed for examining Atomoxetine, Duloxetine, Paroxetine, Fluoxetine and Sertraline (Fig. 1) in pharmaceutical preparations, biological matrices and environmental samples. The analytical methodologies described in the literature consist of HPLC,<sup>7-11</sup> UHPLC,<sup>12-16</sup> GC,<sup>17-20</sup> enabling accurate quantification in pharmaceutical formulations and biological specimens.

Chemical derivatization is a crucial technique in chromatography for improving detection and selectivity, particularly in amine analysis. TCNQ (7,7,8,8-Tetracyanoquinodimethane) serves as an effective derivatization reagent for n-electron donor drugs, forming purple derivatives with secondary aliphatic amines through displacement reactions, thus enabling efficient HPLC analysis.<sup>21-24</sup>

The research objective was to establish and validate an efficient analytical method for the simultaneous determination of five antidepressants – atomoxetine, duloxetine, paroxetine, fluoxetine and sertraline in various sample matrices. The developed method combined UFLC with TCNQ derivatization, resulting in enhanced detection capabilities for these compounds in urine, tap water, and pharmaceutical samples. This analytical approach provided rapid analysis times while

maintaining high sensitivity and accuracy in quantification.

## EXPERIMENTAL

### Material and Chemicals

The standard compounds of antidepressants (ATM, DLX, PRX, FLX and SRT) were supplied from Sigma-Aldrich Chemie (Steinheim, Germany). Commercial pharmaceutical formulations including ATTEX® capsules (40 mg atomoxetine HCl), DYLOXIA® capsules (60 mg duloxetine), PAXERA® film tablets (40 mg paroxetine), DEPREKS® capsules (20 mg fluoxetine HCl), and MISOL® film tablets (100 mg sertraline HCl) were obtained from a drug store. Analytical grade chemicals and reagents were used throughout the experimental work. The derivatization reagent TCNQ was purchased from Fluka (Neu-Ulm, Germany). Urine samples were collected from healthy volunteer researchers, with no ethical approval required due to voluntary self-collection. Water samples were obtained from the municipal tap water system in Istanbul, Turkey.

### Equipment and Chromatographic Condition

The HPLC experiments were carried out using a “Shimadzu LC 20A UFLC (Shimadzu, Kyoto, Japan) Ultra Fast Liquid Chromatography system”. This system was outfitted with a “SIL AT-HT auto-sampler, an LC-20AT pump, and UV-Visible detector, set to 567 nm, along with a CTO 10 AC column oven”. Separation of the analytes was achieved using an Inertsustain C18 (4.0 × 100 mm, 3 μm) analytical column (GL-Sciences, Tokyo, Japan). The mobile phase consisted of acetonitrile and water in a 85:15 ratio, at a flow rate of 1.0 mL/min, with a total run time of 10 minutes at ambient temperature. Prior to use, the mobile phase was degassed in an ultrasonic bath and filtered using a Millipore vacuum filter system equipped with a 0.45 μm HV filter.

### Preparation of Standard Solution

Stock solutions of individual drugs and their mixtures were prepared in water by dissolving their salts giving a concentration of 100 μg/mL as bases. These solutions were further diluted with water to required concentrations for working solutions. A freshly prepared solution of TCNQ was used at a concentration of 2 mg in 10 mL acetonitrile. A 1 M sodium bicarbonate solution was prepared by dissolving 4.2 g of sodium bicarbonate in 50 mL of water.

### Extraction of Urine, Tap Water and Commercial Dosage Form Samples and Sample Derivatization

Urine samples were collected from male and female volunteers. The human urine samples were provided voluntarily, and no ethical approval was required. Urine samples (1 mL) were spiked with working solutions to achieve drug concentrations ranging from 10 to 100 ng/mL and the samples were alkalinized by adding 0.2 mL of 1 mol/L KOH solution. The free bases were liberated and extracted with 5 mL of chloroform by vortex-mixing. After centrifugation at 4500 g, the organic layer was dried using 500 mg of anhydrous sodium sulfate and centrifuged again. The solution (4.5 mL) was transferred into a new tube and evaporated to dryness under nitrogen with gentle heating. Then, 1 mL of TCNQ reagent was added to the residue, and the tube was sealed. The mixture was heated at

80 °C for 20 minutes. Afterward, the solvent was evaporated under nitrogen at 45 °C, and the residue was dissolved in 100 μL of acetonitrile. A 20 μL aliquot was then directly injected into the chromatographic column.

Tap water samples were processed in the same manner. For the pharmaceutical formulations, powdered tablets containing 10 mg of antidepressants were placed into a 50 mL volumetric flask. Approximately 15 mL of water was added to the flask. The extraction was mechanically stirred for 20 minutes, followed by sonication for an additional 20 minutes. Then, water was added to bring the total volume to 50 mL, and the solution was filtered. The derivatization process was carried out as described above.

### Validation of HPLC Method

The developed UFLC method was validated in accordance with FDA guidelines, focusing on several key parameters including selectivity, accuracy, precision, linearity, sensitivity, and intermediate precision of spiked specimens containing the analyte.<sup>25</sup>

To evaluate intraday and interday precision, 1 mL of urine sample was spiked with the drug at concentrations of 10, 50, and 100 ng/mL. The samples were then analyzed (n = 5) for intraday variability and over a period of five different days to assess interday variability, following the described procedure.

Extraction recovery studies were conducted by analyzing tap water samples spiked with the drug at three concentrations (10, 50, and 100 ng/mL). The drug was added before the extraction process. The reaction and chromatographic procedures were then carried out as described. The peak-area ratios of the drug were determined, and extraction recoveries were evaluated using the calibration curve prepared with aqueous drug solutions.

Intra-day and inter-day studies were performed to determine the precision of the UFLC method, while recovery experiments demonstrated the accuracy of the method. Selectivity studies were carried out to confirm that no matrix effects influenced the results.

The robustness of the method was investigated by intentionally modifying key experimental conditions such as the composition of the organic phase, the flow rate of the mobile phase, and key detection parameters such as the selected wavelength. Additionally, the stability of the drug solution in the mobile phase was examined by

keeping the drug solution at ambient temperature for 24 hours.

## RESULTS AND DISCUSSION

The developed HPLC method relies on the formation of a purple chromogen, which is produced through displacement reactions between antidepressants and TCNQ in acetonitrile at 80 °C for 20 minutes. The method involves extracting the drug base from urine (1 mL) using chloroform after alkalization, followed by derivatization with TCNQ, and directly injecting the resulting reaction

mixture onto a reversed-phase C18 column with detection at 567 nm. The mobile phase consisted of acetonitrile and water in a 85:15 ratio, with the derivatives being eluted at 4.76, 6.27, 7.02, 8.09 and 9.34 minutes for ATM, DLX, PRX, FLX and SRT respectively.

### Conditions for chromatography

The developed HPLC method is based on the purple chromogen formed by displacement reactions of antidepressants with TCNQ in acetonitrile at 80 °C for 20 minutes.

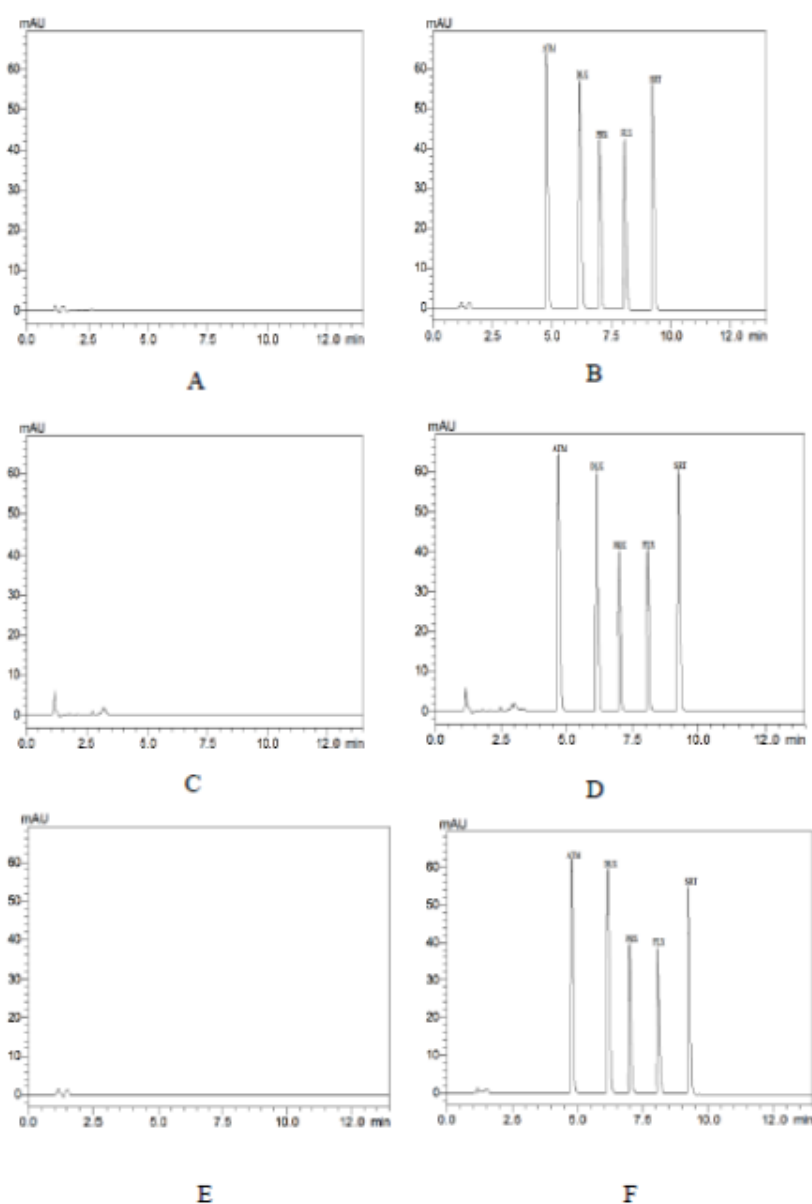


Fig. 2 – Chromatograms of; A) – Blank sample (solvent); B) – Pharmaceutical formulations sample spiked with antidepressants (50 ng/mL); C) – Blank tap water sample; D) – Tap water sample spiked with antidepressants (50 ng/mL); E) – Blank urine sample; F) – Urine sample spiked with antidepressants (50 ng/mL).

The method includes extraction of the drug base from urine, tap water and pharmaceutical formulations (1 mL) with chloroform after alkalization, derivatization with TCNQ, and injection of the reaction mixture directly into a reversed-phase C<sub>18</sub> column. Acetonitrile: water (85:15) was used as the mobile phase, and the derivatives were eluted at 4.76, 6.27, 7.02, 8.09 and 9.34 minutes for ATM, DLX, PRX, FLX and SRT respectively with detection at 567 nm. Figure 2 represents the chromatograms of stated antidepressants in tap water, urine, and pharmaceutical formulations. As illustrated in Fig. 2. The chromatographic performance was satisfactory for the compounds with good shapes, clear of endogenous interference and straight baseline. Although the resolution of FLX was close to 1.0, baseline separation was considered acceptable. The resolution and tailing factor values for each analyte were calculated under the optimized chromatographic conditions. The resolution (Rs) and tailing factor (Tf) values were found to be 2.31 and 1.05 for ATM, 1.44 and 1.08 for DLX, 1.20 and 1.10 for paroxetine (PRX), 1.02 and 1.06 for FLX, and 1.55 and 1.07 for SRT, respectively. These results indicate that all analytes were well separated with sharp and symmetrical peaks, confirming the suitability of the chromatographic method.

### Optimization of derivatization reaction parameters

The chemical reaction agent, (TCNQ), was employed for the derivatization of antidepressants in water, urine, and pharmaceutical samples. Experiments were conducted to investigate the effect of TCNQ concentration. It was determined that 1 mL of a TCNQ solution in acetonitrile was sufficient for the derivatization reaction. The maximum reaction yield was achieved in a water bath at 80 °C for 20 minutes. The reagent volume used for the derivatization reaction ranged from 0.5 mL to 1.5 mL, with the optimum volume found to be 1.0 mL. The reaction temperature varied between 30 °C and 100 °C, with the optimum temperature determined to be 80 °C. The reaction time ranged from 5 to 30 minutes, with the optimal time found to be 20 minutes. The corresponding chromatograms are given in the supplementary material section (additional Figs. 1–3).

### Method validation

Calibration graphs were created for antidepressants in urine, tap water, and pharmaceutical preparations across concentration

ranges of 10–100 ng/mL. The peak area of antidepressants for standard substances, urine, and tap water samples at varying concentrations were recorded and plotted, resulting in the calibration graphs.

The limit of detection (LOD) and the limit of quantitation (LOQ) were determined using the equations  $Kx\sigma/S$ , where  $k$  is a constant value (3.3 for LOD and 10 for LOQ),  $s$  is the standard deviation of the intercept, and  $a$  is the slope. The calculated LOD and LOQ values, along with the parameters of the calibration functions (including the slope and intercept), are provided in Table 1. Although LOD and LOQ values were calculated to estimate the method's sensitivity, the method's practical sensitivity was further supported by accurate quantification of the analytes at low concentration levels (10 ng/mL) in real matrices with acceptable recovery and precision.

Deionized water, spiked urine, and tap water samples at three different antidepressants concentration levels (10, 50, and 100 ng/mL) were analyzed to assess the mean recovery following extraction. Recovery values for the substances in the samples ranged from 95.41 to 100.85%.

To assess the precision of the UFLC method, both intra-day and inter-day precision were evaluated. The results for both intra- and inter-day analyses are presented in Table 1.

The method was assessed for its robustness by deliberately altering specific process parameters, such as the mobile phase flow rate ( $\pm 0.1$  mL/min) and the column oven temperature ( $25 \pm 2^\circ\text{C}$ ). Despite observing slight variations, these adjustments did not significantly influence the overall results. The changes in the mobile phase flow rate resulted in RSD% values ranging from 1.46 to 1.86. The changes in the column temperature resulted in RSD% values ranging from 0.86 to 0.91. Consequently, the findings supported the robustness of the method outlined in this study.

The reaction between antidepressants and TCNQ is simple, and no buffer solution or any other agent is needed to proceed. The formed derivative is stable at room temperature in the dark up to 24 hours. In addition, to evaluate the effect of freezing and heating, spiked samples were subjected to three freeze-thaw ( $-20^\circ\text{C}$  to room temperature) and separate samples were stored at  $45^\circ\text{C}$  for 24 hours. In both cases, no significant changes were observed in peak area or retention time, and recovery values remained within the 95–101% range, indicating that the analytes are stable under these conditions. Antidepressants is not affected by

freezing, thawing, or heating processes. No significant change was observed in urine, tap water and pharmaceutical formulations spiked with the drug and stored at  $-20\text{ }^{\circ}\text{C}$  for a period of 2 months. To further confirm the stability of the spiked formulations, analyses were performed over a 60 days period. The results indicated that no significant degradation or alteration occurred in the spiked samples during this time interval, supporting the stability of the formulations and confirming that the reaction procedure is not expected to cause any harm to the drug. To evaluate long-term stability, spiked samples were stored at  $-20\text{ }^{\circ}\text{C}$  and analyzed at intervals of 0, 30, and 60 days. Chromatographic

responses and recovery percentages were compared at each time point. The results demonstrated no significant degradation, with recovery values between 94.8% and 101.2%, and %RSD values below 2.0%. these results confirm that the spiked formulations remained stable for at least 60 days under storage conditions. Degradation or alteration of the analytes was assessed by monitoring peak areas, retention times, and the overall chromatographic profiles at defined intervals. No significant changes were observed during the analysis period, and recovery values remained within the range of 94.8% to 101.2%, supporting the chemical stability of the compounds in the tested matrices.

Table 1

Validation and regression parameters for antidepressants

	Deionised water					Spiked urine					Tap water				
	ATM	DLX	PRX	FLX	SRT	ATM	DLX	PRX	FLX	SRT	ATM	DLX	PRX	FLX	SRT
<b>Concentration</b>															
<b>Linear Range<sup>a</sup> (ng/mL)</b>	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100	10-100
<b>Regression equation<sup>b</sup></b>															
<b>Slope ± SD</b>	503.45 ±10.23	392.57 ±5.27	350.42 ±6.01	362.46 ±2.98	377.79 ±7.37	498.74 ±18.25	399.56 ±9.78	375.39 ±12.41	374.86 ±5.69	396.74 ±12.41	500.14 ±10.03	389.76 ±6.39	347.19 ±4.22	365.89 ±3.87	370.46 ±8.02
<b>Intercept ± SD</b>	1446.3 ±24.36	2112.9 ±35.36	2298.8 ±39.35	2386.6 ±25.39	1506.1 ±24.87	1521.46 ±29.87	2251.45 ±42.69	2359.74 ±55.61	2478.96 ±29.68	1785.45 ±29.76	1458.42 ±20.37	2126.89 ±30.57	2265.41 ±52.14	2301.2 ±20.65	1514.36 ±27.87
<b>Correlation Coefficient</b>	0.9949	0.9975	0.9994	0.9992	0.9996	0.9938	0.9967	0.9961	0.9974	0.9975	0.9958	0.9941	0.9991	0.9994	0.9992
<b>LOD (ng.mL<sup>-1</sup>)</b>	1.05	1.98	1.87	1.46	1.24	1.08	1.67	1.79	1.32	1.02	1.09	1.89	1.77	1.39	1.02
<b>LOQ (ng.mL<sup>-1</sup>)</b>	3.47	6.54	6.18	4.82	4.09	3.56	5.51	5.90	4.29	3.36	3.59	6.24	5.84	4.59	3.37
<b>Precision</b>															
<b>Intra-day RSD %</b>	0.92	0.84	0.73	0.91	0.87	1.24	1.36	1.42	1.87	1.49	0.90	0.83	0.74	0.98	0.92
<b>Inter-day RSD %</b>	1.14	1.35	0.97	1.16	1.29	1.63	1.84	1.79	1.68	1.99	1.19	1.37	0.93	1.24	1.67

<sup>a</sup> Average of six determinations.

<sup>b</sup>  $y = aC + b$  where  $C$  is the concentrations in ng/mL and  $y$  is the peak area.

### Applicability of the method

To evaluate the method's applicability, experiments were conducted using tap water, urine, and pharmaceutical samples. Accuracy was assessed by performing three replicate measurements at three different spiked concentrations for each sample type.

For deionized water, recoveries were between 99.45% and 100.85%, while for tap water, recoveries ranged from 99.58% and 100.89%, with standard deviations not exceeding 1.74%, as shown in Table 1. For urine samples, the recoveries ranged from 95.25% to 96.84%, with standard deviations not higher than 1.99% (Table 2).

Table 2  
Extraction recovery of antidepressants from deionised water, tap water and urine samples

Added concentration (ng/mL)	ATM		DLX		PRX		FLX		SRT	
	Recovery% ± RSD*% *	±	Recovery% ± RSD*% *	±	Recovery% ± RSD*% *	±	Recovery% ± RSD*% *	±	Recovery% ± RSD*% *	±
Deionised water	10	99.45± 1.16		99.89±1.01		100.85±0.86		100.47± 0.41		99.95± 1.35
	50	99.76± 0.96		100.01±1.32		99.86±1.02		99.89± 1.09		99.97± 0.85
	100	99.97± 0.68		100.17± 0.58		100.34± 0.85		99.83± 1.47		99.90±0.95
Tap water	10	99.58±0.99		99.93± 1.89		100.37± 0.91		100.85± 0.83		99.67± 1.41
	50	99.70±1.01		100.89± 1.74		99.87± 1.56		99.91± 1.58		99.86± 1.27
	100	99.95± 0.73		100.36± 0.96		100.18± 0.81		99.99± 1.74		99.64± 0.99
Urine	10	95.14± 1.99		96.89± 1.87		95.84±1.74		95.65± 1.69		95.54± 1.86
	50	96.83± 1.84		95.57± 1.94		95.41± 1.56		96.24± 1.74		95.25± 1.14
	100	96.17± 1.54		96.24± 1.23		95.71± 1.34		95.78± 1.97		96.84± 1.58

To assess the method's applicability, antidepressants in tablet formulations were also analyzed. The results, summarized in Table 3, showed excellent recovery

percentages and standard deviations (SD) of less than 2. No interference from additives or excipients was observed during the experiment.

Table 3  
Determination of antidepressants in pharmaceutical preparation by the proposed methods (n = 5)

Antidepressants	Label claim (mg/per tablet)	Mean <sup>f</sup> ± S.D	Recovery (%)	RSD (%)
ATM <sup>a</sup>	40	40.09 ± 0.44	100.23	1.10
DLX <sup>b</sup>	60	60.02 ± 0.27	100.03	0.45
PRX <sup>c</sup>	40	40.21 ± 0.56	100.53	1.39
FLX <sup>d</sup>	20	19.99 ± 0.27	99.95	1.35
SRT <sup>e</sup>	100	100.31 ± 1.12	100.31	1.12

<sup>a</sup> ATTEX<sup>®</sup> capsule(containing 40 mg atomoxetine HCl)

<sup>b</sup> DYLOXIA<sup>®</sup> capsule(containing 60 mg duloxetine)

<sup>c</sup> PAXERA<sup>®</sup> film tablet (containing 40 mg paroxetine)

<sup>d</sup> DEPREKS<sup>®</sup> capsule(containing 20 mg fluoxetine HCl)

<sup>e</sup> MISOL<sup>®</sup> film tablet (containing 100 mg sertraline HCl)

<sup>f</sup>five independent analyses.

## CONCLUSION

This study presents a novel, sensitive, and reliable UFLC method for the simultaneous determination of atomoxetine, duloxetine, paroxetine, fluoxetine, and sertraline in various matrices using TCNQ as a derivatization reagent. The developed method demonstrated excellent linearity, precision, and accuracy across all target

analytes, with detection limits ranging in the ng/mL level, suitable for trace analysis in environmental and biological samples. The method's main advantages include minimal sample preparation, rapid analysis time (less than 10 minutes), and high throughput capability. The successful application to tap water, urine samples, and pharmaceutical formulations validates the method's versatility and robustness in different matrices. Moreover, the use

of TCNQ as a derivatization reagent enhanced the selectivity and sensitivity of the analysis, making it a valuable alternative to existing methods. Although the method demonstrated excellent intermediate precision, further studies may be conducted to assess full reproducibility under different laboratory conditions. This technique offers the advantage of a short elution time (approximately 10 minutes), enabling quick analysis, and a low limit of detection (LOD) of  $1.02 \text{ ng}\cdot\text{mL}^{-1}$ . This validated analytical procedure offers a practical solution for routine quality control in pharmaceutical analysis and environmental monitoring of these widely prescribed psychiatric medications. The method's applicability to various matrices makes it particularly valuable for both pharmaceutical quality control and environmental monitoring programs. Compared to previously reported analytical methods for the determination of these antidepressants, the proposed UFLC method with TCNQ derivatization offers several advantages. These include a shorter analysis time (less than 10 minutes), improved sensitivity (LODs as low as  $1.02 \text{ ng/mL}$ ), and simplified sample preparation. Additionally, simultaneous quantification of five analytes at a single detection wavelength enhances the method's efficiency and practicality. Compared to previously reported methods, the proposed method offers shorter analysis time and improved sensitivity.

## REFERENCES

1. A. Rodayan, S. Afana, P. A. Segura, T. Sultana, C. D. Metcalfe, V. Yargeau, *Environ. Toxicol. Chem.*, **2016**, *35*, 843–849.
2. Z. Wang, S. Gao, Q. Dai, M. Zhao, F. Yang, *Environ. Poll.*, **2020**, *261*, 114163.
3. A. J. Scheen, *Diabetes Metab.*, **2020**, *46*, 423–426.
4. M. Ouda, D. Kadadou, B. Swaidan, A. Al-Othman, S. Al-Asheh, F. Banat, S. W. Hasan, *Sci. Total Environ.*, **2021**, *754*, 142177.
5. K. Kümmerer, *Annu. Rev. Environ. Resour.*, **2010**, *35*, 57–75.
6. M. A. Mottaleb, C. Stowe, D. R. Johnson, M. J. Meziani, M. A. Mottaleb, *Food Chem.*, **2016**, *190*, 529–536.
7. W. Guo, W. Li, G. Guo, J. Zhang, B. Zhou, Y. Zhai, C. Wang, *J. Chrom. B*, **2007**, *854*, 128–134.
8. J. W. Chae, H. M. Baek, S. K. Kim, H. I. Kang, K. I. Kwon, *Biomed. Chromatogr.*, **2013**, *27*, 953–955.
9. Z. Zhu, L. Neirinck, *J. Chrom. B*, **2002**, *780*, 295–300.
10. C. Sabbioni, F. Bugamelli, G. Varani, L. Mercolini, A. Musenga, M. A. Saracino, S. Fanali, M. A. Raggi, *J. Pharm. Biomed. Anal.*, **2004**, *36*, 351–356.
11. V. Melis, I. Usach, J. E. Peris, *J. Sep. Sci.*, **2012**, *35*, 3302–3307.
12. T. G. Skaalvik, E. L. Øiestad, R. Trones, S. Pedersen-Bjergaard, S. Hegstad, *J. Chrom. B*, **2021**, *1183*, 122926.
13. N. Kumar, D. Sangeetha, P. Balakrishna, *Pharm. Methods*, **2011**, *2*, 161–166.
14. M. Snamina, R. Wietecha-Postuszny, M. Zawadzki, *Talanta*, **2019**, *204*, 607–612.
15. M. Runfola, D. L. D. Lima, A. P. Fonseca, Z. Barbosa, *J. Sep. Sci.*, **2018**, *41*, 4246–4252.
16. P. Cai, X. Xiong, D. Li, Y. Zhou, C. Xiong, *Bioanalysis*, **2020**, *12*, 35–52.
17. A. Kul, M. Özdemir, A. Önal, O. Sagırlı, *Microchem. J.*, **2021**, *164*, 105953.
18. L. Fernandez-Lopez, M. Pellegrini, M. C. Rotolo, A. Luna Maldonado, M. Falcon, R. Mancini, *Forensic Sci. Int.*, **2019**, *299*, 154–160.
19. P. Cabarcos-Fernández, M. J. Tabernero-Duque, I. Álvarez-Freire, A. M. Bermejo-Barrera, *J. Anal. Toxicol.*, **2021**, *46*, 146–156.
20. E. S. Koçoğlu, S. Bakırdere, S. Keyf, *Bull. Environ. Contam. Toxicol.*, **2017**, *99*, 354–359.
21. A. Oztunc, A. Onal, S. Erturk, *J. Chrom. B*, **2002**, *774*, 149–155.
22. A. Onal, A. Oztunc, *Ther. Drug Monit.*, **2006**, *28*, 180–184.
23. A. Onal, A. Oztunc, *Rev. Anal. Chem.*, **2011**, *30*, 165–169.
23. A. Oztunc, A. Onal, S. E. Toker, *J. AOAC Int.*, **2010**, *93*, 556–561.
25. U.S. Food and Drug Administration (FDA), <http://www.fda.gov/cder/Guidance/4252fnl.pdf> (2001) Access date:2021.