Detection of atomoxetine and its metabolites in the urine by thin-layer chromatography and mass spectrometry

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A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

The aim of the study was the detection of atomoxetine and its biotransformation products in the urine under TLC screening conditions and identification of the metabolites using mass spectrometry method.

Materials and methods. The volunteer's urine samples after taking a single therapeutic dose of atomoxetine (2 capsules of 60 mg each of Strattera®) were studied. Sample preparation included diluting acid hydrolysis followed by the native compound and metabolites extraction with chloroform from the saturated solution of ammonium sulfate at pH of 11–12. Thin-layer chromatography studies of the extracts were carried out in 18 mobile phases including those proposed by The International Association of Forensic Toxicologists for general drug screening, and those widely used in forensic toxicological studies. The color reactions were carried out using a range of chromogenic reagents. A Varian 1200 L mass spectrometer (Netherlands) equipped with a dual quadrupole mass analyzer was applied for analysis of the eluates from chromatograms. Identification was undertaken at the direct introduction of the sample into the ion chamber, electron-impact ionization (70 eV), and full ion scanning mode.

Results. The spot of the native drug on the chromatogram was identified by the R_f value. Two atomoxetine biotransformation products were identified by the molecular weights that correspond to the molecular ion peaks in the mass spectra.

Conclusions. Atomoxetine and its biotransformation products were detected in the urine under TLC screening conditions and identified using mass spectrometry method. Chromatographic mobility of the native compound, hydroxyatomoxetine, and dihydroxyatomoxetine in the TLC screening systems as well as the results of their visualization using chromogenic reagents applied for toxicological drug screening in the systematic toxicological analysis have been determined.

Key words: atomoxetine, hydroxyatomoxetine, dihydroxyatomoxetine, sample preparation, thin layer chromatography, mass spectrometry.

Current issues in pharmacy and medicine: science and practice 2022; 15 (1), 25–30

Виявлення атомоксетину та його метаболітів у сечі за допомогою тонкошарової хроматографії та мас-спектрометрії

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Мета роботи – виявлення атомоксетину та продуктів його біотрансформації в сечі в умовах ТШХ-скринінгу та ідентифікація метаболітів методом мас-спектрометрії.

Матеріали та методи. Досліджено зразки сечі добровольця після приймання одноразової терапевтичної дози атомоксетину (2 капсули по 60 мг Strattera®). Пробопідготовка включала кислотний гідроліз із дальшою екстракцією нативної сполуки та метаболітів хлороформом із насиченого розчину сульфату амонію при pH 11–12. Дослідження екстрактів методом тонкошарової хроматографії проводили у 18 рухомих фазах, які включали ті, що запропоновані Міжнародною асоціацією судових токсикологів для загального скринінгу лікарських речовин, а також фази, котрі широко використовують при судово-токсикологічних дослідженнях. Кольорові реакції виконали з використанням ряду хромогенних реактивів. Для аналізу елюатів із хроматограм використовували мас-спектрометр Varian 1200 L (Нідерланди) з подвійним квадрупольним мас-аналізатором. Ідентифікацію здійснили під час прямого введення зразка в іонну камеру, іонізації електронним ударом (70 еВ) у режимі повного сканування іонів.

Результати. Пляму нативного препарату на хроматограмі ідентифікували за значенням R_f. Два продукти біотрансформації атомоксетину ідентифікували за молекулярними масами, що відповідали пікам молекулярних іонів у мас-спектрах.

Висновки. Атомоксетин і продукти його біотрансформації виявили в сечі в умовах ТШХ та ідентифікували методом мас-спектрометрії. Встановили хроматографічну рухливість нативної сполуки, гідроксиатомоксетину та дигідроксиатомоксетину у скринінгових ТШХ-системах, а також результати їхньої візуалізації за допомогою хромогенних реагентів, що використовують для токсикологічного скринінгу лікарських засобів у систематичному токсикологічному аналізі.

ARTICLE INFO

http://pharmed.zsmu.edu.ua/article/view/252070

UDC 615.065:54.061/062:547.712.22:001.8
DOI: 10.14739/2409-2932.2022.1.252070

Current issues in pharmacy and medicine: science and practice 2022; 15 (1), 25–30

Key words: atomoxetine, hydroxyatomoxetine, dihydroxyatomoxetine, sample preparation, thin layer chromatography, mass spectrometry

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Received: 20.12.2021 // Revised: 10.01.2022 // Accepted: 18.01.2022

Ключові слова: атомоксетин, гідроксиатомоксетин, дигідроксиатомоксетин, пробопідготовка, тонкошарова хроматографія, мас-спектрометрія.

Актуальні питання фармацевтичної і медичної науки та практики. 2022. Т. 15, № 1(38). С. 25–30

Обнаружение атомоксетина и его метаболитов в моче методами тонкослойной хроматографии и масс-спектрометрии

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Цель работы – обнаружение атомоксетина и продуктов его биотрансформации в моче в условиях ТСХ-скрининга и идентификация метаболитов методом масс-спектрометрии.

Материалы и методы. Исследовали образцы мочи добровольца после приёма одноразовой терапевтической дозы атомоксетина (2 капсулы по 60 мг Strattera®). Пробоподготовка включала кислотный гидролиз с последующей экстракцией нативного соединения и метаболитов хлороформом из насыщенного раствора сульфата аммония при pH 11–12. Исследования экстрактов методом тонкослойной хроматографии проводили в 18 подвижных фазах, включавших рекомендованные Международной ассоциацией судебных токсикологов для общего скрининга лекарственных веществ, а также фазы, которые широко используют при судебно-токсикологических исследованиях. Цветные реакции производили с использованием ряда хромогенных реактивов. Для анализа элюатов с хроматограмм использовали масс-спектрометр Varian 1200 L (Нидерланды) с двойным квадрупольным масс-анализатором. Идентификацию проводили при прямом введении образца в ионную камеру, ионизации электронным ударом (70 эВ) в режиме полного сканирования ионов.

Результаты. Пятно нативного препарата на хроматограмме идентифицировали по значению R_f. Два продукта биотрансформации атомоксетина были идентифицированы по молекулярным массам, соответствующим пикам молекулярных ионов в масс-спектрах.

Выводы. Атомоксетин и продукты его биотрансформации обнаружено в моче в условиях ТСХ и идентифицировано методом масс-спектрометрии. Установлена хроматографическая подвижность нативного соединения, гидроксиатомоксетина и дигидроксиатомоксетина в скрининговых ТСХ-системах, а также результаты их визуализации с помощью хромогенных реагентов, используемых для токсикологического скрининга лекарственных средств в систематическом токсикологическом анализе.

Ключевые слова: атомоксетин, гидроксиатомоксетин, дигидроксиатомоксетин, пробоподготовка, тонкослойная хроматография, масс-спектрометрия.

Актуальные вопросы фармацевтической и медицинской науки и практики. 2022. Т. 15, № 1(38). С. 25–30

Atomoxetine ((3R)-*N*-methyl-3-(2-methylphenoxy)-3-phenylpropan-1-amine hydrochloride) is a modern antidepressant drug relating to a class of selective norepinephrine reuptake inhibitors (SNRI). Atomoxetine was the first non-stimulant drug approved by the FDA (USA) in late 2002 year for the treatment of ADHD in both children and adults [1,2]. Unlike traditional psychostimulants, atomoxetine does not have a potential for abuse; it is not classified as a controlled substance. The therapeutic daily dose is 40 mg daily increased to 80 mg after 3 days and up to 100 mg after 2 to 4 weeks [3]. Atomoxetine has also been shown to be effective for treating therapeutically resistant depression [4,5]. It can cause a range of side effects [2,6–8], and episodes of acute and lethal atomoxetine poisoning also were registered [3,9–11].

Atomoxetine is actively metabolized in the body via hydroxylation, *N*-desmethylation, and benzyl oxidation. Less than 3 % of atomoxetine dose is excreted as an unchanged drug [2,3,12]. The postmortem atomoxetine concentration in the urine was 0.1 mg/L. Half-life is 5.2 h and 21.6 h depending on the type of metabolizers: an extensive and a poor type respectively [3].

TLC screening is the most accessible type of screening procedure that is used in forensic toxicology. The International Association of Forensic Toxicologists (TIAFT) has proposed unified TLC systems for which a database of R_f values for more than 16.000 toxicologically important drugs has been created [3]. Studies on the determination of the chromatographic mobility parameters for atomoxetine in the TLC systems recommended by TIAFT have not been carried out.

Aim

The aim of the study was the detection of atomoxetine and its biotransformation products in the urine under TLC screening conditions and identification of the metabolites using mass spectrometry method.

Materials and methods

The pure substance of atomoxetine was isolated from the medicine “Strattera” (7 capsules, 60 mg; “Lilly” (serial number C406168), Czech Republic) according to the method described in the article [13]. Atomoxetine-base was obtained by extraction of the analyte with chloroform from the aqueous solution at pH of 11 saturated with ammonium sulphate. The volunteer’s urine samples after taking a single therapeutic dose of atomoxetine (2 capsules of 60 mg each of Strattera®) were studied. Urine was collected in separate portions of 50 mL for 14 hours, starting after the 5th hour of taking the antidepressant. All chemicals were of analytical grade or better.

Sample preparation. The concentrated hydrochloric acid in the volume of 1 ml (it was based on the ratio of 0.10 mL of the acid for every 2.0 mL of the biological fluid) was added to 20 mL of the urine and the mixture was heated in a boiling water bath for 30 min. The obtained hydrolysate was cooled, placed in a separatory funnel, and shaken with 7 mL of diethyl ether three times for 5 min each time. The organic layer was separated and discarded. The aqueous layer was placed in a separatory funnel again, alkalified with the sodium hydroxide 20 % solution to pH of 11–12, ammonium sulphate

was added until the saturated solution was obtained, and atomoxetine-base was extracted with 10 mL of chloroform three times each time. The extracts obtained were combined, filtered through a pleated blue band paper filter containing 0.2 g of anhydrous sodium sulphate, and evaporated in a water bath at 40 °C to remove chloroform. The dry residue was dissolved in 5 mL of chloroform, mixed thoroughly, transferred to a volumetric flask with a capacity of 25.00 mL adjusting to the specified volume with the same solvent.

Thin-layer chromatography studies of the extracts. The Merk chromatography plates were used. Aliquot of the 1/10 part of the obtained biological extract, aliquot of the 1/10 part of the blank biological extract were concentrated to the minimum volume of ~0.05 mL and applied on four TLC plates as spots. Then 10 μL of the standard solution of atomoxetine in methanol (1.0 mg/mL) was spotted next using microsyringe. The remaining biological extract evaporated to the minimum volume of ~0.05 mL was applied as a band only on the chromatography plate, which then was developed in the mobile phase of chloroform – acetone – 25 % ammonia solution (25:5:0.3). Then the zone in the chromatogram corresponding to this band was not treated by the location reagent.

Chromatograms were developed in chloroform, and then in one of the mobile phases which are listed in *Table 1* using rectangular glass chambers (25 × 25 × 12 cm). UV-light, ninhydrin solution in acetone and Dragendorff–Munier reagent were used for the visualization. Then atomoxetine was eluted from the chromatogram band untreated by the location reagents with 4 mL of methanol (the elution yield was 99.2 %), the eluate was filtered through a blue band paper filter.

The color reactions were carried out with 0.5 mL of the obtained eluates using pieces of chromatographic plates and a range of the chromogenic reagents listed in *Table 2*.

Mass Spectrometry studies of the eluates. A Varian 1200 L mass spectrometer (Netherlands) equipped with a dual quadrupole mass analyzer was applied for analysis. Detection was undertaken at the direct introduction of the sample into ion chamber, electron-impact ionization (70 eV), and full ion scanning mode.

Results

Three spots were detected on the chromatograms of the biological extracts which were obtained after sample preparation of the volunteer's urine after taking a single therapeutic dose of atomoxetine. Blank biological extracts did not give corresponding spots. The spot of the native compound was identified by R_f, the value of which coincided with the specified retention parameter for atomoxetine in the standard solution.

Figure 1 shows atomoxetine-base mas spectrum (EI, 70 eV), m/z (I_{rel},%): 255 $[M]^+$(4), 176 (4), 148 (16), 117 (4), 108 (6), 91 (6), 72 (7), 65 (3), 44 (100.00). Found, m/z: 255.15 $[M]^+$. $C_{17}H_{21}NO$. Calculated, m/z: 255.4 [3]. The compliance with the database of NIST 17 mass spectrum library was of 85–87 %.

Figures 2, 3 show mass spectra of the eluates from chromatograms corresponding to the other two spots that did not match the native drug. Suspected products of atomoxetine biotransformation were identified by the molecular weights that correspond to the molecular ion peaks in the mass spectra.

Table 1. R_f values of atomoxetine and its metabolites extracted from the urine

No.	Mobile phase	Atomoxetine	Hydroxyatomoxetine	Dihydroxyatomoxetine
1	Chlorophorm-acetone (80:20)	0	0	0
2	Ethyl acetate	0	0	0
3	Chlorophorm-methanol (90:10)	0.15	0.08	0.05
4	Ethyl acetate-methanol – 25 % ammonia solution (85:10:5)	0.49	0.31	0.24
5	Methanol	0.05	0.03	0.01
6	Methanol-*n*-buthanol (3:2)	0.11	0.04	0.01
7	Methanol – 25 % ammonia solution (100:1.5)	0.36	0.22	0.19
8	Cyclohexane-toluene-diethyl amine (15:3:2)	0.22	0.07	0.04
9	Acetone	0.01	0	0
10	Toluene-acetone-ethanol – 25 % ammonia solution (45:45:7.5:2.5)	0.46	0.20	0.17
11	Chlorophorm-dioxane -aetone – 25 % ammonia solution (47.5:45:5:2.5)	0.47	0.18	0.12
12	Chloroform-*n*-butanol (3:2)	0.86	0.75	0.72
13	Ethyl acetate-acetone –25 % ammonia solution (50:45:5)	0.55	0.38	0.35
14	Benzene-Methanol-Diethylamine (90:10:10)	0.63	0.46	0.43
15	Chloroform-acetone – 25 % ammonia solution (25:5:0.3)	0.74	0.60	0.56
16	Hexane-ethyl acetate-ethanol – 25 % ammonia solution (30:10:5:1)	0.27	0.10	0.07
17	Chloroform-acetone – 25 % ammonia solution (12:24:1)	0.38	0.22	0.18
18	Chloroform	0	0	0

Hydroxyatomoxetine mas spectrum (EI, 70 eV), m/z (I_{rel},%): 271 $[M]^+$(2), 254 $[M\text{-}OH]^+$(2), 176 (20), 165 (23), 146 (9), 133 (24), 117 (24), 105 (44), 91 (15), 77 (49), 60 (53), 44 (100) (*Fig. 1*). Found, m/z: 271.30 $[M]^+$. $C_{17}H_{21}NO_2$. Calculated, m/z: 271.4.

Dihydroxyatomoxetine mas spectrum (EI, 70 eV), m/z (I_{rel},%): 287 $[M]^+$(1), 271 $[M\text{-}OH]^+$(2), 255 $[M\text{-}OH\text{-}OH]^+$(2),

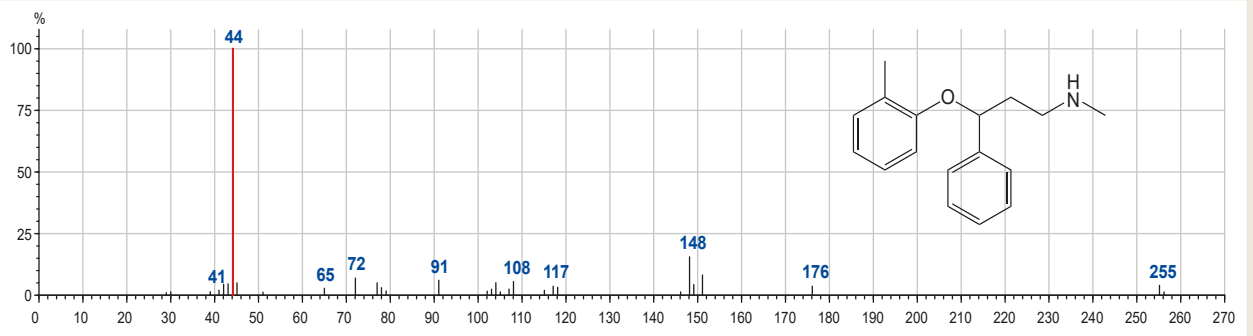

Fig. 1. Mass spectrum of atomoxetine-base obtained from the substance of the drug.

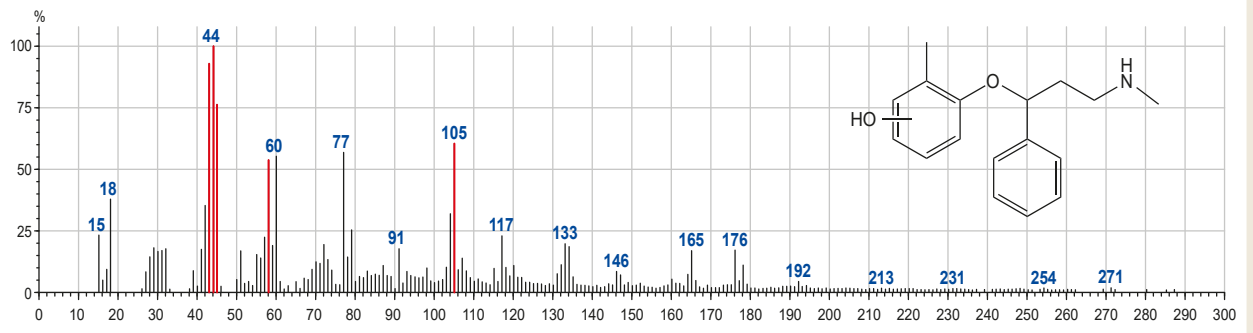

Fig. 2. Mass spectrum of hydroxyatomoxetine extracted from the tested urine samples.

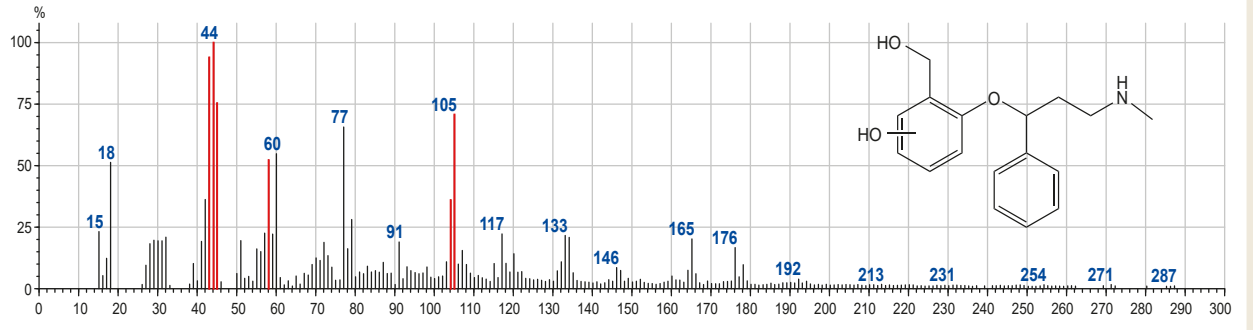

Fig. 3. Mass spectrum of dihydroxyatomoxetine extracted from the tested urine samples.

Table 2. Colour of the interaction products of atomoxetine and its metabolites with the chromogenic reagents

No.	Reagent	Atomoxetine (sensitivity, μg in spot) [13]	Hydroxy-atomoxetine	Dihydroxy-atomoxetine
1	UV light 254 nm/365 nm	fluorescence purple (0.3)/light yellow (0.5)	fluorescence purple/light yellow	fluorescence pink/light yellow
2	Dragendorff–Munier	orange (0.5)	orange	orange
3	Ninhydrin solution	pink-violet (2.0)	pink-violet	pink-violet
4	Mandelin	light-pink (5.0)	yellow	yellow
5	Mandelin followed by formaldehyde vapour	yellow (5.0)	yellow	yellow
6	Marquis	pink → light – pink (2.0)	pink → yellow	red
7	Froehde	blue (1.0)	blue → brown	green
8	Liebermann	Light – pink (10.0)	orange → pink	red → yellow
9	Erdmann	Light – yellow (10.0)	yellow → orange	orange
10	Sulphuric acid concentrated	pink → light – yellow (10.0)	pink	red

176 (17), 165 (20), 146 (9), 133 (22), 117(22), 105 (70), 91 (19), 77 (66), 60 (55), 44 (100) (*Fig. 2*). Found, m/z: 287.30 $[M]^+$. $C_{17}H_{21}NO_3$. Calculated, m/z: 287.4.

R_f values of atomoxetine, hydroxyatomoxetine, and dihydroxyatomoxetine extracted from the urine are shown in *Table 1*. Colour of the interaction products of atomoxetine and its metabolites with the range of chromogenic reagents are presented in *Table 2*.

Discussion

Sample preparation conditions were optimized on the basis of early obtained results of extraction yield of atomoxetine from aqueous solutions with organic solvents depending on pH and nature of salting-out agent [14]. The pre-treatment of the biological fluid using dilute acid hydrolysis was carried out because the drug is excreted mostly urinary as the major oxidative metabolite of 4-hydroxyatomoxetine-*O*-glucuronide (80 % of the administered dose) [2,3,12]. Urine samples were collected according to the literature half-life values of atomoxetine [3].

The obtained results of the identification of atomoxetine metabolites in the studied urine samples are consistent with the literature data: 4-hydroxyatomoxetine was found as the major atomoxetine oxidation product in the urine of persons with both an extensive and poor type of metabolism [15,16] and dihydroxyatomoxetine, namely, 2-hydroxymethyl-4-hydroxyatomoxetine was found for poor metabolizers [16].

The chromatographic mobility of atomoxetine and its metabolites was studied in 18 mobile phases including those proposed by TIAFT (No. 1–9) for general drug screening, and those widely used in forensic toxicological studies (No. 10–18) [3]. According to the TIAFT recommendations, using several chromatographic systems, a minimum of three, preferably with the low correlation of R_f values, significantly increases the reliability of substance identification by TLC [3]. As can be seen from *Table 1*, atomoxetine and its metabolites had low R_f values in mobile phases No. 1–9, only the phase of ethyl acetate–methanol–25 % ammonia (85:10:5) (No. 4) allowed to achieve a satisfactory separation of the native drug and metabolites. The chloroform – acetone – 25 % ammonia solution (25:5:0.3) mobile phase (No. 15) showed higher values of the chromatographic mobility of atomoxetine and its metabolites and a satisfactory separation. So, it was chosen for the TLC-purification in the current study. The mobile phase of chloroform – dioxane – acetone–25 % ammonia solution (47.5:45:5:2.5) (No. 11) also can be recommended for TLC study of the biological extracts containing atomoxetine and its metabolites as a low correlated one with the mobile phases No. 4 and 1 No. 5.

The colors selective with respect to biogenic matrix components were obtained with ninhydrin, Froehde, Liebermann, Erdman's, Marquies reagents, and concentrated sulphuric acid (*Table 2*). The sensitivity of the color reactions for the atomoxetine standard methanol solution was in the range of 0.5–10.0 μg in the spot [13]. This work is a contribution to the toxicological screening of atomoxetine using TLC. Obtained data can be used in the practice of forensic and clinical toxicology.

Conclusions

1. Atomoxetine and its biotransformation products were detected in the urine under TLC screening conditions and identified using mass spectrometry method.

2. Chromatographic mobility of the native compound, hydroxyatomoxetine and dihydroxyatomoxetine in the TLC screening systems as well as the results of their visualization using chromogenic reagents applied for toxicological drug screening in the systematic toxicological analysis have been determined.

Prospects for further research. The developed conditions for detection of atomoxetine and its metabolites in the urine under the conditions of TLC screening will be used for further creation of toxicological examination algorithm of biological samples for the presence of atomoxetine in the cases of lethal intoxications by antidepressants.

Funding
This work was performed in accordance with the plan of the scientific-research work of the National University of Pharmacy "Chemical-toxicological analysis of biologically active substances and drugs", the state registration code is 0114U000958.

Conflicts of interest: authors have no conflict of interest to declare.
Конфлікт інтересів: відсутній.

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References

[1] Childress, A. C. (2015). A critical appraisal of atomoxetine in the management of ADHD. *Therapeutics and Clinical Risk Management, 12*, 27-39. https://doi.org/10.2147/TCRM.S59270

[2] Baselt, C. R. (2020). *Disposition of Toxic Drugs and Chemicals in Man, 12-th edition.* Seal Beach, CA: Biomedical Publications.

[3] Moffat, A. C., Osselton, M. D., & Widdop, B. (2011). *Clarke's analysis of drugs and poisons in pharmaceuticals, body fluids and postmortem material,* 4th ed. London, Chicago: Pharmaceutical Press.

[4] Pilhatsch, M. K., Burghardt, R., Wandinger, K. -P., Bauer, M., & Adli, M. (2006). Augmentation with atomoxetine in treatment-resistant depression with psychotic features – a case report. *Pharmacopsychiatry, 39*(2), 79-80. https://doi.org/10.1055/s-2006-931547

[5] Cesneková, D., Šnircová, E., Nosáľová, G., & Ondrejka, I. (2016). Is atomoxetine effective in some comorbid mental disorders in ADHD? *European Pharmaceutical Journal, 63*(1), 29-32. https://doi.org/10.1515/afpuc-2016-0003

[6] Reed, V. A., Buitelaar, J. K., Anand, E., Day, K. A., Treuer, T., Upadhyaya, H. P., Coghill, D. R., Kryzhanovskaya, L. A., & Savill, N. C. (2016). The Safety of Atomoxetine for the Treatment of Children and Adolescents with Attention-Deficit/Hyperactivity Disorder: A Comprehensive Review of Over a Decade of Research. *CNS drugs*, *30*(7), 603-628. https://doi.org/10.1007/s40263-016-0349-0

[7] Howes, C. F., & Sharp, C. (2018). Delusional infestation in the treatment of ADHD with atomoxetine. *BMJ case repots, 2018*, bcr2018226020. https://doi.org/10.1136/bcr-2018-226020

[8] Başay, Ö., Basay, B. K., Öztürk, Ö., & Yüncü, Z. (2016). Acute Dystonia Following a Switch in Treatment from Atomoxetine to Low-dose Aripiprazole. *Clinical Psychopharmacology and Neuroscience, 14*(2), 221-225. https://doi.org/10.9758/cpn.2016.14.2.221

[9] Aronson, J. K. (2016). Atomoxetine. In *Meyler's Side Effects of Drugs* (16th ed., pp. 738-740.) Elsevier. https://www.sciencedirect.com/topics/medicine-and-dentistry/atomoxetine

[10] Paxton, G. A., & Cranswick, N. E. (2008). Acute suicidality after commencing atomoxetine. *Paediatrics and Child Health, 44*(10), 596-598. https://doi.org/10.1111/j.1440-1754.2008.01389.x

[11] Garside, D., Ropero-Miller, J. D., & Riemer, E. C. (2006). Postmortem tissue distribution of atomoxetine following fatal and nonfatal doses – three case reports. *Journal of Forensic Sciences, 51*(1), 179-182. https://doi.org/10.1111/j.1556-4029.2005.00021.x

[12] Yu, G., Li, G. -F., Markowitz, J. S. (2016). Atomoxetine – A Review of its Pharmacokinetics and Pharmacogenomics Relative to Drug Disposition. *Journal of Child and Adolescent Psychopharmacology, 26*(4), 314-326. https://doi.org/10.1089/cap.2015.0137

[13] Tomarovska, L. Yu., Baiurka, S. V., & Karpushina, S. A. (2017). Development of the methods for atomoxetine identification suitable for the chemical and toxicological analysis. *Vìsnik farmacìï – News of Pharmacy, 2*(90), 13-20. https://doi.org/10.24959/nphj.17.2154

[14] Tomarovska, L. Yu., Baiurka, S. V., & Karpushina, S. A. (2020). Study of Solvent extraction of Atomoxetine from Aqueous solutions and Biological fluids. *Research Journal of Pharmacy and Technology, 13*(9), 4303-4309. https://doi.org/10.5958/0974-360X.2020.00760.X

[15] Dinh, J. C., Pearce, R. E., Haandel, L. V., Gaedigk, A., & Leeder, J. S. (2016). Characterization of Atomoxetine Biotransformation and Implications for Development of PBPK Models for Dose Individualization in Children. *Drug Metabolism & Disposition, 44*(7), 1070-1079. https://doi.org/10.1124/dmd.116.069518

[16] Sauer, J. M., Ponsler, G. D., Mattiuz, E. L., Long, A. J., Witcher, J. W., Thomasson, H. R., & Desante, K. A. (2003). Disposition and metabolic fate of atomoxetine hydrochloride: the role of CYP2D6 in human disposition and metabolism. *Drug Metabolism & Disposition, 31*(1), 98-107. https://doi.org/10.1124/dmd.31.1.98