Antiepileptic Drugs

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For drugs, such as antiepileptic drugs (AEDs), whose therapeutic or toxic effects are more closely related to blood levels than to a specific dose, monitoring of plasma levels plays a crucial role. Many drugs used in epilepsy therapy often cause acute poisonings (carbamazepine, oxcarbazepine, valproic acid, lamotrigine). AEDs do not have an ideal pharmacokinetic profile, which at the same time qualifies them to monitor both in the therapeutic and toxic aspects. Currently, a great benefit for patients using various AEDs is adjusting the dosage to their individual needs and monitoring sufficient blood concentrations. There is still a need to develop new, rapid methods that meet the validation criteria. This trend has been observed in the last few years in the bioanalysis of different type of biological samples, not only blood, serum or plasma, but also saliva and blood/serum/plasma dried spots technique.

Keywords: epilepsy ; Antiepileptic drugs ; analytical methods ; therapeutic monitoring ; chromatography ; developing procedures ; quantitative determination

1. Introduction

About 70 million people worldwide suffer from epilepsy, a neurological disease that negatively affects patients' quality of life and that of their families. Antiepileptic drugs (AEDs), despite many scientists' and the medical community's efforts, are still the basic tool in the treatment of patients with epilepsy, a disease of unknown etiology. To date, about thirty first–third generation AEDs are used in various types of epilepsy. The new AED cenobamate (CNB) was approved by the US FDA in 2019 to reduce uncontrolled partial-onset seizures in adults [1][2]. AEDs possess a long tradition in the treatment of different modes of epilepsy and over the decades many analytical methods have been developed for the therapeutic monitoring of AEDs. More improved liquid chromatography (LC) methods have also appeared in the last years, based mainly on sensitive mass spectrum detection. Generally, the elaborated methods are focused on determining many AEDs and their metabolites in one analytical run. The main problem of recent years has been developing procedures for sample micronization and automatic preparation of clinical material for quantitative determination. Microextraction techniques like solid-phase microextraction (SPME), single-drop microextraction (SDME), or dispersive liquid-liquid microextraction (DLLME) were elaborated for the isolation of many medicines, including AEDs, from biological matrices.

Therapeutic Drug Monitoring (TDM) of AEDs involves measuring the drug concentrations in blood, serum, or plasma to improve the efficacy of applied treatment according to known rule if you can't measure it, you can't improve it. TDM was introduced in the late 1960s to minimize the toxicity effect caused by aminoglycosides. The main aim of TDM is to optimize drug exposure to minimize toxicities and maximize efficacy. The therapeutic tool is particularly essential when a drug that has a narrow therapeutic window and is characterized by a significant correlation between drug concentration and its toxicity, and when the clinical results depend on the drug level—total and/or free in the blood, not on the dose taken [3]. AEDs are monitored mainly due to the high inter-individual variability resulting from non-linear pharmacokinetics and the narrow therapeutic scope. New reference ranges were established for total and free concentrations of AED's [4]. Essentials are interactions of AEDs in the pharmacokinetic phase. Carbamazepine (CBZ), oxcarbazepine (OXC), vigabatrin (VGB) are indicated for epileptic patients with focal seizures. Clobazam (CLB) for myoclonic episodes and valproic acid (VPA) are dedicated to generalized epilepsy. Levetiracetam (LEV), lamotrigine (LTG), rufinamide (RFM), topiramate (TPM), and zonisamide (ZSM) with a broad spectrum of action are used for most types of seizures. They require monitoring also due to the rapid absorption process, short half-life or changing with dose (nonlinearity) and significant changes in concentrations during the day or the degree of binding with blood proteins [5].

Recommendations for TDM in the treatment of epilepsy are the following [5][6][7]:

- small differences between the toxic effect of the drug and the symptoms of the disease
- doses of the drug that do not relieve symptoms
- patient belongs to one of the groups at increased risk: Elderly, children, pregnant, patients with renal and hepatic dysfunction

- required changes in the dosage of a given drug when the patient is in weak condition
- the use of polytherapy of AED's drugs
- a drug with dose-dependent—non-linear pharmacokinetics.

Currently, a great benefit for patients using various AEDs is adjusting the dosage to their individual needs and monitoring the sufficient concentrations. To correctly interpret the results obtained during TDM, pharmacokinetics knowledge on absorption, distribution, biotransformation, drug excretion and an understanding of the physiological, pathophysiological and environmental factors affecting the therapy's success are necessary [6][8].

Initially in TDM colorimetric methods were used, and later more modern techniques were implemented, such as immunochemistry in the early 1980s, or LC. In the 2000s, the LC-MS method allowed for the quantification of free drug concentrations in biological matrices, mainly in plasma and interstitial fluids, with very sensitive detection [9]. The control of drug concentrations has provided a new perspective in the treatment in line with pharmacokinetic principles and clinical observations [7].

The matrix for TDM is often plasma or serum, rarely urine through, and increasingly unstimulated saliva, DBS or dried saliva spot (DSS). For a reliable test, the concentration of free drug in the blood has to be measured that reflects effective levels in the brain, heart tissue. In most clinical trials, the ratio between free and bounded fractions is constant at the equilibrium state. Therefore, a measurement of the total concentration (protein-bounded and free form) is acceptable in drugs with low protein binding. Such analysis is more comfortable, cheaper and requires less time expenditure [5][10].

Sample preparation is a crucial step during the whole process of the quantitative determination of the clinical samples. Simple protein precipitation or different extraction procedures before injection into LC of monitored analytes is equally important as a separation technique before detection. The most useful method for extraction includes liquid-solid, liquid-gas or liquid-liquid extraction, and mechanical separation. It is also essential to precisely determine the degree of reliability of a given method using statistical parameters known as the validation process. Validation determines the suitability of analytical methods based on the evaluation of validation parameters, like selectivity, recovery, calibration curve, determination of the linearity range, sensitivity, accuracy, precision, limit of quantification (LOQ), and limit of detection (LOD). Designed for the TDM bioanalytical method has to fulfill validation criteria established mainly by the European Medicines Agency (EMA) [11] or the Food Drug Administration (FDA) [12].

2. Bioanalytical Methods

Methods that meet the validation criteria, including high sensitivity and selectivity, are the basis for TDM, and finally, for effective and safe pharmacotherapy. This statement also applies to AEDs, for which monitoring of drug concentrations becomes the rule nowadays, not only in terms of total concentration (VPA, PHT, CBZ, GBP, LTG, lithium, TPM, LEV) but also of unbound fractions, especially for those drugs with a high (90%) or even higher protein binding rate. Free concentrations of many AEDs including PHT, CBZ, and VPA are in some laboratories routinely determined. An increase in TDM is expected for the most recent AEDs for which the therapeutic concentration ranges have been established. This concerns BRV (0.2–2 mg/L), PER (0.1–1 mg/L), STM (2–10 mg/L) and STP (4–22 mg/L) [4]. For other AEDs: ESL, LTG, OXC, PGB, TPM, and VPA, the reference ranges have been updated or harmonized. It can be expected that free fraction will be determined for other AEDs: CLB, and its active metabolite N-CLB, PER, TGB, retigabine, STP, medicines with high protein binding [13].

There is still a need to develop new, rapid methods that meet the validation criteria. This trend has been observed in the last few years in the bioanalysis of AEDs, where LC-MS/MS is a dominated technique; however, it is costly. Data presented in the review show that near 50% of the LC methods applied the high-resolution detection despite the technical problems with the stability of MS/MS response. The sensitivity connected with the high-resolution LC-MS/MS causes that deuterated analogues of the AEDs are used as effective internal standards even with very small differences in molecular mass as well as retention time [14][15][16][17][18][19][20][21][22][23][24][25][26][27][28]. There is still an increase in the full automatization of analytical determination of AEDs in order to decrease hands-on time as well as consumption of organic solvents, protect the natural environment and improve the analytical process in terms of accuracy, precision, repeatability, and sensitivity. To realize the purpose also different microextraction techniques for AEDs analysis were developed, including ultrasound-assisted emulsification microextraction [29], MEPS [30][31][32], microextraction combined with micro-derivatization-increased detection (MDID) [17], DLLME [33][34]. Automatization was applied for the DBS extraction of VPA, PHB, PHT, CBZ and its active 10,11 epoxide [14]. The effective online SPE coupled with an analytical column of LC-HRMS system was applied for quantification of the first generation of AEDs: PHB, PHT and CBZ and its active metabolite in clinical samples [35]. The interesting idea seems to be online extraction using RACNTs for analysis of CBZ, PHB and PRM, previously used only for extraction cadmium and lead [36].

However, traditional protocols with steps involving sample preparation using traditional LLE and newer SALLE [37], as well as protein precipitation, are frequently applied in the bioanalysis of AEDs, more often than advanced micro-extraction techniques.

Among the presented LC methods, the columns with different length, dimensions and particle sizes based on popular lipophilic chain C-18 are very often applied. Generally, in LC-MS/MS and UHPLC-MS/MS, the columns are characterized by dimensions of 2–3 mm, and smaller than 5 µm particle size (1.7–3.5 µm). The other types of LC columns, like those containing biphenyl phase [38], chiral column Chiracel with normal phase [39], or columns with cyano stationary phase (polar phase) [40] were rarely used in quantification of AEDs. Less popular methods like MEKC gained attention for TDM of PIR [41], capillary electrophoresis for TPM and PRM [42][43]. Worth mentioning is that HPTLC, not so useful in TDM was also developed for determination of OXC and ESL in biological samples [37]. The GC-MS technique loses its importance compared to LC-MS/MS, although it has been developed in recent years to determine TPR [44], GBP [24], LCM [45][28].

Moreover, alternative specimen sampling is proposed employing non-invasive and patient-friendly techniques, including DBS or collection of saliva. These alternative specimens, appropriate for TDM, are especially valuable in specific clinical situations involving pediatric patients or critically ill patients.

References

1. XCOPRI. Full Prescribing Information. Available online: https://www.accessdata.fda.gov/drugsatfda_docs/label/2019/212839s000lbl.pdf (accessed on 13 July 2020).
2. Cenobamate (XCOPRI). Clinical Pharmacology and Biopharmaceutics Review(s); Application Number 212839Orig1s000. Available online: https://www.accessdata.fda.gov/drugsatfda_docs/nda/2019/212839Orig1s000ClinPharmR.pdf (accessed on 13 July 2020).
3. Cusumano, J.A.; Klinker, K.P.; Huttner, A.; Luther, M.K.; Roberts, J.A.; LaPlante, K.L. Towards precision medicine: Therapeutic drug monitoring–guided dosing of vancomycin and β-lactam antibiotics to maximize effectiveness and minimize toxicity. Am. J. Health Syst. Pharm. 2020, 77, 1104–1112.
4. Reimers, A.; Berg, J.A.; Burns, M.L.; Brodtkorb, E.; Johannessen, S.I.; Johannessen Landmark, C. Reference ranges for antiepileptic drugs revisited: A practical approach to establish national guidelines. DDDT 2018, 12, 271–280.
5. Landmark, C.J.; Johannessen, S.I.; Patsalos, P.N. Therapeutic drug monitoring of antiepileptic drugs: Current status and future prospects. Expert Opin. Drug Metab. Toxicol. 2020, 16, 227–238.
6. Serragui, S.; Lachhab, Z.; Tanani, D.S.; Cherrah, Y. Therapeutic Drug Monitoring of Antiepileptic Drugs: Indications and Modalities. J. Pharm. Pharmacol. Res. 2019, 3, 41–50.
7. Knezevic, C.E.; Marzinke, M.A. Clinical Use and Monitoring of Antiepileptic Drugs. J. Appl. Lab. Med. 2018, 3, 115–127.
8. Perucca, E.; French, J.; Bialer, M. Development of new antiepileptic drugs: Challenges, incentives, and recent advances. Lancet Neurol. 2007, 6, 793–804.
9. Reeves, D.; Lovering, A.; Thomson, A. Therapeutic drug monitoring in the past 40 years of the Journal of Antimicrobial Chemotherapy. J. Antimicrob. Chemother. 2016, 71, 3330–3332.
10. Patsalos, P.N.; Berry, D.J.; Bourgeois, B.F.D.; Cloyd, J.C.; Glauser, T.A.; Johannessen, S.I.; Leppik, I.E.; Tomson, T.; Perucca, E. Antiepileptic drugs best practice guidelines for therapeutic drug monitoring: A position paper by the subcommission on therapeutic drug monitoring, ILAE Commission on Therapeutic Strategies. Epilepsia 2008, 49, 1239–1276.
11. European Medicines Agency Guideline on bioanalytical method validation. Available online: https://www.ema.europa.eu/en/documents/scientific-guideline/guideline-bioanalytical-method-validation_en.pdf (accessed on 16 August 2020).
12. U.S. Department of Health and Human Services Food and Drug Administration; Center for Drug Evaluation and Research (CDER); Center for Veterinary Medicine (CVM). Bioanalytical Method Validation: Guidance for Industry. Available online: https://www.fda.gov/files/drugs/published/Bioanalytical-Method-Validation-Guidance-for-Industry.pdf (accessed on 16 August 2020).
13. Patsalos, P.N.; Zugman, M.; Lake, C.; James, A.; Ratnaraj, N.; Sander, J.W. Serum protein binding of 25 antiepileptic drugs in a routine clinical setting: A comparison of free non-protein-bound concentrations. Epilepsia 2017, 58, 1234–1243.

14. Velghe, S.; Deprez, S.; Stove, C.P. Fully automated therapeutic drug monitoring of anti-epileptic drugs making use of dried blood spots. J. Chromatogr. A 2019, 1601, 95–103.
15. Velghe, S.; Stove, C.P. Volumetric absorptive microsampling as an alternative tool for therapeutic drug monitoring of first-generation anti-epileptic drugs. Anal. Bioanal. Chem. 2018, 410, 2331–2341.
16. D'Urso, A.; Cangemi, G.; Barco, S.; Striano, P.; D'Avolio, A.; de Grazia, U. LC-MS/MS-Based Quantification of 9 Antiepileptic Drugs from a Dried Sample Spot Device. Ther. Drug Monit. 2019, 41, 331–339.
17. Wu, Y.-J.; Li, Y.-S.; Tseng, W.-L.; Lu, C.-Y. Microextraction combined with microderivatization for drug monitoring and protein modification analysis from limited blood volume using mass spectrometry. Anal. Bioanal. Chem. 2018, 410, 7405–7414.
18. Dao, K.; Thoueille, P.; Decosterd, L.A.; Mercier, T.; Guidi, M.; Bardinet, C.; Lebon, S.; Choong, E.; Castang, A.; Guittet, C.; et al. Sultiame pharmacokinetic profile in plasma and erythrocytes after single oral doses: A pilot study in healthy volunteers. Pharm. Res. Perspect. 2020, 8.
19. Linder, C.; Neideman, M.; Wide, K.; von Euler, M.; Gustafsson, L.L.; Pohanka, A. Dried Blood Spot Self-Sampling by Guardians of Children with Epilepsy Is Feasible: Comparison With Plasma for Multiple Antiepileptic Drugs. Ther. Drug Monit. 2019, 41, 509–518.
20. Deeb, S.; McKeown, D.A.; Torrance, H.J.; Wylie, F.M.; Logan, B.K.; Scott, K.S. Simultaneous Analysis of 22 Antiepileptic Drugs in Postmortem Blood, Serum and Plasma Using LC–MS-MS with a Focus on Their Role in Forensic Cases. J. Anal. Toxicol. 2014, 38, 485–494.
21. Milosheska, D.; Roškar, R. A novel LC–MS/MS method for the simultaneous quantification of topiramate and its main metabolites in human plasma. J. Pharm. Biomed. Anal. 2017, 138, 180–188.
22. De La Vega, H.; Fox, K.; Pardi, J.; Santiago-Tirado, W.; Cooper, G. Validation of a High-throughput Screening and Quantification Method for the Determination of Gabapentinoids in Blood Using a Combination of LC-TOF-MS and LC-MS-MS. J. Anal. Toxicol. 2019, 43, 696–702.
23. Nahar, L.; Smith, A.; Patel, R.; Andrews, R.; Paterson, S. Validated Method for the Screening and Quantification of Baclofen, Gabapentin and Pregabalin in Human Post-Mortem Whole Blood Using Protein Precipitation and Liquid Chromatography–Tandem Mass Spectrometry. J. Anal. Toxicol. 2017, 41, 441–450.
24. Sadones, N.; Van Bever, E.; Van Bortel, L.; Lambert, W.E.; Stove, C.P. Dried blood spot analysis of gabapentin as a valid alternative for serum: A bridging study. J. Pharm. Biomed. Anal. 2017, 132, 72–76.
25. Dwivedi, J.; Namdev, K.K.; Chilkoti, D.C.; Verma, S.; Sharma, S. An Improved LC-ESI-MS/MS Method to Quantify Pregabalin in Human Plasma and Dry Plasma Spot for Therapeutic Monitoring and Pharmacokinetic Applications. Ther. Drug Monit. 2018, 40, 610–619.
26. Duhamel, P.; Ounissi, M.; Le Saux, T.; Bienayme, H.; Chiron, C.; Jullien, V. Determination of the R (−) and S (+)-enantiomers of vigabatrin in human plasma by ultra-high-performance liquid chromatography and tandem mass-spectrometry. J. Chromatogr. B 2017, 1070, 31–36.
27. Korman, E.; Langman, L.J.; Jannetto, P.J. High-Throughput Method for the Quantification of Lacosamide in Serum Using Ultrafast SPE-MS/MS. Ther. Drug Monit. 2015, 37, 126–131.
28. Nikolaou, P.; Papoutsis, I.; Spiliopoulou, C.; Voudris, C.; Athanaselis, S. A fully validated method for the determination of lacosamide in human plasma using gas chromatography with mass spectrometry: Application for therapeutic drug monitoring. J. Sep. Sci. 2015, 38, 260–266.
29. Bahmaei, M.; Khalilian, F.; Mashayekhi, H.A. Determination of Carbamazepine in Biological Samples Using Ultrasound-Assisted Emulsification Micro-extraction and Gas Chromatography. J. Chem. Health Risks 2015, 5.
30. Ferreira, A.; Rodrigues, M.; Oliveira, P.; Francisco, J.; Fortuna, A.; Rosado, L.; Rosado, P.; Falcão, A.; Alves, G. Liquid chromatographic assay based on microextraction by packed sorbent for therapeutic drug monitoring of carbamazepine, lamotrigine, oxcarbazepine, phenobarbital, phenytoin and the active metabolites carbamazepine-10,11-epoxide and licarbazepine. J. Chromatogr. B 2014, 971, 20–29.
31. Fortuna, A.; Sousa, J.; Alves, G.; Falcão, A.; Soares-da-Silva, P. Development and validation of an HPLC-UV method for the simultaneous quantification of carbamazepine, oxcarbazepine, eslicarbazepine acetate and their main metabolites in human plasma. Anal. Bioanal. Chem. 2010, 397, 1605–1615.
32. Lourenço, D.; Sarraguça, M.; Alves, G.; Coutinho, P.; Araujo, A.R.T.S.; Rodrigues, M. A novel HPLC method for the determination of zonisamide in human plasma using microextraction by packed sorbent optimised by experimental design. Anal. Methods 2017, 9, 5910–5919.
33. Feriduni, B.; Farajzadeh, M.A.; Jouyban, A. Determination of Two Antiepileptic Drugs in Urine by Homogenous Liquid-Liquid Extraction Performed in A Narrow Tube Combined with Dispersive Liquid-liquid Microextraction Followed by Gas

Chromatography-flame Ionization Detection. Iran. J. Pharm. Res. 2019.

34. Fazeli-Bakhtiyari, R.; Panahi-Azar, V.; Sorouraddin, M.H.; Jouyban, A. Determination of valproic acid in human plasma using dispersive liquid-liquid microextraction followed by gas chromatography-flame ionization detection. Iran J. Basic Med. Sci. 2015, 18, 979–988.
35. Qu, L.; Fan, Y.; Wang, W.; Ma, K.; Yin, Z. Development, validation and clinical application of an online-SPE-LC-HRMS/MS for simultaneous quantification of phenobarbital, phenytoin, carbamazepine, and its active metabolite carbamazepine 10,11-epoxide. Talanta 2016, 158, 77–88.
36. dos Santos, R.C.; Kakazu, A.K.; Santos, M.G.; Belinelli Silva, F.A.; Figueiredo, E.C. Characterization and application of restricted access carbon nanotubes in online extraction of anticonvulsant drugs from plasma samples followed by liquid chromatography analysis. J. Chromatogr. B 2017, 1054, 50–56.
37. Mohamed, F.A.; Ali, M.F.B.; Rageh, A.H.; Mostafa, A.M. A highly sensitive HPTLC method for estimation of oxcarbazepine in two binary mixtures with two metabolically related antiepileptic drugs: Application to pharmaceutical and biological samples. Microchem. J. 2019, 146, 414–422.
38. Mikayelyan, A.; Aleksanyan, A.; Sargsyan, M.; Gevorgyan, A.; Zakaryan, H.; Harutyunyan, A.; Zhamharyan, L.; Armoudjian, Y.; Margaryan, T. Protein precipitation method for determination of c lobazam and N-desmethylclobazam in human plasma by LC–MS/MS. Biomed. Chromatogr. 2020, 34.
39. Loureiro, A.I.; Fernandes-Lopes, C.; Wright, L.C.; Soares-da-Silva, P. Development and validation of an enantioselective liquid-chromatography/tandem mass spectrometry method for the separation and quantification of eslicarbazepine acetate, eslicarbazepine, R-licarbazepine and oxcarbazepine in human plasma. J. Chromatogr. B 2011, 879, 2611–2618.
40. Ni, Y.; Zhou, Y.; Xu, M.; He, X.; Li, H.; Haseeb, S.; Chen, H.; Li, W. Simultaneous determination of phentermine and topiramate in human plasma by liquid chromatography–tandem mass spectrometry with positive/negative ion-switching electrospray ionization and its application in pharmacokinetic study. J. Pharm. Biomed. Anal. 2015, 107, 444–449.
41. Yeh, H.-H.; Yang, Y.-H.; Ko, J.-Y.; Chen, S.-H. Rapid determination of piracetam in human plasma and cerebrospinal fluid by micellar electrokinetic chromatography with sample direct injection. J. Chromatogr. A 2006, 1120, 27–34.
42. Ishikawa, A.A.; da Silva, R.M.; Santos, M.S.F.; da Costa, E.T.; Sakamoto, A.C.; Carrilho, E.; de Gaitani, C.M.; Garcia, C.D. Determination of topiramate by capillary electrophoresis with capacitively-coupled contactless conductivity detection: A powerful tool for therapeutic monitoring in epileptic patients. Electrophoresis 2018, 39, 2598–2604.
43. Tůma, P.; Bursová, M.; Sommerová, B.; Horsley, R.; Čabala, R.; Hložek, T. Novel electrophoretic acetonitrile-based stacking for sensitive monitoring of the antiepileptic drug perampanel in human serum. J. Pharm. Biomed. Anal. 2018, 160, 368–373.
44. Hahn, R.Z.; Antunes, M.V.; Costa Arnhold, P.; Andriguetti, N.B.; Verza, S.G.; Linden, R. Determination of topiramate in dried blood spots using single-quadrupole gas chromatography–mass spectrometry after flash methylation with trimethylanilinium hydroxide. J. Chromatogr. B 2017, 1046, 131–137.
45. Mouskeftara, T.; Alexandridou, A.; Krokos, A.; Gika, H.; Mastrogianni, O.; Orfanidis, A.; Raikos, N. A Simple Method for the Determination of Lacosamide in Blood by GC-MS. J. Forensic Sci. 2020, 65, 288–294.
