DEVELOPMENT AND VALIDATION OF A RAPID AND SIMPLE UPLC/MS METHOD FOR QUANTIFICATION OF THERAPEUTIC AND TOXIC VALPROIC ACID CONCENTRATIONS IN PATIENT PLASMA

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Valproic acid (VPA) is antiepileptic drug with a long history of clinical use, increasingly applied in the treatment of various diseases (bipolar disorder, migraine prophylaxis, etc). Due to significant inter-individual differences VPA concentration is often measured in the purpose of therapeutic drug monitoring (TDM) (1). Acute poisonings with this drug represent a significant problem. According to the American Association of Poison Control Centers, there has been VPA poisoning incidence increase in the last 20 years. Reports from the National Poison Control Center in Belgrade list VPA as one of the three most common causes of antiepileptic drugs intoxications (2). This paper aimed to develop a modern analytical method for VPA quantification in patient plasma. UPLC/MS method with fast and simple sample preparation has been validated. After protein precipitation, VPA was extracted from 100 μL plasma using HLB cartridges. Caprylic acid solution was used as internal standard. Chromatographic separation was achieved on C18 column (1.8 μm, 2.1 X 150 mm) with gradient elution at constant mobile phase flow. Good peak resolution was achieved (Rt_{VPA} - 4.73 min, Rt_{ISTD} - 4.94 min). Validation was performed according European Medicines Agency recommendations. Based on statistical analysism it was shown that the method is precise, accurate, specific, sensitive and linearity is confirmed in a wide range of concentrations (1-250 mg / L). Advantages of this method are simple preparation procedure from a small amount of sample, without prior derivatization and short duration of analysis, which fully satisfies the needs of TDM and urgent toxicological analyses.

References

1. Forooghipour M et al.Therapeutic Drug Monitoring of Valproic Acid in Patients with Monotherapy at Steady State. Iranian Journal of Basic Medical Sciences, 2009; 12(3): 146-149.
2. Gummin D et al. 2016 Annual Report of the American Association of Poison Control Centers’ National Poison Data System (NPDS): 34th Annual Report. Clinical Toxicology. 2017; 55(10):1072-1254.

RAZVOJ I VALIDACIJA BRZE I JEDNOSTAVNE UPLC/MS METODE ZA ODREĐIVANJE TERAPIJSKIH I TOKSIČNIH KONCENTRACIJA VALPROINSKE KISELINE U PLAZMI PACIJENATA

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Valproinska kiselina (VK) je lek iz grupe antiepileptika sa dugom istorijom kliničke upotrebe, koji se, osim u terapiji epilepsije, poslednjih decenija sve više koristi u terapiji različitih bolesti (bipolarni poremećaj, profilaksa migrene itd). Zbog značajnih interindividualnih varijacija, koncentracija VK se često određuje u svrhe terapijskog praćenja leka (TDM – *Therapeutic Drug Monitoring*) (1). S druge strane, akutna trovanja ovim lekom predstavljaju značajan problem. Prema podacima Američkih centara za kontrolu trovanja došlo je do povećanja incidence trovanja VK u poslednjih 20 godina, dok izveštaji Nacionalnog centra za kontrolu trovanja u Beogradu navode VK kao jedan od tri najčešća uzročnika trovanja kada su u pitanju intoksikacije antiepilepticima (2). Cilj ovoga rada je razvoj savremene analitičke metode za određivanje koncentracije VK u plazmi pacijenata. Validovana je UPLC/MS metoda koja podrazumeva brzu i jednostavnu pripremu uzorka. Nakon precipitacije proteina, izvršena je ekstrakcija iz 100 µL plazme uz pomoć HLB kertridža. Kao interni standard korišćen je rastvor kaprilne kiseline. Hromatografsko razdvajanje je postignuto na C18 koloni (1,8 µm, 2,1 X 150 mm) gradijentnim eluiranjem pri konstantnom protoku mobilne faze. Postignuta je dobra rezolucija pikova (Rt_{VK} - 4,73 min, Rt_{ISTD} - 4,94 min). Validacija je izvedena prema preporukama Evropske agencije za lekove, a na osnovu statističke analize je pokazano da je metoda precizna, tačna, specifična, osetljiva i potvrđena je linearnost u širokom opsegu koncentracija (1-250 mg/L). Prednosti ove metode su jednostavan način pripreme iz male količine uzorka, bez prethodne derivatizacije, i kratko vreme trajanja analize, što u potpunosti zadovoljava potrebe TDM-a i urgentnih toksikoloških analiza.

Literatura

1. Forooghipour M et al.Therapeutic Drug Monitoring of Valproic Acid in Patients with Monotherapy at Steady State. Iranian Journal of Basic Medical Sciences, 2009; 12(3): 146-149.
2. Gummin D et al. 2016 Annual Report of the American Association of Poison Control Centers' National Poison Data System (NPDS): 34th Annual Report. Clinical Toxicology. 2017; 55(10):1072-1254.