Effect of Genetic Polymorphism on the Clinical Response to Valproate Therapy in Epileptic Children

Thesis

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AAN	American Academy of Neurology.
ABCB1	ATP-Binding Cassette, Sub-Family B, Member 1
ADR	Adverse Drug Reaction
AEDs	Antiepileptic Drugs
AUC	Area Under the Plasma Concentration-Time Curve
BID	bis in die, twice a day
CDR	Concentration to Dose Ratio
CT	Computerized Tomography
CYP P450	Cytochrome P450
DNTP	Deoxynucleotide Triphosphate
EDTA	Ethylenediaminetetraacetic Acid
EEG	Electroencephalography
EPC	Epilepsia Partialis Continua
FLAIR	Fluid-Attenuated Inversion Recovery
G6PDH	Glucose-6-Phosphate Dehydrogenase
GABA-A	γ-Aminobutyric Acid, Subunit A
GCSE	Generalized Convulsive Status Epilepticus
ILAE	International League Against Epilepsy
LD	Linkage Disequilibrium
MDR	Multi-Drug Resistance Gene
MRI	Magnetic Resonance Imaging
NAD	Nicotinamide Adenine Dinucleotide
NICE	The National Institute For Clinical Excellence In The United Kingdom.
PCR	Polymerase Chain Reaction
PCR-RFLP	Polymerase Chain Reaction-Restriction Fragment Length Polymorphism
PT	Prothrombin Time
PTT	Partial Thromboplastin Time
SCN1A	Sodium Channel Gene
SD	Standard Deviation
SIGN	Scottish Intercollegiate Guidelines Network.
SMA	Supplementary Motor Area
SNP	Single Nucleotide Polymorphism

TAE	Tris-Acetate-EDTA
TIA	Transient Ischemic Attack
TID	ter in die, three times a day
TNF	Tumor Necrosis Factor
UDGPA	Uridine 5´-Diphosphate-Glucuronic Acid
UGT	Uridine Diphosphate Glucuronosyltransferase
VPA	Valproic Acid

Abstract

Background/objective: Valproic acid (VPA) is widely used in pediatric epilepsy. It is mainly eliminated through conjugation by UDP-glucuronosyltransferases which are known to be polymorphic. The aim of the study was to investigate the effect of UGT1A6 polymorphism at 541A>G and 552A>C loci on the serum level of VPA and overall clinical response. Seizure control and incidence of adverse drug reactions (ADR) were investigated in a cohort of Egyptian children with idiopathic epilepsy.

Methods: Genetic polymorphisms were detected in 48 patients receiving VPA monotherapy by PCR-RFLP. Steady state concentrations at trough level were determined by homogenous enzyme immunoassay technique. All patients were monitored for seizure frequency and seizure severity (SS) using Chalfont SS scale as well as for ADRs.

Results: Patients of variant genotype group (AC & CC) had lower concentration dose ratios (CDRs) than those with (AA) genotype for UGT1A6 552A>C (p=0.029). For both UGT1A6 541A>G and 552A>C, the median CDR of variant allele carriers was significantly lower than wild-type allele carriers (p= 0.047 and p= 0.001, respectively). Higher SS scores on Chalfont scale were associated with (AA) genotype of UGT1A6 552A>C than variant genotypes group (AC & CC) (p=0.020). No significant effect was detected on seizure control, while fatigue and cognitive adverse effects were significantly higher in wild genotype group of UGT1A6 552A>C and variant genotype group of UGT1A6 541 A>G, respectively.

Conclusion: UGT1A6 polymorphisms at 541A>G and 552A>C loci may be associated with increased VPA metabolism in Egyptian epileptic children. The severity of seizures as well as susceptibility to certain ADRs may be linked to the presence of certain UGT1A6 genotypic variants.

Introduction

Epilepsy is a chronic neurological condition manifesting as recurrent, unprovoked epileptic seizures. It is one of the most common chronic neurological diseases, with an estimated 50 million people affected worldwide ***(Duncan et al., 2006)***. Epilepsy is an extremely heterogeneous disorder, comprising a large spectrum of different seizure and syndrome types with multiple underlying etiologies ***(Bhalla et al., 2011)***.

Antiepileptic drugs (AED) are the primary form of treatment for seizures and epilepsy, while brain surgery and vagal nerve stimulation are reserved for selected refractory cases ***(Elger and Schmidt, 2008)***. Pharmacotherapy is fraught with problems mainly related to the unpredictability of efficacy, adverse drug reactions (ADRs) and optimal dosing in individual patients ***(Depondt, 2008)***.

The choice of drug and initial dosing is mainly based on factors such as epilepsy type, patient's age, gender, co-medication, and concomitant diseases ***(Elger and Schmidt, 2008)***. Further dose adjustments are based on seizure frequency and occurrence of ADRs. This may consume a considerable time of trial and error before an acceptable balance is achieved between efficacy and toxicity ***(Berg and Chadwick, 2000; Loscher et al., 2009)***.

AED efficacy, toxicity and dosing are influenced by multiple factors including environmental factors, patient-related factors, as well as genetic factors ***(Evans and McLeod, 2003)***. Identification of genetic factors influencing AED response could enable prediction of response in individual patients which is the essense of the pharmacogenetic studies. This could lead to more rapid seizure control with fewer ADRs and thus to an improved quality of life for patients with epilepsy ***(Loscher et al., 2009)***.

Genetic association studies are currently the most widely used approach where correlations between genetic variants and phenotypical differences are assessed on a population scale, to identify genetic variations contributing to variable drug responses ***(Goldstein et al., 2003)***. Increased knowledge of genetic associations with drug response is becoming more available making it possible that genetic information could be commonly used to guide drug therapy decisions in the near future ***(Johnson, 2013)***.

Clinical pharmacists have an established role in the health care setting, including education for both patients and providers, selecting and monitoring drug therapies for individual patients, ensuring safe and appropriate use of medications in populations, as well as conducting clinical research ***(El-Ibiary et al., 2008)***. Pharmacogenetic information serves

as a potential tool providing unique opportunities for clinical pharmacists to expand these roles, thereby, optimizing drug treatment for individual patients ***(Brock et al., 2003)***.

Valproic acid (VPA) is one of the major antiepileptic drugs with high efficacy against multiple seizure types, including both primarily generalized and partial seizure in adults and children ***(Loscher, 2002)***. Serious adverse effects, despite being rare, including fatal hepatotoxicity, acute pancreatitis, encephalopathy as well as bone-marrow suppression, have been associated with VPA treatment ***(Gerstner et al., 2007)***.

Therapy with VPA is clinically complicated by high interindividual variability in both pharmacokinetics and pharmacodynamics giving rise to a wide dosing range and therapeutic plasma level of 50-100 mg/l which necessitates its blood level monitoring during therapy ***(Jiang and Wang, 2004; Ferraro and Buono, 2005)***.

Glucuronidation and β-oxidation are the principal pathways of VPA metabolism, with glucuronidation reaching up to 50% of the total metabolism of the initial dose ***(Ito et al., 1990; Argikar and Remmel, 2009)***. Glucuronidation involves the conjugation of a glucuronic acid moiety to a range of functional groups of a specific substrate increasing their polarity which facilitates their excretion in bile or urine ***(Guillemette, 2003)***. It is carried out by Uridine diphosphate glucuronosyltransferase (UGTs), a superfamily of enzymes (>16) expressed on the inner membrane of the endoplasmic reticulum and are categorized into three subfamilies: UGT1A, UGT2A, and UGT2B ***(Mackenzie et al., 1997)***. Glucuronidation of VPA has been reported to be carried out by UGT1A3, UGT1A6, UGT1A9, UGT2B7, and UGT2B15 **(*Ethell et al., 2003; Argikar and Remmel, 2009)***.

UGT1A6 gene was demonstrated to be highly polymorphic with at least four alleles characterized by three single nucleotide polymorphisms (SNPs) including polymorphisms rs2070959 (541A>G) and rs1105879 (552A>C), rs6759892 (19T>G) in the coding sequence which can lead to both transcriptional and functional changes of its encoded enzymes ***(Ciotti et al., 1997; Nagar et al., 2004)***. Genetic polymorphisms in UGT1A6 are highly prevalent in different racial populations and the frequency of some variant alleles has shown different distribution between Caucasians and Asians ***(Lampe et al., 1999)***.

Studies investigating the clinical implications of these variant alleles are of great importance for clinical practice as they help assess the required dose of VPA in different populations.

Few studies are available on the effect of UGT1A6 gene polymorphisms on the levels of VPA in epileptic patients. All of them were carried out in Asian populations of different ethnic backgrounds; moreover, the results generated from these studies were not entirely