The Effect of Polymorphism on the CYP2B6 ,GABRE and ABCB1 Genes on the Pharmacodynamics of Propofol during General Anaesthesia for Abdominal Hysterectomy

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ABSTRACT

The completion of the human genome project and the development of new technologies for high-throuput genetic analyses has allowed for establishment of the concept of personalized medicine, with special emphasis of individualized therapy. The view that genetic information transforms the utilization of medicinal drugs is getting a wider acceptance among medical community at large. Pharmacogenetics is defined as “the study of genetic variations affecting drug response”. Genetic variation can influence a drug’s pharmacokinetics, pharmacodynamics and therapeutic outcomes.

Propofol (2,6 – disopropylphenol) is intravenously administered for the induction of the maintenance of anesthesia. Propofol was introduced into clinical practice as a general anesthetic agent in 1977 and has become the agent of choice for rapid intravenous induction. This drug is metabolized in the liver by the cytochrome P450 superfamily enzymes (CYPs) and phase II drug-metabolizing enzymes. Important genes associated with metabolism for propofol are as follows: GABRG2, UGT1A9, CYP2C9, GSTP1, SULT1A1 and NQO1.[1]

In clinical practice a large interindividual variability in response to propofol is observed. Studies have demonstrated that individual differences in genetic factors (polymorphisms in selected genes responsible for pharmacokinetics and pharmacodynamics) and nongenetic factors (sex, weight and height) contributed to the variability in dose requirements of propofol.[2;3]

Cytochrome P450 family (CYP450), ATP-binding cassette (ABCB1), serine/threonine-protein kinase 3 (TAOK3), family with sequence similarity 53 member B (FAM53B), and the cannabinoid receptor (CNR1) are postulated to be involved in propofol pharmacokinetics; opioid receptors (OPRM1 and OPRD1), β-adrenoceptor (ADRB1), Catechol-O-methyltransferase (COMT), and ligand-gated ion channel (P2RX7) are postulated to be directly or indirectly involved in the pharmacodynamic response to propofol; nitric oxide synthase (NOS3), GABA type A (GABAA) receptor, NMDA receptors (GR1N3A and GR1N2B), Galanin (GAL), fatty acid amide hydrolase (FAAH), 5-hydroxytryptamine receptor (5HT2A), cholinergic receptors (CHRM2 and CHRNA5), dopamine transporters (DAT and DRD2), casein kinase (CSNK1E), calcium channels, potassium channels (KCNS1 and GIRK) and sodium channels (SCN9A) are also likely involved in the action of propofol. [(4)]

Susceptibility to propofol anesthesia has been shown to be remarkably variable based on clinical observations of responses of patients of the same ethnic origin, and this variability is reflected in different dose requirements and the amount of required recovery time.

In order to understand the reason for this interindividual variability in the clinical response to propofol, we decide to investigate the impact of single-nucleotide polymorphisms (SNP) in CYP2B6, GABRE and ABCB1 on variations seen in clinical response to propofol.

CYP2B6 is one of the most polymorphic CYP genes in humans and variants have been shown to affect transcriptional regulation, splicing, mRNA and protein expression, and catalytic activity. Some variants appear to affect several functional levels simultaneously, thus, combined in haplotypes, leading to complex interactions between substrate-dependent and -independent mechanisms. The c.516>GT (rs3745274, Gln172His) polymorphism causes an altered splicing site that results in the loss of exon 4 to 6, leading to a severe reduction in the normal transcript and active protein levels.[(5)] The hydroxylation reaction of propofol produces 4-hydroxypropofol, which is responsible for 1/3rd hypnotic activity of propofol. In the context of propofol

response the effect this polymorphism was proved to be substrate-specific and usually led to a disturbed gene expression.

GABRA1 gamma-aminobutyric acid type A receptor alpha1 subunit is the specific target of nonbenzodiazepine hypnotic agents and is responsible for their hypnotic and hallucinogenic effects. The GABRA1 gene mutation leads to the formation of an abnormal α1 subunit that reduces $GABA_A$ receptor function. $GABA_A$ receptors containing the abnormal subunit are broken down before they reach the cell membrane. Based on our study, the G-to-A mutation in rs2279020 in GABAA1 may change the pharmacological properties of the receptor by varying the composition and arrangement of subunits. Under propofol anesthesia, the minor A allele of rs2279020 in GABAA1 may induce a stronger inhibition in the brain, as shown by the higher Bispectral Index BIS in those patients after the loss of consciousness.[6]. This result significantly supports the role of GABAA1 in susceptibility to propofol anesthesia. In addition, mutation of the GABAA1 receptor (rs2279020) also contributed to the different effects of propofol on blood pressure.

ABCB1 (MDR1, P-glycoprotein) gene is the first identified and the best characterized gene from ABC transporter family. This gene encodes transmembrane protein, mediates ATP-dependent transport of various molecules and is located in the chromosome 7q21.1 and consists of 28 translated exons and 27 introns with over 100kb. P-gpis a “gate keeper” of the brain and is expressed in the luminal surface of capillary endothelial cells of the blood-brain barrier (BBB) transport the toxic compounds out of the brain and effectively prevent the uptake. The single nucleotide polymorphism (SNP) C3435T in exon 26 is one of more than 100 polymorphic variants of this gene that have been discovered to date. This polymorphism correlates with altered expression levels of P-glycoprotein, range of drug response and clinical conditions. Patients who are homozygous TT have a two to three times lower expression of P-gp than the wild type CC. This change in structure could result in increased drug absorption from the intestine, a reduced renal clearance, or an increased brain concentration [7].

Eighty-two patients undergoing propofol general anesthesia (bolus dose 2 mg/kg) in the Clinical of gynecology and obstetrics in Skopje, Republic of Macedonia were recruited for this study. Written informed consent was obtained for each patient. Genomic DNA was extracted from whole blood, using Genomic DNA Whole Blood Kit and Genomic DNA FFPE One-Step Kit (RBC Bioscience, Taiwan) according to the manufacturer provided protocols on a MagCore HF16 Plus automatic DNA extractor (RBC Bioscience, Taiwan). The presence of the (CYP2B6 - 516G>A; GABRA1 - 1059+15G>A and ABCB1 3435T>C polymorphisms were analyzed by allelic discrimination TaqMan assay (MxPro 3005P, Strategene, La Jolla, Ca, USA) using TaqMan SNP genotyping assay for CYP2B6 (rs3745274 assay ID C___7817765_60), for GABRA1 (rs2279020 assay ID C__15966883_10) and for ABCB1 (rs1045642 assay ID C___7586657_20).

Demographics of the participants were as follows: age 51,68% ± 9,09 years; weight 79 ± 17,24 kg; anesthesia induction time 74,13 ±42,33 sec; weak up time 14,50 ± 6,85. Our results for CYP2B6 - 516G>T polymorphism showed a prevalence of 73% for the G allele and 27% for the T allele. This was distributed as 58.43% for the GG genotype, 37.07% for the GT genotype and 4.5 % for the TT genotype. The frequency of TT-type of rs 1045642 was 25,84% while for TC and CC genotypes were 49,43% and 24,73% respectively. The detected genotype frequencies for GABRA1 (rs2279020 assay ID C__15966883_10) were 35,2% for GG, 46,6% AG and 19,2% for AA. Our results showed that the frequency of these polymorphisms in our patients is similar to that in other population studies. The influence of these variants on interindividual variability in the clinical response to propofol in our group of patients will be presented at the Congress.

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