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POLYMORPHISM OF THE CYP2C19 ISOENZYME AS A RISK FACTOR FOR GASTROPATHIES INDUCED BY THE USE OF NONSTEROIDAL ANTI-INFLAMMATORY DRUGS IN PATIENTS WITH PAIN SYNDROME

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ABSTRACT

Nonsteroidal anti-inflammatory drugs (NSAIDs) are one of the most widely used medicines: more than 30 million people take them every day in the world. The popularity and widespread use of NSAIDs is explained by their significant analgesic and anti-inflammatory effect in pain syndromes of different genesis. **The aim** of the study was to evaluate the role of genetic polymorphism of the CYP2C19 isoenzyme in predisposition to NSAID-gastropathies. **Research materials and methods:** the study is based on the examination data of 69 patients with pain syndrome (27 men, 42 women aged 56.4±9.1 years) who underwent inpatient treatment at the 3rd TMA clinic. 11 patients (15.9%) with gastropathies were identified among the examined. All patients necessarily underwent upper endoscopy and determination of Hp status by performing a ^{13}C-urea breath test. **Results of the study:** as a result of the study, we found that the frequency of carrying allele A in patients taking NSAIDs was 97.1%, in the control group – 98.9%. Whereas the frequency of the G allele was 2.6 times more common among patients with pain syndrome and corresponded to the expected Hardy-Weinberg equilibrium, $\chi 2=7.0$, $p=0.008$. Carriers of the G allele were found in 30.4% of patients taking NSAIDs, whereas in the control this allele was found in 1.1% of volunteers. **Conclusion:** The presence of the CYP 2C19 G allele is significantly associated with endoscopically confirmed NSAID-induced gastropathy and can be considered as a risk factor for their development, which is presumably explained by the participation of the CYP 2C19 isoenzyme in the metabolism of arachidonic acid, which plays a role in gastrocytoprotection; Patients with CYP2C19 polymorphism have accelerated metabolism of PPIs, which significantly reduces their clinical effectiveness.

Key words: NSAID, PPIs, Cytochrome P450, gastropathy, gastroduodenal damage.

INTRODUCTION

Nonsteroidal anti-inflammatory drugs (NSAIDs) are one of the most widely used medicines: more than 30 million people take them every day in the world. The popularity and widespread use of NSAIDs is explained by their significant analgesic and anti-inflammatory effect in pain syndromes of different genesis. Unfortunately, the use of NSAIDs is significantly limited due to their undesirable complications, primarily gastrointestinal [6].

NSAID-gastropathy is an erosive and ulcerative lesion of the gastroduodenal zone of the gastrointestinal tract (gastrointestinal tract) that occurs when using NSAIDs and acetylsalicylic acid and has a characteristic clinical and endoscopic picture. Its diagnostic criteria are chronological connection with the use of NSAIDs, asymptomatic / erased clinical picture, high risk of bleeding manifestation, acute often multiple injuries, predominant localization in the antrum of the stomach, absence of an inflammatory shaft around ulcers, foveolar hyperplasia of the mucous membrane and sufficiently rapid healing with the abolition of NSAIDs. Gastroduodenal toxicity of NSAIDs is explained by the blockade of the production of cytoprotective prostanoids mediated by cyclooxygenase (COX)-1, such as prostaglandin E2 and prostacyclin. Highly selective COX-2 inhibitors cause less pronounced gastroduodenal damage than non-selective NSAIDs that inhibit COX-1 and -2, but they do not completely solve the problem of gastrotoxicity [6]. The extreme urgency of the problem is caused by a large number of hospitalizations and deaths associated with the use of NSAIDs, as well as high economic costs for the treatment of NSAID-gastropathy, steadily increasing every year. Ulceration and bleeding induced by the use of NSAIDs are still one of the main clinical problems of internal medicine.

According to various studies, approximately 25-40% of chronic NSAID users have erosions and peptic ulcers (PI) of the gastroduodenal zone, and 2-4% have bleeding or perforation. In many patients, especially the elderly, the use of NSAIDs can do more harm than good. The relative risk (RR) of bleeding, perforation and death due to NSAID-induced ulcers is 3; 6 and 7.6, respectively [6]. Pathologies associated with the use of NSAIDs are the causes of the development of diseases and deaths in many countries of the world. For example, in the USA, the side effects of NSAIDs are the 15th most common cause of death, and on average, gastrointestinal complications are noted in 30% of patients using NSAIDs even in the absence of ulcers on the mucous membrane. In the UK, NSAIDs have become the main class of drugs that cause side effects, which are

noted in 30% of over 18 thousand. hospitalized patients, and NSAID-induced ulcers and bleeding account for 61% of deaths associated with adverse drug reactions (NLR) [6]. Recently, scientific papers have appeared on the frequency and severity of NLR from the use of NSAIDs and their effectiveness depending on the polymorphism of various isoenzymes of the cytochrome P450 hepatic system (CYP 2C8, CYP 2C9, CYP 2C19), responsible for the metabolism of many drugs [2, 3, 4, 6, 11]. The CYP 2C cytochrome system isoenzyme subfamily includes 20% of the CYP 450 content in the liver and metabolizes 25-30% of commonly used drugs, in particular such clinically important ones as NSAIDs, proton pump inhibitors (PPIs), antidepressants, benzodiazepines and clopidogrel [7, 8]. CYP 2C isoenzymes also metabolize endogenous substances such as arachidonic acid and estrogens [9]. Three members of the CYP 2C subfamily (CYP 2C8, CYP 2C9, CYP 2C19) are highly polymorphic isoenzymes and have numerous single nucleotide polymorphisms (SNPs) with different frequency in different ethnic populations [13, 15]. Today, 14 CYP2C8-, 35 CYP 2C9- and 28 CYP2C19-coding ONPS are known, some of them are clinically significant because they can significantly alter the metabolism of various drugs.

Data from previous studies indicate that people with ONP in the genes encoding enzymes that metabolize NSAIDs have a higher risk of developing peptic ulcer and/or upper gastrointestinal bleeding, although the results obtained are quite controversial [2, 3, 12]. The results of a systematic review of the problem showed that it is currently very difficult to assess whether there is an interaction between the effects of NSAIDs and the presence of coding variants in the main NSAID-metabolizing systems of cytochrome P450, such as CYP 2C9, CYP 2C8 and CYP 2C19, and whether these variants increase the risk of NSAID gastropathy independently of each other [2, 3, 4].

In recent years, data have been obtained that ONP is possible not only with loss of function, but also with its enhancement, in particular in the CYP 2C19 isoenzyme. Relatively recently, it has been established that ONP in the CYP 2C19 family (CYP 2C19*17) can predispose to peptic ulcer by means of effects independent of the use of NSAIDs, in particular as a result of changes in the metabolism of arachidonic acid [12]. In addition, ONP in the CYP2C19 family may predispose to the development of peptic ulcer indirectly by altering the metabolism of NSAIDs [5, 8, 10].

Cytochrome P450 (P-450) enzymes are the main participants in the metabolism of xenobiotics. The genetic variability of the genes encoding these enzymes plays an important role in the manifestation of individual sensitivity to drugs [14]. CYP2C19 participates in the metabolism of a number of drugs, in

particular proton pump inhibitors (omeprazole, pantorazole, lansoprazole, rabeprazole and esomeprazole) [1].

Drug interactions are the leading cause of a decrease or complete absence of the effect of therapy and adverse drug reactions.

The aim of the study was to evaluate the role of genetic polymorphism of the CYP2C19 isoenzyme in predisposition to NSAID-gastropathies.

Research materials and methods: the study is based on the examination data of 69 patients with pain syndrome (27 men, 42 women aged 56.4±9.1 years) who underwent inpatient treatment at the 3rd TMA clinic. 11 patients (15.9%) with gastropathies were identified among the examined. All patients necessarily underwent upper endoscopy and determination of *Hp* status by performing a ^{13}C-urea breath test.

All patients were divided into two groups:

11 patients with gastropathies who developed during 2 weeks of NSAID use (main group);

58 patients without gastropathy who used NSAIDs before endoscopy (comparison group).

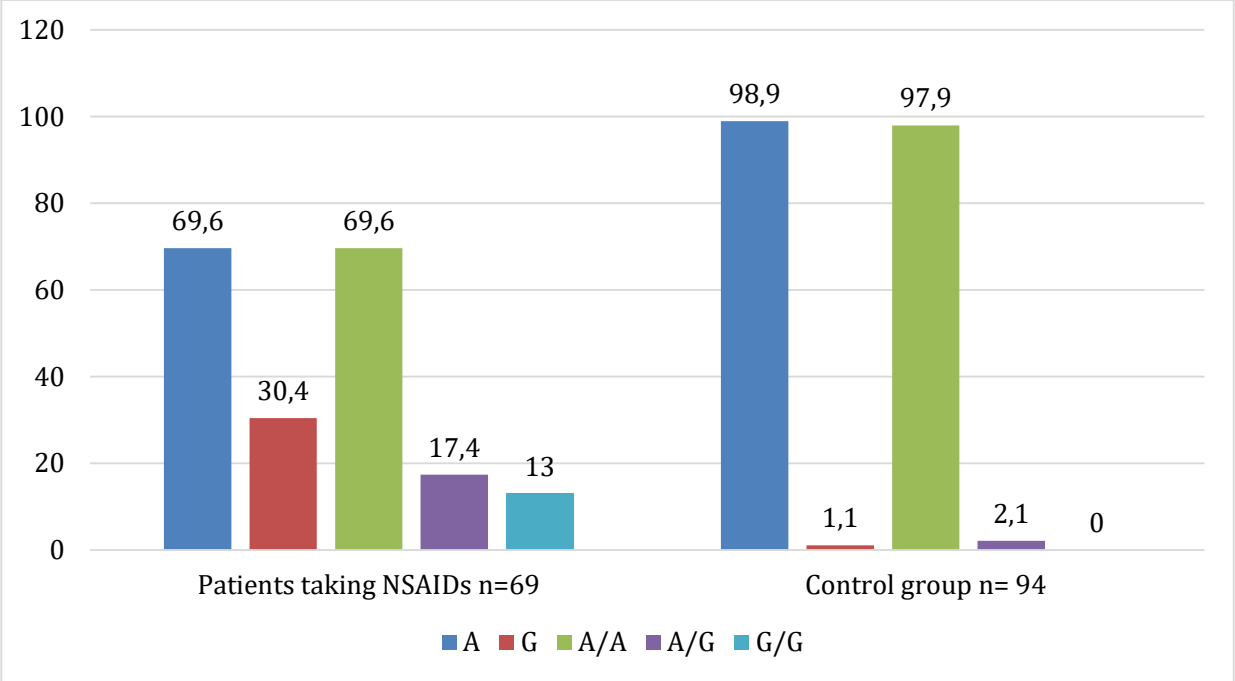
The control group included 94 healthy volunteers (control group)

Molecular genetic studies were carried out on the basis of the Laboratory of medical genetics of NIIG and the PC of the Ministry of Health of the Republic of Uzbekistan. Genotyping after isolation of genomic DNA from whole blood with the addition of ethylenediaminetetraacetate was carried out by multiplex polymerase chain reaction with a fluorescent scheme for detecting products in real time using LITECH CYP 2C19 ACE test systems (St. Petersburg, Russia).

Statistical processing of the results of the study was carried out: using the online calculator open api [https://www.openepi.com / TwobyTwo.htm]. The correspondence of the distribution of the observed frequencies of the genotypes of the studied genes in the control group, theoretically expected by the Hardy-Weinberg equilibrium, was evaluated by the criterion $\chi 2$. The calculation was carried out using an online calculator: http:// www.oege.org/software / hw em r-calc.shtml.

Results of the study: as a result of the study, we found that the frequency of carrying allele A in patients taking NSAIDs was 97.1%, in the control group – 98.9%. Whereas the frequency of the G allele was 2.6 times more common among patients with pain syndrome and corresponded to the expected Hardy-Weinberg equilibrium, $\chi 2=7.0$, $p=0.008$. Carriers of the G allele were found in 30.4% of patients taking NSAIDs, whereas in the control this allele was found in 1.1% of volunteers. The carriage of the heterozygous A/G allele in the CYP2C19 gene in

patients taking NSAIDs was noted in 17.4% of cases, whereas in the control in 2.1% of cases (Fig. 1).



OR=6,9 (CI 1,44-33,0); $\chi 2$=58,8, p<0,001

Fig. 1. Frequency of distribution of alleles and genotypes of polymorphism in the CYP2C19 gene in groups of patients depending on gender and control

A relationship was established between the G allele, the G/G genotype and the presence of NSAID gastropathy at both allele and genotype levels (Table 1).

Table 1

Identification of alleles and genotypes of polymorphism in the CYP2C19 gene depending on the presence of gastropathies among patients with pain syndrome taking NSAIDs

Allele/genotype	Gastropathies + (n=11)		Gastropathies - (n=58)		
	n	%	n	%	
A	14	63,6	82	70,7	$\chi 2$ =12,1; p=0,05; OR=0,25; 95% CI 0,11-0,56; df=0,014
G	8	36,4	34	29,3	
AA	6	54,5	42	72,4	$\chi 2$ =14,2; p=0,01; OR=8,25; 95% CI 2,56-26,6; df=0,030
AG	2	18,2	11	19,0	
G/G	2	18,2	5	8,6	

Why CYP2C19 is associated with gastropathies at the allele and genotype levels is not fully understood. As is known, CYP 2C19 is a common polymorphism with enhanced function, whose carriers have higher metabolic rates of some clinically important drugs (PPIs, escitalopram, sertraline, clopidogrel, etc.), followed by a decrease in their concentration in blood plasma and a weakening of the clinical effect (Goldstein J.A., 2001; Ingelman-Sundberg M. et al., 2007; Rosemary J., Adithan C., 2007; Baldwin R.M. et al., 2008; Hunfeld N.G. et al., 2008; Li-Wan-Po A. et al., 2010; Pedersen R.S. et al., 2010; Sibbing D. et al., 2010; Scott S.A. et al., 2012b; Zabalza M. et al., 2012; Musumba C.O. et al., 2013).

Nevertheless, based on a review of data on the functional and clinical consequences of carrying the CYP2C19 G allele, it was concluded that CYP2C19 has only a minor effect, which is unlikely to be clinically significant, with the exception of CYP 2C19* G homozygotes, and only for drugs with a narrow "therapeutic window" (Ingelman-Sundberg M. et al., 2007; Li-Wan-Po A. et al., 2010; Scott S.A. et al., 2012b).

On the other hand, in some recent studies, scientists have concluded that the carriage of the CYP2C19 allele in patients taking clopidogrel is associated with lower platelet reactivity, a reduced risk of cardiovascular complications and stent thrombosis, but with a higher risk of intense bleeding (Harmsze A.M. et al., 2012; Zabalza M. et al., 2012). Therefore, one of the possible explanations for the revealed connection may be that CYP 2C19 carriers have accelerated metabolism and a decrease in the clinical efficacy of PPIs, which cause a decrease in the gastroprotective ability of the mucous membrane to resist aggressive factors (for example, NSAIDs and *Hp* infections), thus predisposing to the occurrence of gastropathies.

In addition, CYP2C isoenzymes are involved not only in the metabolism of xenobiotics, but also in various endogenous substances (Ingelman-Sundberg M. et al., 2007; Scott S.A. et al., 2011; 2012a). For example, arachidonic acid is metabolized by three main enzymatic pathways: cyclooxygenase, lipoxygenase, and CYP 450-monooxygenase with CYP 2C19 (Kaspera R., Totah R.A., 2009; Musumba C.O. et al., 2013). The CYP 2C19 isoenzyme effectively metabolizes arachidonic acid into four types of epoxyeicosatrienic acids (EETC): 5,6-EETC; 8,9-EETC; 11,12-EETC and 14,15-EETC, which are species- and organ-specific and have a variety of physiological functions (including control of vascular tone, angiogenesis, cell migration, proliferation, inflammation) (Kaspera R., Totah R.A., 2009).

Although the effect of EETCS on the human gastrointestinal tract has not been fully studied, however, they presumably inhibit the production of prostaglandin E2 in smooth muscle cells and participate in the formation of reactive oxygen species in vascular endothelial cells, which contributes to ischemic lesions (Pilotto A. et al., 2007; Kaspera R., Totah R.A., 2009; Musumba C. et al., 2009). Therefore, one of the possible explanations for the relationship between gastropathies and CYP 2C19 can be considered that the latter alters the metabolism of arachidonic acid, as a result of which the protective properties of the mucous membrane of the gastroduodenal zone of the gastrointestinal tract decrease due to a combination of a decrease in the production of gastroprotective prostaglandin E2, increased vasoconstriction in the microcirculatory bed of the mucous membrane and the production of damaging reactive oxygen species (Musumba C. et al., 2009; 2012).

CONCLUSION

1. The presence of the CYP 2C19 G allele is significantly associated with endoscopically confirmed NSAID-induced gastropathy and can be considered as a risk factor for their development, which is presumably explained by the participation of the CYP 2C19 isoenzyme in the metabolism of arachidonic acid, which plays a role in gastrocytoprotection.

2. Patients with CYP2C19 polymorphism have accelerated metabolism of PPIs, which significantly reduces their clinical effectiveness.

REFERENCES

1. Klyaritskaya I.L., Rabotyagova YU.S. Polimorfizm gena CYP2C19 i klinicheskoye znacheniye yego opredeleniya. Krymskiy terapevticheskiy zhurnal 2013;1:19-25.

2. Blanco G., Martínez C., Ladero J.M. Interaction of CYP2C8 and CYP2C9 genotypes modifies the risk for nonsteroidal anti-inflammatory drugs-related acute gastrointestinal bleeding. // Pharmacogenet Genomics. – 2008. - vol.18(1). – P. 37–43.

3. Carbonell N., Verstuyft C., Massard J. CYP2C9*3 Loss-of-Function Allele Is Associated With Acute Upper Gastrointestinal Bleeding Related to the Use of NSAIDs Other Than Aspirin // Clin. Pharmacol. Ther. – 2010. - vol.87(6). – P. 693–698.

4. Estany-Gestal A., Salgado-Barreira A., Sánchez-Diz P., Figueiras A. Influence of CYP2C9 genetic variants on gastrointestinal bleeding associated with nonsteroidal anti-inflammatory drugs: a systematic critical review. // Pharmacogenet. Genomics. – 2011. -vol. 21(7). – P. 357–364.

5. Hunfeld N.G., Mathot R.A., Touw D.J. Effect of CYP2C19*2 and *17 mutations on pharmacodynamics and kinetics of proton pump inhibitors in Caucasians // Br. J. Clin. Pharmacol. – 2008. – vol. 65(5). – P. 752–760.

6. Hunt R.H., Lanas A., Stichtenoth D.O., Scarpignato C. Myths and facts in the use of anti-inflammatory drugs //Ann. Med. – 2009. - Vol. 41(6). – P. 423–437.

7. Increased omeprazole metabolism in carriers of the CYP2C19*17 allele; a pharmacokinetic study in healthy volunteers Baldwin R.M., Ohlsson S., Pedersen R.S. et al. //Br. J. Clin. Pharmacol. – 2008. – vol. 65(5). – P. 767–774.

8. Ingelman-Sundberg M., Sim S.C., Gomez A., Rodriguez-Antona C. Influence of cytochrome P450 polymorphisms on drug therapies: pharmacogenetic, pharmacoepigenetic and clinical aspects // Pharmacol. Ther. – 2007. – vol.116(3). – P. 496–526.

9. Kaspera R., Totah R.A. Epoxyeicosatrienoic acids: formation, metabolism and potential role in tissue physiology and pathophysiology // Expert Opin. Drug Metab. Toxicol. – 2009. – vol.5(7). – P. 757–771.

10. Li-Wan-Po A., Girard T., Farndon P. Pharmacogenetics of CYP2C19: functional and clinical implications of a new variant CYP2C19*17. // Br. J. Clin. Pharmacol. – 2010. – vol. 69(3): 222–230.

11. Ma J., Yang X.Y., Qiao L. CYP2C9 polymorphism in non-steroidal anti-inflammatory drugs-induced gastropathy // J. Dig. Dis. – 2008. – vol. 9(2): 79–83.

12. Musumba C.O., Jorgensen A., Sutton L. et al. (2013) CYP2C19*17 gain-of-function polymorphism is associated with peptic ulcer disease // Clin. Pharmacol. Ther. 2013. - 93(2): 195–203.

13. Pedersen R.S., Brasch-Andersen C., Sim S.C. Linkage disequilibrium between the CYP2C19*17 allele and wildtype CYP2C8 and CYP2C9 alleles: identification of CYP2C haplotypes in healthy Nordic populations // Eur. J. Clin. Pharmacol. – 2010. – vol. 66(12): 1199–1205.

14. Preissner S.C., Hoffmann M.F., Preissner R. Polymorphic cytochrome P450 enzymes (CYPs) and their role in personalized therapy // PLoS One 2013;8(12): e82562.

15. Scott S.A., Martis S., Peter I. et al. Identification of CYP2C19*4B: pharmacogenetic implications for drug metabolism including clopidogrel responsiveness // Pharmacogenomics J. – 2012. – vol. 12(4): 297–305.