

UDC 616.311.2

DESCRIPTION OF POLYMORPHIC VARIANTS OF NUCLEAR TRANSCRIPTION FACTOR NF- κ B1 AS PREDICTORS OF GENERALISED PERIODONTITIS DEVELOPMENT

N. V. Gasyuk

"Ternopil State Medical University", Ternopil, Ukraine

Summary. In the article results of molecular genetic studies of pathogenetic mechanisms of inflammatory diseases periodontal tissues through polymorphism nuclear transcription factor NF- κ B1, which controls the expression of immune response genes, apoptosis and cell cycle. The data make it possible to state that certain polymorphic variants of the gene NF- κ B1 enable the formation of groups at risk of morbidity of generalized parodontitis.

The contingent of people with genotype (Del/Del) to recommend under medical observation. Persons with polymorphic variant of NF- κ B1 (Del/Ins) constitute risk in the event of bad habits and concomitant somatic pathology that are predictors for the development of general parodontitis induced vascular disorders against the background of lamina propria gums. Persons with polymorphic variant of NF- κ B1 (Ins/Ins), constitute a risk group in case of adverse impact of local factors and existing teeth-jaw abnormalities and deformities.

Adjusted distribution of polymorphic variants enables timely prediction and prevention of generalized periodontitis

Key words: genotype, periodontitis, polymorphism, nuclear faktor transcription.

Introduction. The problem of prognostic criteria emergence and early diagnosis of inflammatory diseases of periodontal tissues is extremely important, as determined by the clinical course of pathological process, data of laboratory diagnostics, accompanied by severe and destructive changes in periodontal tissues and early tooth loss [2].

The etiology of various forms of generalized periodontitis is associated with aggressive gram-negative and anaerobic microorganisms on the background of lowering defense of organism, including the leading role played by the functional status of neutrophils and nonspecific protection factors [6, 8, 9].

For today it is established that the destruction of the periodontal tissues in the course of generalized periodontitis occur as the result of destruction of tissue elements by proteases, free radicals and by inhibiting reparative processes by cytokines that produce polymorphonuclear leukocytes due to violation of their functional activity caused by either genetic or acquired secondary factors [1, 3].

Tactics of complex early diagnosis of generalized periodontitis is questionable issue among experts and have a number of features in the selection of diagnostic measures.

In recent time, in the literature data an attention is focused on necessity of development of a special set of diagnostic and treatment periodontal interventions [2, 6], which includes professional oral hygiene with grinding and polishing of necks and roots of teeth, surgical interventions in periodontal tissues using tissue-compatible osteoplastic

preparations for restoration of the structure of alveolar bone, orthodontic and orthopedic treatment following certain indications [5, 7].

However, traditional methods of diagnosis and prognosis of various forms of generalized periodontitis is not always effective and timely, don't allow to solve the problem by clinicians due to cyclical and unpredictable pathological process, highly progressed periodontal destruction, toxic and allergic reactions.

The aim of this study was to determine predictors of development of generalized periodontitis based on the present characteristics of polymorphic variants of nuclear transcription factor NF- κ B1.

Objects and methods. Dental examination of 45 people with intact periodontium was conducted in accordance with the standards of diagnosis and treatment of dental patients (MOH Ukraine "On approving of the protocols of medical care in specialties "orthopedic dentistry", "therapeutic dentistry", "surgical dentistry", "orthodontics", "pediatric dentistry", "pediatric surgical dentistry" from 28.12.2002 №507 and MOH Ukraine "On approval of standards of medical care and quality of medical care" from 28.12.2002 №507).

Determination of polymorphic gene section NF- κ B1 was performed from cells of buccal epithelium of examined people by means of polymerase chain reaction. Collection of material was performed with sterile disposable dental brush, followed by the introduction of a reagent in ependorph with reagent "DNA Express" (NPF "LyTeh", Russia). Genome deoxyribonucleic acid was isolated by a set of "DNA Express" ("LyTeh", Moscow).

Polymorphic gene area NF- κ B1 (rs28362491) was amplified by polymerase chain reaction. The final volume of the reaction mixture was 25 μ l and contained:

- specific oligonucleotide primers – by 66 ng each, direct – 5'-TGGGCACAAGTCGTTTATGA-3'; reverse – 5'-CTCGAGCCGGTAGGGAAG-3';
- 2,5 μ l 10 times buffer for amplification;
- 2 mM magnesium chloride;
- 0,2 mM deoxy nucleotide triphosphate (dNTP);
- 2,5 units. Taq DNA polymerase;
- 20-50 ng of genomic DNA.

In the tube from above was placed 25 mg mineral oil.

Amplification was performed on thermocyclers "Tertsyk" (OOO "DNA Technology", Russia).

To identify alleles was performed restriction analysis of amplicons using restriction enzyme PflMI ("SybEnzym", Russia) at 37 °C.

Products cleavage of polymorphic gene section NF- κ B1 was detected by horizontal electrophoresis in 2% agarose gel in single TBE (50 mM Tris-H₃BO₃ and 2 mM EDTA, pH 8,0), for 2 hours at a voltage of 2 V per 1 cm gel. As a molecular weight marker of DNA was used pBR322/Alu I. Gels were stained by ethidium bromide, followed by visualization of the results in the UV light.

The results of clinical and complex morphological studies were analyzed using a number of methods of biostatistics. Statistical research was performed at the Department of Statistical Research of SHEI "I. Horbachevsky Ternopil State Medical University". Processing data was carried out using the license program "Statistica" of "StatSoft".

In the statistical processing of results, molecular genetic studies, in accordance with certain genotype (Del/Del), (Del/Ins), (Ins/Ins), as a basis for understanding of the genetic structure of the population took the law of genetic equilibrium by Hardy-Weinberg. On the basis of this law, according to data on the frequency of recessive phenotype in a population that have homozygous genotype was calculated prevalence of polymorphic variants of NF- κ B1 in the surveyed groups. Statistical justification of probability differences distribution of genotypes was performed using χ^2 test adjusted for continuity by Yates [4].

Results and its discussion. In the examined group that is consisted of patients with intact periodontium genotype (Del/Del) – homozygotes had 5 people (11%), which allows the formation of risk group for morbidity of generalised periodontitis, and recommend to take the present contingent of individuals under medical supervision.

Since the most pronounced clinical manifestations of inflammatory and degenerative changes in periodontal

tissues, the characteristics of which corresponds to the clinical picture of rapidly progressive periodontitis was observed in patients with this genotype.

The results of correlation analysis, definition of Spearman correlation coefficient ($p < 0,05$), the data about the lack of correlation between certain parameters in patients with polymorphic variant (Del/Del). The lack of correlation in this case shows that no matter how changing parameters (PMA, present or absent concomitant somatic pathology, etc.), relatively major, in this case genotype (Del / Del) is unchanged and the determining factor that causes the development of generalized periodontitis, clinical picture of which corresponds to quickly progressing.

In people with genotype (Del/Ins) was identified the presence of a direct correlation of Spearman ($p < 0,05$) between the immunohistochemical profile of individual and present concomitant somatic pathology of the correlation coefficient ($r = +0,60$), between harmful habit (smoking) and PMA index ($r = +0,17$), which indicates a direct role of these parameters in the emergence and development of generalized periodontitis. 23 (51%) individuals in the control group of polymorphic variant NF- κ B1 (Del/Ins) form risk group in the case of bad habits of concomitant somatic pathology, that in the future cause development of induced generalized periodontitis on the background of vascular disorders of the lamina propria of gums (Table 1).

There is no reliable as direct or reverse correlation by Spearman ($p < 0,05$) in genotype between the parameters (Ins/Ins), which also allows the assumption that the pathogenic mechanisms of generalized periodontitis in patients with this variant have a polymorphic genetic component, but the features of clinical manifestations are more benign and are characterized by chronic long course compared to the polymorphic variant (Del/Del).

17 people (33%) of control group with polymorphic variant of NF- κ B1 (Ins/Ins), constitute a risk group in the case of influence both local adverse factors and local teeth-jaw abnormalities and deformities

Conclusion. Thus, defined polymorphic variants of genes NF- κ B1 enable the formation of risk groups in the incidence of generalized periodontitis. The contingent of people with genotype (Del/Del) is recommended to take under medical supervision. Persons with polymorphic variant NF- κ B1 (Del/Ins) constitute a risk group in case of bad habits and concomitant somatic diseases, which are predictors for the development of induced generalized periodontitis on the background of vascular disorders of the lamina propria of gums. Persons with polymorphic variant of NF- κ B1 (Ins/Ins), constitute a risk group in the case of

Table 1.

Distribution of polymorphic variants of genes and- κ B1y patients with intact periodontium

Characteristics of examined person	Homozygotes (D/D)	Heterozygotes (D/I)	Homozygotes (I/I)
Patients with intact periodontium (45 people)	5 (11%)	23 (51%)	17 (38%)
Probability of development of generalized periodontitis	High	Moderate	Low

Note: – the distribution of genotypes polymorphisms (Ins/Ins, Ins/Del, Del/Del) determined in accordance with the law of genetic equilibrium Hardy-Weinberg;

– The likelihood of differences defined by criterion χ^2 adjusted for continuity Yates.

impact of unfavorable local factors and presence of tooth-jaw abnormalities and deformities.

Declaration of interest. The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of this article.

Funding. This article did not receive any specific grant from any funding agency in the public, commercial or not-for-profit sectors.

REFERENCES

1. Айала Ф. Современная генетика / Ф. Айала, Дж. Кайгер. – М.: Книга по требованию, 2012. – 294 с.
2. Григорян А. С. Болезни пародонта / Григорян А. С., Грудянов А. И., Рабухина Н. А. – М.: “Медицинское информационное агентство”. – 2004. – 320 с.
3. Григорян А. С. Ключевые звенья патогенеза заболеваний пародонта в свете данных цитоморфометрического метода исследования / А. С. Григорян, А. И. Грудянов // *Стоматология*. – 2001. – № 1. – С. 5–8.

4. Реброва О. Ю. Статистический анализ медицинских данных. Применение пакета прикладных программ STATISTIKA / Реброва О. Ю. – М.: Медиа Сфера, 2002. – 312 с.

5. Kinane D. F. Periodontal manifestations of systemic disease / D.F. Kinane, G. J. Marshall // *Aust. Dent. J.* – 2001. – № 46 (1). – P. 2–12.

6. The interleukin-1 promoter haplotype ATA is a putative risk factor for aggressive periodontitis / S. Reichert, H. K. Machulla, J. Klapproth, U. Zimmermann // *J. Periodontol. Res.* – 2008. – Vol. 43, № 1. – P. 40–47.

7. Schulz S. Single nucleotide polymorphisms in interleukin-1 gene cluster and subgingival colonization with *Aggregatibacter actinomycetemcomitans* in patients with aggressive periodontitis // S. Schulz, J.M. Stein, U. Zimmermann // *Hum. Immunol.* – 2011. – № 72 (10). – P. 940–946.

8. The del/del genotype of the nuclear factor-kappaB -94ATTG polymorphism and its relation to aggressive periodontitis / S. Schulz, L. Hierse, W. Altermann [et al.] // *J. Periodontol. Res.* – 2010. – № 45 (3). – P. 396–403.

9. The effect of periodontal therapy on TNF-alpha, IL-6 and metabolic control in type 2 diabetics // J. Talbert, J. Elter, H.L. Jared [et al.] // *J. Dent. Hyg. Spring.* – 2006. – P. 802 – 807.

ХАРАКТЕРИСТИКА ПОЛІМОРФНИХ ВАРІАНТІВ ЯДЕРНОГО ФАКТОРА ТРАНСКРИПЦІЇ NF-κB1 ЯК ПРЕДИКТОРІВ РОЗВИТКУ ГЕНЕРАЛІЗОВАНОГО ПАРОДОНТИТУ

Гасюк Н.В.

Державний вищий навчальний заклад
“Тернопільський державний медичний
університет імені І.Я. Горбачевського
МОЗ України”, м. Тернопіль, Україна

Резюме. В статті приведені результати молекулярно-генетичного дослідження патогенетичних механізмів виникнення запальних захворювань тканин пародонта через поліморфізм ядерного фактора транскрипції NF-κB1, який контролює експресію генів імунної відповіді, апоптозу і клітинного циклу. Отримані дані дають можливість стверджувати, що визначені поліморфні варіанти гену NF-κB1 дають можливість формування груп ризику на захворюваність генералізованим пародонтитом. Контингент осіб з генотипом (Del/Del) рекомендуємо взяти під диспансерний нагляд. Особи з поліморфним варіантом NF-κB1 (Del/Ins) складають групу ризику у випадку наявності шкідливих звичок та супутньої соматичної патології, що є предикторами для розвитку індукованого генералізованого пародонтиту на тлі судинних розладів власної пластинки ясен. Особи із поліморфним варіантом NF-κB1 (Ins/Ins), складають групу ризику у випадку впливу місцевих несприятливих факторів та наявних зубо-щелепних аномалій і деформацій. Приведений розподіл поліморфних варіантів дає можливість прогнозування та своєчасного попередження виникнення генералізованого пародонтиту

Ключові слова: генотип, пародонтит, поліморфізм, ядерний фактор транскрипції.

ХАРАКТЕРИСТИКА ПОЛІМОРФНЫХ ВАРИАНТОВ ЯДЕРНОГО ФАКТОРА ТРАНСКРИПЦИИ NF-κB1 КАК ПРЕДИКТОРОВ РАЗВИТИЯ ГЕНЕРАЛИЗОВАНОГО ПАРОДОНТИТА

Гасюк Н.В.

Государственное высшее учебное заведение
“Тернопольский государственный медицинский
университет имени И.Я. Горбачевского МЗ
Украины”, г. Тернополь, Украина

Резюме. В статье приведены результаты молекулярно-генетического исследования патогенетических механизмов возникновения воспалительных заболеваний тканей пародонта через полиморфизм ядерного фактора транскрипции NF-κB1, который контролирует экспрессию генов иммунного ответа, апоптоза и клеточного цикла. Полученные данные дают возможность утверждать, что определенные полиморфные варианты гена NF-κB1 дают возможность формирования групп риска на заболеваемость генерализованным пародонтитом.

Контингент лиц с генотипом (Del/Del) рекомендуем взять под диспансерное наблюдение. Лица с полиморфным вариантом NF-κB1 (Del/Ins) составляют группу риска в случае наличия вредных привычек и сопутствующей соматической патологии, являются предикторами для развития индуцированного пародонтита на фоне сосудистых расстройств собственной пластинки десен. Лица с полиморфным вариантом NF-κB1 (Ins/Ins), составляют группу риска в случае влияния местных неблагоприятных факторов и имеющихся зубо-челюстных аномалий и деформаций. Приведенное распределение полиморфных вариантов дает возможность прогнозирования и своевременного предупреждения возникновения генерализованного пародонтита

Ключевые слова: генотип, пародонтит, полиморфизм, ядерный фактор транскрипции.