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Allelic polymorphism of glucocorticoid receptor NR3C1 (GR): from molecular biology to clinical implications

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Polymorphism of stress-related genes is a key factor determining difference in the stress reactivity and resistance among humans. Glucocorticoid receptors are important actors of stress responses. This review is focused on the molecular biology and clinical implications of glucocorticoid receptor gene polymorphism.

Keywords: glucocorticoid receptors, allelic polymorphism, SNP, NR3C1, GR, stress-induced pathology.

The diseases of civilization (arterial hypertension, atherosclerosis, sugar diabetes, immunodeficiency, autoimmune pathology, bipolar disorders and depression) are of significant economic and humanitarian damage to modern humankind. They are based on the systemic down-regulation, occurring as a response to the allostatic load, distress and disadaptation. Glucocorticoid hormones as stress effectors and agents of the adaptation to stress take instant part in the regulation of metabolism, homeostasis, immune reactions, endocrine state and psychiatric processes, which influences the mood, memory, and general psychological activity. Among genes-stress regulators, the genes of glucocorticoid receptors should be paid special attention. The current review presents the analysis of the genetics and molecular biology of the receptors of glucocorticoid hormones (NR3C1), the spectrum of activation effects which includes the range from epigenetic modifications of the genome to the impact on psychiatric processes.

Gene structure and expression regulation. The glucocorticoid hormone receptor gene *NR3C1* (nuclear receptor subfamily 3, group C, class 1) is present in the human genome in the only copy, located in the locus 5q31.3 (long arm of chromosome 5, site 3, band 1, subband 3). The length of the gene is 157581 base pairs, it contains nine exons, coding for the sequences of 777 aminoacid residues [2]. The gene structure is presented in Fig. 1 and 2.

The expression of *NR3C1* gene is under the control of several alternative first exons (according to different data, from seven to nine), preceding CpG-rich sequence (promotor region) [3, 4]. Seven tissue-specific promotors, promoting different mechanisms of genetic control over the gene expression in organs and tissues, are localized in introns between these alternative exons. The methylation of alternative exons and promotors is the epigenetic mechanism of the regulation of gene activity [4]. The activity of different promotors is likely to have impact on the choice of a specific mechanism of splicing, leading to the formation of some isoforms of receptors. The genetic polymorphism of the regulatory sites is known for its association with the development of depressive disorders [5].

Splicing and translation. Receptor isoforms. The formation of eight splice-variants, coding for four main isoforms of the receptor – GR-α, GR-β, GR-γ, as well as little-studied isoform GR-P is possible after the

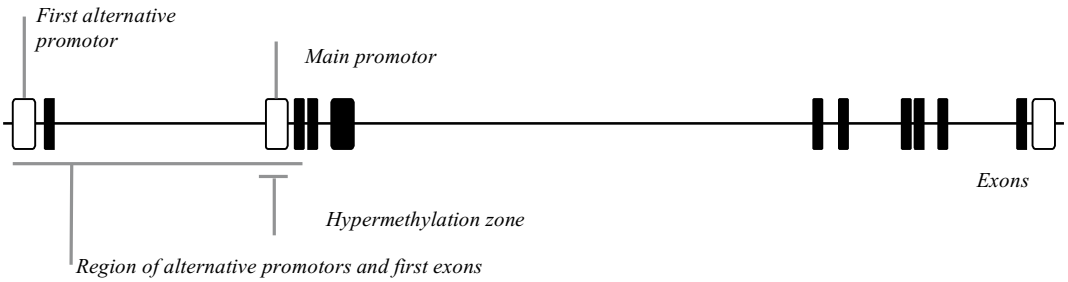

Fig. 1 *NR3C1* gene structure

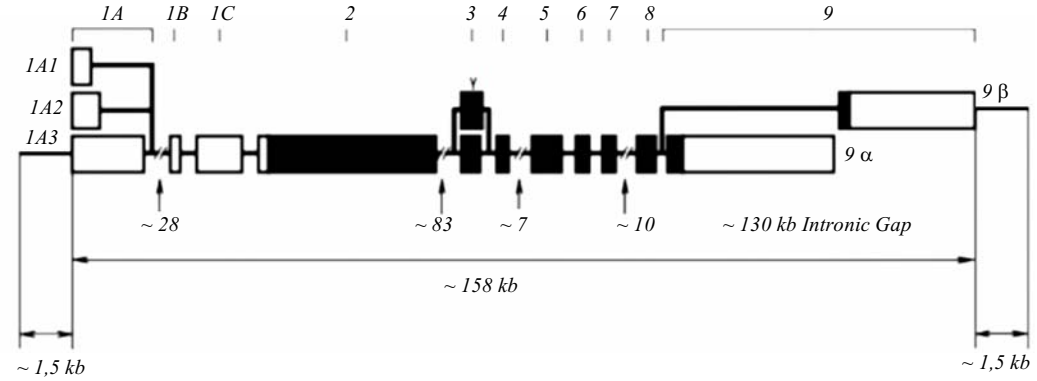

Fig. 2 Non-translational regions (white blocks) and the intron-exon structure of *NR3C1* gene (according to [31])

alternative splicing of pro-mRNA, read from the sequence of *NR3C1* gene (Table 1). Besides, the shift of the reading frame in the process of the initiation of translation leads to a number of additional isoforms of the receptor GR-α (GR-αA, GR-αB, GR-αC and GR-αD; Table 2). This mechanism of the alternative initiation of translation is typical for the synthesis of protooncogenes, the transcription regulators, kinases and growth factors in eukaryotes [7]; it allows increasing the number of protein products of the same gene, thus enlarging the range of its physiological functions and the number of tissue-specific variants of the response [6].

The receptor isoforms are remarkable for C-terminal site which influences the mechanisms of cytoplasmic-nuclear traffic and the activation of target genes [6]. The most wide-spread form of the receptor is GR-α [6], the aminoacid sequence of which is presented in Fig. 1. α-Isoform is presented by a number of shorter subisoforms with some absent aminoacids at C-terminal end (Table 2) which does not influence the binding to the hormone, but modifies the processes of translocation of the activated receptors into the nucleus [6].

The main distinction of the less prevailing variant, GR-β, the level of expression of which is about 1 % of GR-α one [8], is the absence of specific glucocorticoid-binding domain [9]. β-isoform is predominantly localized in the nucleus and is likely to serve as a dominantly negative regulator of GR-α and MR, it is also capable of competing with the latter for the sites of promotors as well as forming heterodimers [9, 8]. Thus, lower level of sensitivity of neutrophils to glucocorticoid-mediated apoptosis, compared to T-lymphocytes, is likely to be conditioned by the increased expression of GR-β in these cells. At the same time some authors doubt the capability of GR-β to inhibit the effects of GR-α in the physiological and pathological conditions, as due to the low level of GR-β basic expression, at least 500-fold increase in its synthesis is required to obtain a significant effect [8]. It was established that GR-β inhibits only the transactivational effects of GR-α, not affecting the induced transrepression [8]. In addition, the effect of GR-β on more high-affine MR is also possible in the physiological conditions [8]. It should be noted that the isoform GR-β is not observed in rats as they do not have the corresponding site of the active splicing in *NR3C1* gene [11]. The physiological significance of other isoforms of the receptor, in particular, GR-γ and GR-P, is yet to be studied in detail.

The isoform GR-γ was described by Rivers et al. [12] in 1999. It contains an additional arginine in the

Table1
Human gene NR3C1 mRNA spilce variant

Identifier in the catalog NCBI RefSeq	Characteristics	Resulting protein	Reference
NM_000176.2	Main transcript, coding the most common receptor isoform	GR-α isoforms, obtained after alternative variants of mRNA translation initiation: GR-A, GR-B, GR-C, GR-D and their subtypes	[6]
NM_001204258.1			
NM_001204259.1			
NM_001204260.1			
NM_001204261.1			
NM_001204262.1			
NM_001204263.1			
NM_001204264.1			
NM_001018074.1	Variants with alternative 5'-non-coding sequences		
NM_001018075.1			
NM_001018076.1			
NM_001018077.1			
NM_001020825.1	Formed due to alternative site of splice-acceptors in 3'-terminal exon	GR-β	[9]
NM_001024094.1	Formed due to alternative splicing in the site of one of the encoding exons	GR-γ	[12]
NM_001204265.1	Variant without two exons at 3'-end	GR-P	[40, 41]

structure of DNA-binding domain, occurring as a result of alternative splicing of the intron between the 3rd and 4th exons. This modification is known to induce the reduction in the transcriptional activity of the receptor by 48% [13]. GR-γ is highly expressed in different tissues, its share is in the range of 3.8–8.7 % in the total population of GR receptors [12].

Receptor structure. The receptor consists of four domains, three of which are presented by highly conservative sequences, present in other proteins (Table 3, Fig. 3). The effect of GR is achieved via dimers, which may exist in the form of homo- and heterodimers with MR. Homo- and heterodimers induce different subgroups of genes, providing fine regulation depending on changing levels of hormones in stress conditions [14, 15]. In this respect high significance is attributed to the fact that the character and magnitude of GR/MR-mediated responses is determined not only by binding receptors to ligands, but also by the presence of nonsteroid coregulators, coactivators and corepressors [15]. The dimer configuration of a GR molecule differs from that of the receptors of other steroid hormones and contains an additional intermolecular β-layer [16].

Receptor activation. Under basic cellular conditions the receptor exists in the composition of a multiprotein complex with one GR molecule and heat shock proteins – two hsp90 molecules, one hsp70, one hsp56 and immunomodulin [8].

The hormone binding to the receptor leads to a number of events, including the dissociation of heat shock proteins, immunomodulin and phosphorylation of GR, inducing the transportation of the

Table2
Human GR receptor isoforms (gene NR3C1)

Isoform	Structure	Localization and function
GR-α (α-A, GR-A)	Main receptor isoform, containing DNA- and hormone-binding domains	Located in the cytoplasm; is transported into the nucleus after ligand-binding, where it operates. Isoforms differ in the mechanisms of transportation into the nucleus [6]
GR-αB	Terminated at codon 571–573	Short receptor isoforms, different in the mechanisms of transportation into the nucleus [6]
GR-αC1	Terminated at codon 748–750	
GR-αC2	Terminated at codon 760–762	
GR-αC3	Terminated at codon 784–786	
GR-αD1	Terminated at codon 1438–1440	
GR-αD2	Terminated at codon 1483-1485	
GR-αD3	Terminated at codon 1498-1500	
GR-β	Shorter isoform, compared to GR-α ,with prominent C-terminal part, non-containing the hormone-binding domain [9]. Not observed in rats [11].	Predominantly localized in the nucleus, a dominantly-negative regulator of GR- activity [9].
GR-γ	DNA-binding domain contains the additional amino acid (arginine)	Lower affinity to GRE compared to GR-α [12]
GR-P	Shorter C-terminal area, not observed in other isoforms	Unknown function. Highly expressed in glucocorticoid-resistant cell lines, may serve as a modulator of susceptibility to glucocorticoids [40, 41]

Table3
GR-αA proteins' domains

Localization	Domain	Function
1–420	Transactivational	Interaction with transcription factors
419–486	DNA-binding by “zinc fingers” type	Interaction with GRE-sequence of DNA and modulation of transcription initiation frequency
487–527	Hinged	Molecular structure motility
528–777	Ligand-binding	Hormone-binding, translocation into the nucleus

hormonereceptor complex in the nucleus [8]. The half-life of the hormone-receptor complex for GR is 5 min (compared to 45 min for MR) [14]; K_D dissociation constant is ≈0.5 nM [8].

The level of receptor-activating glucocorticoid hormones in blood is susceptible to pulse fluctuations in the course of 24 hours. The average duration of one secretion pulse for rats is approximately one hour. In the evening the secretion peaks are more frequent, they follow each other almost without any intervals and have high amplitude; in the morning their amplitude is decreased and they are divided by intervals of half an hour or an hour [14, 17]. In the blood the glucocorticoids interact with corticosteroid-binding proteins (CBP) [8]; their penetration via hematoencephalic barrier is regulated by P-glycoprotein mdr1A (multiple drug resistance) [18].

The peak concentrations of glucocorticoids in blood induce the activation and translocation of GR molecules into the nucleus as early as after 30 min (in the basic conditions GR receptors in the nucleus are practically not present) [14, 19]. Then their level

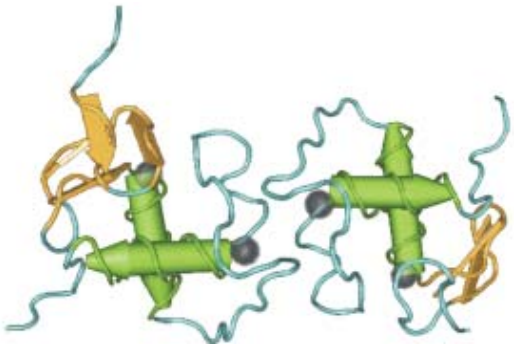

Fig. 3 DNA-binding domain of the glucocorticoid receptor

diminishes to the initial values rather fast (in the course of 90 min) [14, 19]. Therefore, the total duration of the activation cycle of receptors, their translocation and elimination from the nucleus is 120 min. The elimination of receptors from the nucleus occurs due to their proteasome degradation in the nucleus; it is blocked by the inhibitors of 26S-subunit of the proteasome [14]. The cytoplasmic concentration of receptors in the course of this cycle does not decrease considerably which testifies to only a small amount of receptors, passing in each recruiting cycle [14].

Post-translational modification. Currently 14 different variants of post-translational modification are described for human GR molecule, including phosphorylation, sumoylation and ubiquitination (Table 4). The phosphorylation and dephosphorylation of GR are of considerable impact on the function of the receptor, its intracellular traffic and affinity to ligands [20]. In a free state the receptor is phosphorylated (sites Tre171 and Ser246 in GR of rats [21]), however, the binding of the hormone (not the antagonists) leads to its further phosphorylation, which is likely required for the physiological activity [22]. This phosphorylation seems to occur in sites Ser 203 and Ser211 with the participation of cyclin-dependent kinases [20, 22]. These phosphorylation sites are related to the regulation of the traffic of the activated receptor into various cellular compartments: GR-Ser211P is found in the nucleus, while GRSer203-P and twice phosphorylated GR-Ser 203/Ser211-P are present only in the cytosol [22]. The phosphorylation of Ser226 site with the involvement of JNK and MAPK leads to the accelerated withdrawal of the receptor from the nucleus, inhibiting the transcription of glucocorticoid-dependent genes [23, 21]. The replacement of serine by alanine (Ser266A1a) in this locus induces the loss of sensitivity of GR receptor to the effect of JNK [23]. The activation of JNK occurs with the participation of cytokines, lipopolysaccharides and osmotic stimuli [23] and the phosphorylation by Ser226 residue may be the mechanism of contra-regulation of the systemic immune and stress response.

Besides the phosphorylation, GR protein is affected by the recently discovered process of sumoylation, conjugated with specific small ubiquitin-related modifier (SUMO-1) [24]. The process is catalyzed by E3-ligases, which are different from the ligases, participating in the ubiquitination. The sumoylation modifies the traffic and the activity of a number of proteins. GRs may be sumoylated in three loci, related to the transactivational domain (Lys277 and Lys293) and the ligand-binding domain in the C-terminal region (Lys703) which leads to the modification of the transcriptional activity of the receptor, and the observed effect is promotor-dependent [24]. The sites of the transactivational domain are more susceptible to this type of modification compared to those of the ligand-binding domain. The glucocorticoid hormones are likely to stimulate the sumoylation of the receptor via its translocation into the nucleus with the corresponding E3-ligase (Ubc9) [24].

The glucocorticoid receptor may function in two modes: as a transcription factor, binding to the glucocorticoid-responsive elements of DNA (GRE) and as a regulator of the activity of other transcriptional factors. In 2009 it was established that different GR-binding nucleotide sequences of DNA serve as cofactors in the activity of the receptor, specifically and differentially changing its conformation and thus affecting the expression of some genes [25].

Receptor degradation. Similar to the majority of other nuclear receptors, GR is a substrate of the ubiquitin-dependent proteasome degradation [14, 26, 27]. CHIP complexes [26] and the protein Mdm2 (E3-ligase) participate in the processes of the proteasome degradation of receptors [28].

The recruiting of various subunits of the proteasome and E3-ligases into the promotor region during the activation of the steroid hormone receptors is a required condition for their effect, and the blockade of proteasome proteolysis may cause the association of the receptors and chromatin [26]. On the other hand, the

Table4
Postranslational modification GR-αA sites

№	Position	Aminoacid	Modification	Kinase	Significance	Reference
1	8	Threonine	Phosphorylation	–	–	–
2, 4	45, 134	Serine	Phosphorylation	–	–	–
3, 5	133, 141	Serine	Phosphorylation	–	–	[20, 42]
7, 8	203, 211	Serine	Phosphorylation	Cdk	Activates the receptor after hormone-binding	[22]
9	226	Serine	Phosphorylation	ERK2, MAPK-8, JNK	Decreases receptor activity	[21, 23]
10	234, 267	Serine	Phosphorylation	–	–	–
11	277, 293	Lysine	Sumoylation	Ubc9 (E3-лигаза)	–	[24]
12	419	Lysine	Ubiquitination in DNA-binding domain	CHIP, Mdm2 (E3-лигаза)	Possible PEST-marker of degradation	[26–28]
13	508	Serine	Phosphorylation of the hinged domain	DNA-dependent protein kinase	Effects the transcriptional activity	[43]
14	703	Lysine	Sumoylation	Ubc9 (E3-лигаза)	–	[24]

inhibition of proteasome degradation of proteins leads to the enhanced transcriptional activity in tissues which may be related to the impaired degradation and persistent activity of the transcription factors in the nucleus. Another mechanism of the participation of proteasome degradation in the regulation of the activity of the glucocorticoid receptors is the stimulating effect of various agents, estrogens, in particular, on the expression of the specific E3-ligase Mdm2, inducing the decrease in GR activity [28]. Therefore, the activity of the proteasome is a key element in the dynamic regulation of the activity of steroid hormones.

Genetic polymorphism of *NR3C1* gene. The polymorphism of genes, participating in the implementation of the stress response, is likely to play the key role, determining the discrepancies between the stress-responsiveness and resistance in the human population [1]. According to the data of the National Institutes of Health (USA) there are currently 2,571 known polymorphisms of human *NR3C1* gene of the single nucleotide polymorphism type (SNP, Fig. 4); among these polymorphisms, the incidence degree of the minor allele for 167 and 127 of them is over 10% and 1%, respectively. We have discovered 42 missence-mutations among all SNPs, inducing the substitution of some amino acids in the structure of different isoforms of GR receptors. In addition to SNP, there are scientific literature data on the familial mutation of *NR3C1* gene with the deletion of four nucleotides, related to the intron and exon sites and leading to the termination of the expression of the damaged gene [29].

Missence polymorphisms. There are data on 42 missence SNPs in each of four domains of the proteins (Table 5). This Table demonstrates that the highest clinical significance is attributed to the polymorphisms, related to the transactivational and ligand-binding domains. The main impairments of the receptor function for these polymorphisms are as follows:

1) retardation of the activated receptor traslocation into the nucleus (proven for rs104893912, rs121 909727 and rs104893908);

2) decreased affinity to the hormone (proven for rs121909727, rs104893908) or co-activators (proven for rs104893912, rs121909727);

3) the receptor instability (proven for rs104893908);

4) decrease in the transactivational activity (proven for rs104893912, rs121909727 and rs104893914; assumed for rs148967394).

Among the polymorphisms given in Table 5, rs121909726, rs56149945, rs104893909, rs104893910, rs104893911, rs104893912,

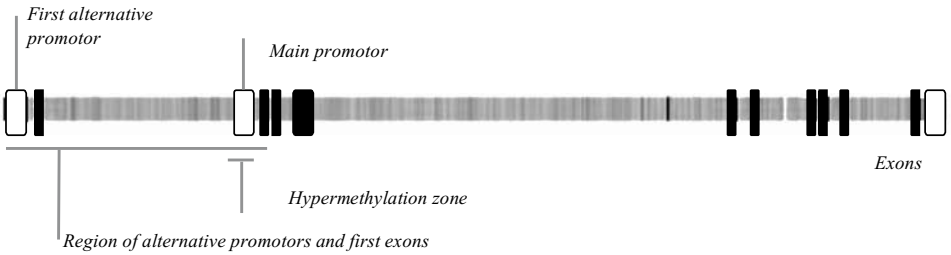

Fig. 4 Regional density of SNPs in various sites of *NR3C1* gene

rs104893913, rs104893914, rs121909727 have been described in humans. All of them are the consequences of missence mutations, impairing the protein structure, and each of them induces the development of generalized familial forms of the glucocorticoid resistance. In case of rs104893911 the resistance is accompanied by the hypokalemia and pseudohermaphroditism [30]. The main symptom of the glucocorticoid resistance is the high level of cortisone and ACTH in the blood plasma which may be related to the increase in the concentration of mineral-corticoid hormones and sex steroids [30].

The majority of the missence polymorphisms, studied in the human population, are remarkable for rather low frequency of incidence; there are rare homozygotes of mutant alleles, which may be explained by their inviability (see Table 5). In its turn, the low frequency of heterozygotes indicates a considerable effect of the negative natural selection regarding the mutations of *NR3C1* gene [31] which may be related to its significant role in the stress conditions and adaptation processes.

Polymorphisms of non-coding sites of the gene and other forms of non-missence mutations. The majority of SNPs in humans belong to intron sequences of the gene, which are adjoining the exons [31]. There are also some known mutations in the alternative exons, coding for the GR receptor isoform and the mutations, adding some more amino acids to the existing exons. The general description of these SNPs is presented in Table 6.

Among all the polymorphisms, listed in Table 6, noteworthy are two polymorphisms of the highest clinical relevance: rs6198 and rs41423247.

The polymorphism rs6198 is the replacement of nucleotide A by G in position 3669 (exon 9β) [31]. The mutant variant leads to the change in the stability of variant GR-β mRNA, and thus to its intensified synthesis. This GR isoform plays the role of dominant inhibitor of the active alpha-form of the receptor, inducing the reduced response to glucocorticoids. Therefore, the mutant form of the gene exhibits in a lower basal level of arterial pressure [31] as well as in an increased risk of rheumatoid arthritis and systemic lupus erythematosus [32] which might be explained by enhanced anti-inflammatory response, caused by the decreased sensitivity to glucocorticoids [31]. In addition, the formation of SNP by rs10482605 is facilitated by the haplotype, related to the increased risk of developing the major depression [5]. In total, the frequency of the mutant form in the Caucasian population of the USA is 19 %. SNP is remarkable for its dominant character which allows considering heterozygotes as a risk group for the described diseases.

Another widespread polymorphism rs41423247, also called Bc1I, is related to the replacement of C by G in position 41503, corresponding to the intron region of the gene [33]. It suggests the involvement of the mutation into the mechanisms of alternative splicing of mRNA and the changed products of receptor isoforms. The incidence degree for the minor allele is 26.4 % which defines this polymorphism as one of the most widespread variants of *NR3C1* gene in the human population. SNP causes enhanced sensitivity to glucocorticoid hormones [33]. Many studies revealed that the carriers of Bc1I have increased risk of recurrent miscarriage [34], bronchial asthma [35], juvenile idiopathic arthritis [36] which may be caused by the decreased level of corticosterone, observed due to the enhanced sensitivity of GR receptors and the activated negative feedback loop of the regulation of HPA activity [37].

Other forms of polymorphisms with clinical description. Among all the known polymorphisms of *NR3C1* gene a special place is reserved to the mutations, revealed either very rarely or in families and remarkable for their vivid clinical picture. These

Table5
Missens SNPs

dbSNP ID	Replacement	Position in GR-α protein	Protein domain	Frequencies (wild/hetero/mutant)	Clinical state	Mechanism
1	2	3	4	5	6	7
rs61759024	Pro → Ser	9	TAD	99,9/0,1/0; *N* = 4428	n/d	Close of Tre8 phosphorylation site
rs6190 ER22/23EK	Arg → Lys	23	TAD	90,3/9,7/0; *N* = 226 (европ.)	Associated with glucocorticoid resistance at Kron's disease [44]; one work demonstrates the association of cortisone products at social stress [45], while another one does not [46]	
rs72481829	Asp → Asn	25	TAD	0,6/0/99,4; *N* = 356	n/d	n/d
rs148102613	Phe → Leu	29	TAD	100/0/0; *N* = 4550	n/d	n/d
rs143711342	Tyr → His	30	TAD	99,0/0,1/0; *N* = 4550	n/d	n/d
rs148967394	Ser → Pro	44	TAD	100/0/0; *N* = 4550	n/d	Adjoins Ser45 phosphorylation site, which may result in the decrease in transactivational ability of receptors
rs79138720	Val → Gly	50	TAD	Non-clinical source	n/d	n/d
rs6192	Phe → Val	65	TAD	99,2/0,8/0; *N* = 4450	No effect of the basal level of arterial pressure [31]	
rs145020010	Met → Val	98	TAD	100/0/0; *N* = 4222	n/d	n/d
rs72542740	Gly → Arg	99	TAD	99,8/0,2/0; *N* = 526	n/d	n/d
rs72481830	Asn → Ser	130	TAD	99,4/0,6; *N* = 360	n/d	n/d
rs141093427	Phe → Leu	156	TAD	100/0/0; *N* = 4548	n/d	n/d
rs186831584	Val → Ile	163	TAD	n/o	n/d	n/d
rs146524172	Asn → Ser	180	TAD	100/0/0; *N* = 4550	n/d	n/d
rs140309412	Asn → Ile	222	TAD	100/0/0; *N* = 4224	n/d	n/d
rs72542742	Ala → Thr	229	TAD	99,7/0,3/0; *N* = 4132	n/d	n/d
rs183372229	Leu → Phe	286	TAD	n/o	n/d	n/d
rs72542743	Ile → Val	292	TAD	Alleles: 99,8/0,2; *N* = 526	Close to Lys419 sumoylation site	
rs72542745	Ser → Gly	325	TAD	Alleles: 99,8/0,2; *N* = 526	n/d	n/d
rs72558022	Asp → His	346	TAD	94,4/5,6/0; *N* = 18	n/d	n/d
rs148470701	Gln → Glu	347	TAD	n/o	n/d	n/d
rs6195N363S	Asn → Ser	363	TAD	95,8/4,2/0; *N* = 48		
rs56149945	Asn → Ser Asn → Ile	363	TAD	95,9/4,0/0/0; *N* = 4550	Known clinical case	
rs1800445	Asn → Ser	365	TAD	n/o	n/d	n/d

Окончание табл. 5

1	2	3	4	5	6	7
rs147136661	Ser → Phe	370	TAD	100/0/0; *N* = 4544	n/d	n/d
rs145046100	Thr → Ala	413	TAD	n/o	n/d	n/d
rs113048309	Ser → Pro	425	DBD	n/o	n/d	n/d
rs104893913	Arg → His	477	DBD	n/o	n/d	n/d
rs72542747	Thr → Ser	504	HD	n/o	n/d	n/d
rs72481843	Gly → Ala	516	HD	Single case	n/d	n/d
rs33391	Asn → Lys	517	HD	Eight cases, not observed in the population		
rs104893911	Val → Ala	571	LBD	?		Possible pathogenecity
rs104893909	Ile → Asn	559	LBD	Single case	n/d	n/d
rs104893908	Asp → Val	641	LBD	Single case	PGR	Reduced affinity to the hormone, impaired transportation into the nucleus, receptor instability [39]
rs113100205	Val → Ile	658	LBD	Одна семья	n/d	n/d
rs104893914	Gly → Ser	679	LBD	n/o	PGR	Reduced transactivational effect [47]
rs68012717	Asp → Glu	687	LBD	n/o		
rs121909727	Phe → Leu	737	LBD	Single case	TGR	Reduced affinity to the hormone, delay in nuclear translocation and impaired interaction with NCOA2 (GR-interacting protein-1 co-activator) [48]
rs104893910	Ile → Met	747	LBD	?		
rs121909726	Leu → Phe	753	LBD	n/o		
rs186936077	Asn → Asp	766	LBD	n/o		
rs104893912	Leu → Pro	773	LBD	Single case	TGR	Two-fold reduction in transactivational ability, dominant-negative effect on wild type of the receptor, slowed-down translocation into the nucleus, impaired interaction with NCOA2 [49]

Note. DBD – DNA-binding domain; LBD – ligand-binding domain; TAD – transactivational domain; HD – hinged domain; PGR and TGR – primary and total glucocorticoid resistance; n/o – no observation; n/d — no data.

mutations promote deeper understanding of the functional relevance of the receptor and the mechanisms of controlling its activity. Below is a short description of two mutations, described in scientific literature.

Table 6
The main clinically significant SNP in noncoding regions of the NR3C1gene

dbSNP ID	TypeSNP	Minor allele frequency	Mutation region	clinical significance
rs7701443	A/G	G: 46,8 %; *N* = 1022	Intron	Resistance to corticosteroids in children with Crohn's disease [44]
rs9324924	G/T	T: 44,8 %; *N* = 978	Intron	Data on the relationship with glucocorticoid resistance have not been identified [50]
rs4912911	A/G	G: 44,8 %; *N* = 1000	Intron	Recessive resistance to corticosteroids [44]
rs4607376	A/G	G: 43,8 %; *N* = 957	Intron	Data on the relationship with glucocorticoid resistance have not been identified [50]
rs6191	G/T	G: 42,3 %; *N* = 923	Exon 9β/3'-flanking region	Possible relation with lithium resistance in patients with bipolar disorder [51] due to depression [51]
rs33388	A/T	A: 41,6 %; *N* = 908	Intron	Possible relation with lithium resistance in patients with bipolar disorder [51] due to depression [51], there is no association with rheumatoid arthritis [51] is not shown due to depression in women in pre-menopausal women [52]
rs6198	A/G	G: 9,2 %; *N* = 201	Exon 9β/3'-flanking region	See description in text
rs41423247	C/G	C: 27,7 %; *N* = 604	Intron	See description in text
rs6196	A/G	G: 13,8 %; *N* = 302	Additional Asn in exon 5	Resistance to corticosteroids in children with Crohn's disease [44], communication with overweight [54], there is no inter-connection with the response to psychosocial stress in children, the association to high blood pressure [31]

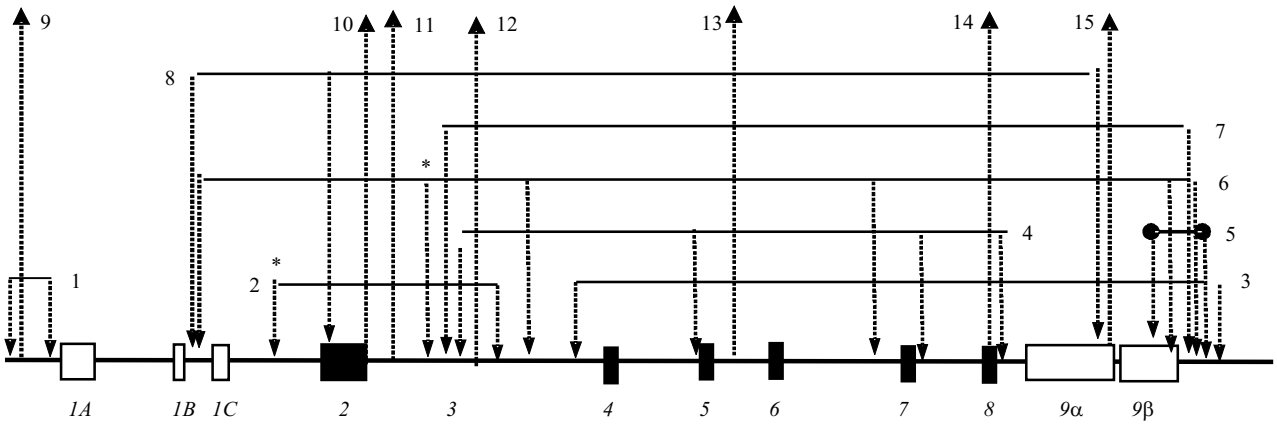

Fig. 5 The scheme of SNPs in the composition of the main haplotypes of *NR3C1* gene (according to [31]). Detailed description of SNP and haplotypes is presented in Table 7.

In 1976 the authors of [38] described a medical case, when a patient had a high level of cortisone and ACTH associated with hypertension and hypokalemia with no other deviations. The proband's DNA demonstrated the mutation of SNP type, consisting in the substitution of valine for asparagine in position 641 of GR-α, corresponding to the hormone-binding domain (rs104893908) [39]. The mutation induced the decrease in the affinity to cortisone, the impaired transportation of the receptor into the nucleus and its total instability [39].

Noteworthy are three medical cases, revealed by the authors of [29] in one Danish family, caused by the deletion of four nucleotides in the donor splice site (here the pairs of nucleotides belonged to the sites of sixth intron and exon). The carriers of the clinical pathology were the father and three of five children; all these patients had evident hypercortisolism, caused by the functioning of only half of GR receptors. The proband was the daughter with hyperandrogenism, caused by the compensatory hyperproduction of the corticosteroid hormones [29].

Haplotypes of *NR3C1* gene in humans. The work [31] presents the detailed study of the *NR3C1* gene haplotypes, consisting of various combinations of separate SNP, widespread among the main ethnic groups of American population. There were eight main revealed haplotypes, consisting of two and more SNP, which occur in the associations at the values of correlation coefficient 0.8 and over (Fig. 5, Table 7). In addition, the authors distinguished seven additional haplotypes, containing only one SNP, which are either rather frequent among Americans or of considerable relevance. It was established that haplotypes No. 2 and 3 are notable for their statistically reliable relation to the increased levels of the arterial pressure [31].

Unfortunately, at present we are unsuccessful in finding other studies on the *NR3C1* gene haplotypes with sufficient representation.

Conclusions. Modern medical science is moving towards combining the results of genetic and molecular biological investigations with the clinical practice. The focus of the scientists tends to shift from the study on the monogenic diseases of low incidence level, but with a vivid and specific clinical picture, to the analysis of molecular mechanisms of polygenic, multifactorial diseases, which are widespread and remarkable for many clinical variants and types of the clinical course. In this regard there is a special interest to the glucocorticoid hormone receptors, which are one of the key regulators of immunological, stress and adaptation processes. The study on the relation of molecular biology and genetics of the GR receptors to the development of various forms of clinical pathological states and diseases offers new opportunities for further research in the sphere of pathophysiology and therapy of the diseases of civilization.

Table7
Description of the main gene NR3C1 haplotypes , in the African Americans, Mexican and European Americans population ([31])

haplotype	SNP ID	SNP region
1	–1225	Distal promoter
	rs6868190	Distal promoter
2	rs10482616	Intron 1C
	rs10482672	Intron 3
3	rs852978	Intron 3
	rs6196	Exon 9α (additional Asn)
	rs258748	3'-flanking region
4	rs852979	Intron 3
	rs6188	Intron 4
	rs258813	Intron 7
	rs258750	Intron 8
5	rs6191	Exon 9β/3'-flanking region
	rs258747	3'-flanking region
6	rs10482605	Proximal promoter
	rs10482605	Intron 2
	rs123324	Intron 3
	rs10482689	Intron 6
	rs6198	Exon 9β/3'-flanking regionь (AUUUA-sequence)
	rs17287758	3'-flanking region
7	rs4986593	Intron 2
	rs17209237	Flanking region
8	rs10482604	Proximal promoter
	rs6192	Exon 2 (Phe → Val)
	rs10043662	Exon 9α/3'-flanking region
9	–921	Distal promoter
10	rs6195	Exon 2 (Asn → Ser)
11	rs41423247	Intron 2 (BclI)
12	rs10482669	Intron 3
13	rs10482682	Intron 5
14	rs258751	Exon 8 (additional Asp)
15	rs6193	Exon 9α/3'-flanking region

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Аллельный полиморфизм рецептора глюкокортикоидных гормонов NR3C1 (GR):
от молекулярной биологии к клинике

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Резюме

Полиморфизм генов, участвующих в реализации стрессорного ответа, является одним из ключевых фак- торов, определяющих различия в стресс-реактивности и резистентности в человеческой популяции. Среди генов – регуляторов стресса в первую очередь следует выделить гены рецепторов глюкокортикоидов. В обзоре дана детальная характеристика их молекулярной биологии, на основании чего проведен анализ возможной связи наиболее распространенных вариантов SNP с альтерацией стрессорных реакций и развитием клинической патологии.

Ключевые слова: глюкокортикоидные рецепторы, аллельный полиморфизм, SNP, NR3C1, GR, болезни адаптации.

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Алельний поліморфізм рецептора глюкокортикоїдних гормонів NR3C1 (GR): від молекулярної біології до клініки

Резюме

Поліморфізм генів, які беруть участь у реалізації стресорної відповіді, є одним із ключових факторів, що визначають розбіжності в стрес-реактивності і резистентності в людській популяції. Серед генів – регуляторів стресу у першу чергу варто відзначити гени рецепторів глюкокортикоїдів. В огляді надано детальну характеристику молекулярної біології даних генів, на підставі чого зроблено аналіз можливого зв'язку найрозповсюдженіших варіантів SNP з альтерацією стресорних реакцій та розвитком клінічної патології.

Ключові слова: глюкокортикоїдні рецептори, алельний поліморфізм, SNP, NR3C1, GR, захворювання адаптації.

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