THE IMPACT OF POLYMORPHISMS IN ANTIOXIDANT GENES ON THE RISK OF MALIGNANT NEOPLASM DEVELOPMENT IN EXPOSED INDIVIDUALS

Blinova EA ✉, Korechenkova AV, Yanishevskaya MA, Akleyev AV

Urals Research Center for Radiation Medicine of the Federal Medical Biological Agency, Chelyabinsk, Russia

In the context of additional radiation exposure, single nucleotide polymorphisms in the genes encoding the antioxidant system enzymes can contribute to the oxidative stress enhancement, damage to DNA, and therefore lead to the increase in the risk of malignant neoplasm (MN) development. The study was aimed to determine the association of the *CYBA* (rs4673), *GPX1* (rs1050450), *MPO* (rs2333227), *CAT* (rs7943316), *SOD2* (rs4880) polymorphic loci with the risk of MN development in individuals affected by low dose rate chronic radiation exposure considering intergenic interactions and the radiation dose. Two groups of individuals were included in the study: exposed individuals with no MNs — 384 people with the mean accumulated dose to the red bone marrow (RBM) of 796.95 ± 35.97 mGy; exposed individuals with the history of MNs — 227 people with the mean accumulated dose to RBM of 520.06 ± 38.72 mGy. Amplification of the rs4880, rs2333227, rs7943316, rs4673, rs1050450 polymorphic loci was performed with real time PCR. Compliance with the Hardy–Weinberg equilibrium was reported for all gene polymorphisms. It has been found that the rs4880*C (SOD2) and rs1050450*T (*GPX1*) alleles are associated with the risk of MN development in accordance with the dominant (OR = 1.49 (1.02–2.18), p = 0.04) and recessive (OR = 2.00 (1.11–3.62), p = 0.02) inheritance modes, respectively. An interfactor interaction model with the 100% reproducibility and 66% accuracy (p = 0.001) has been obtained that includes the *SOD2* (rs4880), *CYBA* (rs4673) polymorphisms and the factor of accumulated dose to RBM. Thus, polymorphic loci of the genes regulating the oxidative status of the cells are associated with the increased risk of MN development in individuals, who have experienced chronic radiation exposure with predominant exposure of RBM.

Keywords: single nucleotide polymorphism, chronic radiation exposure, Techa River, antioxidant system, malignant neoplasm

Funding: the paper was prepared during implementation of a part of the research project (contract No. 635/FMBC/23 dated 29.11.2023).

Author contributions: Blinova EA — developing the method, manuscript writing; Korechenkova AV — statistical data processing, manuscript writing; Yanishevskaya MA — laboratory tests, manuscript writing; Akleyev AV — study conception and design, manuscript writing, academic advising.

Compliance with ethical standards: all the subjects submitted the form of the informed consent to study participation and biomaterial sampling and storage in the tissue bank finalized in the study protocol approved by the Ethics Committee of the Urals Research Center for Radiation Medicine of FMBA of Russia (protocol No. 2 dated 13 April 2023) before inclusion in the study.

✉ **Correspondence should be addressed:** Eugenia A. Blinova
Vorovsky, 68-A, Chelyabinsk, 454141, Russia; blinova@urcrm.ru

Received: 25.03.2024 **Accepted:** 03.06.2024 **Published online:** 26.06.2024

DOI: 10.47183/mes.2024.022

ВЛИЯНИЕ ПОЛИМОРФИЗМА В ГЕНАХ АНТИОКСИДАНТОВ НА РИСК РАЗВИТИЯ ЗЛОКАЧЕСТВЕННЫХ НОВООБРАЗОВАНИЙ У ОБЛУЧЕННЫХ ЛЮДЕЙ

Е. А. Блинова ✉, А. В. Кореченкова, М. А. Янишевская, А. В. Аклеев

Уральский научно-практический центр радиационной медицины Федерального медико-биологического агентства, Челябинск, Россия

На фоне дополнительного радиационного воздействия однонуклеотидные полиморфизмы в генах, кодирующих ферменты антиоксидантной системы, могут способствовать усилению окислительного стресса, возникновению повреждений ДНК и, как следствие, приводить к повышению риска развития злокачественных новообразований (ЗНО). Целью работы было установить связи полиморфных локусов *CYBA* (rs4673), *GPX1* (rs1050450), *MPO* (rs2333227), *CAT* (rs7943316), *SOD2* (rs4880) с риском развития ЗНО у лиц, подвергшихся хроническому низкоинтенсивному радиационному воздействию, с учетом межгенных взаимодействий и дозы радиационного облучения. В исследование были включены две группы людей: облученные лица без ЗНО — 384 человека со средней накопленной дозой облучения красного костного мозга (ККМ) 796,95 ± 35,97 мГр; облученные лица с ЗНО в анамнезе — 227 человек со средней накопленной дозой облучения ККМ 520,06 ± 38,72 мГр. Амплификацию полиморфных локусов rs4880, rs2333227, rs7943316, rs4673, rs1050450 проводили методом ПЦР в реальном времени. Для всех полиморфных участков генов выявлено соответствие равновесию Харди–Вайнберга. Обнаружено, что аллели rs4880*C (SOD2) и rs1050450*T (GPX1) ассоциированы с повышенным риском развития ЗНО согласно доминантной (ОШ = 1,49 (1,02–2,18), p = 0,04) и рецессивной (ОШ = 2,00 (1,11–3,62), p = 0,02) моделям наследования соответственно. Получена модель межфакторных взаимодействий со 100%-й воспроизводимостью и точностью 66% (p = 0,001), включающая в себя полиморфизмы *SOD2* (rs4880), *CYBA* (rs4673) и фактор накопленной дозы облучения ККМ. Таким образом, полиморфные локусы генов, регулирующих оксидантный статус клеток, связаны с повышенным риском развития ЗНО у лиц, подвергшихся хроническому радиационному воздействию с преимущественным облучением ККМ.

Ключевые слова: однонуклеотидный полиморфизм, хроническое облучение, река Теча, антиоксидантная система, злокачественное новообразование

Финансирование: статья подготовлена в рамках выполнения составной части научно-исследовательской работы (контракт № 635/ФМБЦ/23 от 29.11.2023).

Вклад авторов: Е. А. Блинова — постановка методики, написание статьи; А. В. Кореченкова — статистическая обработка данных, написание статьи; М. А. Янишевская — лабораторные исследования, написание статьи; А. В. Аклеев — концепция исследования, написание статьи, научное руководство.

Соблюдение этических стандартов: все участники добровольно подписали форму информированного согласия на участие в исследовании и забор биологического материала в банк тканей, утвержденную в протоколе исследования, одобренном этическим комитетом ФГБУН УНПЦ РМ ФМБА России (протокол № 2 от 13 апреля 2023 г.), до включения в исследование.

✉ **Для корреспонденции:** Евгения Андреевна Блинова
ул. Воровского, д. 68, корп. А, г. Челябинск, 454141, Россия; blinova@urcrm.ru

Статья получена: 25.03.2024 **Статья принята к печати:** 03.06.2024 **Опубликована онлайн:** 26.06.2024

DOI: 10.47183/mes.2024.022

The mechanisms underlying the damaging effects of ionizing radiation are closely related to the oxidative stress enhancement in exposed cells [1]. The increase in the levels of reactive oxygen species (ROS) contributes to damage to macromolecules, including proteins, nucleic acids, and lipids, which results in DNA dysfunction and damage, as well as in the apoptotic cell death [2]. The main role in antioxidant defense is played by glutathione peroxidase (*GPX* gene), catalase (*CAT* gene), manganese-dependent superoxide dismutase (*SOD2* gene), myeloperoxidase (*MPO* gene), and cytochrome b-245 (*CYBA* gene). Single nucleotide polymorphisms (SNPs) in the genes encoding antioxidant enzymes can contribute to alteration of the enzyme activity and enzyme function impairment [3]. In particular, alteration of superoxide dismutase and glutathione peroxidase activity and the decrease in their ability to neutralize free radicals is observed in individuals having unfavorable alleles in *SOD2* (rs4880) and *GPX1* (rs1050450) polymorphic loci, respectively [4]. Catalase is an important endogenous antioxidant enzyme that catalyzes decomposition of hydrogen peroxide into oxygen and water, thereby neutralizing the ROS harmful effects. The rs7943316 polymorphic locus in the promoter region of the *CAT* gene can modify the transcription factor binding affinity. Due to the presence of mutant allele T, abnormal transcription factor binding can result in the promoter activity, gene expression alteration, and the decrease in the enzyme catalyst activity [5]. The decrease in the catalyst activity of antioxidant enzymes, in its turn, increases susceptibility to oxidative stress. The rs2333227 polymorphic locus located in the promoter region of the gene encoding myeloperoxidase downregulates the *MPO* gene, thereby disrupting the SP1 transcription factor binding site. It has been found that substitution of the G base with A base is associated with the decreased expression of the *MPO* gene mRNA and the decrease in the amount of enzyme, while the G allele, in contrast, is associated with the increased MPO production [6]. A number of polymorphisms in the *CYBA* promoter and exon regions affect the gene expression and the NADPH oxidase activation, which results in the increased production of free radicals along with the detected antioxidant deficiency [7]. All of these testify to the fact that the presence of SNP in the genes encoding the antioxidant system enzymes can have an effect on both quantitative and functional characteristics of the enzyme, as well as modify the radiation effects in case of ionizing radiation exposure.

The study was aimed to determine the association of the *CYBA* (rs4673), *GPX1* (rs1050450), *MPO* (rs2333227), *CAT* (rs7943316), *SOD2* (rs4880) polymorphic loci with the risk of malignant neoplasm (MN) development in individuals with chronic low dose rate exposure considering intergenic interactions and the radiation dose.

METHODS

Genotyping based on the *CYBA* (rs4673), *GPX1* (rs1050450), *MPO* (rs2333227), *CAT* (rs7943316), *SOD2* (rs4880) polymorphic markers was performed in individuals, who lived in the radioactively contaminated areas along the Techa River and were affected by chronic low dose rate exposure in the low to medium dose range [8]. All the patients enrolled were admitted to the Clinical department of the Urals Research Center for Radiation Medicine of FMBA of Russia (URCRM) in 2003–2023; blood samples collected from these patients were stored in the URCRM tissue bank. The inclusion criteria for all the examined individuals were as follows: residence in one of 41 villages located in the territory adjacent to the Techa River at any time between 1 January 1950 and 31 December 1960; availability of the individual accumulated dose to the red bone marrow (RBM) calculated using the Techa River Dosimetry System-2016 (TRDS-2016) [9]. The exclusion criteria for all the examined individuals were as follows: hematological disorders and the lack of information about the past medical history.

The examined individuals (611 people) were divided into two groups: those, who were chronically exposed on the Techa River and had no MNs, — 384 individuals; those, who were chronically exposed on the Techa River and had a history of MNs of various localization, — 227 individuals. The detailed characteristics of the studied groups are provided in Table 1.

The accumulated dose to RBM of the examined individuals was within the range of 0.85–3 715.72 mGy, and the mean dose did not exceed 659 mGy. No significant differences in the accumulated dose to RBM between the groups of exposed individuals with and without MNs were revealed ($p > 0.05$). The studied groups were matched by sex, ethnicity, and age.

The exposed residents of the Techa riverside villages had the following solid cancers: MNs of the digestive system — 49 individuals (ICD-10 codes: C00, C02, C04, C15, C16, C18.4, C19, C22.7, C25.9, C26), respiratory system — 28 individuals (ICD-10 codes: C30, C32.9, C34), skin — 44 individuals (ICD-10 codes: C43.9, C44), female reproductive system — 70 individuals (ICD-10 codes: C50, C53, C54, C56), male reproductive system — eight individuals (ICD-10 codes: C61, C63), urinary system — 16 individuals (ICD-10 codes: C64, C67), endocrine system — 13 individuals (ICD-10 code: C73). Furthermore, MNs of bone and articular cartilage — one individual (ICD-10 code: C40), brain and nervous system — two individuals (ICD-10 code: C71, C72), eye and adnexa — two individuals (ICD-10 code: C69), MNs without specification of site — four individuals (ICD-10 code: C80), carcinoma *in situ* unspecified — three individuals (ICD-10 code: D09), and MNs of uncertain or unknown behavior of other and unspecified

Table 1. Characteristics of the studied groups

Parameter		Individuals exposed on the Techa River with no MNs ($n = 384$)	Individuals exposed on the Techa River with the history of MNs ($n = 227$)
Sex, *n* (%)	Male Female	124 (32.29) 260 (67.71)	83 (36.56) 144 (63.44)
Ethnicity, *n* (%)	Slavs Turkic people	138 (35.94) 246 (64.06)	104 (45.81) 123 (54.19)
Age at the time of examination, years; mean ± SD (min–max)		73.91 ± 9.04 (43.00–97.00)	73.42 ± 8.82 (47.00–95.00)
Accumulated dose to RBM, mGy; mean ± SE (min–max)		796.95 ± 35.97 (1.69–3715.72)	520.06 ± 38.72 (0.85–3507.07)

Note: mean ± SD (min–max) — mean ± standard deviation (min–max); mean ± SE (min–max) — mean ± standard error of the mean (min–max).

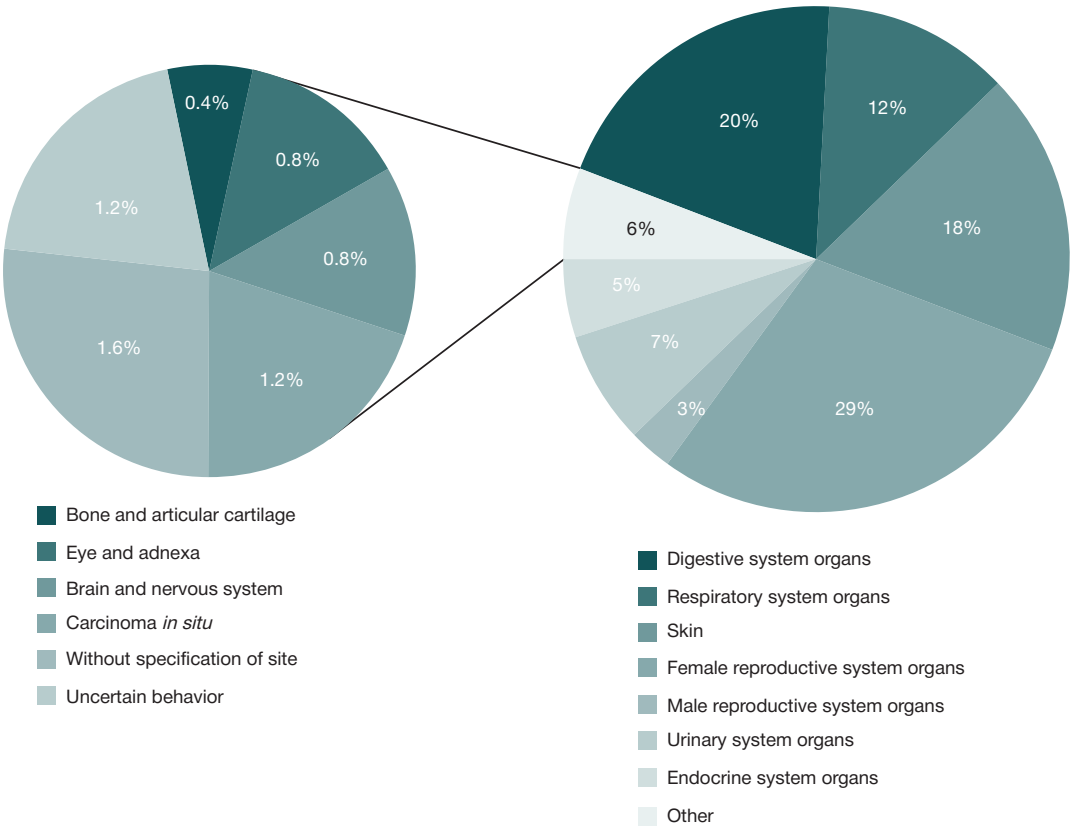

Fig. 1. Distribution of MNs in the group of exposed individuals depending on the cancer localization

sites — three individuals (ICD-10 code: D48) were reported in the studied cohort. The distribution of MNs in the group of exposed individuals is presented in Fig. 1.

Peripheral blood for testing was collected to the 9 mL Vacuette test tubes (Greiner Bio-One; Austria) covered with the K3-EDTA blood anticoagulant. Genomic DNA was isolated on the spin columns using the ExtractDNA Blood & Cells kit (Evrogen; Russia) in accordance with the manufacturer's protocol.

SNPs of the candidate genes were selected for the study based on the location of the polymorphic locus in the gene, as well as the association with MN revealed by analysis of the HapMap (URL: hapmap.ncbi.nlm.nih.gov), NCBI (URL: https://www.ncbi.nlm.nih.gov/), SNPedia (URL: https://snpedia.com/) databases. Table 2 provides characteristics of the studied gene polymorphic regions.

Amplification of the studied polymorphic loci was performed by real-time PCR on the StepOnePlus Real-Time PCR System (Applied Biosystems; USA) using the reagent kits synthesized by TestGen OOO (Russia) in accordance with the manufacturer's protocol.

Deviation of the frequency distribution for genotypes of the studied polymorphisms from the expected Hardy–Weinberg equilibrium distribution was estimated using the chi-squared test (χ^2) in the Gene Calc online calculator (URL: https://gene-calc.pl/hardy-weinberg-page). The odds ratio (OR) with the 95% confidence interval was calculated to estimate the relationship of alleles in the polymorphic loci with the risk of MN development. The relationship was considered significant at $p < 0.05$.

Non-parametric Multifactor Dimensionality Reduction method (MDR software package v. 3.0.2, available at http://sourceforge.net/projects/mdr) [10] was used to estimate intergenic interactions and radiation doses. During such analysis the multilocus genotypes and factors are combined into groups with the increased and decreased risk of the disease development to reduce dimensionality of the number of parameters calculated. Thus, the optimal factor interaction model allowing one to predict susceptibility/no susceptibility to certain disorders is selected among all the proposed variants of models constructed based on the entered primary data through repeated verification [11]. Optimality of the resulting models was assessed based on their reproducibility based on the findings of the cross validation consistency (CVC) and the testing balanced accuracy (TBA). The model should be reproduced at least 9 times out of 10 and the model accuracy should exceed 55%. P-value for the testing balanced accuracy

Table 2. Characteristics of polymorphic regions

Gene	SNP	Alleles	Minor allele	Position	Location
SOD2	rs4880	T/C	C	chr6:159692840	Missense variant
MPO	rs2333227	C/T	T	chr17:58281401	2KB Upstream Variant
CAT	rs7943316	A/T	T	chr11:34438925	2KB Upstream Variant
CYBA	rs4673	C/T	C	chr16:88646828	Missense variant
GPX1	rs1050450	C/T	T	chr3:49357401	Missense variant

Note: 2KB Upstream Variant — a sequence variant located within 2KB 5' of a gene.

Table 3. Abundance of the studied SNP genotypes in exposed individuals

Gene/SNP	Genotype	Exposed individuals without MNs				Exposed individuals with MNs			
		Number (%)	Ho	He	*p*	Number (%)	Ho	He	*p*
SOD2/rs4880	C/C C/T T/T	70 (19) 174 (47) 125 (34)	0.47	0.49	0.5	41 (20) 110 (54) 52 (26)	0.54	0.5	0.2
MPO/rs2333227	C/C C/T T/T	276 (72) 101 (27) 4 (1)	0.27	0.25	0.1	122 (74) 42 (25) 1 (1)	0.26	0.23	0.2
CAT/rs7943316	A/A A/T T/T	60 (16) 179 (48) 134 (36)	0.48	0.48	0.9	25 (13) 89 (46) 75 (39)	0.47	0.47	0.8
CYBA/rs4673	C/C C/T T/T	201 (54) 139 (37) 34 (9)	0.37	0.4	0.2	96 (49) 78 (40) 21 (11)	0.4	0.43	0.4
GPX1/rs1050450	C/C C/T T/T	182 (49) 162 (44) 28 (8)	0.44	0.41	0.3	73 (46) 62 (40) 22 (14)	0.4	0.45	0.1

Note: Ho — observed heterozygosity; He — expected heterozygosity; *p* — value for the Hardy–Weinberg test.

was determined using a 1000-fold permutation test. The differences were considered significant at $p < 0.05$. The factor interaction analysis results were visualized using the graphs plotted using the Fruchterman–Rheingold force-directed layout algorithm. The contribution of each factor and/or interaction of factors is measured using the enthropy value (H) expressed in %. Thus, the factor with the 100% enthropy unambiguously determines the class the individual belongs to (healthy/unhealthy individuals); therefore, the factor with 0% plays no role in susceptibility to the disease. To evaluate the effect of the dose the examined individuals were subdivided into three dose subgroups: 1 — 0.85–99 mGy; 2 — 100–999 mGy; 3 — ≥ 1000 mGy.

RESULTS

The results of assessing the distribution of SNP of the antioxidant system genes in the chronically exposed individuals are provided in Table 3.

No deviation from Hardy–Weinberg equilibrium was observed for all SNPs of the genes. Furthermore, the observed and expected heterozygosity values were similar for all SNPs, which indicated the random nature of the sample.

In the next phase of the study we performed analysis of the relationship of each gene polymorphism with the risk of MN development based on the recessive and dominant inheritance modes (Table 4).

The analysis showed that the rs4880*C and rs1050450*T alleles were associated with the increased risk of MNs (OR = 1.49 (1.02–2.18), $p = 0.04$) and OR = 2.00 (1.11–3.62), p = 0.02, respectively). Considering the fact that the complex cascade of interactions between the antioxidant system gene products and the contribution of the radiation exposure factor to the risk of MN development are not taken into account when assessing the associations of single alleles and genotypes, we performed the analysis of intergenic interactions with the accumulated dose yielding the 1n-, 2n-, 3n-, and n-factor models (Table 5). Simultaneous testing of all five SNPs and the

Table 4. Association of SNPs with the risk of MN development

Gene/SNP	Mode	Genotype		OR (95% CI)	*p*
		Exposed individuals without MNs	Exposed individuals with MNs		
SOD2/rs4880	Dominant	T/T (125) C/T-C/C (244)	T/T (52) C/T-C/C (151)	1.00 1.49 (1.02–2.18)	0.04
	Recessive	T/T-C/T (299) C/C (70)	T/T-C/T (162) C/C (41)	1.00 1.08 (0.70–1.66)	0.72
MPO/rs2333227	Dominant	C/C (276) C/T-T/T (105)	C/C (122) C/T-T/T (43)	1.00 0.93 (0.61–1.40)	0.72
	Recessive	C/C-C/T (377) T/T (4)	C/C-C/T (164) T/T (1)	1.00 0.57 (0.06–5.18)	0.6
CAT/rs7943316	Dominant	T/T (134) A/T-A/A(239)	T/T (75) A/T-A/A (114)	1.00 0.85 (0.59–1.22)	0.38
	Recessive	T/T-A/T (313) A/A (60)	T/T-A/T (164) A/A (25)	1.00 0.80 (0.48–1.32)	0.37
CYBA/rs4673	Dominant	C/C (201) C/T-T/T (173)	C/C (96) C/T-T/T (99)	1.00 1.20 (0.85–1.69)	0.31
	Recessive	C/C-C/T (340) T/T (34)	C/C-C/T (174) T/T (21)	1.00 1.21 (0.68–2.14)	0.52
GPX1/rs1050450	Dominant	C/C (182) C/T-T/T (190)	C/C (73) C/T-T/T (84)	1.00 1.10 (0.76–1.60)	0.61
	Recessive	C/C-C/T (344) T/T(28)	C/C-C/T (135) T/T(22)	1.00 2.00 (1.11–3.62)	0.02

Note: OR (95% CI) — odds ratio with the 95% confidence interval.

Table 5. Models of intergenic interactions with the accumulated dose in chronically exposed individuals

Model	Testing balanced accuracy	Cross validation consistency	*p*	Se	Sp
Dose to RBM	0.53	5/10	0.275	0.304	0.827
SOD2 rs4880, dose to RBM	0.55	6/10	0.162	0.761	0.454
SOD2 rs4880, dose to RBM, *CYBA* rs4673	0.66	10/10	0.001	0.75	0.601
SOD2 rs4880, dose to RBM, *CYBA* rs4673, *GPX1* rs1050450	0.57	10/10	0.062	0.837	0.578

Note: *p* — value for the testing balanced accuracy obtained in the 1000-fold permutation test; Se — sensitivity; Sp — specificity.

factor of accumulated dose to RBM was carried out. Table 5 presents the four best combinations of factors based on the modeling results. Further increase in the number of parameters significantly reduced the accuracy of the models presented.

Among all the models revealed, the 3-factor model including the *SOD2* (rs4880), *CYBA* (rs4673) polymorphisms and the accumulated dose to RBM was the most accurate (66%) and had 100% reproducibility ($p = 0.001$). The rs4880*C (genotypes C/C and C/T) and rs4673*T (genotypes T/T and T/C) alleles were reported as the ones associated with the increased risk of MN development. The *GPX1* (rs1050450) polymorphism was included in the 4n-factor model. However, specificity of such model decreased with increasing sensitivity, which affected accuracy. Other polymorphisms were considered insufficiently informative. The graph of interaction between the elements of the 3-factor model is provided in Fig. 2.

The *SOD2* (rs4880) polymorphism explaining 0.96% of phenotypic entropy (uncertainty) and the factor of the absorbed dose to RBM are of the highest informational value (1.24%). The *CYBA* (rs4673) informational contribution of 0.06% is the least, which is likely to play no significant role in the risk of MN development. However, it is worth considering interactions between all elements of the model that are of synergistic nature. Thus, the pair of rs4880 and rs4673 account for 0.61% of entropy; rs4673 and the accumulated dose to RBM account for 0.83%; rs4880 and the accumulated dose to RBM account for 0.21%.

DISCUSSION

ROS being the products of normal cell metabolism play an important role in stimulation of the intracellular signaling pathway in response to changes in the intra- and extracellular environment. The majority of ROS are generated in the mitochondrial respiratory chain [12]. However, imbalance between the free radical and reactive metabolite formation and elimination by means of the antioxidant system enzymes results in the development of oxidative stress. The factors contributing to oxidative stress are diverse: from lifestyle (smoking, alcohol consumption) to environmental exposures (chemical and radiation exposure). Furthermore, chronic diseases and inflammation can also be associated with oxidative stress. Eventually, oxidative stress results in damage to the important biomolecules and cell structures with potential consequences for the whole body [13]. In case of persistent oxidative stress, ROS are produced for a long time. This is how a significant impairment of the cell structure and functions can occur, which may lead to somatic mutations and neoplastic transformation [14]. The ability of antioxidants to neutralize the effects of free radicals may constitute an important component of the body's antitumor defense. The antioxidant system effectiveness is genetically determined. Overexpression or decreased activity of the antioxidant enzymes can modify the effects of radiation exposure [15].

Our study has shown that the *SOD2* rs4880*C and *GPX1* rs1050450*T alleles are associated with the increased risk of MN development in the individuals, who had chronic radiation exposure in a wide dose range, which is generally in line with the data reported for non-exposed people. According to the genomic assessment of oncogenicity based on the regBase prognostic model, rs4880 is considered to be likely pathogenic. According to the published data, rs4880 is associated with the increased risk of prostate cancer [16]. Furthermore, in individuals with the *SOD2* (rs4880) C/T and T/T genotypes, the measured *SOD2* enzyme activity was 33% lower than in carriers of the C/C genotype [17]. However, the study [18], in contrast, showed a decrease in the enzyme activity in individuals with the T variants in the codon 16 resulting in the oxidative stress enhancement, which is a probable cause of damage to the cell structures. A number of studies have shown the association of the *GPX1* (rs1050450) genetic variant with the susceptibility to MNs in non-exposed individuals [19]. The complex meta-analysis including 31 published papers has demonstrated that rs1050450 can contribute to the susceptibility to MN development due to disrupted antioxidant balance. Carriers of the T allele variant have the increased risk of developing various types of MNs, especially in Asian subgroups, based on the dominant genetic model [20]. The decrease in the gene functional activity can be a possible mechanism underlying such effects. For example, according to the ClinVar data, rs1050450 is associated with glutathione peroxidase deficiency.

With due account of the fact that many factors, including intergenic interactions and environmental factors, influence the malignant transformation of the cell, we have analyzed the role of intergenic interactions of SNPs and the dose to RBM in the development of MNs in exposed individuals. As a result of the analysis a 3-factor model having the highest accuracy (66% ($p = 0.001$)) and 100% reproducibility was identified. According

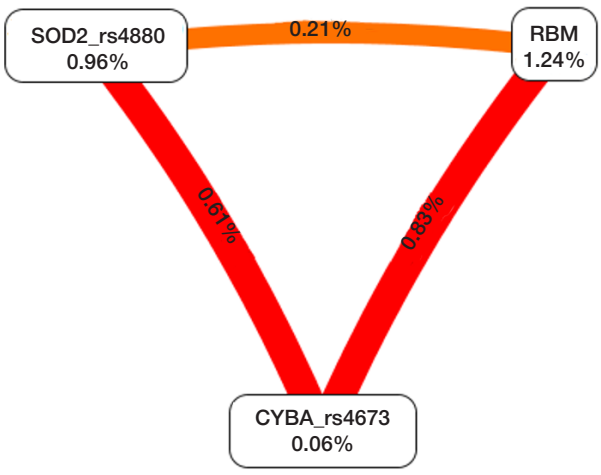

Fig. 2. Graph of interaction between the elements of the 3-factor model

to this model, increased risk of MN development was registered for the combination of the *SOD2* C*rs4880 allele, *CYBA* T*rs4673, and the accumulated dose to RBM. It should be noted that the most accurate model includes the *SOD2* (rs4880) polymorphic variant. Its association with the risk of MN development has been identified in our study. At the same time, the *GPX1* (rs1050450) polymorphism also associated with the risk of MN development was not included in the model: *CYBA* (rs4673) was identified instead of it based on the testing results. However, considering its insignificant contribution (0.06%), it is difficult to unambiguously determine its role at this phase of our research. The research results [21] show that rs4673 is associated with the increased risk of breast cancer. Individuals with the rs4673 C/T and T/T genotype have 1.42 times higher risk of breast cancer, than individuals with the C/C genotype.

Apparently, the increased production of ROS due to radiation exposure against the background of reduced superoxide dismutase activity could possibly contribute to the oxidative stress enhancement, damage to the cells components and DNA, and therefore lead to an increase in the risk of MN development.

CONCLUSIONS

The study has shown that polymorphic loci of the genes regulating the oxidative status of the cells, such as the *SOD2* rs4880*C (OR = 1.49; 95% CI = 1.02–2.18; p = 0.039) and *GPX1* rs1050450*T (OR = 2.00; 95% CI = 1.11–3.62; p = 0.024) alleles, are associated with the increased risk of MN development in the chronically exposed individuals. The model of interfactor interactions has also made it possible to determine the increased risk of MN development in carriers of the rs4880*C, rs4673*T minor alleles and the dose to RBM. Further research is required to reveal the modifying effect of exposure in individuals with unfavorable alleles in SNPs of the antioxidant system genes.

References

1. Buonanno M, de Toledo SM, Pain D, Azzam EI. Long-term consequences of radiation-induced bystander effects depend on radiation quality and dose and correlate with oxidative stress. Radiat Res. 2011; 175 (4): 405–15. DOI: 10.1667/RR2461.1.
2. Sage E, Shikazono N. Radiation-induced clustered DNA lesions: repair and mutagenesis. Free Radic Biol Med. 2017; (107): 125–35. DOI: 10.1016/j.freeradbiomed.2016.12.008.
3. Abd El Azeem RA, Zedan MM, Saad EA, Mutawi TM, Attia ZR. Single-nucleotide polymorphisms (SNPs) of antioxidant enzymes SOD2 and GSTP1 genes and SLE risk and severity in an Egyptian pediatric population. Clin Biochem. 2021; (88): 37–42. DOI: 10.1016/j.clinbiochem.2020.11.010.
4. Nikic P, Dragicevic D, Jerotic D, Savic S, Djukic T, Stankovic B, et al. Polymorphisms of antioxidant enzymes SOD2 (rs4880) and GPX1 (rs1050450) are associated with bladder cancer risk or its aggressiveness. Medicina (Kaunas). 2023; 59 (1): 131. DOI: 10.3390/medicina59010131.
5. Nawab SN, Zehra S, Fawwad A, Azhar A. A study on catalase gene promoter polymorphism-21 A/T (rs7943316) in healthy Pakistani population. Pak J Med Sci. 2017; 33 (6): 1521–4. DOI: 10.12669/pjms.336.13188.
6. Taioli E, Benhamou S, Bouchardy C, Cascorbi I, Cajas-Salazar N, Dally H, et al. Myeloperoxidase G-463A polymorphism and lung cancer: a HuGE genetic susceptibility to environmental carcinogens pooled analysis. Genet Med. 2007; 9 (2): 67–73. DOI: 10.1097/gim.0b013e31803068b1.
7. Kumar R, Kohli S, Ali Z, Duhan K, Ram R, Gupta M, et al. CYBA (p22phox) variants associate with blood pressure and oxidative stress markers in hypertension: a replication study in populations of diverse altitudes. Hypertens Res. 2015; 38 (7): 498–506. DOI: 10.1038/hr.2015.31.
8. Akleev AV, Kiselev MF, redaktory. Mediko-biologicheskie i jekologicheskie posledstvija radioaktivnogo zagrjaznenija reki Techa. M.: Vtoraja tipografija FU "Medbiojekstrem", 2001; 531 p. Russian.
9. Degteva MO, Nape BA, Tolstyh EI, Shishkina EA, Bugrov NG, Krestinina LJu, et al. Raspredelenie individual'nyh doz v kogorte ljudej, obluchennyh v rezul'tate radioaktivnogo zagrjaznenija reki Techi. Medicinskaja radiologija i radiacionnaja bezopasnost'. 2019; 64 (3): 46–53. DOI: 10.12737/article_5cf2364cb49523.98590475. Russian.
10. Hahn LW, Ritchie MD, Moore JH. Multifactor dimensionality reduction software for detecting gene-gene and gene-environment interactions. Bioinformatics. 2003; 19 (3): 376–82. DOI: 10.1093/bioinformatics/btf869.
11. Burmistrova AL, redaktor. Metaorganizm. Stress i adaptacija. Cheljabinsk: Cheljabinskij gosudarstvennyj universitet, 2019; 239 p. Russian.
12. Ďuračková Z. Some current insights into oxidative stress. Physiol Res. 2010; 59 (4): 459–69. DOI: 10.33549/physiolres.931844.
13. Reuter S, Gupta SC, Chaturvedi MM, Aggarwal BB. Oxidative stress, inflammation, and cancer: how are they linked? Free Radic Biol Med. 2010; 49 (11): 1603–16. DOI: 10.1016/j.freeradbiomed.2010.09.006.
14. Khandrika L, Kumar B, Koul S, Maroni P, Koul HK. Oxidative stress in prostate cancer. Cancer Lett. 2009; 282 (2): 125–36. DOI: 10.1016/j.canlet.2008.12.011.
15. Córdoba EE, Abba MC, Lacunza E, Fernánde E, Güerci AM. Polymorphic variants in oxidative stress genes and acute toxicity in breast cancer patients receiving radiotherapy. Cancer Res Treat. 2016; 48 (3): 948–54. DOI: 10.4143/crt.2015.360.
16. Burri RJ, Stock RG, Cesaretti JA, Atencio DP, Peters S, Peters CA, et al. Association of single nucleotide polymorphisms in SOD2, XRCC1 and XRCC3 with susceptibility for the development of adverse effects resulting from radiotherapy for prostate cancer. Radiat Res. 2008; 170 (1): 49–59. DOI: 10.1667/RR1219.1.
17. Bastaki M, Huen K, Manzanillo P, Chande N, Chen C, Balmes JR, et al. Genotype-activity relationship for Mn-superoxide dismutase, glutathione peroxidase 1 and catalase in humans. Pharmacogenet Genomics. 2006; 16 (4): 279–86. DOI: 10.1097/01.fpc.0000199498.08725.9c.
18. Sutton A, Imbert A, Igoudjil A, Descatoire V, Cazanave S, Pessayre D, et al. The manganese superoxide dismutase Ala16Val dimorphism modulates both mitochondrial import and mRNA stability. Pharmacogenet Genomics. 2005; 15 (5): 311–9. DOI: 10.1097/01213011-200505000-00006.
19. Zhao Y, Wang H, Zhou J, Shao Q. Glutathione peroxidase GPX1 and its dichotomous roles in cancer. Cancers (Basel). 2022; 14 (10): 2560. DOI: 10.3390/cancers14102560.
20. Chen J, Cao Q, Qin C, Shao P, Wu Y, Wang M, et al. GPx-1 polymorphism (rs1050450) contributes to tumor susceptibility: evidence from meta-analysis. J Cancer Res Clin Oncol. 2011; 137 (10): 1553–61. DOI: 10.1007/s00432-011-1033-x.
21. Tupurani MA, Padala C, Puranam K, Galimudi RK, Kupsal K, Shyamala N, et al. Association of CYBA gene (-930 A/G and 242 C/T) polymorphisms with oxidative stress in breast cancer: a case-control study. PeerJ. 2018; (6): e5509. DOI: 10.7717/peerj.5509.

Литература

1. Buonanno M, de Toledo SM, Pain D, Azzam EI. Long-term consequences of radiation-induced bystander effects depend on radiation quality and dose and correlate with oxidative stress. Radiat Res. 2011; 175 (4): 405–15. DOI: 10.1667/RR2461.1.
2. Sage E, Shikazono N. Radiation-induced clustered DNA lesions: repair and mutagenesis. Free Radic Biol Med. 2017; (107): 125–35. DOI: 10.1016/j.freeradbiomed.2016.12.008.
3. Abd El Azeem RA, Zedan MM, Saad EA, Mutawi TM, Attia ZR. Single-nucleotide polymorphisms (SNPs) of antioxidant enzymes SOD2 and GSTP1 genes and SLE risk and severity in an Egyptian pediatric population. Clin Biochem. 2021; (88): 37–42. DOI: 10.1016/j.clinbiochem.2020.11.010.
4. Nikic P, Dragicevic D, Jerotic D, Savic S, Djukic T, Stankovic B, et al. Polymorphisms of antioxidant enzymes SOD2 (rs4880) and GPX1 (rs1050450) are associated with bladder cancer risk or its aggressiveness. Medicina (Kaunas). 2023; 59 (1): 131. DOI: 10.3390/medicina59010131.
5. Nawab SN, Zehra S, Fawwad A, Azhar A. A study on catalase gene promoter polymorphism-21 A/T (rs7943316) in healthy Pakistani population. Pak J Med Sci. 2017; 33 (6): 1521–4. DOI: 10.12669/pjms.336.13188.
6. Taioli E, Benhamou S, Bouchardy C, Cascorbi I, Cajas-Salazar N, Dally H, et al. Myeloperoxidase G-463A polymorphism and lung cancer: a HuGE genetic susceptibility to environmental carcinogens pooled analysis. Genet Med. 2007; 9 (2): 67–73. DOI: 10.1097/gim.0b013e31803068b1.
7. Kumar R, Kohli S, Ali Z, Duhan K, Ram R, Gupta M, et al. CYBA (p22phox) variants associate with blood pressure and oxidative stress markers in hypertension: a replication study in populations of diverse altitudes. Hypertens Res. 2015; 38 (7): 498–506. DOI: 10.1038/hr.2015.31.
8. Аклеев А. В., Киселев М. Ф., редакторы. Медико-биологические и экологические последствия радиоактивного загрязнения реки Теча. М.: Вторая типография ФУ «Медбиоэкстрем», 2001; 531 с.
9. Дегтева М. О., Напье Б. А., Толстых Е. И., Шишкина Е. А., Бугров Н. Г., Крестинина Л. Ю. и др. Распределение индивидуальных доз в когорте людей, облученных в результате радиоактивного загрязнения реки Течи. Медицинская радиология и радиационная безопасность. 2019; 64 (3): 46–53. DOI: 10.12737/article_5cf2364cb49523.98590475.
10. Hahn LW, Ritchie MD, Moore JH. Multifactor dimensionality reduction software for detecting gene-gene and gene-environment interactions. Bioinformatics. 2003; 19 (3): 376–82. DOI: 10.1093/bioinformatics/btf869.
11. Бурмистрова А. Л., редактор. Метаорганизм. Стресс и адаптация. Челябинск: Челябинский государственный университет, 2019; 239 с.
12. Ďuračková Z. Some current insights into oxidative stress. Physiol Res. 2010; 59 (4): 459–69. DOI: 10.33549/physiolres.931844.
13. Reuter S, Gupta SC, Chaturvedi MM, Aggarwal BB. Oxidative stress, inflammation, and cancer: how are they linked? Free Radic Biol Med. 2010; 49 (11): 1603–16. DOI: 10.1016/j.freeradbiomed.2010.09.006.
14. Khandrika L, Kumar B, Koul S, Maroni P, Koul HK. Oxidative stress in prostate cancer. Cancer Lett. 2009; 282 (2): 125–36. DOI: 10.1016/j.canlet.2008.12.011.
15. Córdoba EE, Abba MC, Lacunza E, Fernánde E, Güerci AM. Polymorphic variants in oxidative stress genes and acute toxicity in breast cancer patients receiving radiotherapy. Cancer Res Treat. 2016; 48 (3): 948–54. DOI: 10.4143/crt.2015.360.
16. Burri RJ, Stock RG, Cesaretti JA, Atencio DP, Peters S, Peters CA, et al. Association of single nucleotide polymorphisms in SOD2, XRCC1 and XRCC3 with susceptibility for the development of adverse effects resulting from radiotherapy for prostate cancer. Radiat Res. 2008; 170 (1): 49–59. DOI: 10.1667/RR1219.1.
17. Bastaki M, Huen K, Manzanillo P, Chande N, Chen C, Balmes JR, et al. Genotype-activity relationship for Mn-superoxide dismutase, glutathione peroxidase 1 and catalase in humans. Pharmacogenet Genomics. 2006; 16 (4): 279–86. DOI: 10.1097/01.fpc.0000199498.08725.9c.
18. Sutton A, Imbert A, Igoudjil A, Descatoire V, Cazanave S, Pessayre D, et al. The manganese superoxide dismutase Ala16Val dimorphism modulates both mitochondrial import and mRNA stability. Pharmacogenet Genomics. 2005; 15 (5): 311–9. DOI: 10.1097/01213011-200505000-00006.
19. Zhao Y, Wang H, Zhou J, Shao Q. Glutathione peroxidase GPX1 and its dichotomous roles in cancer. Cancers (Basel). 2022; 14 (10): 2560. DOI: 10.3390/cancers14102560.
20. Chen J, Cao Q, Qin C, Shao P, Wu Y, Wang M, et al. GPx-1 polymorphism (rs1050450) contributes to tumor susceptibility: evidence from meta-analysis. J Cancer Res Clin Oncol. 2011; 137 (10): 1553–61. DOI: 10.1007/s00432-011-1033-x.
21. Tupurani MA, Padala C, Puranam K, Galimudi RK, Kupsal K, Shyamala N, et al. Association of CYBA gene (-930 A/G and 242 C/T) polymorphisms with oxidative stress in breast cancer: a case-control study. PeerJ. 2018; (6): e5509. DOI: 10.7717/peerj.5509.