Dermatology Practical & Conceptual

Familial Melanoma Phenotype With Xeroderma Pigmentosum Group C (XP-C) Genotype - The Putative Role of *MC1R* Polymorphism as Modifier

Franciele Antonieta Bianchi Leidenz[1], Flavia Vasques Bittencourt[2], Williana Garcia Braga[1], Elio Magno de Sá Araújo[1], Carolina Cavalieri Gomes[3], Vanessa de Fatima Bernardes[3], Eitan Friedman[4], Luiz De Marco[1]

1 Departments of Surgery, School of Medicine, Universidade Federal de Minas Gerais, Belo Horizonte, Brazil
2 Departments of Medicine, School of Medicine, Universidade Federal de Minas Gerais, Belo Horizonte, Brazil
3 Departments of Pathology, School of Medicine, Universidade Federal de Minas Gerais, Belo Horizonte, Brazil
4 The Preventive Personalized Medicine Center, Assuta Medical Center and the Sackler School of Medicine, Tel-Aviv University, Tel-Aviv, Israel

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Corresponding author: L. De Marco, Department of Surgery, School of Medicine, Universidade Federal de Minas Gerais, Av Alfredo Balena 190, Belo Horizonte 30130-100, Brazil; e-mail: Ldemarco@ufmg.br.

ABSTRACT

Introduction: Xeroderma pigmentosum (XP), a rare inherited condition, hallmarked by extreme sensitivity to sun exposure resulting in multiple skin cancers and non-malignant skin alterations is attributed to homozygous inactivating pathogenic variants (PVs) in DNA repair genes, predominantly the XPC gene.

Objectives: Report a unique phenotypic expression of mutant XPC allele that may be compatible with a putative modifier role for MC1R polymorphism.

Methods: A family of 13 siblings, seven of whom were diagnosed with at least one cutaneous melanoma (N = 53) and non-melanoma skin cancers (N = 9) was studied. Of seven melanoma-affected cases, five consented for genetic analysis. CDKN2A revealed no PV in any case and subsequent whole-exome sequencing (WES) identified a rare homozygous missense PV (c.919C>T; p.Arg307Trp) in exon 8 of the XPC gene in all affected individuals. Notably, XPC PV carriers who co-harbored the p.I155T MC1R variant (N = 3) exhibited larger number of tumors, deeper Breslow indexes, higher rates of invasive melanomas and earlier age at diagnosis compared with non MC1R variant carriers (N = 2).

Conclusions: Familial malignant melanoma phenotype may, in fact, be an unusual clinical presentation of XPC, and MC1R may be a genetic modifier of penetrance and phenotype of mutant XPC alleles.

Introduction

Xeroderma Pigmentosum (XP) is a rare autosomal recessive disorder whose estimated rates of 2.3/1,000,000 in Western countries clinically hallmarked by multitude of skin anomalies [1]: hyperpigmentation, premature skin aging, cutaneous (and ocular) photosensitivity, and an increased risk for developing a host of skin tumors [2]. Genetically, XP is associated with homozygous pathogenic variants (PVs) in seven nucleotide excision repair pathway genes: XPA, ERCC3, XPC, ERCC2, DDB2, ERCC4, ERCC5 and one clinical variant (XPV, Xeroderma Pigmentosum Variant) attributed to a mutated POLH gene [2].

The most commonly mutated gene underlying XP in Caucasians is XPC group C (MIM #278720) [3]. The clinical phenotype in XP due to mutant XPC alleles dependent on sun exposure and the mutational background, hence on the geographical origin [4]. The incidence of skin cancers is 1,000 times higher in XP patients compared with average risk population and a major contributor to the substantially decreased life expectancy in XP cases, around 30 years [5]. These patients have an estimated 10,000-fold increased risk of non-melanoma skin cancer, a 2,000-fold increased risk of melanoma under the age of 20 [1] and the first skin lesions may appear as early as eight years of age [5].

Familial melanoma syndrome (FMS) is a term used to describe the presence of two or more cases of cutaneous melanoma in first- or second-degree relatives [6]. An alternative classification [7] refers to these cases as Melanoma dominant syndrome when melanoma is the first or the predominant clinical manifestation in a family.

Objectives

Here we report a family with cutaneous melanomas with a seemingly autosomal dominant inheritance pattern without clear clinical characteristics of XP, where a homozygous *XPC* inactivating PV co-segregated with the phenotype, and a mutant *MC1R* allele seemingly affected the clinical melanoma phenotype.

Methods

The proband (Figure 1), individual II.11, was referred in 2000 at age 28, after a diagnosis of melanoma and a history of death of his older brother from metastatic melanoma of the lower lip at age 38. Since then, a total of 24 melanomas have been confirmed in the proband. These melanomas were mostly located in the head (N = 19) and neck (N = 2) regions, including the scalp (Figure 2), and upper trunk (N = 3). The majority (19/24) were in situ, three were thin melanomas (Breslow 0.45 mm, 0.47 mm and 0.9 mm), one was 1.4 mm, and one was 2.0 mm. Physical examination revealed brown hair and brown eyes, Fitzpatrick phototype III, sun damage on exposed areas with some pigmented changes, such as solar lentigines. Throughout 20 years of follow-up, he also presented two basal cell carcinomas (BCC) and one basosquamous carcinoma of the head and neck. History of unprotected excessive sun exposure was reported. Dermoscopy of pigmented lesions did not show any specific patterns. All consenting siblings (12) were examined, and five of these siblings were diagnosed with melanoma, (a total of 7/13 melanoma affected siblings). Of melanoma patients, the majority had multiple atypical nevi (three had more than 20, one had fewer than 10 and one had none), all have a relevant history of unprotected sun exposure throughout life. They did not get sunburned easily and did not have a recollection of many blistering despite having a long history of excessive unprotected sun exposure. Most of them have freckles or solar melanosis, confirming the sun-damaged skin. Clinical characteristics of affected individuals are shown in Table 1. All melanoma cases reported a history of significant sun exposure, Fitzpatrick type III, and solar lentigines in sun exposed areas. At the time of reporting, five of the six living affected siblings have had multiple primary melanomas, varying from three to 24 tumors per person. Age range at first melanoma

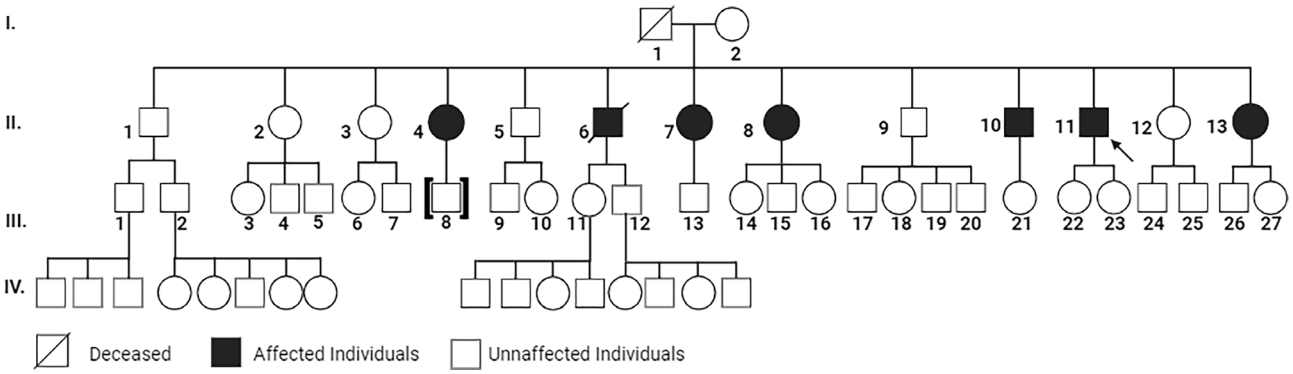

Figure 1. Pedigree of proband (II.11) family. Individuals affected by cutaneous melanoma are represented in black-filled shapes, while white shapes represent individuals without history of melanoma.

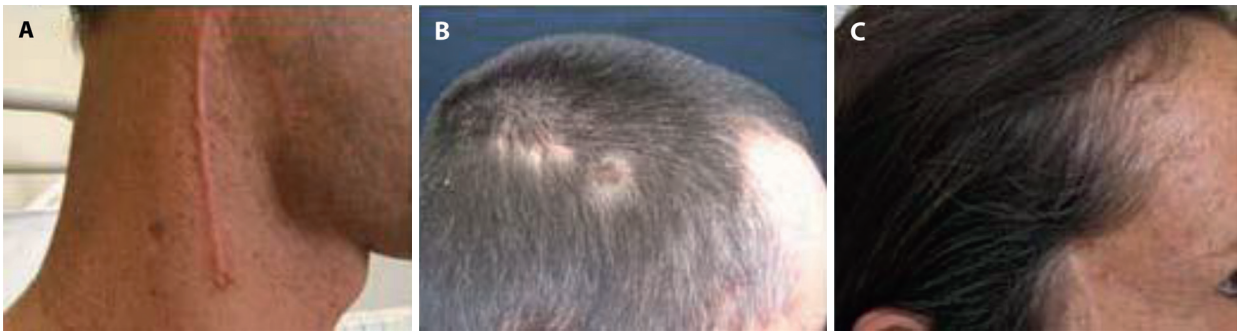

Figure 2. Clinical images of affected individuals. (A) Note skin photoaging and solar lentigines on sun exposed areas of the proband with a large scar from a previous removed melanoma and a new melanoma on the left side of the scar. (B) Melanoma on the scalp in the proband, in a relatively sun protected area. (C) Two melanomas on the forehead of individual II.8. (D) One melanoma in the left cervical region of individual II.13 on exposed area showing solar lentigines, surrounded by pigmentary changes.

Table 1 Familial melanoma and their characteristics

Affected individuals	Age of first melanoma	Number of melanomas	Tumor(s) localization(s)	Breslow thickness	Sun exposure	Total nevus body count	Other skin cancers
II.4	45	3	Head (3)	All *in situ*	Excessive unprotected	<20	None
II.7	33	11	Head (3), neck (1), limbs (2) and upper trunk (5)	All *in situ*	Excessive unprotected	20-100	Two BCC
II.8	35	7	Head (3), neck (2), and chest (2)	0.24mm and 6 *in situ*	Excessive unprotected	20-100	Three BCC
II.10	48	1	Head (scalp)	*in situ*	Excessive unprotected	20-100	None
II.11	28	24	Head (19), neck (2), and upper trunk (3)	0.45mm; 0.47mm; 0.9mm; 1.4mm; 2.0mm and 19 *in situ*	Excessive unprotected	>100	Two BCC, one basosquamous carcinoma
II.13	22	7	Head (3), neck (3) and upper trunk (1)	0.23mm; 0.36 mm; 2.3 mm and 4 *in situ*	Excessive unprotected	>100	One BCC

diagnosis was 22-45 years of age. Neither visceral metastasis nor lymph node disease have been reported in any of the six affected siblings, and no adjuvant therapies were given. Interestingly, no relevant ophthalmologic condition has been diagnosed in the affected family members. The siblings were born to consanguineous (second-degree cousins) phenotypically healthy parents; the father died at age 58 without any history of skin cancer. The mother is still alive (age 86), in good health and never had any skin cancer. Occupational exposure to sun was suggested by the fact that they were mostly farm laborers. Figure 2 demonstrates some of their clinical features. This study was approved by the Institutional Review Board of Universidade Federal de Minas Gerais (CAAE 0472.0.203.240-11). Prior to enrolment, written informed consent was obtained from all patients.

Sanger sequencing for the CDKN2A gene, the most common high penetrance melanoma susceptibility gene, was initially performed. Subsequently, peripheral blood DNA from individual II.11 was submitted to Whole-Exome Sequencing (WES). The VCF file was analyzed using Mendel, MD software (http://mendel.medicina.ufmg.br) [8] as well as by Ingenuity® Variant Analysis™ software (www.ingenuity.com/variants). Captured sequences were aligned with the human reference genome GRCh37 and a sequence of filters applied. A flow chart is shown in Figure 3. Given the autosomal recessive inheritance pattern in the studied family we focused

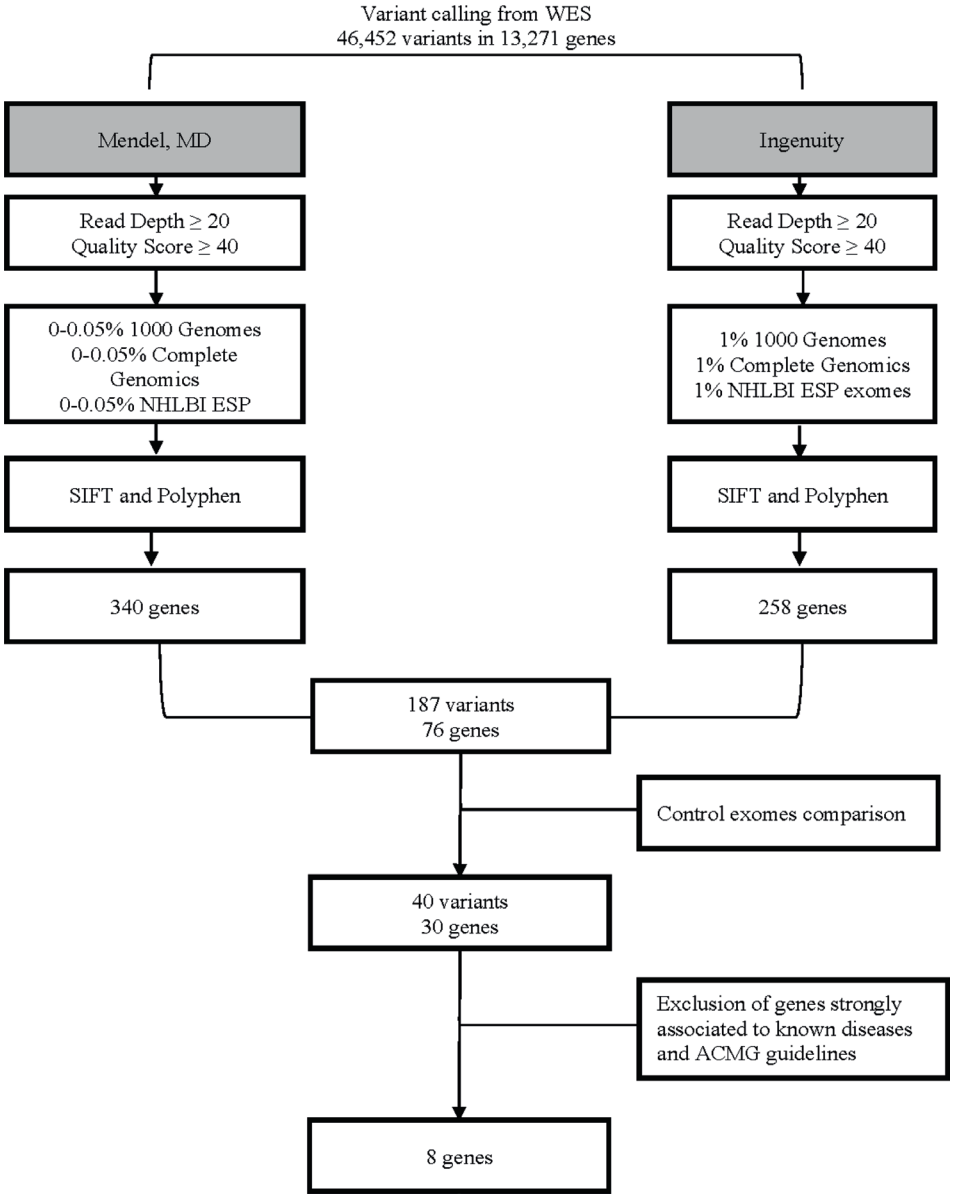

Figure 3. Flowchart of variant analyses.

on homozygous variants. All seemingly candidate variants were confirmed by Sanger sequencing and co- segregation was subsequently carried out in all consenting cases (N = 15) using Sanger sequencing.

To further assess the putative clinical significance of the detected XPC sequence variant and the possibility of residual protein function, we performed immunohistochemical analysis using XPC antibody (D-10: sc-74410, Santa Cruz Biotechnology) both on melanoma tissue from the proband and on normal mucosal tissue from the same individual mucosa (serving as control).

Results

CDKN2A gene genotyping yielded no PVs. WES showed 46,452 variants and, after a series of filtering, exclusion of variants in our control exomes bank and selection of homozygous variants only, it was narrowed down to eight genes, of whom XPC was the strongest candidate.

Sequencing of XPC demonstrated that the clinically unaffected mother carried the heterozygous XPC variant p.Arg307Trp (c.919C>T) in exon 8. All five affected individuals studied were homozygous (T/T) for this sequence variant.

Another known pathogenic variant (c.464T>C; p.Ile155Thr) was detected in the MC1R gene (NM_002386.4) by WES. Three (II.7, II. 11 and II.13) of the five affected melanoma cases who carried the XPC homozygous variant harbored this heterozygous PV. These three co-carriers (XPC and MC1R) carriers displayed significantly worse disease phenotype compared with the two non-co-carriers: higher number of melanomas per person (14 versus 5), earlier age of melanoma diagnosis (average 27.6 years versus 40 years) and thicker melanomas (three invasive in co-carriers versus noninvasive melanomas in non-co-carriers, with mean

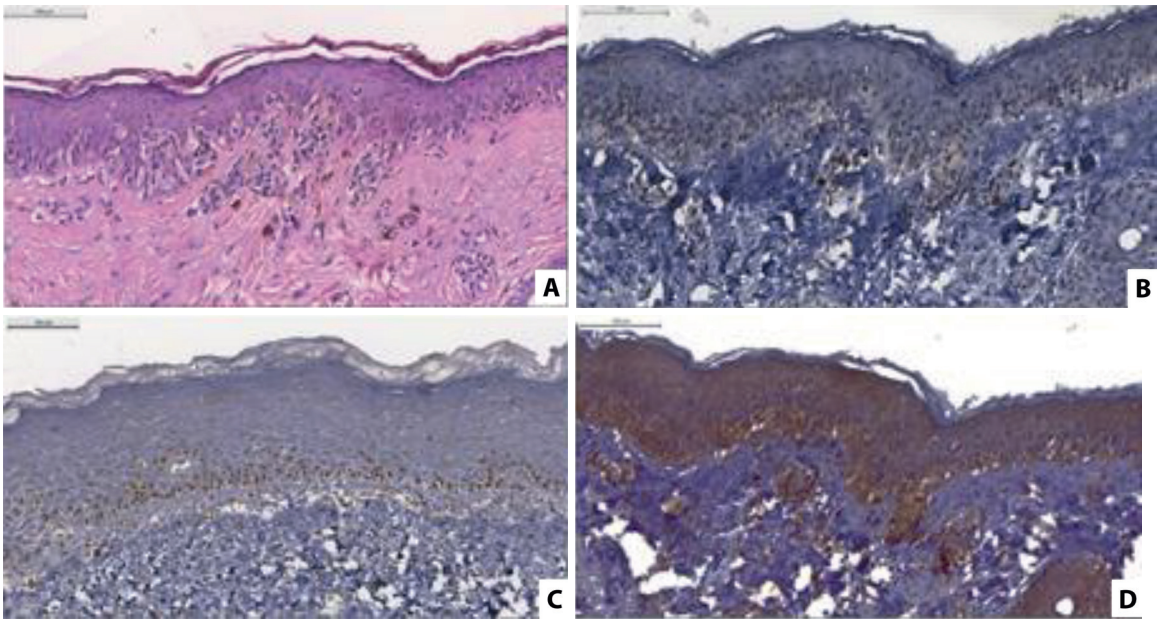

Figure 4. Immunohistochemistry assay. (A) Melanoma tissue from proband stained with H&E. (B) Melanoma tissue from proband very lightly stained by the XPC antibody, suggesting there is no significant expression of this protein in this tissue. The dark color is mostly due to melanin. (C) XPC antibody in a control tissue (mucosa) showing high positivity, therefore, significant quantity of XPC protein in this normal tissue. (D) the same melanoma tissue from proband highly colored by a control anti-tubulin antibody, showing that this tissue is indeed viable.

Breslow thickness of 0.17 mm in carriers, versus 0.017 mm in non-carriers). Our data shows that the median number of non-melanoma skin cancer (NMSC) per person among MC1R wild-type XP-C individuals was 1.5 (three BCC in two subjects). In contrast, among carriers of the I155T variant, the median number of NMSC was 2.0, with six tumors occurring in three individuals.

The XPC c.919C>T;p.Arg307Trp variant is predicted to be pathogenic by several different bioinformatics tools: disease causing by MutationTaster (http://www.mutationtaster.org/), probably damaging by PolyPhen-2 with a score of 1.0 (http://genetics.bwh.harvard.edu/pph2/) and deleterious by the Protein Variation Effect Analyzer (PROVEAN, http://provean.jcvi.org/index.php), with a score of –6.872. The pathogenicity of the c.919C>T;p.Arg307Trp sequence variant is also inferred by its rarity (prevalence in GnomAD (https://gnomad.broadinstitute.org/) 0.00002422 with none reported in Brazilian databanks (https://abraom.ib.usp.br/). Variant classification according to 2015 ACMG guidelines is PM2 (**moderate evidence of pathogenicity**) **and with additional supporting evidence of pathogenicity (PP1).**

Immunohistochemistry analysis demonstrated no XPC staining in the melanoma tissue compared with normal non cancer surrounding dermal tissue, as is predicted by functional studies [9] (Figure 4).

Conclusions

XP is usually diagnosed in childhood heralded by the presence of typical skin lesions [5]. The current study describes a family with atypical XP presentation age in the proband. Late onset of tumors and late diagnosis of XP are uncommon but there are a few previously reported cases with distinct phenotypes [10,11]. To our knowledge, there are two reports of *XPC*-related XP phenotype being diagnosed in late adulthood: an 83-year-old French woman with multiple melanomas whose unusual long-term survival was attributed to a lower UV-radiation exposure and regular clinical follow-up and a 42-year-old Caucasian man with multiple melanomas and a missense PV in the XPC gene that still retained some XPC protein function and, seemingly contributing to late XP diagnosis [4,11].

XP typically leads to a shortened life span, with cancer related death usually occurring at 30-40 years of age [12]. In the case described herein all but one patient are alive aged 41-62 years, at the time of reporting (Figure 1). The less severe phenotype and the less pronounced effect on early age mortality in this family suggests that the XPC mutated protein is still capable of exerting some residual activity and repair DNA damage, as previously suggested [4].

Being a recessive condition, it is expected that around 25% of the offspring would be affected. In this family, seven of the 13 siblings are affected (53.8%), with an extraordinary higher number of melanomas (N = 43) compared with non-melanoma skin cancers (n= 9), an unusual tumor distribution to the one usually seen in XP patients – carcinomas are five times more common than melanomas [1]. Additional clinically unique features in this family that have led to XP not even being considered as a possible diagnosis prior to genetic analysis, were the fact that melanomas were not only

diagnosed in chronically sun exposed areas, such as scalp of hairy individuals, lack of typical XP-associated skin changes such as actinic keratosis, atrophy, telangiectasias and marked skin photoaging.

In the present study, having both the XPC homozygous variant and a variant MC1R allele (p.I155T) may have deleteriously affected the phenotype, with those harboring both variants exhibiting seemingly a more aggressive clinical and pathological phenotype. Few studies reported the effect of co-harbouring MC1R alleles in genetically proven XP cases. A previous study of 17 Nepalese XP patients suggested that the p.R163Q MC1R variant in XPC patients was associated with younger age at first carcinoma and more numerous cutaneous carcinomas, although this latter difference was not statistically significant [1]. Additionally, it has been speculated that the coexistence of XPC mutation with another MC1R variant (p.V60L) was associated with multiple primary melanoma phenotype in a single French patient who had 10 melanomas and six BCC before being diagnosed with XP [12]. Although the putative modifier effect on MC1R variant on the phenotype of the homozygous deleterious variant in the XPC gene is intriguing, it remains currently speculative at best and awaits further functional studies and/or additional families displaying similar effect.

In conclusion, a familial melanoma phenotype with a presumed autosomal recessive inheritance pattern was shown to be atypical in a genetically proven XP. The role, if any, of MC1R alleles in modifying the clinical tumor phenotype in XPC mutation carriers is suggested. Validation and extension of these preliminary observations are needed.

Ethics statement: This study was approved by the Institutional Review Board of Universidade Federal de Minas Gerais (CAAE 0472.0.203.240-11). Prior to enrolment, written informed consent was obtained from all patients.

References

1. Espi P, Parajuli S, Benfodda M, et al. Clinical and genetic characteristics of xeroderma pigmentosum in Nepal. *J Eur Acad Dermatol Venereol*. 2018;32(5):832-839. DOI: 10.1111/jdv.14717. PMID: 29178624.
2. Ijaz A, Basit S, Gul A, et al. XPC gene mutations in families with xeroderma pigmentosum from Pakistan; prevalent founder effect. *Congenit Anom (Kyoto)*. 2019;59(1):18-21. DOI: 10.1111/cga.12281. PMID: 29569758.
3. Ali MZ, Blatterer J, Khan MA, et al. Identification of a novel protein truncating mutation p.Asp98* in XPC associated with xeroderma pigmentosum in a consanguineous Pakistani family. *Mol Genet Genomic Med*. 2020;8(2):e1060. DOI: 10.1002/mgg3.1060. PMID: 31923348. PMCID: PMC7005610.
4. Meneses M, Chavez-Bourgeois M, Badenas C, et al. Atypical Clinical Presentation of Xeroderma Pigmentosum in a Patient Harboring a Novel Missense Mutation in the XPC Gene: The Importance of Clinical Suspicion. *Dermatology*. 2015;231(3):217-221. DOI: 10.1159/000433527. PMID: 26278556.
5. Ribeiro MG, Zunta GL, Santos JS, Moraes AM, Lima CSP, Ortega MM. Clinical features related to xeroderma pigmentosum in a Brazilian patient diagnosed at advanced age. *Appl Clin Genet*. 2018;11:89-92. DOI: 10.2147/TACG.S155083. PMID: 30127633. PMCID: PMC6089094.
6. Sá BCS, Moredo LF, Gomes EE, Araújo ESS, Duprat JP. Hereditary melanoma: a five-year study of Brazilian patients in a cancer referral center - phenotypic characteristics of probands and pathological features of primary tumors. *An Bras Dermatol*. 2018;93(3):337-340. DOI: 10.1590/abd1806-4841.20186201. PMID: 29924249. PMCID: PMC6001076.
7. Abdo JF, Sharma A, Sharma R. Role of Heredity in Melanoma Susceptibility: A Primer for the Practicing Surgeon. *Surg Clin North Am*. 2020;100(1):13-28. DOI: 10.1016/j.suc.2019.09.006. PMID: 31753108.
8. G C C L Cardenas R, D Linhares N, L Ferreira R, Pena SDJ. Mendel,MD: A user-friendly open-source web tool for analyzing WES and WGS in the diagnosis of patients with Mendelian disorders. *PLoS Comput Biol*. 2017;13(6):e1005520. DOI: 10.1371/journal.pcbi.1005520. PMID: 28594829. PMCID: PMC5464533.
9. Budden T, Davey RJ, Vilain RE, et al. Repair of UVB-induced DNA damage is reduced in melanoma due to low XPC and global genome repair. *Oncotarget*. 2016;7(38):60940-60953. DOI: 10.18632/oncotarget.10902. PMID: 27487145. PMCID: PMC5308628.
10. Fassihi H, Sethi M, Fawcett H, et al. Deep phenotyping of 89 xeroderma pigmentosum patients reveals unexpected heterogeneity dependent on the precise molecular defect. *Proc Natl Acad Sci U S A*. 2016 1;113(9):E1236- E1245. DOI: 10.1073/pnas.1519444113. PMID: 26884178. PMCID: PMC4780618.
11. Macke EL, Morales-Rosado JA, Gupta A, et al. A novel missense variant and multiexon deletion causing a delayed presentation of xeroderma pigmentosum, group C. *Cold Spring Harb Mol Case Stud*. 2020;6(4):a005165. DOI: 10.1101/mcs.a005165. PMID: 32843428. PMCID: PMC7476405.
12. Jacobelli S, Soufir N, Lacapere JJ, et al. Xeroderma pigmentosum group C in a French Caucasian patient with multiple melanoma and unusual long-term survival. *Br J Dermatol*. 2008;159(4):968-973. DOI: 10.1111/j.1365-2133.2008.08791.x. PMID: 18717677.