A clinical case of multiple primary cancers in a carrier of rare SDK2 and NOTCH2 gene mutations

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Abstract

Background: Genetic predisposition is one of the risk factors for the development of multiple primary cancers (MPCs), the frequency of which increases and ranges from 2 to 17%. This study describes a combination of rare mutations, rs746551843 in the NOTCH2 gene and rs144933006 in the SDK2 gene, in a woman with breast cancer and leiomyosarcoma without a clearly burdened family history.

Case presentation: A 55-year-old Caucasian woman received complex treatment on the basis of the National Medical Research Centre for Oncology for left breast cancer and leiomyosarcoma of soft tissues of the left thigh. The patient was referred for consultation with a geneticist. Among direct relatives, a maternal aunt with a history of kidney cancer was not a carrier of the studied single nucleotide polymorphisms (SNPs). The healthy son of the patient inherited both mutations.

Conclusion: Thus, perhaps in the described case, there is a synergistic effect of two alleles of moderate and low penetrance, which led to the phenotype of multiple primary cancers.

Keywords: Breast cancer, Leiomyosarcoma, Multiple primary cancers, SDK2, NOTCH2

Background

The identification of risk factors, as well as the definition of multiple primary cancers (MPCs), remain unsolved problems at present [1]. Depending on the accepted term, the studied patient population, and the time of observation, the total frequency of MPCs ranges from 2 to 17% [2]. Among the main causes of MPCs, factors related to lifestyle, the environment, treatment conditions, and genetic determinants are noted. In this case, the identification of the germline mutation in a patient with a primary form of cancer portends an increased risk of developing additional primary tumours. This case report describes the clinical case of MPCs (breast cancer and leiomyosarcoma) in a patient without a clearly burdened family history who is a carrier of rare germline mutations identified as a result of whole exome sequencing (WES).

Case presentation

A 55-year-old Caucasian patient received complex treatment for left breast cancer and leiomyosarcoma of soft tissues of the left thigh. From the history of the disease, the patient sought medical help in 2014. On examination, she was diagnosed with left breast cancer and leiomyosarcoma of soft tissues of the left thigh. Radical Madden mastectomy and a wide excision of the soft tissue tumour of the left thigh was chosen as the first step of the complex treatment. Postoperative diagnosis was left breast cancer, cT2N1M0, pT2N1M0, stage IIB, and the condition after surgical treatment was clinical group 2. Leiomyosarcoma of soft tissues of the left thigh was
pG3T1aN0M0, stage IIB, and the condition after surgical treatment was clinical group 2.

In the adjuvant mode, she received 6 courses of chemotherapy according to the FAC (fluorouracil, Adriamycin and cytoxan) scheme, as well as a course of remote gamma therapy in the area of postoperative scars of the mammary gland and thigh and the lymphatic drainage zone from the mammary gland with a total radiation dose of 50 Gy. Subsequently, she was observed for 3 years without recurrence. In October 2017, the patient noted an increase in the inguinal lymph nodes on the left, and an inguinal-femoral lymphadenectomy was performed. Postoperative histological findings were breast cancer metastasis. The patient received an additional 4 courses of second-line adjuvant chemotherapy with taxanes, and the postoperative scar of the inguinal region on the left was irradiated with a total radiation dose of 40 Gy. At the end of treatment, she continued to be observed by an oncologist, and disease recurrence was not determined. The last known visit to an oncologist was November 16, 2019, at the time of treatment without recurrence.

The patient was referred for consultation with a geneticist. When collecting a family history (Fig. 1), we became aware of a mother's sister with kidney cancer, diagnosed at the age of 72 years, and in remission for more than 3 years. An allegedly benign tumour of the reproductive system was removed from the patient's mother at the age of 46. The maternal grandmother had a benign tumour on her face. The patient has one son, 30 years old, who is conditionally healthy. The patient's diagnosis was based on a histopathological conclusion, while other diagnoses in her family were based on a self-reporting.

Given the family history, the primary multiple localization of different types of cancer and the absence of mutations in the $BRCA1$, $BRCA2$, and $CHEK2$ genes prevailing in the Caucasian European population, the patient was asked to undergo WES as part of the study. For the NGS study, genomic DNA extracted from blood using the Cobas DNA Sample Preparation Kit (Roche, Anderlecht, Belgium) was used. Library preparation and subsequent sequencing on the NextSeq550 Illumina platform (San Diego, USA) was performed according to the manufacturer's recommendations. The following kits were used: using Nextera DNA Exome Kits (Illumina, San Diego, USA), xGen Exome Research Panel v2 (Integrated DNA Technologies, San Diego, USA), xGen Blocking Oligos (Integrated DNA Technologies, San Diego, USA) and the xGen Hybridization and Wash Kit (Integrated DNA Technologies, San Diego, USA). Screening and description of identified variants was carried out using two databases of germline mutations (HGMD, ClinVar) [3, 4], two cancer databases of mutations (COSMIC, ICGC) [5, 6], four population databases (dbSNP, 1000 Genome, ExAC, GnomAD) [7–10], and in silico prediction software (SIFT, PROVEAN, LRT, Mutation Assessor, PolyPhen-2 HDIV, PolyPhen-2 HVAR, Mutation Taster, FATHMM, GERP, Cancer Genome Interpreter, CSpage) [11–20]. Identified variants were

![Fig. 1 Sanger sequencing of A NOTCH2 and B SDK2 variants in proband, son and aunt by maternal lineage](image-url)
classified in accordance with the recommendations of the Association for Molecular Pathology, American Society of Clinical Oncology, College of American Pathologists and American College of Medical Genetics and Genomics (ACMG) [21, 22].

WES genotyping of DNA extracted from the blood of the patient with MPCs revealed two heterozygous mutations: one in the NOTCH2 gene, c.6178C>T p.Arg2060Cys rs746551843 and one in the SDK2 gene c.3778A> G p. Ser1260Gly rs144933006 (Fig. 1). Most in silico prediction programs (SIFT, PROVEAN, LRT, Mutation Assessor, PolyPhen-2 HDIV, PolyPhen-2 HVAR) indicated the damaging effect of the identified variants (Table 1). The driver properties of both variants are predicted: rs746551843—Cancer Genome Interpreter and CScape; rs144933006—CScape. The localization of the detected single nucleotide polymorphisms (SNPs) was highly conservative: the Genomic Evolutionary Rate Profiling (GERP) score was 5.37 for rs144933006 and 5.72 for rs746551843. ClinVar lacks information on these SNPs. In HGMD, rs746551843 is associated with hereditary breast cancer.

The living direct relatives were genotyped for the presence of the 2 identified mutations. The maternal aunt was not a carrier of these SNPs. The healthy son of the patient inherited both mutations in the heterozygous state (Fig. 2).

**Discussion**

The described case of malignant neoplasms of two localizations and two types can be associated with the identified mutations. It should be noted that the frequency of both mutations was low in different human populations. According to HGMD, a missense mutation in NOTCH2 causes a predisposition to hereditary breast cancer syndrome. According to GnomAD, the frequency of the described SNP is extremely low: 0.000036 in the Caucasoid population and 0.00003249 in South Asia. NOTCH2 is a known oncogene associated with the development and progression of tumours. In the study by Torrezan et al. [23], SNP rs746551843 was identified by WES results in Brazilian families with a family history of breast cancer. In addition to bioinformation algorithms for selecting candidate genes, the authors analysed mutations in an independent cohort of 42 Brazilian women with a risk of developing hepatitis C and negative for pathogenic variants of the BRCA1/2, TP53 and ATM genes. Earlier, in a WES study in one of three families with BRCA-negative familial breast cancer, an inherited rs746551843 was also found in a pool of several pathogenic mutations [24].

SNP rs144933006 is not widespread (0.000006462) and has been identified in several cases in the European population. This variant is not described in the literature. However, Rajendran and Deng, using various computational approaches, discovered 65 new candidate genes of breast cancer, among which the SDK2 gene was noted [25]. The authors suggest that SDK2 mutations can make certain changes to the network of protein-protein interactions of the main gene pool characteristic of breast cancer. According to the ICGC portal, the most common carriers of genetic variants of SDK2 are

**Table 1** Effect of genetic variants according to in silico prediction programs

| In silico prediction programs | rs746551843 | rs144933006 |
|------------------------------|-------------|-------------|
| SIFT                         | Deleterious | Deleterious |
| PROVEAN                      | Deleterious | Deleterious |
| LRT                          | Unknown     | Deleterious |
| FATH-MM                      | Tolerated   | Tolerated   |
| Mutation Assessor            | Medium impact | Medium impact |
| MutationTaster               | Disease causing | Disease causing |
| PolyPhen-2 HDIV              | Probably damaging | Probably damaging |
| PolyPhen-2 HVAR              | Probably damaging | Probably damaging |
patients with leiomyosarcoma – 65 out of 67 people (including unvaldated cases).

Summarizing the available data, in accordance with the recommendations of ACMG, we classified the SNPs found as variants with VUS (variant of unknown significance), the status of which can be upgraded to pathogenic or likely pathogenic in event of such cases are detected and with proper functional analysis.

Conclusion
A polygenic disease model is a possible explanation for the observed clinical findings. In accordance with this concept, which was proposed and considered earlier [26], alleles of moderate and low penetrance will act synergistically and play a dominant role. Unfortunately, we can only rely on the patient’s self-report, and many of the affected members of this family have died, which limits the interpretation of our data.

The diagnosis and treatment of multiple oncological pathologies remains a challenge due to the different definitions of MPCs, the absence of specific screening guidelines and established treatment guidelines. The treatment of such patients should be individualized using genetic data and an interdisciplinary approach.

Abbreviations
ACMG: American College of Medical Genetics and Genomics; FAC: Fluorouracil, adriamycin and cytoxan; GERP score: The Genomic Evolutionary Rate Profiling score; MPCs: Multiple primary cancers; SNPs: Single nucleotide polymorphisms; WES: Whole exome sequencing; VUS: Variant of unknown significance

Acknowledgements
We are grateful to the representatives of the Albiogen company, Nazar Samarow, Valery Kharlamov, Nikolai Egorov, as well as Oksana Mazur, scientific researcher at the Institute of Genetics and Cytology of the National Academy of Sciences of Belarus, for their help in organizing the WES study.

Authors’ contributions
NNT was responsible for writing original manuscript and data visualization. DYG was responsible for investigation, methodology, formal analysis and writing original manuscript. EPO was responsible for data curation and methodology. LNV was responsible for supervision, review and editing manuscript. TVA and EEK were responsible for methodology, review and editing manuscript. OK was responsible for conceptualization and project administration. The authors have read and approved the manuscript.

Funding
None.

Availability of data and materials
All data used during this report are included in this published article. Further data are available from the corresponding author on reasonable request.

Declarations
Ethics approval and consent to participate
The manuscript had the approval of the Ethical Committee of the National Medical Research Centre for Oncology (Protocol 9/1, 4th March 2019). Written consent was taken from the patient of the index case for publication.

Consent for publication
Written consent was taken from the patient of the index case for publication.

Competing interests
The authors declare no competing interest.

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Received: 25 February 2021 Accepted: 13 May 2021
Published online: 16 June 2021

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