Egyptian Journal of Medical Human Genetics

CASE REPORT Open Access

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

Nataliya N. Timoshkina[1], Dmitry Y. Gvaldin[1*], Ekaterina P. Omelchuk[1], Larisa N. Vashhenko[2], Tatjana V. Ausheva[2], Emma E. Kechedzhieva[2] and Oleg I. Kit[3]

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 tissue 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, cT2NxM0, pT2N1M0, stage IIB, and the condition after surgical treatment was clinical group 2. Leiomyosarcoma of soft tissues of the left thigh was

* Correspondence: dgvaldin@gmail.com
[1]Laboratory of Molecular Oncology, National Medical Research Centre for Oncology, Rostov-on-Don, Russia
Full list of author information is available at the end of the article

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, CScape) [11–20]. Identified variants were

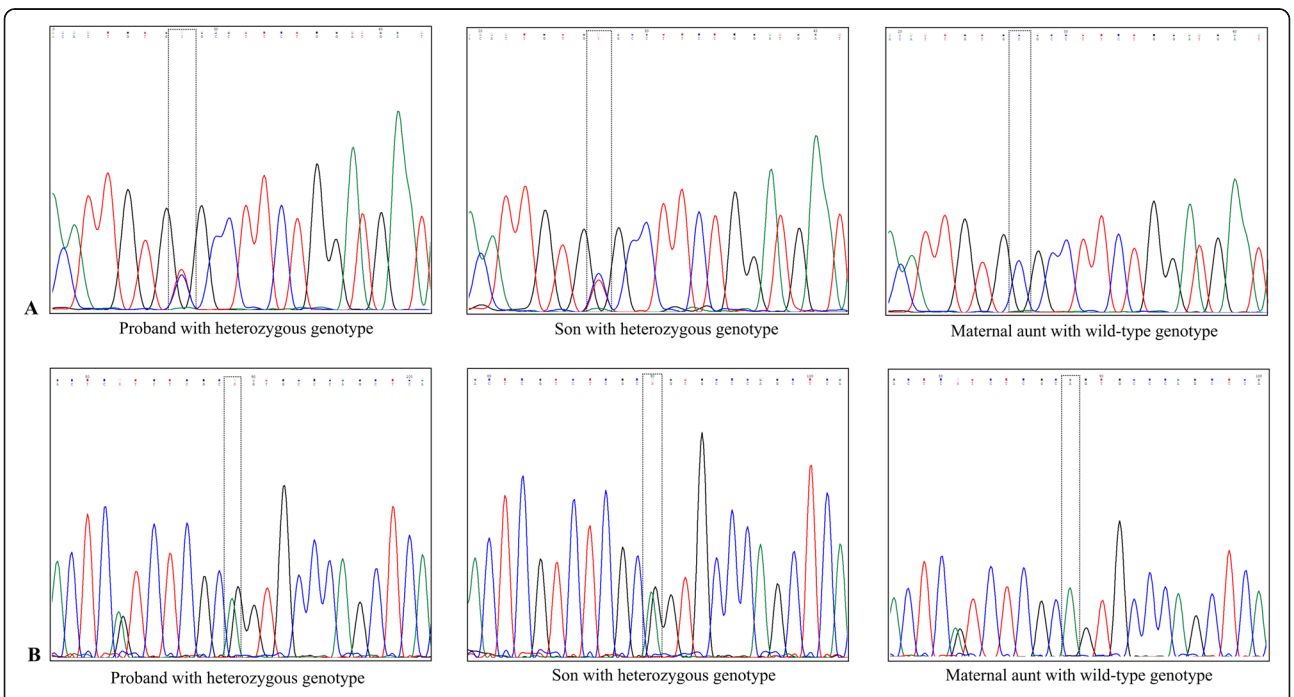

Fig. 1 Sanger sequencing of **A** *NOTCH2* and **B** *SDK2* variants in proband, son and aunt by maternal lineage

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).

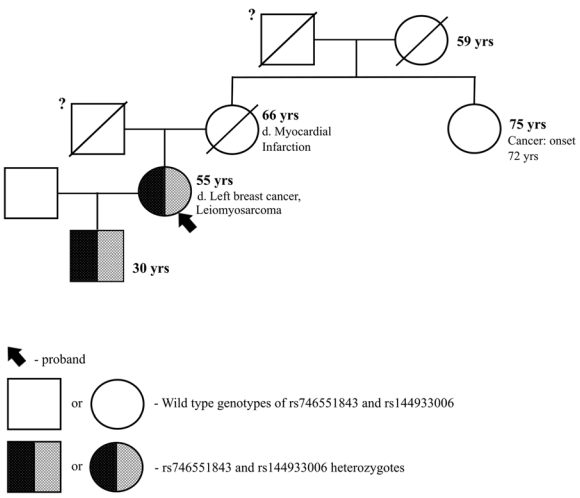

Fig. 2 Pedigree. Genetic variants that were identified in the patient were tested in the aunt by maternal lineage and son

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 (0.00006462) 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
FATHMM	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 unvalidated 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 Samarov, 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. OIK 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.

Author details

[1]Laboratory of Molecular Oncology, National Medical Research Centre for Oncology, Rostov-on-Don, Russia. [2]Department of Bone, Skin, Soft Tissue and Breast Tumors, National Medical Research Centre for Oncology, Rostov-on-Don, Russia. [3]National Medical Research Centre for Oncology, Rostov-on-Don, Russia.

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

References

1. Vogt A, Schmid S, Heinimann K, Frick H, Herrmann C, Cerny T, Omlin A (2017) Multiple primary tumours: challenges and approaches, a review. ESMO Open 2(2):e000172. https://doi.org/10.1136/esmoopen-2017-000172
2. Wood ME, Vogel V, Ng A, Foxhall L, Goodwin P, Travis LB (2012) Second malignant neoplasms: assessment and strategies for risk reduction. J Clin Oncol 30(30):3734–3745. https://doi.org/10.1200/JCO.2012.41.8681
3. Stenson PD, Ball EV, Mort M, Phillips AD, Shiel JA, Thomas NST, Abeysinghe S, Krawczak M, Cooper DN (2003) Human gene mutation database (HGMD): 2003 update. Hum Mutat 21(6):577–581. https://doi.org/10.1002/humu.10212
4. Landrum MJ, Chitipiralla S, Brown GR, Chen C, Gu B, Hart J, Hoffman D, Jang W, Kaur K, Liu C, Lyoshin V, Maddipatla Z, Maiti R, Mitchell J, O'Leary N, Riley GR, Shi W, Zhou G, Schneider V, Maglott D, Holmes JB, Kattman BL (2020) ClinVar: improvements to accessing data. Nucleic Acids Res 48(D1):D835–D844. https://doi.org/10.1093/nar/gkz972
5. Tate JG, Bamford S, Jubb HC, Sondka Z, Beare DM, Bindal N, Boutselakis H, Cole CG, Creatore C, Dawson E, Fish P, Harsha B, Hathaway C, Jupe SC, Kok CY, Noble K, Ponting L, Ramshaw CC, Rye CE, Speedy HE, Stefancsik R, Thompson SL, Wang S, Ward S, Campbell PJ, Forbes SA (2019) COSMIC: the catalogue of somatic mutations in cancer. Nucleic Acids Res 47(D1):D941–D947. https://doi.org/10.1093/nar/gky1015
6. Zhang J, Bajari R, Andric D, Gerthoffert F, Lepsa A, Nahal-Bose H, Stein LD, Ferretti V (2019) The international cancer genome consortium data portal. Nat Biotechnol 37(4):367–369. https://doi.org/10.1038/s41587-019-0055-9
7. Sherry ST, Ward MH, Kholodov M, Baker J, Phan L, Smigielski EM, Sirotkin K (2001) dbSNP: the NCBI database of genetic variation. Nucleic Acids Res 29(1):308–311. https://doi.org/10.1093/nar/29.1.308
8. 1000 Genomes Project Consortium, Auton A, Brooks LD et al (2015) A global reference for human genetic variation. Nature. 526(7571):68–74. https://doi.org/10.1038/nature15393
9. Karczewski KJ, Weisburd B, Thomas B, Solomonson M, Ruderfer DM, Kavanagh D, Hamamsy T, Lek M, Samocha KE, Cummings BB, Birnbaum D, The Exome Aggregation Consortium, Daly MJ, MacArthur DG (2017) The ExAC browser: displaying reference data information from over 60 000 exomes. Nucleic Acids Res 45(D1):D840–D845. https://doi.org/10.1093/nar/gkw971
10. Wang Q, Pierce-Hoffman E, Cummings BB et al (2020) Landscape of multi-nucleotide variants in 125,748 human exomes and 15,708 genomes. Nat Commun 11(1):2539. https://doi.org/10.1038/s41467-019-12438-5
11. Ng PC, Henikoff S (2003) SIFT: predicting amino acid changes that affect protein function. Nucleic Acids Res 31(13):3812–3814. https://doi.org/10.1093/nar/gkg509
12. Choi Y, Chan AP (2015) PROVEAN web server: a tool to predict the functional effect of amino acid substitutions and indels. Bioinformatics. 31(16):2745–2747. https://doi.org/10.1093/bioinformatics/btv195
13. Chun S, Fay JC (2009) Identification of deleterious mutations within three human genomes. Genome Res 19(9):1553–1561. https://doi.org/10.1101/gr.092619.109
14. Reva B, Antipin Y, Sander C (2011) Predicting the functional impact of protein mutations: application to cancer genomics. Nucleic Acids Res 39(17):e118. https://doi.org/10.1093/nar/gkr407

15. Ramensky VE, Gerasimova A, Bork P, Kondrashov AS, Sunyaev SR (2010) A method and server for predicting damaging missense mutations. Nat Methods 7(4):248–249
16. Schwarz JM, Cooper DN, Schuelke M, Seelow D (2014) MutationTaster2: mutation prediction for the deep-sequencing age. Nat Methods 11(4):361–362. https://doi.org/10.1038/nmeth.2890
17. Shihab HA, Gough J, Cooper DN, Stenson PD, Barker GLA, Edwards KJ, Day INM, Gaunt TR (2013) Predicting the functional, molecular, and phenotypic consequences of amino acid substitutions using hidden Markov models. Hum Mutat 34(1):57–65. https://doi.org/10.1002/humu.22225
18. Huber CD, Kim BY, Lohmueller KE (2020) Population genetic models of GERP scores suggest pervasive turnover of constrained sites across mammalian evolution. PLoS Genet 16(5):e1008827. https://doi.org/10.1371/journal.pgen.1008827
19. Tamborero D, Rubio-Perez C, Deu-Pons J, Schroeder MP, Vivancos A, Rovira A, Tusquets I, Albanell J, Rodon J, Tabernero J, de Torres C, Dienstmann R, Gonzalez-Perez A, Lopez-Bigas N (2018) Cancer genome interpreter annotates the biological and clinical relevance of tumor alterations. Genome Med 10(1):25. https://doi.org/10.1186/s13073-018-0531-8
20. Rogers MF, Shihab HA, Gaunt TR, Campbell C (2017) CScape: a tool for predicting oncogenic single-point mutations in the cancer genome. Sci Rep 7(1):11597. https://doi.org/10.1038/s41598-017-11746-4
21. Li MM, Datto M, Duncavage EJ, Kulkarni S, Lindeman NI, Roy S, Tsimberidou AM, Vnencak-Jones CL, Wolff DJ, Younes A, Nikiforova MN (2017) Standards and guidelines for the interpretation and reporting of sequence variants in cancer: a joint consensus recommendation of the Association for Molecular Pathology, American Society of Clinical Oncology, and College of American Pathologists. J Mol Diagn 19(1):4–23. https://doi.org/10.1016/j.jmoldx.2016.10.002
22. Richards S, Aziz N, Bale S et al (2015) Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. Genet Med 17(5):405–423. https://doi.org/10.1038/gim.2015.30
23. Torrezan GT, de Almeida FG, Figueiredo MC et al (2018) Complex landscape of germline variants in Brazilian patients with hereditary and early onset breast cancer. Front Genet 9:161. https://doi.org/10.3389/fgene.2018.00161
24. Wen H, Kim YC, Snyder C, Xiao F, Fleissner EA, Becirovic D, Luo J, Downs B, Sherman S, Cowan KH, Lynch HT, Wang SM (2014) Family-specific, novel, deleterious germline variants provide a rich resource to identify genetic predispositions for BRCAx familial breast cancer. BMC Cancer 14(1):470. https://doi.org/10.1186/1471-2407-14-470
25. Rajendran BK, Deng CX (2017) Characterization of potential driver mutations involved in human breast cancer by computational approaches. Oncotarget 8(30):50252–50272. https://doi.org/10.18632/oncotarget.17225
26. Shiovitz S, Korde LA (2015) Genetics of breast cancer: a topic in evolution. Ann Oncol 26(7):1291–1299. https://doi.org/10.1093/annonc/mdv022

Publisher's Note