Η συχνότητα του κληρονομικού καρκίνου μαστού στον Κρητικό πληθυσμό.

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ΠΕΡΙΛΗΨΗ

Εισαγωγή: η γενετική εξέταση γονιδίων που σχετίζονται με κληρονομικό καρκίνο του μαστού συστήνεται όλο και συχνότερα στην κλινική πρακτική. Έχει δειχθεί ότι παραλλαγές σε 9 γονίδια (*ATM, BRCA1, BRCA2, CHEK2, PALB2, BARD1, RAD51C, RAD51D* και *TP53*) σχετίζονται με σημαντικό κίνδυνο εμφάνισης καρκίνου του μαστού. Στόχος της εργασίας είναι η εξέταση των γονιδίων αυτών σε πληθυσμό γυναικών Κρητικής καταγωγής με καρκίνο μαστού και υγιείς μάρτυρες.

Μέθοδοι: γυναίκες με καρκίνο μαστού ανεξάρτητα από την ηλικία διάγνωσης και το οικογενειακό ιστορικό καρκίνου και υγιείς μάρτυρες, όλες κρητικής καταγωγής, ήταν επιλέξιμες. Γαμετικό DNA απομονώθηκε από το περιφερικό αίμα και παραλλαγές σε 34 γονίδια εξετάστηκαν με το panel γονιδίων BRIDGES.

Αποτελέσματα: 475 ασθενείς και 275 υγιείς μάρτυρες εντάχθηκαν στη μελέτη. Η μέση ηλικία διάγνωσης για τις ασθενείς ήταν 55.8 έτη (±12.4) και ένταξης για τις υγιείς 61.40 (16.92). Σε περίπου 4,2% των ασθενών ανιχνεύθηκαν παραλλαγές που προκαλούν αποκοπή του άκρου της πρωτεΐνης (pathogenic truncating variants: PTV), έναντι 1,34% στις υγιείς ($p < 0.05$).

Επιπλέον σε 3 ασθενείς ανιχνεύθηκαν άλλες παθογόνες παραλλαγές (non PTV), (*ATM* n=2 και *BRCA1* n=1), έναντι δύο στον υγιή πληθυσμό (*ATM* n=2). Σε 19 από τα υπόλοιπα 25 γονίδια, από ένα PTV ανιχνεύθηκε στα γονίδια *MRE11A, MUTYH, NF1, MSH6, RINT1, RAD50* και *PIK3CA* στις ασθενείς και στα γονίδια *BRIP1, EPCAM* and *MUTYH* στις υγιείς.

Συμπέρασμα: τα αποτελέσματα αναδεικνύουν τα γονίδια που σχετίζονται με αυξημένο κίνδυνο εμφάνισης καρκίνου μαστού στον Κρητικό πληθυσμό. Η πληροφορία είναι σημαντική για το γενετικό έλεγχο και τη συμβουλευτική.

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INTRODUCTION

In a recent study by Dorling et al. [1], 34 associated and suspect breast cancer susceptibility genes were assessed for their relation to increased risk of developing breast cancer in the general population. Of these, protein-truncating variants in five genes (*ATM, BRCA1, BRCA2, CHEK2* and *PALB2*) were convincingly associated with breast cancer at $p<0.0001$ whereas protein-truncating variants in four other genes (*BARD1, RAD51C, RAD51D* and *TP53*) were associated at the nominal significant level $p<0.05$.

In this study we aimed to explore the protein truncating variants in these genes in a series of case/control samples from the Cretan population.

METHODS

Raw VCF files were generated using the VarDict variant caller [2]. The following filters were applied at VCF level; phred scaled sequencing quality score (QUAL) >= 30, allele fraction (AF) >= 0.2 and mean mapping quality (MQMEAN) >= 60, mean number of mismatches per read (NM) <= 2.0, AFxBase Depth >= 7.5. Variants with amplicon bias and/or failing any of these filters were removed.

Three main variant categories defined as classified by the ANNOVAR annotation tool [3], were examined; protein-truncating variants (PTVs, frameshifting insertions/deletions, stop/gain or canonical splice variants), non-PTVs classified as pathogenic or likely pathogenic (by either ClinVar, https://www.ncbi.nlm.nih.gov/clinvar/; ENIGMA *BRCA1/2* expert panel guidelines, https://enigmaconsortium.org/; *TP53* American College of Medical Genetics (ACMG) guidelines; *TP53* published quantitative model for *TP53* missense variant classification [4]) and rare unclassified missense variants (population frequency (by the Exome Aggregation Consortium (ExAC) of <0.001).

MATERIALS

Women diagnosed with breast cancer irrespective of age or family history were eligible for inclusion in the study. Cases and controls were recruited from the University Hospital of Heraklion, Crete.

475 cases and 275 controls passed all quality control filters and were used in the analyses. Mean (SD) age of diagnosis for cases was 55.8(12.4) and mean age of interview for controls 61.4(16.9).

The BRIDGES panel was used for the sequencing which consists of 34 genes associated or suspected to be associated with increased risk of breast cancer.

RESULTS

The most prevalent PVT in cases were identified in *BRCA1* (1.26%), followed by *ATM, BRCA2, CHEK2, PALB2*, *RAD51C* and *RAD51D*. No PTV were identified in *BARD1* or *TP53*. In controls, we identified PTVs in *CHEK2, ATM* and *PALB2*, no other PTVs were identified in any of the other breast cancer susceptibility genes.

In cases, 76 unique rare missense variants in established BC susceptibility genes were detected in 85 samples (17.9%), whereas in controls, 31 unique rare missense variants in established BC susceptibility genes, were detected in 32 samples (11.6%). We also identified pathogenic variants in 19 of the remaining 25 suspected genes, but non in excess in cases compared to controls.

	Cases		Controls	
	PTV number	PTV %	PTV number	PTV %
BRCA1	6	1.26	-	-
BRCA2	3	0.63	-	-
PALB2	2	0.42	1	0.27
ATM	4	0.84	2	0.54
CHEK2	3	0.63	2	0.54
BARD1	-	-	-	-
RAD51C	1	0.21	-	-
RAD51D	1	0.21	-	-
TP53	-	-	-	-
Total	20	4.21	5	1.34

Table 1. Pathogenic truncating variants identified in the nine associated genes

DISCUSSION

Here we present the results of nine genes associated with increased risk of developing breast cancer in the Cretan population. The prevalence of PTVs in this large series of unselected cases and controls provide important information as to the frequency of the variants in the general population and could guide further clinical genetic testing.

The BRIDGES study provided information on which genes are most clinically useful for use in clinical practice. Of the 34 genes on the panel five were shown to be strongly associated with breast cancer (*ATM, BRCA1, BRCA2, CHEK2* and *PALB2)* and another four (*BARD1 RAD51C, RAD51D* and *TP53*) to a lesser extend. For the rest of the genes there were no evidence of association with increased breast cancer risk. Here we present the frequencies of the PTVs identified in these genes in the Cretan population of cases and controls unselected for family history.

These results are comparable between our population and the general European Ancestry population.

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