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Genetic Variation of *hTERT*, Leukocyte Telomere Length and the Risk of Breast Cancer: A Case-Control Study in Egyptian Females

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Abstract

Background: *hTERT* is a key player in telomere biology and its activity is directly related to cell senescence and development of many health-related problems including cancer. Although previous studies investigated this association, the results greatly vary among populations. This study aimed to investigate the association of *hTERT* gene SNPs and the risk of breast cancer (BC) in Egyptian females and their impact on telomere length (TL). **Methods**: 218 BC patients and 178 age-matched healthy females were genotyped for *hTERT* variants rs2736098G > A, rs2735940C > T using PCR-RFLP and for MNS16A tandem repeat using PCR to determine their association with breast cancer risk. Telomere length was measured using *qPCR*. **Results**: *hTERT* rs2736098G > A results indicated that both AG and GG genotypes and G allele were associated with an increased risk of BC. The rs2735940 TT genotype was significantly associated with BC risk, however, the MNS16A tandem repeat region polymorphism didn't show any correlation with the risk of developing BC. TL showed a significant reduction in BC patients with age < 40 years compared with controls. However, it didn't show a significant difference above the age of 40 years. **Conclusions**: *hTERT* rs2736098 and rs27365940, not MNS16A may be associated with an increased risk of developing BC in Egyptian females. Also, telomere length can be a promising screening marker of BC especially in young population.

Keywords

Human Telomerase Reverse Transcriptase (*hTERT*), Single Nucleotide

Polymorphism, Telomere Length, Breast Cancer Risk

1. Introduction

Breast cancer (BC) is a serious health problem that women face around the world. In 154 of 185 countries, BC is the most frequently diagnosed cancer according to the GLOBOCAN 2018 and is the highest mortality cause in women in more than 100 countries [1]. The most rapid increase in incidence rates of BC shows an incline in countries that have been historically relatively low like South America, Africa, and Asia [2]. BC pathogenesis is affected by several factors, including genetic, family history, lifestyle, endocrine factors, as well as social and economic development [3]. However, there is a compelling need to clarify its exact etiology.

Telomeres are (TTAGGG)n repetitive sequences of DNA that can be found at the ends of chromosomes. Telomeres function is to maintain genetic stability by protecting chromosome during repetitive DNA replication cycles [4]. Due to the end-replication problem, telomeres are shortened in most somatic tissues, and are proposed physiological markers of aging and age-related pathology as well as cancer [5]. The function of telomeres and its role in cancer development is mainly determined by telomere length (TL) [6]. Although shortened telomeres can induce apoptosis, a critical length threshold is required. Otherwise, cells may bypass such signals and lead to what is known as telomere crisis. During telomere crisis, genomes experience an extensively unstable state that is a typical event in cancer progression [7] [8].

Telomeres are maintained by the telomerase complex, which consists of the Telomerase reverse transcriptase (TERT) catalytic subunit encoded by gene positioned at chromosome 5p15.33, and the Telomerase RNA component (TERC) encoded by the gene positioned on chromosomal region 3q26, with several other associated proteins required for proper telomerase assembly and recruitment to chromosomes [9]. While TERC is widely expressed, *hTERT* is downregulated in most human somatic cells leading to progressive telomere shortening [10].

Given the key role of *hTERT* in maintaining telomere stability, several common functional single nucleotide polymorphisms (SNPs) of the *hTERT* gene promoter region may impact the risk of different types of cancers [11] [12]. The relationship between *hTERT* polymorphisms and BC risk has been investigated in a number of studies [13] [14] [15]. Downstream of the *hTERT* gene, a polymorphic region known as the MNS16A minisatellite region was also found to be linked with the *hTERT* activity in many types of cancers [16] [17], however, the results were inconclusive.

The association of *hTERT* genomic variation with tumor biology draws attention to the need to verify this relation in different populations with diverse ancestries. In this case-control study, we genotyped a number of *hTERT* variants

(rs2736098G > A, rs2735940C > T and MNS16A) since the previous studies reached paradoxical conclusions regarding their association with cancer risk. We also measured the leukocyte telomere length and analyzed their association with BC risk in a sample of Egyptian females.

2. Methods

2.1. Patients

This case-control study enrolled 218 Egyptian breast cancer female patients who were referred to the Medical Research Institute and Ayadi Al-Mostakbal hospitals (Egypt) and 178 age-matched healthy females; they were unrelated to the patients and had no history of any type of cancer. Ethical approvals for recruitment were obtained from the local Ethics Committee of Medical Research Institute, University of Alexandria, and informed consent was obtained from each patient and healthy individual.

2.2. DNA Extraction

Whole blood samples were collected from breast cancer patients and healthy controls into EDTA-coated Vacutainer tubes and stored at −80˚C till genomic DNA was extracted. Genomic DNA was extracted using Invitrogen PureLink™ Genomic DNA Mini Kit (Thermo Fischer Scientific, USA) according to the manufacturer's instructions. The purity and concentration of DNA were evaluated by NanoDrop® ND-1000 UV-Visible Spectrophotometer (Thermo Fischer Scientific, USA). The extracted DNA was stored at −20˚C for further use.

2.3. *hTERT* Genotyping

Polymerase chain reaction-restriction fragment length polymorphism (PCR–RFLP) analysis was performed for genotyping of *hTERT* rs2736098G > A and rs2735940C > T polymorphisms. The primers' sequences, length of amplified DNA fragments, restriction pattern, and restriction enzymes used are presented in **Table 1**. PCR reactions were performed in a final volume of 25 µl contained 12.5 µl of 2x Dream Taq green master mix with dual dye (Thermo Fischer Scientific, USA), 100 ng genomic DNA, 0.2 µmol of each appropriate primer. The PCR cycling conditions were: a denaturation step of 5 min at 95˚C; 30 cycles of 30 s at 94˚C, 30 s at 57˚C (for rs2736098G > A) or 56˚C (for rs2735940C > T), and 30 s at 72˚C; a final elongation step of 7 min at 72˚C. Each PCR product was digested using 10 U of the corresponding restriction enzyme Bsp120I at 37˚C for 1 hour then the reaction was stopped by incubation at 80˚C for 10 minutes. After digestion, then the products were electrophoresed on 3% agarose gel containing 0.5 µg/mL ethidium bromide and visualized under UV illumination.

The MNS16A variable number of tandem repeat polymorphisms was genotyped using PCR as Wang *et al.* [18] reported previously. The primers' sequences and length of amplified DNA fragments were also mentioned in **Table 1**. The PCR conditions started with an initial denaturation step of 5 min at 95˚C; 30

Table 1. Primer sequences, DNA fragment length and restriction enzyme and restriction pattern (if available) for *hTERT* rs2736098, rs2735940 and MNS16A polymorphisms.

Polymorphism	Primer Sequence	DNA fragment Length	Restriction Enzyme	Restriction pattern (bp)
rs2736098 (G > A)	F: 5'AGGACGCGTGGACCGAGTGA-3' R: 5'GGAACCCAGAAAGATGGTCTC-3'	324 bp	Bsp120I (PspOMI)	AA: 324, AG: 324, 187, 137 GG: 137, 187
rs2735940 (T > C)	F: 5' ATCTTCTGCTTCCATTTCTTCTC 3' R: 5'TCGTCTTGTAAATACTTAGGATTCC 3'	235 bp	MspI (HpaII)	TT: 235 TC: 235, 211, 24 CC: 211, 24.
MNS16A	F: 5'-AGGATTCTGATCTCTGAAGGGTG-3' R: 5'-TCTGCCTGAGGAAGGACGTATG-3'	S: 243, 272 bp L: 333, 302 bp	--	--

cycles of 30 s at 94˚C, 30 s at 57˚C, and 30 s at 72˚C; a final elongation step of 7 min at 72˚C. The PCR product was then electrophoresed on 3% agarose gel containing 0.5 μg/mL ethidium bromide and visualized under UV illumination.

2.4. Telomer Length Determination

Leukocyte telomere length measurement was carried out according to the quantitative PCR method reported previously by Gil *et al.* [19]. The rhodopsin gene was selected as single copy gene to normalize the telomere PCR. The primer sequences for both telomere length and rhodopsin were as follows: Telomere Length 1 5'-CGGTTTGTTTGGGTTTGGGTTTGGGTTTGGGTTTGGGTT-3'; Telomere Length 2 5'-GGCTTGCCTTACCCTTACCCTTACCCTTACCCTTACCCT-3' Rhodopsin Forward 5'-CGTGGCCTGGTACATCTTCA-3' and Rhodopsin Reverse 5'-ACGCCGGCAGGGTCAT-3'. All samples were run in an Applied Biosystems Real-Time PCR System (Applied Biosystems, Foster City, CA). PCR reactions were performed in a final volume of 25 μl contained 12.5 μl of SYBR Green master mix (Thermo Fischer Scientific, USA), 100 ng genomic DNA, 2.5 μl primers, 8 μl DNase-free water. The PCR cycling conditions were a denaturation step of 10 min at 95˚C followed by 25 cycles of 15 s at 95˚C, 120 s at 54˚C. The Ct was calculated for both telomere and Rhodopsin were calculated. Telomere length was represented as a T/S ratio where (T) represented the telomere repeat copy number and (S) represented the single copy gene number according to the equation reported previously [20].

2.5. Statistical Analysis

Statistical analysis was performed using the Statistical Package for Social Sciences Software (SPSS 21; SPSS Inc.; Chicago, IL, USA). The frequencies of genotypes, alleles between breast cancer patients and healthy control as well as the clinicopathological characteristics were analyzed using Person chi-square test. The association between genotypes and alleles and the risk of breast cancer were assessed by computing the odds ratio (OR) and 95% confidence intervals

(95% CI) from unconditional logistic regression analyses. The statistical analysis of telomere length was performed using Mann-Whitney U nonparametric test. A p value of <0.05 was considered statistically significant. The Hardy-Weinberg equilibrium was tested for all polymorphisms.

3. Results

3.1. Characteristics of Study Population

The age of breast cancer patients and healthy controls enrolled in the study are presented in Table 2. There was no significant difference between patients and controls regarding age (p = 0.464). Table 2 also presents the distribution of clinical characteristics of breast cancer patients including histological grade, axillary lymph node involvement and receptors' status.

Table 2. Description of the Clinicopathological Characteristics of Breast Cancer Patients.

Characteristic	Breast Cancer Patients n-218	
Age	Mean ± S.D.	49.68 ± 1.39
Menstrual status	Perimenopausal	83 (38%)
	Postmenopausal	135 (62%)
Family History	Positive	47 (21.6%)
	Negative	169 (77.5%)
	Unknown	2 (0.9%)
Tumor type	Invasive Ductal Carcinoma	213 (97.7%)
	Invasive Lobular Carcinoma	5 (2.3%)
Histological Grade	II	165 (75.7%)
	III	51 (23.4%)
	Unknown	2 (0.9%)
Vascular Invasion	Positive	145 (66.6%)
	Negative	71 (32.5%)
	Unknown	2 (0.9%)
Axillary Lymph Node Involvement	Positive	161 (73.9%)
	Negative	55 (25.2%)
	unknown	2 (0.9%)
Estrogen Receptor (ER)	Positive	146 (66.7%)
	Negative	72 (33.3%)
Progesterone Receptor (PR)	Positive	131 (60%)
	Negative	87 (40%)
Her2/neu Receptor	Positive	66 (30.3%)
	Negative	142 (65.1%)
	Unknown	10 (4.6%)

3.2. The Impact of *hTERT* rs2736098G > A, rs2735940C > T and MNS16A on Breast Cancer Risk

The different genotypes of studied *hTERT* polymorphisms are illustrated in Figure 1. The genotype and allele frequencies of *hTERT* polymorphisms for breast cancer cases and controls as well as their association with BC risk are presented in Table 3. The statistical analysis of rs2736098G > A frequency distribution showed that the GG increased the risk of breast cancer (OR = 7.484, 95% CI = 2.593 - 21.597, $p \leq 0.001$ and OR = 1.146, 95% CI = 1.080 - 1.216, $p \leq 0.001$ respectively) compared to AA genotype which significantly lowered the risk of BC (OR = 0.332, 95% CI = 0.195 - 0.563, $p \leq 0.001$). Our results also indicated that the G allele was associated with higher BC risk compared to the A allele (OR = 1.743, 95% CI = 1.168 - 2.600, p = 0.006). The rs2735940C > T genotyping showed that the TT genotype was associated with a significant increase in the risk of BC compared to the rest of genotypes (OR = 1.519, 95% CI = 1.008 - 2.289, p = 0.045). However, neither the MNS16A genotypes nor its alleles showed any association with BC risk or protection. The association between the studied *hTERT* polymorphisms and BC risk was further examined by stratifying the subjects according to tumor histology (Table 4).

3.3. Telomere Length

Subjects in both BC and control groups were stratified into two subgroups according to their age with cut-off 40 years of age. For estimating the leukocyte telomere length, the telomere/single copy gene (T/S) ratio was calculated for all subjects. The statistical analysis revealed that the T/S ratio of BC patients with age less than 40 years showed a significant reduction in TL compared with controls with matched age, however, the TL in both patients and controls with age older than 40 years did not show a significant difference ($p \leq 0.001$ and 0.963 respectively) (Figure 2). In our study, the mean TL changed with different *hTERT* polymorphisms genotypes. For rs2736098G > A, the TL decreased with every G allele, and the T/S ratio was 1.14 for AA, 0.71 for AG, and 0.40 for GG, respectively. The GG genotype was significantly shorter than the AA genotype (p = 0.045). For the rs2735940C > T polymorphism, and T/S ratio was 1.22 for CC, 0.88 for TC, and 0.56 for TT. Both CT and TT genotypes were significantly shorter than the CC genotype (p = 0.002 and < 0.001 respectively). Regarding the MNS16A tandem repeat, the mean T/S ratio was 1.62 for LL, 0.83 for LS and 0.54 for SS genotypes. The LS and SS genotypes were also significantly shorter than the LL genotype (p = 0.012 and 0.062 respectively) (Figure 3).

4. Discussion

Given the fact that telomeres are maintained by telomerase complex, it's been proposed that genetic polymorphisms in its catalytic subunit *hTERT* can have a significant influence on the predisposition to developing cancer [21]. In the current case-control study, we investigated the association of three SNPs in the

hTERT gene with the risk of BC in a sample of Egyptian females. Our results indicated that the GG genotype and G allele of *hTERT* rs2736098G > A and TT genotype of rs2735940C > T were associated with increased risk for BC, while MNS16A didn't show any significant association.

Table 3. Genotype and allele frequencies distribution of *hTERT* polymorphisms (rs2736098, rs2735940 and MNS16A) in breast cancer patients and healthy control groups and their association with breast cancer risk.

	BC patients	Control	X^2	p-value	Odds ratio	95% CI
	n = 218	n = 178				LL-UL
rs2736098						
Genotypes						
AA	25 (11.5%)	48 (27.3%)	15.456	**<0.001**	1	-
AG	161 (73.8%)	124 (70.5%)	0.999	0.318	1.230	0.792 - 1.909
GG	32 (14.7%)	4 (2.2 %)	14.774	**<0.001**	**7.484**	2.593 - 21.597
Alleles						
A	(48.4%)	(62.5%)	7.447	**0.006**	1	-
G	(51.5%)	(37.5%)			**1.743**	1.168 - 2.600
rs2735940						
Genotypes						
CC	28 (12.8%)	30 (16.9%)	1.255	0.263	1	-
TC	95 (43.6%)	88 (49.4%)	1.447	0.229	0.790	0.531 - 1.175
TT	95 (43.6%)	60 (33.7%)	4.223	**0.040**	**1.519**	1.008-2.289
Alleles						
C	(36%)	(40%)	2.567	0.109	1	-
T	(65%)	(60%)			1.41	0.926 - 2.147
MNS16A						
Genotypes						
LL	80 (36.7%)	55 (30.9%)	1.353	0.245	1	-
LS	86 (39.4%)	68 (38.2%)	0.095	0.758	1.054	0.702 - 1.582
SS	52 (23.9%)	55 (30.9%)	2.458	0.117	0.701	0.449 - 1.093
Alleles						
L	56%	50%	0.091	0.763	1	-
S	44%	50%			0.941	0.634 - 1.397

Table 4. Association of the *hTERT* polymorphisms (rs2736098, rs2735940 and MNS16A) genotypes with clinicopathological characteristics.

Characteristics	*hTERT* rs2736098			*hTERT* rs2735940			*hTERT* MNS16A		
	AA	AG	GG	CC	TC	TT	LL	LS	SS
Family History									
Positive	9	41	2	9	20	18	10	22	14
Negative	16	118	30	19	75	75	69	63	38
X^2	2.200	0.966	6.529	1.693	0.023	0.555	5.545	1.759	1.294
p-value	0.138	0.326	**0.011**	0.193	0.880	0.463	**0.019**	0.185	0.255
Grade									
II	20	114	29	24	79	56	58	63	42
III	4	46	2	4	15	38	21	22	10
X^2	0.315	5.916	6.394	2.426	9.323	19.122	0.281	0.137	1.041
p-value	0.575	**0.015**	**0.011**	0.119	**0.002**	**<0.001**	0. 596	0.711	0.307
Axillary Lymph Nodes Involvement									
Positive	19	125	17	18	79	63	70	58	40
Negative	5	35	15	9	16	31	10	27	11
X^2	0.305	4.186	9.074	1.007	7.879	4.754	6.949	7.384	0.017
p-value	0.581	**0.041**	**0.003**	0.316	**0.005**	**0.029**	**0.008**	**0.007**	0.898
Vascular Invasion									
Positive	18	105	22	15	67	63	54	55	36
Negative	7	54	10	14	27	30	25	30	16
X^2	0.304	0.020	0.118	3.603	1.297	0.028	0.085	0.373	0.137
p-value	0.581	0.887	0.731	0.058	0.255	0.868	0.771	0.541	0.711
ER Status									
Positive	13	110	24	23	63	58	63	50	34
Negative	12	51	8	5	31	36	17	35	17
X^2	3.062	0.223	0.978	1.747	0.069	1.397	7.571	4.700	0.343
p-value	0.080	0.637	0.616	0.186	0.793	0.237	**0.006**	**0.030**	0.558
PR Status									
Positive	113	92	26	20	58	49	56	41	35
Negative	12	69	6	8	37	46	24	44	17
X^2	0.771	2.233	7.001	1.577	0.541	3.088	4.472	9.302	1.205
p-value	0.380	0.135	**0.008**	0.209	0.462	0.079	**0.034**	**0.002**	0.272
Her2/neu Status									
Positive	5	48	16	8	23	38	20	30	12
Negative	18	107	14	18	69	52	55	56	33
X^2	0.077	1.335	6.427	0.077	4.971	5.860	0.660	1.608	0.322
p-value	0.781	0.248	**0.011**	0.781	**0.026**	**0.015**	0.417	0.205	0.570

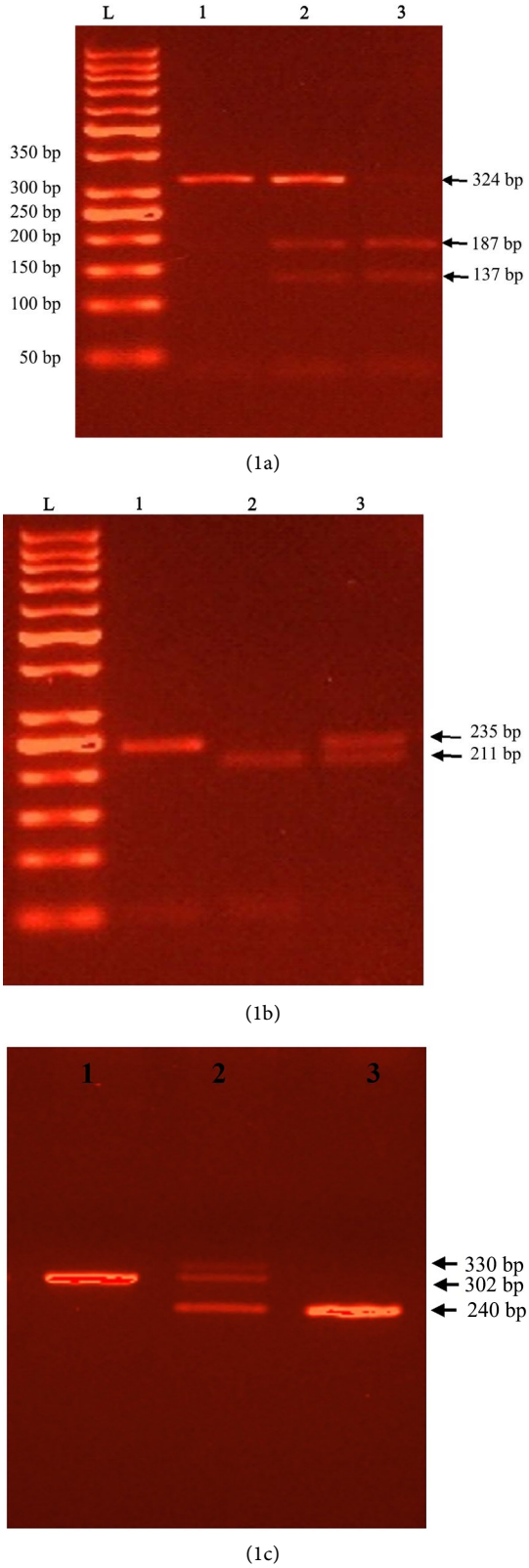


Figure 1. Photographs of DNA gel electrophoresis for the detection of *hTERT* polymorphisms. 1a: rs2736098. Lane 1, AA; Lane 2, AG; lane 3, GG. 1b: rs2735940. Lane 1, TT; lane 2, CC, lane 3, TC. 1c: MNS16A. Lane 1 LL; lane 2 LS; lane 3 SS. L: 50 bp DNA ladder.

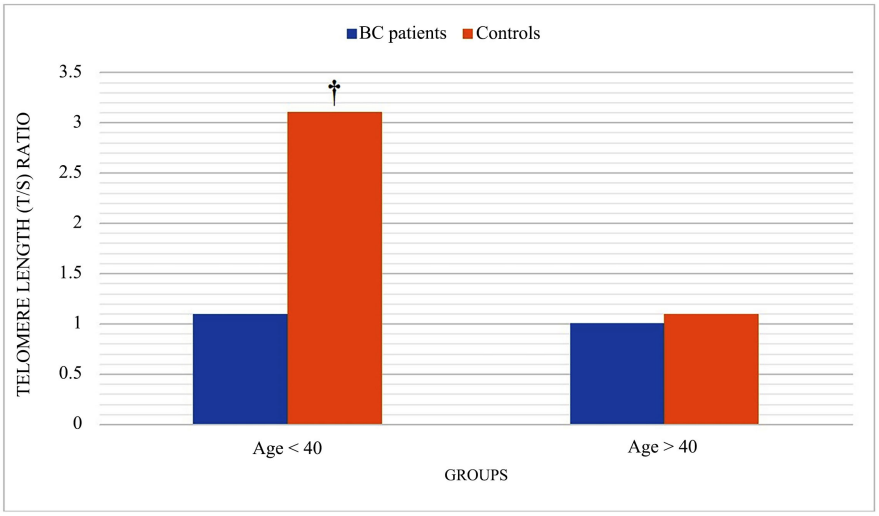
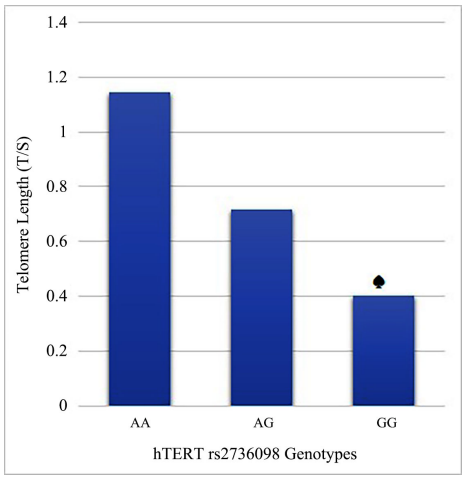
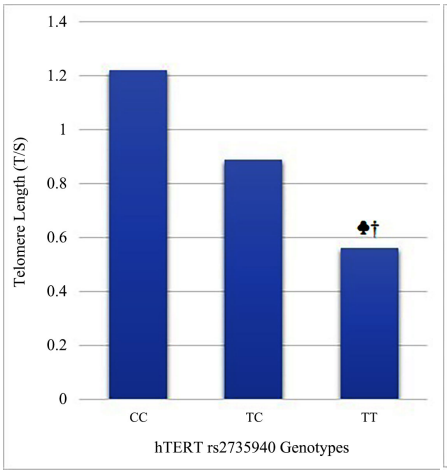


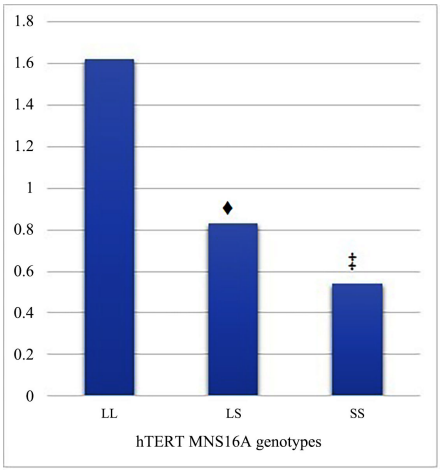
Figure 2. Statistical comparison between telomere in BC patients and control groups according to their age group ($p < 0.05$).



(3a)



(3b)



(3c)

Figure 3. Mean telomere length of BC patients stratified according to (3a) rs2736098, (3b) rs2735940 and (3c) MNS16A genotypes respectively.

One of the most commonly studied SNPs in the TERT gene is the rs2736098G > A polymorphism, it is associated with the risk of many types of cancer [22]. Although this SNP results in silent or synonymous mutation, thus the encoded amino acid remains unchanged (Asn305Asn), it may affect both telomerase activity and TL because of its location within the gene regulatory elements resulting in alteration of transcription factor binding [23].

The results of the present work are supported by many previous studies, Oztas *et al.* [24], reported that rs2736098 GG genotype is associated with increased risk of BC among Turkish women (OR = 1.88). Furthermore, G allele of *hTERT* rs2736098G > A is reported to be associated with BC risk in Iranian population (OR = 1.38). Hashemi *et al.* [25] reported that the *hTERT* rs2736098 G > A variant is associated with BC risk. Their results showed that GA as well as GG and GA + GG increased the risk of BC compared with the AA genotype. The rs2736098 G allele increased the risk of BC compared with the A allele. A meta-analysis released in 2018 indicated the association between BC susceptibility and rs2736098 variant, where A allele had a protective role against BC in allelic, homozygous, and recessive models [26]. Nevertheless, the association of *hTERT* rs2736098 variant with BC risk is inconclusive in some populations. In the Polish population, Savage *et al.* [27] genotyped different SNPs including rs2736098G > A in the *hTERT* genomic region, and no significant association with BC risk was found. Although, his results suggested that *hTERT* variants may be associated with the risk in women having a positive family history of BC. On the other hand, in women of African descent, no association was reported [28].

The association between *hTERT* rs2735940C > T polymorphism and cancer susceptibility is contentious. Pande *et al.* and Walsh *et al.* reported that either T allele or TT genotype of *hTERT* rs2735940C > T polymorphism may be significantly associated with the risk of developing lung cancer in ethnic Koreans, non-Hispanic white population and subjects of African–American ethnicity [29] [30]. In addition, TT genotype was linked with the risk of childhood acute lymphoblastic leukemia in Chinese population [31]. Moreover, Bayram *et al.* [32] indicated that carriers of TT genotype and/or T allele of *hTERT* rs2735940C > T polymorphism are at higher risk of developing gastric cancer in codominant, dominant and log-additive inheritance models. In accordance with the aforementioned data, a meta-analysis involved 19,385 cancer patients and 17,558 healthy subjects indicated that cancer risk significantly increased for T allele and/or TT genotype of *hTERT* rs2735940 polymorphism [33]. On the other hand, few studies reported different results. In Caucasian population, *hTERT* rs2735940C > T polymorphism has no role in colorectal cancer etiology [34]. Iizuka *et al.* have reported the T allele of *hTERT* rs2735940C > T polymorphism may be associated with a decreased risk of epithelial cancer, particularly prostate, lung and prostate cancer [35].

The MNS16A minisatellite region polymorphism in our study didn't show any correlation with neither the risk of developing BC. However, the long allele of

MNS16A was found to be significantly associated with negative family history and ER+/PR+ receptor pattern. Although MNS16A found to be linked with the *hTERT* activity in many types of cancers, a meta-analysis suggests that MNS16A is associated with the risk of developing gliomas, not other types of cancer including lung, colorectal, nasopharyngeal and breast cancer in Asian and Caucasian population [36]. This variation may be attributed to the differences in microenvironment of tumors of different sites. This may lead to different consequences for the same polymorphism. Collectively, the results of our chosen polymorphisms are comparable to those reported by Hashemi *et al.* in Iranian population. However, the MNS16A short allele not genotype was significantly associated with cancer risk [25].

Quantitative measurement of leukocyte TL of cancer patients with age less than 40 years showed a significant reduction in TL compared with controls with matched age, however, the TL in both patients and controls with age older than 40 years did not show a significant difference. There was a significant association between *hTERT* polymorphisms and telomere length. Telomere plays an essential part in protecting chromosomes and their progressive shortening in somatic cells induces chromosomal instability [37] [38]. Telomere shortening is associated with diverse age-related diseases [39], yet there are inconsistent reports on the relationship between telomere length and cancer incident. Liu *et al.* have reported that short leukocyte TL contributes to increased susceptibility of gastric cardia adenocarcinoma and, along with lifestyle risk factors, can be considered as a promising prognostic marker [40]. Moreover, the risk of developing lung cancer risks was related to short telomeres lengths in patients with breathing problems, with age, sex, and smoking adjustment [41]. In 2018, Pavanello *et al.* proposed that telomeres length was shorter in BC patients than age-adjusted controls however their study failed to link it with the risk of hereditary BC [42]. Excessive telomeres shortening drive too many cells into senescence. Senescent cells acquire a senescence-associated secretory phenotype which has deleterious effects on the tissue microenvironment and promotes tumor progression through proinflammatory signaling [43]. Still, other studies showed inconsistent results. Julin *et al.* findings suggested an association between the risk of overall prostate cancer, including more aggressive disease, with longer leukocyte TL in men with a positive family history of prostate cancer [44]. Similarly, there is an association between longer leukocyte TL and colorectal carcinoma, but not for its premalignant precursors [45]. In light of the results of these studies, further investigations were required to evaluate the possibility of using TL as a biomarker to evaluate the cancer risk.

From the results of the current study, we can conclude that *hTERT* rs2736098 and rs27365940, not MNS16A may be associated with increased risk of developing BC in Egyptian females. To our knowledge, this is the first study to investigate these polymorphisms in Egyptian population and their role in breast cancer risk. Also, we can conclude that BC patients younger than 40 years old have

shorter leukocyte TL when compared to age-matched controls. Thus, telomere length can be a promising screening marker of breast cancer especially in young population.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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