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Association of Adiponectin Gene Functional Polymorphisms (+45T/G and 276G/T) with Obese Breast Cancer

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Abstract

Adiponectin is a naturally occurring, active protein produced by white adipose tissue circulating levels of which have been associated with breast cancer risk. The ADIPOQ +45T/G and 276G/T polymorphisms are likely to play an important role in the susceptibility to breast cancer. To evaluate the prevalence of polymorphism of Adiponectin gene a case-control study was performed in 154 breast cancer patients and 142 controls in South India. We utilized PCR- RFLP based assay to evaluate the association between the +45T/G and 276G/T polymorphism of the ADIPOQ gene and breast cancer risk in a case control study. Frequencies of ADIPOQ +45T/G were 74%, 24% and 2% in the breast cancer patients and 83%, 15% and 2% in the controls, respectively. ADIPOQ +45T/G genotype showed a 1.7-fold increased risk for breast cancer and the 276G/T genotype showed a 1.6-fold increased risk for breast cancer. In the compound genotypes, T45G//G276T and T45G/T276T showed a 1.668 and a 1.791-fold increased risk for breast cancer. Our findings suggest that the ADIPOQ +45T/G and 276G/T polymorphism may be a useful biomarker associated with the risk of breast cancer in obese women in Indian population.

Keywords: Adiponectin; Breast cancer; Single nucleotide polymorphism (SNP); Restriction fragment length polymorphism (RFLP); Polymerase chain reaction (PCR)

Introduction

Obesity is a major health problem and is positively associated with breast cancer incidence and mortality [1,2]. In India obesity is a pandemic, and it leads to increased levels of fat in the body. Adipose tissue derived signaling molecules, including the adipokines, are emerging as key candidate molecules that link obesity with cancer. Increased adiposity tends to be associated with higher circulating levels of estrogens through greater peripheral conversion of androgens to estrogens by aromatase in adipose tissue; hence exogenous estrogen could increase the risk of breast cancer. Adiponectin gene is the most abundant human adipose-specific protein [3]. It belongs to a family of adipocytokines. The ADIPOQ gene consists of three exons and two introns spanning a 17-kb region and has been located on chromosome 3q27 [4]. Adipokines, particularly Adiponectin therefore has important roles in breast cancer biology. Polymorphisms of ADIPOQ have been associated with the risk for cancers, probably by affecting adiponectin plasma levels. Many studies have demonstrated that obesity could increase the risk of cancer, including breast cancer reduced plasma adiponectin levels were observed in patients with obesity. In obese patients, adiponectin, a protein secreted by adipose tissue, has lower expression than that in non-obese subjects this may be associated with an increased risk for developing breast cancer [5].

Various Single Nucleotide Polymorphisms (SNPs) have been identified in the adiponectin gene. One of these SNPs, 45T/G, in exon 2 of the adiponectin gene, has been frequently reported in association with obesity, and breast cancer, and second, rs1501299 (G276T) is in intron 2 of the ADIPOQ gene and does not have a known function. It is probably a marker of some other variant affecting adiponectin expression. To our knowledge, genetic polymorphisms of the adiponectin gene in south Indian population have not yet been studied. In addition, there is very little information about the genetic susceptibility to breast cancer in Indian population. Therefore, in this study, we investigated two SNPs (rs1501299 (G276T) and rs2241766 (T45G)) in ADIPOQ gene and their possible association with breast cancer susceptibility in obese women.

Patients and Methods

Study population

Breast cancer patients were assessed by pathologists on the basis of clinical examinations as well as mammography and clinical-pathological examinations. The breast cancer study is a Hospital-based case-control study conducted in South Indian population. All incident breast cancer cases were newly diagnosed during the study period. Ethical Committee approved the study for the benefit of humans in general. The procedures followed were in accordance with the ethical standards of the responsible committee of the Institutes/Hospitals, to participate in a face-to-face interview using a structured questionnaire.

Selection criteria

Senior pathologists confirmed all diagnoses. We interviewed and collected the data about the patient demographic factors; collected the information on age, menopausal status, smoking, usual alcohol intake, BMI and previous cancer diagnoses. Participants were also asked about their family history of cancer, and the clinical information in these cases was obtained from medical records like tumor size, Grade, Axillary nodes, and whether they were receiving chemotherapy, hormonal therapy or radiotherapy. Patients were recruited following certain inclusion and exclusion criteria, which were determined before the beginning of the study.

Inclusion and exclusion criteria: Patients included in the study were women with breast cancer who were found to have positive

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(tumor-involved) axillary lymph nodes and BMI of >27 kg/m^2 at the time of surgery. Patients must have adequate blood counts, and adequate kidney and liver function. Underweight Patients were excluded from the study.

Sample collection

Based on the above criteria, a total of 154 breast cancer patients and 142 age-matched controls were enrolled in the study. Sampling was done from two major Hospitals in Hyderabad, Andhra Pradesh (India) between the period 2008 to 2012. Those were Bhagwan Mahavir Hospital and Research Centre, Hyderabad, India and MNJ Cancer Hospital, Hyderabad, India.

Collection of biopsy and blood samples

Tumor breast tissues (biopsies) were collected in normal saline from the pathology lab after diagnosis. The tissues were, immediately transferred and stored at -80°C till further processing was done. The tumor samples obtained were of various tumor sizes and diagnosed mainly as invasive Ductal Carcinoma (IDLS). About 3 ml Blood samples were collected from healthy women (Voluntarily) by venipuncture. These samples were used as controls, in various experiments.

Collection of demographic factors and clinical data

All patients were interviewed for recording their demographic factors like age, BMI, and other details as listed in table 1. Clinical records for ER/PR status were obtained from medical records of the patients. Similarly we could record some demographic parameters of control subjects. Wherever possible, this data was correlated to obtain meaningful information to determine the risk factors of the disease. We also determined the significance of all the parameters for statistical evaluations.

Genotyping methodology

Genomic DNA was isolated by the salting-out method from the tissue samples of the cases and peripheral blood samples of the control group. The quantity of recovered DNA was determined spectrophotometrically. Polymerase Chain Reaction (PCR)-based Restriction Fragment Length Polymorphism (RFLP) was done to identify the Adiponectin 45T/G and 276G/T gene polymorphism, as described previously.

Detection details of SNP Adiponectin 45T/G

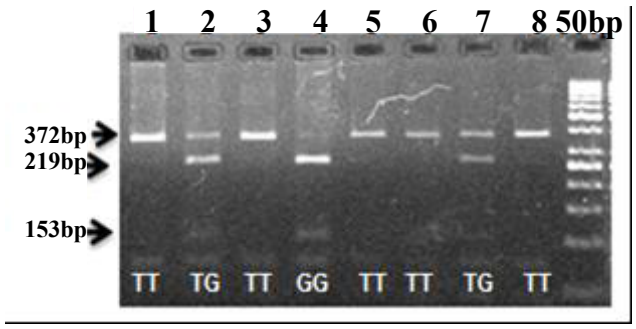
The PCR primer sequences used for 45T/G were 5- GAA GTA GAC TCT GCT GAG ATG G -3 (forward) and 5- TAT CAG TGT AGG AGG TCT GTG ATG -3 (reverse) (Bioserve Biotechnologies, Hyderabad, India). A three-step PCR was standardized using a Biorad system and carried out with an initial denaturation at 95°C for 5 minutes followed by cycling at 95°C for 45 seconds; 60.2°C annealing for 45 seconds, 72°C for 45 seconds extension and a final extension at 72°C for 7 minutes was carried out for about 35 cycles. Amplified PCR products were separated on 2% agarose gel, visualized with ethidium bromide stain. The amplified PCR products were subjected to RFLP using the *SmaI* (Fermentas, Hanover, MD, USA) restriction enzyme for 37°C overnight for enzyme digestion, and visualized on 3% agarose gel stained with ethidium bromide. Three different combinations of fragment lengths were obtained, The Presence of *Hha1* restriction sites which results 182 and 60 bp correspond to samples homozygous wild type, 242 bp representing samples homozygous mutant and a combination of three bands 242, 182 and 60 bp for heterozygous samples (Figure 1).

Detection details of SNP Adiponectin 276G/T

PCR primer sequences used for 276G/T were 5'- GGC CTC TTT CAT CAC AGA CC (forward) and 5'- AGA TGC AGC AAA GCC AAA GT -3' (reverse) (Bioserve Biotechnologies, Hyderabad, India). A three-step PCR was standardized using a Biorad system and carried out with an initial denaturation at 95°C for 5 minutes followed by cycling at 95°C for 60 seconds; 59°C annealing for 45 seconds, 72°C for 45 seconds extension and a final extension at 72°C for 5 minutes was carried out for about 35 cycles. Amplified PCR products were separated on 2% agarose gel, visualized with ethidium bromide stain. The amplified PCR products were subjected to RFLP using the *MVA1269I* (Fermentas, Hanover, MD, USA) restriction enzyme for 37°C overnight for enzyme digestion, and visualized on 3% agarose gel stained with ethidium bromide. Three different combinations of fragment lengths were obtained, The Presence of *MVA1269I* restriction sites which results 148 and 48 bp correspond to samples homozygous wild type, 196 bp representing samples homozygous mutant and a combination of three bands 196, 148 and 48 bp for heterozygous samples (Figure 2).

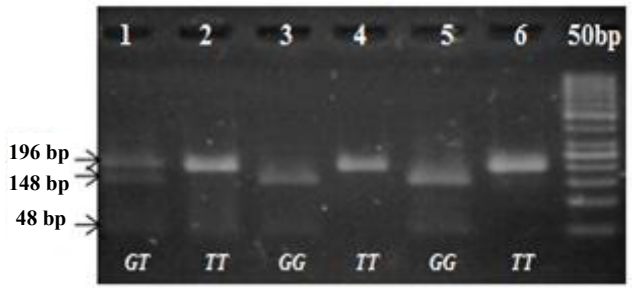
Statistical analysis

Genotyping experiments are presented as allelic frequencies and Genotype distribution with those expected from Hardy-Weinberg Equilibrium (HWE) were made using chi square test, and Values of P (two-tailed) less than 0.05 were considered statistically significant.



Lanes 2 and 7: Heterozygous mutant (TG)
Lane 4: Homozygous mutant (GG)
Lanes 1,3,5,6 and 8: Homozygous wild type (TT)
Lane 9: 50 bp Ladder

Figure 1: RFLP Gel Picture.



Lane 1: Heterozygous mutant (GT)
Lanes 2,4 and 6: Homozygous mutant (TT)
Lanes 3 and 5: Homozygous wild type (GG)
Lane 7: 50 bp Ladder

Figure 2: RFLP Gel Picture.

Odds ratio, were calculated using MedCalc for Windows, version 7.4.1.0 (MedCalc Software, Mariakerke, Belgium).

Results

Epidemiological studies

A total of 154 breast cancer patients and 142 age matched controls formed our study group. The distribution of biological characteristics and selected risk factors of the case controls are presented in table 1. The age range for breast cancer patients was 20-70 years. The mean age at which breast cancer was generally identified was 49.87 years. Breast cancer patients were divided into 5 groups according to age at diagnosis; these were 21-30, 31-40, 41-50, 51-60 and 61-70 years. Incidence of breast cancer cases and a control group was higher in the age groups 41-50 (32.4% and 28.2%) years when compared to other age groups, and the incidence was very low in the age group 61-70 years. The frequency of overweight and obese patients (25.9%, and 54.5%) and control (35.2%, 53.5%) showed a high Body Mass Index (BMI). Age at menarche of BC patients showed that about 54.5% of them had attained menarche between 13 -14 years, while 18% attained it by 15-16 years. This is similar to what was observed in the normal population. Depending on the menopausal status, breast cancer patients and controls were also categorized into premenopausal (48% and 57.8%). and postmenopausal groups (52% and 42.2%). It was observed the majority of sporadic breast cancer cases were higher in postmenopausal group when compared to premenopausal group.

Analyses of ADIPOQ T45G and G276T polymorphisms

We observed frequencies of the ADIPOQ T45G polymorphisms among 154 breast cancer patients and 142 controls. We found the ADIPOQ T45T genotype present among 114 (74%) breast cancer cases and 118 (83%) controls. The T45G genotype among 38 (24%) breast cancer cases and 22 (15%) controls and the G45G genotype among 2 (2%) breast cancer cases and 2 (1.5%) controls. Table 2 Shows that the genotype frequency and the allele frequency distribution of 154 obese breast cancer patients together with 142 controls. The frequencies of ADIPOQ T45T, T45G and G45G genotypes were 74%, 24% and 2% of breast cancer patients and 83%, 15%, 1.5% in the controls, respectively (Table 1). The T45G genotype showed 1.7 folds increased risk for breast cancer (OR 1.78, 95% CI=0.99-3.20, Chi square=3.30, p=0.05). The allele frequency of ADIPOQ 45G was 0.08% in the breast cancer patients and 0.13% in the controls. The frequencies of ADIPOQ GG, GT and TT genotypes were 55.5%, 41.6% and 2.6% of breast cancer patients and 83%, 15%, 1.5% in the controls, respectively (Table 3). The T45G genotype showed 1.6 folds increased risk for breast cancer (OR 1.63, 95% CI=1.01-2.64, Chi square=3.59, p=0.04). The allele frequency of ADIPOQ 276T was 0.23% in the breast cancer patients and 0.15% in the controls. The distributions of the alleles among the breast cancer patients and controls were in accordance with Hardy-Weinberg equilibrium. Further we analyzed the joint effects of the two polymorphisms ADIPOQ T45G and G276T polymorphisms as shown in table 2. In our study we found TT/GT and TG/TT compound genotypes showed that 1.668 and 1.791folds increased risk of breast cancer in obese women. (95% CI 1.15-2.40, χ^2=08.223, Odds Ratio 1.66, p=0.005 and 95% CI 1.04-3.06, χ^2=4.087, Odds Ratio 1.791, p=0.03) (Table 4).

Correlation of demographic factors and genotype frequencies of +45G/T and 276G/T polymorphisms

Data presented in tables 5 and 6 exposed associations between the ADIPOQ genotypes and seven individual demographic factors to understand the influence of ADIPOQ (+45T/G) polymorphism on the breast cancer risk. In the present study age group between 41-50 (32.4%) showed a high frequency, whereas Heterozygous T/G genotype was significantly associated with 31- 40 age (P=0.03) Group when compared with other age groups. The frequency of postmenopausal women was high 80% (52%), when compared with premenopausal women. In this study, total 38 cases were shown T/G genotype, out of this 15 cases were shown TG genotype in postmenopausal and 23 cases were shown in premenopausal women. 2 cases were shown GG genotype in premenopausal women, whereas no cases were found with GG genotype in postmenopausal women. Therefore, the difference in the distribution of genotypes between pre and postmenopausal showed that TG and GG genotypes were not statistically significant (p=0.06). To further evaluate the association between the +T/G polymorphism and BMI, participants were grouped according to BMI range. 40 (25.9%) cases were overweight and 42 (27.3%) cases were obese. In these study total 38 cases were shown TG genotype, 6 obese cases and 11 overweight cases showed TG genotype. Body Mass Index (BMI) status, age at menarche and her2 status distributions were not statistically significant with genotypes. Estrogen and progesterone receptors served as prognostic markers and predicted response to endocrine therapy. In the present study genotypes were correlated with ER/PR status to find whether these polymorphisms showed any association with ER/PR status. It was observed that patients with ER/PR negative tumors showed significant association with +45T/G and 276G/T genotypes (p=0.006).

Discussion

The World Health Organization declared obesity as one of the greatest public health challenges of the 21st century and projects that by 2015; more than 700 million adults will be obese. Obesity is on the rise and is associated with increased risk and progression of a number of cancers including colon, prostate, breast, and liver cancers. It has been well known that obesity is one of the major risks for breast cancer. The prevailing theory behind the association between obesity and breast cancer is based on the increased levels of estrogens observed in obese women [6]. As obesity has been associated with the development of breast cancer, adipocytokines, a group of polypeptide growth factors and cytokines which are produced exclusively by adipose tissue, may underlie the association between obesity and breast cancer risk.

Characteristics	Cases N=154(%)	Controls N=142(%)
Age of Diagnosis		
21-30	16(10%)	32(23%)
31-40	38(25%)	36(25%)
41-50	50(32%)	40(28%)
51-60	38(25%)	20(14%)
61-70	12(8%)	14(10%)
Body mass index(BMI) status		
Normal weight >18.50 to <24.99	30(19%)	16(11%)
Overweight >25 to <29.99	40(26%)	50(35%)
Obese >30.00	84(55%)	76(54%)
Age at Menarche		
<12	52(34%)	38(27%)
13-14	84(54%)	78(55%)
15-16	18(12%)	24(17%)
>17	0(0%)	2(1%)
Menopausal Status		
Post Menopausal	80(52%)	60(42%)
Pre Menopausal	74(48%)	82(58%)

Table 1: Demographic characteristics of patient and control individuals.

Genotype	Cases N=154(%)	Controls N=142(%)	95% CI	χ2 test	Odds ratio	P-Value
TT	114(74%)	118(83%)	0.32 to 1.02	3.07	0.57	0.05
TG	38(25%)	22(16%)	0.99 to 3.20	3.30	1.78	0.05
GG	2(1%)	2(1%)	0.17 to 9.01	0.17	1.25	0.82

Table 2: Distribution of Genotype frequencies of ADIPOQ +45T/G Polymorphism in obese breast cancer cases and controls.

Genotype	Cases N=154(%)	Controls N=142(%)	95% CI	χ2 test	Odds ratio	P-Value
GG	86(56%)	98(69%)	0.35 to 0.91	4.90	0.56	0.02
GT	64(42%)	43(30%)	1.01 to 2.64	3.59	1.63	0.04
TT	4(2%)	1(1%)	0.41 to 34.04	0.65	3.76	0.23

Table 3: Distribution of Genotype frequencies of ADIPOQ 276G/T Polymorphism in obese breast cancer cases and control.

Genotypes	Cases	Controls	95% CI	χ2 test	Odds ratio	P-Value
TT/GG	200	216	0.40 to 0.83	8.22	0.58	0.003
TT/GT	178	161	0.75 to 1.44	0.35	1.04	0.78
TT/TT	118	119	0.61 to 1.19	0.65	0.86	0.37
TG/GG	124	120	0.66 to 1.27	0.16	0.92	0.62
TG/GT	102	65	1.15 to 2.40	7.13	1.66	0.005
TG/TT	42	23	1.04 to 3.06	4.08	1.79	0.03
GG/GG	88	100	0.52 to 1.04	2.70	0.73	0.08
GG/GT	66	45	0.95 to 2.20	2.66	1.44	0.08
GG/TT	6	3	0.46 to 7.51	0.30	1.86	0.38

Table 4: Combined effect of both +45T/G and 276G/T Polymorphism in obese breast cancer cases and controls.

Characteristics	Cases N=154(%)	Wild type TT N=114(%)	Heterozygous TG N=38(%)	Homo mutant GG N=2(%)	P-Value
Age of Diagnosis					
21-30	16(10%)	10(63%)	6(37%)	0(0%)	0.22
31-40	38(25%)	22(58%)	14(37%)	2(5%)	0.03
41-50	50(32%)	42(84%)	8(16%)	0(0%)	0.07
51-60	38(25%)	28(74%)	10(26%)	0(0%)	0.82
61-70	12(8%)	12(100%)	0(0%)	0(0%)	0.12
Body mass index(BMI) status					
Normal weight >18.50 to <24.99	30(19%)	20(67%)	9(30%)	1(3%)	0.17
Overweight >25 to <29.99	40(26%)	29(73%)	11(27%)	0(0%)	0.38
Obese >30.00	84(55%)	65(86%)	18(14%)	1(0%)	0.06
Age at Menarche					
11-12	52(34%)	37(71%)	13(25%)	2(4%)	0.84
13-14	84(54%)	64(76%)	20(24%)	0(0%)	0.70
15-16	18(12%)	13(72%)	5(28%)	0(0%)	0.77
Menopausal Status					
Post Menopausal	80(52%)	65(81%)	15(19%)	0(0%)	0.06
Pre Menopausal	74(48%)	49(66%)	23(31%)	2(3%)	0.06
Estrogen Receptor status					
ER+ /PR+	48(31%)	38(79%)	10(21%)	0(0%)	0.42
ER+/PR-	10(7%)	4(40%)	6(60%)	0(0%)	0.01
ER-/PR+	6(4%)	6(100%)	0(0%)	0(0%)	0.30
ER-/PR-	90(58%)	66(73%)	22(25%)	2(2%)	1.00
Her2 status					
Her2+	135(88%)	100(74%)	33(24%)	2(2%)	0.88
Her2-	19(12%)	14(74%)	5(26%)	0(0%)	0.88
Chemotherapy					
5FU,Adriamycin,Endoxane	108(70%)	84(78%)	22(20%)	2(2%)	0.06
Hormonal therapy	11(7%)	9(82%)	2(18%)	0(0%)	0.59
RD	28(18%)	15(54%)	13(46%)	0(0%)	0.005
No Data	7(5%)	6(86%)	1(14%)	0(0%)	0.51

Table 5: Correlation of breast cancer patient's demographic factors and ADIPOQ +45G/T genotype frequencies.

Understanding of the adiponectin hormone plays an important role in obesity-related cancer, including breast cancer.

The present study makes an important contribution to our understanding of adiponectin gene in relation to breast cancer because it is the first to comprehensively evaluate known variations in adiponectin gene. To the best of our knowledge, this is the first study that investigated the association between the rs2241766 (+45T/G) and rs1501299 (276G/T) polymorphisms of ADIPOQ gene and breast cancer in south Indian population. The synonymous variation +45T>G (rs2241766) at exon 2 is most widely reported to be associated with breast cancer. In this study, significant difference was found in the distribution of ADIPOQ gene polymorphisms between breast cancer

Characteristics	Cases N=154(%)	Wild type GG N=86(%)	Heterozygous GT N=64(%)	Homo mutant TT N=4(%)	P-Value
Age of Diagnosis					
21-30	16(10%)	6(38%)	10(62%)	0(0%)	0.16
31-40	38(25%)	20(53%)	16(42%)	2(5%)	0.35
41-50	50(32%)	32(64%)	18(36%)	0(0%)	0.005
51-60	38(25%)	22(58%)	14(37%)	2(5%)	0.06
61-70	12(8%)	6(50%)	6(50%)	0(0%)	1.00
Body mass index(BMI) status					
Normal weight >18.50 to <24.99	30(19%)	17(57%)	13(43%)	0(0%)	0.14
Overweight >25 to <29.99	40(26%)	22(55%)	18(45%)	0(0%)	0.37
Obese >30.00	84(55%)	47(57%)	33(33%)	4(10%)	0.03
Age at Menarche					
11-12	52(34%)	34(65%)	16(31%)	2(4%)	0.0005
13-14	84(54%)	48(57%)	36(43%)	0(0%)	0.06
15-16	18(12%)	4(22%)	12(67%)	2(11%)	0.01
Menopausal Status					
Post Menopausal	80(52%)	48(60%)	30(38%)	2(2%)	0.004
Pre Menopausal	74(48%)	38(51%)	34(46%)	2(3%)	0.51
Estrogen Receptor status					
ER+ /PR+	48(31%)	28(58%)	20(42%)	0(0%)	0.10
ER+/PR-	10(7%)	8(80%)	2(20%)	0(0%)	0.01
ER-/PR+	6(4%)	4(67%)	2(33%)	0(0%)	0.25
ER-/PR-	90(58%)	46(51%)	40(44%)	4(5%)	0.37
Her2 status					
Her2+	135(88%)	78(58%)	54(40%)	3(2%)	0.003
Her2-	19(12%)	8(42%)	10(53%)	1(5%)	0.50
Chemotherapy					
5FU,Adriamycin,Endoxane	108(70%)	62(57%)	42(39%)	4(4%)	0.006
Hormonal therapy	11(7%)	6(55%)	5(45%)	0(0%)	0.67
RD	28(18%)	16(57%)	12(43%)	0(0%)	0.28
No Data	7(4%)	2(29%)	5(71%)	0(0%)	0.12

Table 6: Correlation of breast cancer patients demographic factors and ADIPOQ +45G/T genotype frequencies.

patients and controls. ADIPOQ +45T/G genotype showed a 1.7-fold and G276T genotype showed 1.6 folds increased risk for breast cancer. In our study compound genotypes TT/GT and TG/TT showed that 1.668 and 1.791folds increased risk of breast cancer in obese women. This suggests that these polymorphisms may modulate expression of adiponectin receptor 1 mRNA, was also associated with breast cancer risk. Our findings are consistent with three previous studies reported in different populations. In 2011, Al Khaldi et al. found that Adiponectin gene 45T>G was associated with breast cancer, Researcher demonstrate that adiponectin gene (+45T/G) and rs1501299 (276G/T) may be the predisposing factors in some cancers in the Kuwait population [7]. The *ADIPOQ* +45G allele (TG+GG) had a significant association with the risk of the metabolic syndrome [8], whereas in another study Wang et al. [9] reported that Distributions of 45T>G (rs2241766) genotypes were not significantly different between type 2 diabetes cases and controls both in Yi and Han people in China (p>0.05). Ranjzad et al. [10] in case-control study, showed significant association between the ADIPOQ 45T>G polymorphism and risk of polycystic ovary syndrome risk (PCOS). In 2011, Nyante et al. found that +45T/G and 276G/T were associated with breast cancer in whites and African Americans [11], whereas Kaklamani et al. in 2008 found that two functionally relevant adiponectin polymorphisms, +45T→G (rs2241766) and +276G→T (rs 1501299), were significantly associated with breast cancer risk. These polymorphisms increase circulating levels of adiponectin [12]. The genetic variations in the adiponectin gene can affect the circulating adiponectin level and stimulation of adiponectin receptor that may affect the activity of adiponectin. Previous studies have shown that Adiponectin concentrations are inversely correlated with breast cancer risk [13,14], whereas Lanos et al. in 2012 [15] reported that plasma adipokine concentrations may not be good surrogates for breast concentrations among all women. Duggan et al. in 2011 [16] performed a prospective investigation of the fasting serum adiponectin and insulin concentrations and the HOMA-insulin resistance index in stage I and II breast cancer patients and found that higher than median adiponectin concentrations were associated with a decreased risk of breast cancer mortality. A clear understanding is necessary of the impact of these hormones on breast cancer risk. Therefore, mechanisms other than alteration of serum adiponectin levels should be taken into consideration for the observed effect of this SNP. Several molecular links exist between adiponectin and breast carcinogenesis. Adiponectin is inversely correlated with insulin levels and is also associated with IGF-binding proteins, which have been associated with breast cancer [17]. Adiponectin also inhibits the activation of nuclear factor-nB, which is involved in breast cancer development [18]. Adiponectin has also been shown to inhibit the production of TNF-α in macrophages and its actions in endothelial cells [19], and has been found to bind several growth factors, which can induce cell proliferation.

Adiponectin is secreted by adipose tissue and its levels were inversely correlated with Body Mass Index. In the present study overweight and BMI and overweight cases are significantly associated with +45T/G and G276T genotype, suggesting that ADIPOQ gene polymorphism may contribute to weight gain and lead to breast cancer risk. In 2002 Menzaghi et al. observed that the +45 T/G and +276 G/T polymorphisms of the adiponectin gene contribute to weight gain and the development of insulin resistance [20]. Further we analyzed the association of menopausal status with TG genotype; we found that the frequency of TG genotype is significantly associated with premenopausal women, suggesting that weight gain in the later premenopausal years may lead to greater risk of breast cancer. Hormones play a critical role in breast carcinogenesis, determining the associations between hormone levels and breast cancer risk may provide insight into the etiology of this disease and may help identify women who are at high risk.

Conclusion

The present study investigated the association between +45T/G and 276G/T polymorphisms in the adiponectin gene and breast cancer in a South Indian population. Our results demonstrated that the presence of the TG genotype at the position +45 and GT genotype at the position 276 of the adiponectin gene may be associated with the risk of breast cancer. Further studies are needed to explore the complicated interaction between environmental factors and ADIPOQ gene polymorphisms in terms of susceptibility to breast cancer. To the best of our knowledge, ours is the first study to provide information on the role of ADIPOQ +45G/A polymorphism in breast cancer risk in South Indian women.

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