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Association Study between Single Nucleotide Polymorphisms of Vascular Endothelial Growth Factor and Risk of Breast Cancer Among Iranian Population

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ABSTRACT

Background: Vascular endothelial growth factor (VEGF) is a key mediator of angiogenesis which could act as an invasion factor. The aim of this study was to compare the frequency of alleles and genotypes of three polymorphisms in the *VEGF* gene, the 460T/C, 1154G/A, and 634G/C, between female breast cancer patients and healthy women as controls.

Methods: In this hospital-based case-control study, we recruited 214 pathologically proven female breast cancer cases and 161 healthy women subjects. The candidate *VEGF* gene polymorphisms were -634G/C (rs2010963), -460T/C (rs833061) and -1154G/A (rs1570360). Frequency of genotypes was determined by TaqMan real-time PCR allelic discrimination assay. Univariable and multivariable logistic regressions model were used to assess the odds ratios and corresponding confidence intervals. Relative excess risk due to interaction (RERI) was used to check potential additive interactions.

Results: The frequency of *VEGF* -1154G/A genotype in the case and control groups was 39.1 and 37.8%, respectively (P-value = 0.94). The frequency of combined -1154 AA/GA variant genotypes compared with the *VEGF* -1154 GG genotype in case and control groups was 48.8 and 49.4%, respectively (P-value = 0.94). Similarly, for the other two *VEGF* SNPs, -460T/C and -1154G/A, no significant differences were observed in genotype distributions between patients and controls. Moreover, we did not find interaction between *VEGF* SNPs and age for occurrence of breast cancer: RERI was 0.78 (95% CI: 0.47-2.03), attributable proportion (AP) due to interaction was 0.37 (95% CI: -0.13-0.88); and synergy index was 3.58 (95% CI: 0.12-106).

Conclusion: The results suggest that none of the studied polymorphisms in the *VEGF* gene were associated with occurrence of breast cancer in a sample of female Iranian adults. Larger studies are warranted to confirm the results and evaluate gene-environment interactions.

Keywords: Breast cancer, Single nucleotide polymorphisms, Vascular endothelial growth factor, Association study

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INTRODUCTION:

Breast cancer is one of the most frequently diagnosed cancers worldwide. Although it is considered a cancer of the Western lifestyle, nowadays more than half of new breast cancer cases (52%) and 62% of breast cancer deaths are reported from developing countries[1-3]. The molecular mechanism that induces or develops breast cancer remains to be fully understood. It has been demonstrated that genetic factors have significant effects on the development of breast cancer[4-6]. In addition, other risk factors such as environmental, occupational and even infectious agents have been suggested to influence this disease[2].

Angiogenesis plays a central role in both local tumor growth and distant metastasis[7,8]. Newly formed blood vessels supply the tumor with nutrients and oxygen, dispose of the metabolic waste products of tumor cells, and provide potential routes for tumor dissemination[9]. Tumors promote angiogenesis by secreting or activating angiogenic factors such as VEGF[10], therefore VEGF acts as a survival signal for the tumor cells. It is suggested that VEGF serves as an important prognostic biomarker in different tumors including breast cancer[11]. The *VEGF* gene is located on chromosome 6p21.3 and comprises a 14-kb coding region with eight exons[12]. Several SNPs have been identified in the *VEGF* gene[13-15]. Polymorphisms in the *VEGF* gene could be associated with altered expression and secretion of this factor or may correlate with the VEGF protein expression in cancer cells[16-18]. It is also reported that SNPs in the 5'- and 3'-untranslated region (UTR) of the *VEGF* gene could affect protein translation efficiently and tumor tissue expression of VEGF-A; thus, it may be associated with cancer risk development[19]. There is also evidence showing association of VEGF polymorphisms and the risk of breast cancer in different populations[20-22].

We investigated the association between three SNPs of the *VEGF* gene: –460T/C (rs833061), –1154G/A (rs1570360) localized in the promoter region, and –634G/C (rs2010963) in the 5'-UTR region and breast cancer risk in Iranian women.

METHODS:

Study population

A total of 375 subjects, consisting of 214 histopathologically confirmed female breast cancer patients and 161 healthy women as controls were recruited consecutively from July 2011 to February 2014 at the Cancer Institute of Iran, Tehran, Iran. The diagnosis of breast cancer was based on pathological examination of the tissue or biopsies of the tumor. The control group consisted of healthy women who were visiting non-cancer patients at Imam Khomeini hospital. The controls were selected from the same region to represent the target population (Iranian women). The study was approved by the Ethics Committee of Tehran University of Medical Sciences (code No. 12853) and written informed consent was obtained from all subjects for participation in the study.

DNA extraction and genotyping analysis

Genomic DNA was extracted from peripheral blood leukocytes using proteinase K phenol-chloroform extraction procedure[23]. The extracted DNA was stored at −20 °C until analyzed. DNA purity and concentrations were determined by spectrophotometric measurement of absorbance at 260 and 280 nm by UV spectrophotometer.

The three selected *VEGF* SNPs [–460T/C (rs833061), –1154G/A (rs1570360), –634G/C (rs2010963)] were genotyped using a TaqMan

real-time PCR allelic discrimination assay. An ABI Prism 7300 Sequence Detection System (Applied Biosystems, Foster City, USA) was used in accordance with the instructions provided by the manufacturer. The allelic call was performed by the analysis of allelic discrimination plots using, ABI SDS V 1.4 software.

Statistical analysis

Chi-square test was used to evaluate deviations from Hardy-Weinberg Equilibrium. To assess the difference in genotype and allele frequencies between the cases and controls, odds ratio and logistic regressions (univariable and multivariable) were used. Interaction was evaluated using interaction term (multiplicative interaction) and RERI was assessed to check for additive interaction. RERI can be interpreted as the excess risk due to interaction relative to the risk without exposure[24]. P-value ≤ 0.05 was considered significant. All analyses were performed using STATA12.

RESULTS:

A total of 214 breast cancer patients and 161 healthy individuals entered in the final analysis. All the research subjects were female and predominantly middle-aged at the time of diagnosis (mean age: 48.38 ± 10.65 years in the patients and 44.54 ±10.65 years in the control group) **(Table 1)**.

The distribution of genotypes for the three polymorphisms studied here conformed to the Hardy-Weinberg Equilibrium for both patients and healthy controls (P>0.05). The genotype frequencies for the *VEGF* –460T/C, –1154G/A and –634G/C SNPs are shown in **Table 2**. The number of samples analyzed for each polymorphism was not exactly equal, because the Taqman assay did not work for a few samples.

The frequency of *VEGF* -1154A/G genotype in case and control groups was 39.1 and 37.8%, respectively (P-value = 0.94). The frequency of combined -1154 AA/GA variant genotypes compared with the *VEGF* -1154 GG genotype in case and control groups was 48.8 vs. 49.4% (P-value =0.94). The frequency of *VEGF* -460T/C genotype in case and control groups was 15.2 and 20.6%, respectively (P-value =0.24). The frequency of *VEGF* -634C/G genotype in case and control groups was 42.3 and 47.2%, respectively (P-value =0.44). There was not any significant difference in genotype distributions of the studied *VEGF* polymorphisms between patients and controls. No significant difference was observed for the genotype distributions of all the studied polymorphisms between women aged less than 50 years and those above that. Furthermore, no significant association was found between the SNPs and breast cancer in the age categories **(Table 3).**

Additive interaction between age and the -460T/C polymorphism was also tested, but no statistical significance interaction was observed; RERI was 0.78 (95% CI: 0.47, 2.03), attributable proportion (AP) due to interaction was 0.37 (95% CI: -0.13, 0.88) and synergy index was 3.58 (95% CI: 0.12, 106).

The results did not change significantly when we adjusted for breast cancer risk factors including marital status, age at menarche, age at menopause, family history of breast cancer, educational level, body mass index (BMI) and estrogen or progesterone receptor status.

DISCUSSION:

The association of *VEGF* gene polymorphisms with disease risk has attracted much attention because VEGF is a major mediator of angiogenesis

Table 1. Demographic characteristics of study subjects at diagnosis		
Variables	**Patients (214)**	**Controls (161)**
Age	50.61 ± 11.46	46.85 ±10.70
First pregnancy age	20.38±5.84	20 ±4.5
BMI*		
Underweight	0.96%	1.86%
Normal	29.95%	21.11%
Overweight	41.06%	39.13%
Obese	28.01%	37.88%
Marital status		
Single	178 (83.17%)	18 (11.18%)
Married	16 (7.47%)	128 (79.50%)
Divorced	6 (2.8%)	15 (9.31%)
Widow	14 (6.54%)	0 (0%)
Education		
Illiterate	45 (21.13%)	29 (18.13%)
Primary	68 (31.92%)	104 (65%)
Secondary	28 (13.15%)	23 (14.38%)
High school	46 (21.60)	4 (2.5%)
Academic	26 (12.21%)	0 (0%)
Family History of Breast Cancer		
Yes	41 (20.60%)	11 (6.88%)
No	156 (78.39%)	149 (93.13%)
Missing	4 (1.01%)	0 (0%)
Age at Menarche		
<12 years	30 (14.29%)	12(7.59%)
12-15 years	134(63.81%)	132(83.54%)
>15 years	18(8.57%)	14(8.86%)
Missing	28 (13.33%)	0(0%)
*BMI, body mass index.		

Table 2. Genotype frequencies of *VEGF* gene, the 460T/C, 1154G/A, and 634G/C polymorphisms among women with breast cancer and healthy female individuals.

Polymorphism/ genotype	Cases (%)	Controls (%)	OR*	95%CI**	P
1154G/A					
G/G	63 (51.2)	39 (50.6)	Reference§		
A/A	12 (9.7)	9 (11.6)	0.825	0.318, 2.138	0.693
G/A	48 (39.1)	29 (37.8)	1.024	0.556, 1.885	0.938
1154G/A combined variants					
G/G	63 (51.2)	39 (50.6)	Reference§		
A/A & G/A	60 (48.8)	38 (49.4)	0.977	0.552, 1.727	0.937
460T/C					
T/C	92 (48.5)	59 (45.1)	Reference§		
C/C	29 (15.2)	27 (20.6)	0.983	0.597, 1.617	0.237
T/T	69 (36.3)	45 (34.3)	0.688	0.371, 1.277	0.947
460T/C combined variants					
C/C	27 (20.6)	29 (15.2)	0.693	0.388, 1.238	0.216
T/T & T/C	104 (79.4)	161 (84.8)	Reference§	0.807, 2.572	
634G/C					
G/C	86 (42.3)	67 (47.2)	0.827	0.511, 1.38	0.440
C/C	42 (20.5)	26 (18.3)	1.041	0.567, 1.910	0.896
G/G	76 (37.2)	49 (34.5)	Reference§		
634G/C combined variants					
C/C	42 (20.5)	26 (18.3)	1.156	0.671, 1.993	0.600
G/G & G/C	162 (79.5)	116 (81.7)	Reference§		

*OR, odds ratio;**CI, confidence interval.

and plays a significant role in development of solid tumors[25]. Several studies have evaluated the association of *VEGF* polymorphisms with breast cancer risk, but the results have been inconsistent[26-28].

We investigated the association between three *VEGF* gene polymorphisms (the 634G/C, 460T/C, and 1154G/A) with breast cancer risk in a group of female breast cancer patients and healthy controls. We found almost similar frequencies of the studied *VEGF* SNPs genotypes in our patients and controls. This finding is consistent with some previous studies[26, 29, 30]. In a case-control study of 1,093 Chinese women with breast cancer and 1,184 age-matched controls, Kataoka et.al. did not observe any significant difference between patients and controls in the distribution of *VEGF* -460T/C polymorphism which is in line with our findings. They suggested that the *VEGF* 936 C/T polymorphism might be a suscepti-

Table 3. Genotype frequencies of *VEGF* gene, the 460T/C, -634G/C and 1154G/A polymorphisms among women aged <50 and ≥50 years with breast cancer and healthy female controls.

SNP	Age<50				Age≥50			
	Case (%)	Controls (%)	OR* (95%CI**)	P	Cases (%)	Controls (%)	OR* (95%CI**)	P
-460T/C (rs833061)								
TT	25 (29.76)	27 (34.62)	0.65	0.238	44 (41.51)	18 (33.96)	1.42	0.333
TC	(0.30-1.41)	0.238	44 (41.51)		48 (45.28)	28 (52.83)	(0.65-3.13)	
CC	44 (52.38)	31 (39.74)	Reference	0.121	14 (13.21)	7 (13.21)	Reference	0.767
-634G/C(rs2010963)								
GG	40 (44.44)	33 (39.29)	Reference		(0.38-3.83)	16 (27.59)	Reference	
CG	32 (35.56)	36 (42.86)	0.73	0.358	1.16	31 (53.45)	0.77	0.495
CC	(0.35-1.49)	0.767	0.99 (0.40-2.46)	0.981		(0.34-1.71)		0.948
-1154G/A (rs1570360)								
GG	22 (44)	22 (47.83)	Reference		41 (56.16)	17 (54.84)	Reference	
GA	23 (46)	17 (36.96)	1.35	0.491	25 (34.25)	12 (38.71)	0.86	0.747
AA	(0.52-3.50)	45 (34.3)	0.688	0.608	(0.32-2.34)			0.660

*OR, odds ratio;**CI, confidence interval.

bility factor for breast cancer among Chinese women[26]. In another large case-control study involving 571 familial breast cancer cases, 974 unselected breast cancer cases together with ethnically and geographically selected controls, the associations between four *VEGF* SNPs (the -1154G/A, -634G/C, -2578C/A, and 936C/T) and breast cancer susceptibility were investigated in Swedish breast cancer patients. The findings suggested that VEGF is more likely to alter the aggressiveness of the tumor rather than susceptibility to cancer[29]. In another study performed in India, no association was observed between individual polymorphisms of *VEGF* -1154G/A or -634G/C SNP and breast cancer risk among 200 cases and 200 controls[30].

In the present study, no association was found between *VEGF* -634G/C polymorphism and occurrence of breast cancer. In contrast to our study, a few studies reported a positive significant association of -634G/C genotype or VEGF production with different cancer types[19, 31, 32]. Maltese et al reported the association of -634G/C genotype with reduced risk of colorectal cancer in an Italian study population consisting of 302 cases and 115 controls[31]. A hospital-based case-control study from Brazil on 235 sporadic female breast cancer patients and 235 controls investigated the influence of the *VEGF* 936CT and 634GC polymorphisms on the incidence and clinicopathologic characteristics of sporadic breast cancer. Their data indicated that the *VEGF*

variant 634CC and wild 936CC genotypes constitute inherited elements of sporadic breast cancer and sporadic breast cancer aggressiveness in Brazil[19]. Watson et al. demonstrated that the genotype for the -634G/C polymorphism in the *VEGF* gene is significantly correlated with VEGF production from stimulated peripheral blood mononuclear cells[32]. They reported that a G allele at position -634 affects transcriptional activity and increases VEGF production in peripheral blood mononuclear cells in response to lipopolysaccharide[32]. -634G/C is located in the 5'-untranslated regions of *VEGF* gene; considering the existence of different transcriptional binding factors in the 5'-untranslated region of VEGF, the regulation of gene transcription would be complicated[33]. In addition, it was reported that different SNPs in the 5'-untranslated region of the *VEGF* gene affect inter-individual variation in expression of VEGF and might influence the pathogenesis of tumors[34].

Our results showed no differences in the frequencies of -1154G/A SNP between breast cancer and control group. The -1154G/A and -460T/C SNPs are located in the promoter region of *VEGF* gene. A number of studies have indicated that the presence of these two SNPs in the promoter region of the *VEGF gene* is related to VEGF protein production [18, 32, 35]. Some reports indicated that high levels of VEGF expression and increased micro-vessel density in tumors are associated with advanced stage disease and worse prognosis for various types of tumors[36, 37]. Furthermore, it has been shown that different SNPs in the *VEGF* gene do not affect diseases such as type 1 diabetic retinopathy and cardiomyopathy, as in these diseases, angiogenesis does not play an important role in pathogenesis[38].

Few epidemiological studies have addressed the gene-environment interaction of breast cancer, as most of them have assessed the interaction of smoking and some genes. Here, the interaction of age (more and less than 50 years old) and -460T/C SNP was assessed and no additive or multiplicative interaction was observed. However, this study was small, and the results of interaction have not been confirmed or refuted. The most recent investigation from the Breast Cancer Association Consortium (BCAC) worked on few SNPs in 58573 subjects including 26968 cases and 31605 controls. The authors tested the interaction of 8q23-rs13267382 and age of at the first full-term pregnancy and indicated significant interaction, particularly at the ages of 20 to 24 years old and over 30 years ($P_{int}=2.6\times10^{-4}$)[39]. A large population-based case-control study conducted on Chinese women in Shanghai (1,093 cases and 1,184 age-matched controls) indicated no statistically significant association or apparent interaction for -460T/C in relation to breast cancer risk[26]. The main limitation of this study was the sample size and thus, the observed results could be due to type II error.

In conclusion, we failed to find any significant association between polymorphisms in the *VEGF* gene and breast cancer in Iran. Larger studies are needed to confirm the results of this study and to evaluate the gene-environment interaction.

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REFERENCES:

1. DeSantis CE, Bray F, Ferlay J, Lortet-Tieulent J, Anderson BO, Jemal A. International variation in female breast cancer incidence and mortality rates. Cancer Epidemiology and Prevention Biomarkers. 2015;24(10):1495-506.

2. Kamangar F, Dores GM, Anderson WF. Patterns of cancer incidence, mortality, and prevalence across five continents: defining priorities to reduce cancer disparities in different ge-

ographic regions of the world. Journal of clinical oncology. 2006;24(14):2137-50.
3. Porter P. "Westernizing" women's risks? Breast cancer in lower-income countries. New England Journal of Medicine. 2008;358(3):213-6.
4. Ambrosone CB, Hong C-C, Goodwin PJ. Host factors and risk of breast cancer recurrence: genetic, epigenetic and biologic factors and breast cancer outcomes. Improving Outcomes for Breast Cancer Survivors: Springer; 2015. p. 143-53.
5. Mahmoodi M, Nahvi H, Mahmoudi M, Kasaian A, Mohagheghi M-A, Divsalar K, et al. HLA-DRB1,-DQA1 and-DQB1 allele and haplotype frequencies in female patients with early onset breast cancer. Pathology & Oncology Research. 2012;18(1):49-55.
6. Rudolph A, Chang-Claude J, Schmidt MK. Gene–environment interaction and risk of breast cancer. British journal of cancer. 2016;114(2):125-33.
7. Bertolini F, Shaked Y, Mancuso P, Kerbel R. Tumor angiogenesis. N Engl J Med. 2008;359:763-4.
8. Hu Z, Fan C, Livasy C, He X, Oh DS, Ewend MG, et al. A compact VEGF signature associated with distant metastases and poor outcomes. BMC medicine. 2009;7(1):9.
9. Carmeliet P, Jain RK. Angiogenesis in cancer and other diseases. nature. 2000;407(6801):249-57.
10. Roskoski R. Vascular endothelial growth factor (VEGF) signaling in tumor progression. Critical reviews in oncology/hematology. 2007;62(3):179-213.
11. Carpini JD, Karam AK, Montgomery L. Vascular endothelial growth factor and its relationship to the prognosis and treatment of breast, ovarian, and cervical cancer. Angiogenesis. 2010;13(1):43-58.
12. Vincenti V, Cassano C, Rocchi M, Persico MG. Assignment of the vascular endothelial growth factor gene to human chromosome 6p21. 3. Circulation. 1996;93(8):1493-5.
13. Zhao ZZ, Nyholt DR, Thomas S, Treloar SA, Montgomery GW. Polymorphisms in the vascular endothelial growth factor gene and the risk of familial endometriosis. Molecular human reproduction. 2008;14(9):531-8.
14. Li Y, Wang L, Li X, Li S, Wang J, Wu Z, et al. Vascular endothelial growth factor gene polymorphisms contribute to the risk of endometriosis: an updated systematic review and meta-analysis of 14 case-control studies. Genet Mol Res. 2013;12(2):1035-44.
15. Metzger CS, Koutsimpelas D, Brieger J. Transcriptional regulation of the VEGF gene in dependence of individual genomic variations. Cytokine. 2015;76(2):519-26.
16. Al-Habboubi HH, Sater MS, Almawi AW, Al-Khateeb GM, Almawi WY. Contribution of VEGF polymorphisms to variation in VEGF serum levels in a healthy population. European cytokine network. 2011;22(3):154-8.
17. Mohammadi M, Ollier WE, Hutchinson IV. A functional association study of VEGF gene promoter polymorphisms with VEGF expression by stimulated pbm cells. Human Immunology. 2003;64(10):S125.
18. Koukourakis MI, Papazoglou D, Giatromanolaki A, Bougioukas G, Maltezos E, Siviridis E. VEGF gene sequence variation defines VEGF gene expression status and angiogenic activity in non-small cell lung cancer. Lung cancer. 2004;46(3):293-8.
19. Oliveira C, Lourenço GJ, Silva PM, Cardoso-Filho C, Favarelli MH, Gonçales NS, et al. Polymorphisms in the 5′-and 3′-untranslated region of the VEGF gene and sporadic breast cancer risk and clinicopathologic characteristics. Tumor Biology. 2011;32(2):295-300.
20. Krippl P, Langsenlehner U, Renner W, Yazdani-Biuki B, Wolf G, Wascher TC, et al. A common 936 C/T gene polymorphism of vascular endothelial growth factor is associated with decreased breast cancer risk. International journal of cancer. 2003;106(4):468-71.
21. Langsenlehner U, Wolf G, Langsenlehner T, Gerger A, Hofmann G, Clar H, et al. Genetic polymorphisms in the vascular endothelial growth factor gene and breast cancer risk. The Austrian "tumor of breast tissue: incidence, genetics, and environmental risk factors" study. Breast cancer research and treatment. 2008;109(2):297-304.
22. Beeghly-Fadiel A, Shu X-O, Lu W, Long J-R, Cai Q, Xiang Y-B, et al. Genetic variation in VEGF family genes and breast cancer risk: a report from the Shanghai Breast Cancer Genetics Study. Cancer Epidemiology and Prevention Biomarkers. 2010:cebp. 0793.2010.
23. Roe BA, Crabtree J, Khan A. Methods for DNA isolation. Part III. Protocols for recombinant DNA isolation, cloning, and sequencing [Internet edition] Norman, OK: University of Oklahoma. 1995:2488-98.
24. Skrondal A. Interaction as departure from additivity in case-control studies: a cautionary note. American Journal of Epidemiology. 2003;158(3):251-8.
25. Hicklin DJ, Ellis LM. Role of the vascular endothelial growth factor pathway in tumor growth and angiogenesis. Journal of clinical oncology. 2005;23(5):1011-27.
26. Kataoka N, Cai Q, Wen W, Shu X-O, Jin F, Gao Y-T, et al. Population-based case-control study of VEGF gene polymorphisms and breast cancer risk among Chinese women. Cancer Epidemiology and Prevention Biomarkers. 2006;15(6):1148-52.
27. Rodrigues P, Furriol J, Tormo E, Ballester S, Lluch A, Eroles P. The single-nucleotide polymorphisms+ 936 C/T VEGF and− 710 C/T VEGFR1 are associated with breast cancer protection in a Spanish population. Breast cancer research and treatment. 2012;133(2):769-78.

28. Kapahi R, Guleria K, Sambyal V, Manjari M, Sudan M, Uppal MS, et al. Vascular endothelial growth factor (VEGF) gene polymorphisms and breast cancer risk in Punjabi population from North West India. Tumor Biology. 2014;35(11):11171.
29. Jin Q, Hemminki K, Enquist K, Lenner P, Grzybowska E, Klaes R, et al. Vascular endothelial growth factor polymorphisms in relation to breast cancer development and prognosis. Clinical cancer research. 2005;11(10):3647-53.
30. James R, Ramesh G, Krishnamoorthy L, Bhagat R, Chadaga S, Deshmane V, et al. Prevalence of+ 405G> C,− 1154G> A vascular endothelial growth factor polymorphism in breast cancer. Indian Journal of Clinical Biochemistry. 2014;29(1):21.
31. Maltese P, Canestrari E, Ruzzo A, Graziano F, Falcone A, Loupakis F, et al. VEGF gene polymorphisms and susceptibility to colorectal cancer disease in Italian population. International journal of colorectal disease. 2009;24(2):165-70.
32. Watson CJ, Webb NJ, Bottomley MJ, Brenchley PE. Identification of polymorphisms within the vascular endothelial growth factor (VEGF) gene: correlation with variation in VEGF protein production. Cytokine. 2000;12(8):1232-5.
33. Akiri G, Nahari D, Finkelstein Y, Le S-Y, Elroy-Stein O, Levi B-Z. Regulation of vascular endothelial growth factor (VEGF) expression is mediated by internal initiation of translation and alternative initiation of transcription. Oncogene. 1998;17(2):227-36.
34. Bhanoori M, Arvind Babu K, Pavankumar Reddy N, Lakshmi Rao K, Zondervan K, Deenadayal M, et al. The vascular endothelial growth factor (VEGF)+ 405G> C 5′-untranslated region polymorphism and increased risk of endometriosis in South Indian women: a case control study. Human Reproduction. 2005;20(7):1844-9.
35. Stevens A, Soden J, Brenchley PE, Ralph S, Ray DW. Haplotype analysis of the polymorphic human vascular endothelial growth factor gene promoter. Cancer research. 2003;63(4):812-6.
36. Perrone G, Vincenzi B, Santini D, Verzı A, Tonini G, Vetrani A, et al. Correlation of p53 and bcl-2 expression with vascular endothelial growth factor (VEGF), microvessel density (MVD) and clinico-pathological features in colon cancer. Cancer letters. 2004;208(2):227-34.
37. Iordache S, Saftoiu A, Georgescu C-V, Ramboiu S, Gheonea D-I, Filip M, et al. Vascular endothelial growth factor expression and microvessel density—two useful tools for the assessment of prognosis and survival in gastric cancer patients. J Gastrointestin Liver Dis. 2010;19(2):135-9.
38. Rogers MS, D'amato RJ. The effect of genetic diversity on angiogenesis. Experimental cell research. 2006;312(5):561-74.
39. Barrdahl M, Rudolph A, Hopper JL, Southey MC, Broeks A, Fasching PA, et al. Gene-environment interactions involving functional variants: Results from the Breast Cancer Association Consortium. International Journal of Cancer. 2017.