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ORIGINAL PAPER

Genetic polymorphisms associated with the prevalence of retinal vein occlusion in a Greek population

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Abstract

Purpose To investigate possible associations of single-nucleotide polymorphisms (SNPs) from five genes with branch retinal vein occlusion (BRVO) and central retinal vein occlusion (CRVO).

Methods A total of 69 patients with retinal vein occlusion-RVO (24 with BRVO and 45 with CRVO), and 82 controls, were enrolled in this study. All subjects were screened for hypertension, diabetes mellitus, hyperlipidemia, glaucoma, anticoagulant medication, smoking status and history of stroke. The genotyping of *AGTR1*-A1166C, *adiponectin* + 276 G/T, *MMP2*-1306C/T, *Gpla/lla*-C807T/G873A and *VKORC1*-G1639A polymorphisms was performed using restriction fragment length polymorphism or allele-specific polymerase chain reaction.

Results The percentage of the *AGTR1*-A1166C C allele carriers and *Gpla/lla*-C807T/G873A T/A carriers was significantly higher in the CRVO patients than in the controls ($P = 0.00001$ and $P = 0.0004$, respectively). At the multiple logistic regression analysis, the *AGTR1*-A1166C C allele carrier status and the *Gpla/lla*-C807T/G873A T/A allele carrier status were found to be associated with an increased risk of CRVO. Moreover, *adiponectin* + 276 G/T T allele carriers had a significantly increased risk of RVO in subjects $\geq$ 75 years old. There was no significant difference between the BRVO patients and controls concerning the genotype or the allele frequency distributions of these SNPs. The genotype distributions or allelic frequencies of the other evaluated polymorphisms did not significantly differ between the patients with RVO and the control subjects.

Conclusions *AGTR1* A1166C and *Gpla/lla* C807T/G873A polymorphisms are likely to be risk factors for CRVO. *Adiponectin* + 276 G/T SNP is likely to predispose to RVO in older subjects.

Keywords AGTR1 · Adiponectin · GpIa/IIa · MMP2 · RVO · VKORC1

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Introduction

Retinal vein occlusion (RVO) is the second most common type of retinal vascular disorder, after diabetic retinopathy. RVO is also one of the most common causes of sudden unilateral vision loss because of retinal ischemia and macular edema [1]. It has been shown that its prevalence varies from 0.7 to 1.6% [2]. It is usually unilateral and affects principally patients older than 50 years old (y/o). Depending on the location of the obstruction, RVO can be divided into branch retinal vein occlusion (BRVO) and central retinal vein occlusion (CRVO). The obstruction in BRVO is located in one of the branches of retinal vein, typically at arteriovenous crossings, whereas in CRVO, it is located in the retinal vein in close proximity to the optic nerve at or behind the lamina cribrosa, and it affects most of the retina [3, 4].

The etiology of RVO is multifactorial, and it includes hemostasis, endothelial damage and thrombophilia [5]. Many systemic and ophthalmic disorders that cause hypercoagulability and increase the probability of a thrombotic event may play a role. Risk factors for RVO include diabetes, hypertension, hyperlipidemia, aging, cardiovascular disease, open-angle glaucoma, coagulation disorders, thromboembolic events, diuretic drugs, contraceptives and trauma [2]. Although the precise pathogenetic mechanism remains unclear, it may be attributable to compression of the vein at the arteriovenous crossings, degenerative changes of the venous wall, abnormal hematological factors [5] or genetic factors (gene polymorphisms, polygenic disorders, interaction between different gene mutations) [6].

In RVO studies, the probable pathogenic role of the common causes of thrombophilia is controversial [6]. Gene polymorphisms that increase the risk of having a thrombotic event, by affecting proteins involved in the coagulation cascade, are believable candidates as risks factors for RVO [7]. Moreover, given that atherosclerosis seems to be an important contributor to the prevalence of RVO [8], polymorphisms of genes related to atherosclerosis, hypertension and arterial stiffness, such as *AGTR1* (*Angiotensin II type I receptor*) and *adiponectin*, might be predisposing risk factors for RVO as well [9]. Previous studies, however, either obtained conflicting results or they were limited regarding the role of coagulation and arteriosclerosis-related gene variants of *GpIa/IIa (Platelet glycoprotein Ia/IIa),VKORC1* (*Vitamin K epoxide reductase complex subunit 1*), *MMP2* (*Matrix metalloproteinase-2), AGTR1* and *adiponectin* in RVO [9–15]. Moreover, no polymorphism of these genes has been investigated in the Greek population concerning a potential association with RVO.

In light of the above, the aim of the current study was to estimate the existing genetic predisposition in RVO and particularly to investigate possible associations of single-nucleotide polymorphisms (SNPs) from five genes relevant to coagulation and arteriosclerosis including *AGTR1* A1166C, *adiponectin* + 276G/T, *VKORC1* G-1639A, *GpIa/IIa* C807T/G873A and *MMP2*-1306C/T polymorphisms, with CRVO and BRVO in a Greek population.

Materials and methods

Study population

A total of 69 patients with RVO, who were presented to the Eye Clinic of the University General Hospital of Ioannina, and 82 age-matched controls consisted the study population. The study was approved by the hospital ethics committee, and written informed consent was obtained from each patient. The study was accomplished in accordance with the Declaration of Helsinki. There were 45 CRVO patients and 24 BRVO patients. Each subject underwent baseline eye examination, using indirect ophthalmoscopy examination and fundus examination by a slit lamp after pupil dilation. Color fundus photography and fundus fluorescein angiography were taken using a fundus camera (Topcon).

The presence of CRVO was determined based on widespread superficial or deep retinal hemorrhages, with or without optic disk hyperemia or edema, venous dilation and tortuosity, retinal and macular edema or occluded veins. BRVO was described by retinal hemorrhages within the retinal sector corresponding to the sector of the occluded venule and by scattered superficial and deep retinal hemorrhages venous dilation and tortuosity, intraretinal microvascular abnormalities and occluded and sheathed retinal venules. Ocular trauma, uveitis, cancer and abnormal liver or renal function were exclusive criteria for this study. Subjects were not eligible as controls, if there was a history of retinal vascular occlusion, deep vein

thrombosis, pulmonary embolism, myocardial infarction and stroke. All subjects were screened for age, gender, hypertension, hyperlipidemia, history of stroke, glaucoma, diabetes mellitus (DM), anticoagulants medication and current smoking status. All participants were evaluated for obstructive sleep apnea (OSA) indirectly by answering a simple questionnaire (Epworth Sleepiness Scale-ESS), which measures subjective daytime sleepiness [16]. Subjects were asked to rate how likely they were to fall asleep in different situations. Every question was answered on a scale of 0 to 3. ESS values range from zero (unlikely to fall asleep in any situation) to 24 (high chance of falling sleep in all eight situations). Based on the ESS final score, participants were divided into the ones with score < 11 (low risk of sleepiness) and the ones with score ≥ 11 (high risk of sleepiness) [17].

Genotyping

Blood specimens were taken into EDTA-containing tubes and stored at − 20 °C until further analysis. DNA was extracted from peripheral blood using QIAamp DNA Blood mini kit (Qiagen) as per manufacturer's instructions.

The genotype *AGTR1* A1166C (rs5186) was determined by performing PCR-RFLP using sequence-specific primers as previously described [18]. The primers used were F: 5′-GCACCATGTTTT-GAGGTTG-3′ and R: 5′-CGACTACTGCTTAG-CATA-3′. The resulting PCR product was subject to restriction digestion using 0.5 U of *Dde*I enzyme (NEB) at 37 °C overnight. The digested product was run on 2% agarose gel and visualized using ethidium bromide. The presence of the polymorphism resulted in 412 bp, 118 bp and 10 bp bands; however, its absence was indicated by the presence of only a 530 bp band.

The adiponectin + 276 G/T (rs1501299) polymorphism was also determined by performing PCR-RFLP. The primers used were F: 5′-GTCTAGGCCTTAGT-TAATAATGAAGG-3′ and R: 5′-GTGAGAAAGGA-GATCCAGGTAA-3′ as previously described [19]. The resulting PCR product was subject to restriction digestion using 0.5 U of *Stu*I enzyme (NEB) at 37 °C overnight. The digested product was run on 2% agarose gel and visualized using ethidium bromide. The presence of the T allele resulted in a 106 bp band; however, the presence of the G allele was indicated by the presence of 80 bp and 26 bp bands.

The genotyping for *MMP2*-1306 C/T (rs243865) was performed via PCR-RFLP-based methods as it was described previously [20]. The sequences of the primers were as it follows: F: 5′-CTTCCTAGG CTGGTCCTTACTGA-3′; R: 5′-CTGAGACCT GAAGAG CTAAAGAGCT-3′. The restriction reaction of each PCR product was carried out with 5 U *Xsp*I (Fermentas). The digestion resulted in two fragments with a length of 188 bp and 5 bp for allele C and in three fragments with a length of 162 bp, 26 bp and 5 bp for allele T.

The GpIa/IIa PCR was performed according to the method of Dodson et al [14]. Briefly, a two-stage multiplex PCR was performed, the first using primer sequences intron G, exon 8, 807C and 807T, and the second using intron G, exon 8, 873A and 873G. Intron GF: 5′-GATTTAACTTCCCAGCTGCCTTC-3′, Exon 8 f: 5′-CTCAGTATATTGTCATGGTTGCATTG-3′, 807 CF: 5′-GTGGGGACCTCACAAACACATGC-3′, 807 TF: 5′-ATGGTGGGGACCTCAACAAACACA-TAT-3′., 873 GF: 5′-GGTGGGCGACGAAGTGC-TAGG-3′ and 873 AF: 5′-GGTGGGCGACGAAGT GCTAGA-3′. Electrophoresis was carried out on a 2% agarose gel and stained with ethidium bromide.

VKORC1 promoter G-1639A (rs9923231) polymorphism genotyping was performed using PCR-RFLP. The sequence of nucleotides in specific primers for *VKORC1* promoter G-1639A SNP was as follows: F: 5′-GCCAGCAGGAGAGGGAAATA-3′ and R: 5′-AGTTTGGACTACAGGTGCCT-3′. [21]. For restriction analysis, the amplification products were incubated at 37 °C for overnight with 5 U *Msp*I (*Hpa*II) (Thermo Fisher Scientific, USA). In the case of G at position-1639 of the ter amplified fragment, which consisted of 290, bps was cut into two fragments of 168 and 122 bps. G to A substitution resulted in the loss of the restriction site, and fragment of the promoter (290 bps) could not be cleaved.

Statistical analysis

Data were analyzed using SPSS 22.0 (SPSS, Inc). Continuous data were expressed as mean ± SD. Normality tests were carried out for age distribution, and the Mann–Whitney test was applied for median comparison between control subjects and patients with RVO. Categorical data were presented as counts and

percentages. The Chi-squared test or the Fisher exact test was also used to compare the distribution of *AGTR1* A1166C, *adiponectin* + 276 G/T, *MMP2*-1306C/T, *GpIa/IIa* C807T/G873A, *VKORCI* G-1639A polymorphisms between patients with BRVO or CRVO and control subjects. Odds ratios (OR) and 95% confidence intervals (CIs) were calculated with the corresponding Chi-squared or Fisher exact distribution test, whenever there was statistical significance. Significant probability values were also corrected for multiple testing (Bonferroni correction; Pc). The Hardy–Weinberg equilibrium regarding the distribution of the genotypes in the control group was evaluated using the Chi-squared test. In order to determine (CC and AC vs. AA) genotype of *AGTR1* A1166C, (CT/GA + TT/AA vs. CC/GG) genotype of *Gpla/lla* C807T/G873A, DM and anticoagulants medication association with CRVO, a multivariate logistic regression model was applied. It was also implemented to determine (CC and AC vs. AA) genotype of *AGTR1* A1166C, (CT/GA + TT/AA vs. CC/GG) genotype of *Gpla/lla* C807T/G873A, (GT and TT vs. GG) genotype of *adiponectin* + 276 G/T and anticoagulation medication association with RVO in subjects ≥ 75 y/o. The *P* values obtained were two-tailed and determined to be significant at $P < 0.05$.

Results

The clinical characteristics of the study subjects are presented in Table 1. No statistical difference was found between groups in age or sex. The frequencies of DM and anticoagulants medication were higher in the CRVO group than in the control group. There were no significant differences between the BRVO or CRVO patients and the control group regarding hypertension, hyperlipidemia, history of stroke, smoking status, or daytime sleepiness. There were statistically significant more glaucomatous patients among the BRVO patients compared to controls, while there was no statistically significant difference regarding glaucoma between CRVO and control groups.

The distribution of the genotype and allele frequency of *AGTR1* A1166C,*adiponectin* + 276 G/T, *MMP2*-1306C/T, *Gpla/lla* C807T/G873A, *VKORC1* G-1639A polymorphisms in the BRVO or CRVO patients and controls are presented in Table 2. The observed and expected frequencies of the polymorphism in control groups were within the Hardy–Weinberg equilibrium.

The frequencies of the AA, AC and CC genotypes of *AGTR1* A1166C in the CRVO patients were 40, 35.5 and 24.4%, whereas in the controls they were 73, 25.6 and 1%. The A and C allele frequencies were 57.8% and 42.2% in the CRVO group and 86% and 14% in the control group, respectively. The *AGTR1* A1166C C allele carriers had a significantly increased risk of CRVO compared to the A allele carriers ($P = 0.00001$, OR 4.48; 95% CI 2.44–8.23). The frequencies of the CC/GG, CT/GA and TT/AA genotypes of *Gp la/lla* C807T/G873A were 6.6, 84.4 and 8.8% in the CRVO patients, 16.6, 79.1 and 4.1% in the BRVO patients, whereas in the controls they were 41.4, 47.5 and 10.9%. The C/G and T/A allele frequencies were 48.9% and 51.1% in the CRVO group and 65.3% and 34.7% in the control group, respectively. The *Gp la/lla* C807T/G873A T/A allele

Table 1 Clinical characteristics of the patients with RVOs and control volunteers

	Control (82)	BRVO (24)	*	CRVO (45)	†
Age (year)	71.3±13.3	71.9±8.9	0.861	71.5±9.5	0.819
Sex:F/M	33/49	11/13	0.625	20/25	0.646
Hypertension	71(86.6)	19 (79.2)	0.352	44 (97.8)	0.055
Hyperlipidemia	68 (82.9)	18 (75)	0.365	43 (95.5)	0.057
Diabetes mellitus	2 (2.4)	4 (16.7)	0.007	11 (24.4)	0.00001
Stroke	1 (1.1)	0 (0)	1.0	4 (8.9)	0.053
Glaucoma	2 (2.2)	5 (20.8)	0.0046	5 (11.1)	0.053
Anticoagulant medication	1 (1.1)	7 (29.2)	0.0001	13 (28.9)	0.0001
Smoking	3 (3.4)	3 (12.5)	0.109	5 (11.1)	0.599
Sleepiness (ESS)	9.27±1.45	9.42±2.2	0.695	9.38±1.40	0.952
ESS ≥ 11	13 (15.85)	6 (25)	0.351	9 (20)	0.555

*Statistical significance comparing BRVO to control subjects

†Statistical significance comparing CRVO to control subjects

Data are expressed as mean ± SD or number of subjects (percentage)

Table 2 Genotypic and allelic distribution of the five gene polymorphisms between controls and RVO patients

	Control subjects (n = 82)	RVO patients (n = 69)	BRVO patients (n = 24)	CRVO patients (n = 45)
AGTR1 A1166C				
AA	60 (73)	33 (47.8)	15 (62.5)	18 (40)
AC	21 (25.6)	24 (34.8)	8 (33)	16 (35.5)
CC	1 (1.2)	12 (17.4)	1 (4)	11 (24.4)
P(Pc)		0.000282 (0.000846)	0.321	0.000011 (0.000033)
A	141 (86)	90 (65.2)	38 (79.2)	52 (57.8)
C	23 (14)	48 (34.8)	10 (28.2)	38 (42.2)
P		0.000023	0.2624	0.00001
OR (95% CI)		3.27 (1.86–5.74)		4.48 (2.44–8.23)
VKORC1 G-1639A				
AA	11 (13.4)	6 (8.7)	3 (12.5)	3 (6.6)
GA	38 (46.3)	33 (47.8)	12 (0.5)	21 (46.6)
GG	33 (40.2)	30 (43.5)	9 (37.5)	21 (46.6)
P		0.653	0.522	0.948
A	60 (36.6)	45 (32.6)	18 (37.5)	27 (30)
G	104 (63.4)	93 (67.4)	30 (62.5)	63 (70)
P		0.470	0.908	0.290
Adiponectin + 276 G/T				
GG	41 (50)	29 (42)	9 (37.5)	20 (44.4)
GT	32 (39)	33 (47.8)	14 (58.3)	19 (42.2)
TT	9 (10.9)	7 (10.2)	1 (4.1)	6 (13.3)
P		0.545	0.214	0.839
G	114 (69.5)	91 (65.9)	32 (66.7)	59 (65.6)
T	50 (30.5)	47 (34.1)	16 (33.3)	31 (34.4)
P		0.508	0.708	0.517
MMP2-1306C/T				
CC	40 (48.7)	33 (47.8)	10 (41)	23 (51.1)
CT	35 (42.6)	34 (49.3)	13 (54.1)	21 (46.6)
TT	7 (8.5)	2 (2.9)	1 (4.1)	1 (2.2)
P		0.307	0.629	0.444
C	115 (70.1)	100 (72.4)	33 (68.7)	67 (75.3)
T	49 (29.9)	38 (27.6)	15 (31.3)	23 (24.7)
P		0.654	0.855	0.465
Gpla/lla C807T/G873A				
CC/GG	34 (41.4)	7 (10.1)	4 (16.6)	3 (6.6)
CT/GA	39 (47.5)	57 (82.6)	19 (79.1)	38 (84.4)
TT/AA	9 (10.9)	5 (7.3)	1 (4.1)	4 (8.8)
P(Pc)		0.000023 (0.000069)	0.024 (0.072)	0.00001 (0.00003)
C/G	214 (65.3)	142 (51.4)	54 (58.1)	88 (48.9)
T/A	114 (34.7)	134 (48.6)	39 (41.9)	92 (51.1)
P		0.0006	0.2048	0.0004
OR (95% CI)		1.77 (1.28–2.46)		1.96 (1.35–2.84)

Data are expressed as number of subjects (percentage)

P values as compared with control subjects

Odds ratio values as compared to control subjects in allele distributions

OR odds ratio, *Pc* P after Bonferroni correction, *CI* confidence interval

carriers had a significantly increased risk of CRVO, when compared to the C/G allele carriers ($P = 0.0004$, OR 1.96; 95% CI 1.35–2.84). No significant difference was found between the *adiponectin* + 276 G/T, *MMP2*-1306C/T, *VKORC1* G-1639A genotype distributions, and allele frequencies in the BRVO, CRVO patients and the control group.

A multivariate logistic regression analysis was also performed using CRVO as the independent variable, and the results are presented in Table 3. After adjusting for DM and anticoagulants medication, *AGTR1* A1166C C allele carriers (AC + CC), compared to the AA homozygotes, had a significantly increased risk of CRVO ($B = 1.14$; $P = 0.016$; OR 3.13; 95% CI 1.24–7.89). In addition, *Gp la/lla* C807T/G873A T/A allele carriers (CT/GA + TT/AA) compared to CC/GG had a significantly increased risk of CRVO ($B = 2.47$; $P = 0.001$; OR 11.88; 95% CI 2.71–52.05).

In order to analyze further the association of age with the incidence of RVO (CRVO and BRVO), as well as with the five studied gene polymorphisms, we transformed continuous data to categorical by stratifying both control subjects and RVO patients according to their age. Specifically, because the median regarding age was 74 y/o (range 43–87) for RVO group and 75 y/o (range 36–89) for the control group, we chose to separate RVO and control subjects into two age subgroups (< 75, ≥ 75 y/o). In that way, each of the subgroups included similar number of subjects. The main clinical characteristics of the study subjects segregated by age are shown in Table 4. The frequency of DM and the frequency of anticoagulant medication were statistically significant higher in RVO patients compared to control subjects in the younger age group (< 75 y/o) and in the older age group (≥ 75 y/o), respectively.

The distribution of the genotype and allele frequency of *AGTR1* A1166C, *MMP2*-1306C/T, *Gpla/lla* C807T/G873A, *VKORC1* G-1639A polymorphisms in the RVO and control subjects segregated by age are shown in Table 5. Specifically, the frequency of *AGTR1* A1166C (AC + CC) and *Gp la/lla* C807T (CT/GA + TT/AA) genotypes were 52.8% and 88.9% in patients with RVO compared to controls (26.3% and 63.2%) in the younger subgroup ($P = 0.020$ and $P = 0.010$, respectively). Similarly, in the older subgroup the frequency of *AGTR1* A1166C (AC + CC) and *Gpla/lla* C807T (CT/GA + TT/AA) genotypes were 51.5% and 90.9% in patients with RVO compared to controls (27.3% and 54.5%) ($P = 0.030$ and $P = 0.001$, respectively). *Adiponectin* + 276 G/T polymorphism, however, was found to be associated with the incidence of RVO only in the older age group (≥ 75y/o). In particular, the frequency of *adiponectin* + 276 G/T (GT + TT) genotypes was 69.7% in RVO subjects compared to 43.2% in control subjects ($P = 0.021$). Moreover, T allele carriers had a significantly increased risk of RVO compared to the G allele carriers ($P = 0.0003$, OR 3.25; 95% CI 1.68–6.30) in the older subgroup. Analysis of the *MMP2*-1306C/T and *VKORC1* G-1639A polymorphisms revealed no association with the occurrence of RVO in both age subgroups. A multivariate logistic regression analysis (Table 6) was also conducted in the older subgroup, using RVO as the independent variable. After adjusting to anticoagulant medication, *Gpla/lla* C807T/G873A T/A allele carriers (AA/TT + GA/CT) compared to GG/CC had a significantly increased risk of RVO ($B = 2.65$; $P = 0.003$; OR 14.10; 95% CI 2.54–78.39). *Adiponectin* + 276 G/T T allele carriers (GT + TT) compared to GG had also a significantly increased risk of RVO ($B = 2.07$; $P = 0.004$; OR 7.96; 95% CI 1.94–32.66).

Table 3 Multivariate logistic regression analysis between CRVO patients and control subjects

Variable	*B*	*P*	Odds ratio	95% CI	
				Lower	Upper
AGTR1 A1166C (CC + AC vs. AA)	1.14	0.016	3.13	1.24	7.89
Gp la/lla C807T/G873A (CT/GA+TT/AA vs. CC/GG)	2.47	0.001	11.88	2.71	52.05
Diabetes mellitus	2.31	0.027	10.04	1.30	77.93
Anticoagulant medication	2.76	0.004	15.77	2.44	102.05

CI confidence interval

Table 4 Clinical characteristics of the controls and patients with RVO segregated by age

	< 75 y/o			≥ 75 y/o		
	Control subjects (n = 38)	RVO patients (n = 36)	*	Control subjects (n = 44)	RVO patients (n = 33)	†
Sex:F/M	13/25	14/22	0.676	20/24	16/17	0.792
Hypertension	27 (71)	30 (83.3)	0.209	44 (100)	33 (100)	1.0
Hyperlipidemia	27 (71)	30 (83.3)	0.209	42 (95.4)	31 (93.9)	1.0
Diabetes mellitus	0 (0)	9 (25)	0.001	2 (4.5)	6 (18.2)	0.068
Stroke	0 (0)	2 (5.6)	0.233	1 (2.3)	2 (6.1)	0.573
Glaucoma	1 (2.6)	2 (5.6)	0.610	1 (2.3)	3 (9.1)	0.308
Anticoagulant medication	1 (2.6)	6 (16.7)	0.053	1 (2.3)	14 (42.4)	0.0001
Smoking	4 (10.5)	3 (8.3)	1.0	5 (11.4)	5 (15.2)	0.737
Sleepiness (ESS)	8.95 ± 1.23	8.86 ± 1.46	0.73	9.55 ± 1.58	9.97 ± 1.63	0.44
ESS ≥ 11	4 (10.52)	6 (16,67)	0.44	9 (20.45)	9 (27)	0.48

*Statistical significance comparing RVO to control subjects (< 75 y/o)

†Statistical significance comparing RVO to control subjects (≥ 75 y/o)

Data are expressed as mean ± SD or number of subjects (percentage)

Multivariate logistic regression analysis regarding the younger subgroup, using RVO as the independent variable, revealed that *Gpla/lla* C807T/G873A T/A allele carriers, as well as *AGTR1* A1166C C allele carriers, are at increased risk of RVO (Table 1 in Online Resource 1).

Discussion

In the current study, we investigated whether *AGTR1* A1166C, *adiponectin* + 276 G/T, *MMP2*-1306C/T, *Gpla/lla* C807T/G873A and *VKORC1* G-1639A polymorphisms were risk factors for CRVO and BRVO in a Greek population.

We found that *AGTR1* A1166C and *Gpla/lla* C807T/G873A polymorphisms were associated with the incidence of CRVO. Specifically, *AGTR1* A1166C allele carriers (AC + CC), compared to the AA homozygotes and *Gpla/lla* C807T/G873A T/A carriers (CT/GA + TT/AA) compared to CC/GG homozygotes, had a significantly increased risk of CRVO. Moreover, *adiponectin* + 276 G/T polymorphism was found to be associated with the incidence of RVO, but only in the older subgroup (≥ 75 y/o). In particular, *adiponectin* + 276 G/T T allele carriers compared to G allele carriers had a significantly increased risk of RVO, if they were ≥ 75 y/o. Analysis of the *MMP2*-1306C/T and *VKORC1* G-1639A polymorphisms revealed no association with the occurrence of RVO.

Retinal veins and arteries share the same adventitial sheath within the lamina cribrosa. Therefore, arterial stiffness affects neighboring vein, and a combination of compression of the vein, hemodynamic changes, degenerative changes within the venous walls and hypercoagulability may be causative for RVO, according to Virchow's triad [22]. In this context, blood disorders that affect clotting and arteriosclerosis are considered the leading etiologic factors for the RVOs [5].

When a vascular injury takes place, platelets adhere to the basement membrane or other connective tissue components, which are located underneath endothelial cells, and these platelet–collagen interactions are believed to be the most thrombogenic constituents of the vessel walls [23]. Glycoprotein Ia/IIa is a platelet surface glycoprotein and a receptor for collagen type I and IV. *Gp la/lla* C807T polymorphism has been shown to increase the density of the receptor in the platelets and thereby enhance its interaction with collagen and the subsequent stabilization of the

Table 5 Genotypic distribution and allele frequency of five different genetic polymorphisms between RVO patients and control subjects segregated by age

	< 75 y/o				≥ 75 y/o			
	Control subjects (n = 38)	RVO patients (n = 36)	*	OR (95% CI)	Control subjects (n = 44)	RVO patients (n = 33)	†	OR (95% CI)
AGTR1 A1166C								
AC + CC: AA	10 (26.3): 28 (73.7)	19 (52.8): 17 (47.2)	0.020	3.13 1.18–8.30	12 (27.3): 32 (72.7)	17 (51.5): 16 (48.5)	0.030	2.83 1.09–7.34
A	65 (85.5)	46 (63.9)	0.004	0.30 0.13–0.67	76 (86.4)	44 (66.7)	0.0055	0.32 0.14–0.70
C	11 (14.5)	26 (36.1)		3.34 1.50–7.43	12 (13.6)	22 (33.3)		3.17 1.43–7.02
VKORCI G-1639A								
AA + GA: GG	19 (50) 19 (50)	21 (58.3) 15 (41.7)	0.472		30 (68.2): 14 (31.8)	18 (54.5): 15 (45.6)	0.222	
A	22 (28.9)	25 (34.7)	0.451		38 (43.2)	20 (30.3)	0.103	
G	54 (71.1)	47 (65.3)			50 (56.8)	46 (69.7)		
Adiponectin + 276 G/T								
GT + TT: GG	22 (57.9): 16 (42.1)	17 (47.2): 19 (52.8)	0.358		19 (43.2): 25 (56.8)	23 (69.7): 10 (30.3)	0.021	3.03 1.17–7.84
G	48 (68.6)	55 (76.4)	0.349		66 (75)	36 (48)	0.0003	0.31 0.16–0.60
T	22 (31.4)	17 (23.6)			22 (25)	39 (52)		3.25 1.68–6.30
MMP2 1306C/T								
CC versus CT + TT	19 (50) 19 (50)	16 (44.4) 20 (55.6)	0.632		21 (47.7) 23 (52.3)	17 (51.5) 16 (48.5)	0.742	
C	53 (69.7)	50 (69.4)	0.969		62 (70.4)	50 (75.8)	0.465	
T	23 (30.3)	22 (30.6)			26 (29.6)	16 (24.2)		
Gp la/lla C807T/G873A								
CT/GA + TT/AA:CC/GG	24 (63.2): 14 (36.8)	32 (88.9): 4 (11.1)	0.010	4.67 1.36–15.98	24 (54.5): 20 (45.5)	30 (90.9): 3 (9.1)	0.001	8.33 2.21–31.41
C/G	50 (65.8)	37 (51.4)	0.075		57 (64.8)	34 (51.5)	0.097	
T/A	26 (34.2)	35 (48.6)			31 (35.2)	32 (48.5)		

Data are expressed as number of subjects (percentage)

*Statistical significance comparing RVO to control subjects (< 75 y/o)

†Statistical significance comparing RVO to control subjects (≥ 75 y/o)

Odds ratio values as compared to control subjects

CI confidence interval

platelets [24, 25]. *GpIa/IIa* C807T polymorphism has been studied previously as a potential risk factor for RVO [12–14]; however, these studies came up with conflicting conclusions. We found that *Gp la/lla* C807T/G873A T/A carriers are on increased risk of CRVO and this finding aligns with the results reported by Dodson et al [14]. Two other studies did not confirm the association of this polymorphism with

Table 6 Multivariate logistic regression analysis between patients with RVO and control subjects ≥ 75 y/o

Variable	*B*	*P*	Odds ratio	95% CI	
				Lower	Upper
AGTR1 A1166C (CC + AC vs. AA)	1.17	0.086	3.21	0.85	12.17
Gp la/lla C807T/G873A (CT/GA + TT/AA vs. CC/GG)	2.65	0.003	14.10	2.54	78.39
Adiponectin + 276 G/T (GT + TT vs. GG)	2.07	0.004	7.96	1.94	32.66
Anticoagulant medication	4.56	0.001	95.69	6.02	1520.64

CI confidence interval

RVO [12, 13], but they included only patients with BRVO, whereas we did not observe statistically significant association as well.

Angiotensin II (AngII), an acute vasoconstrictor, stimulates mainly AGT1R and regulates systemic blood pressure and vascular tone. The activation of AGT1R induces also endothelial dysfunction, augments vascular inflammation, and it is implicated in the atherogenic process [26, 27]. The A1166C polymorphism was identified in the non-translated region, and it has been associated with obesity, hypertension, left ventricular mass, arterial stiffness and atherosclerosis [28–31]. In particular, it has been reported that the *AGT1R* A1166C CC and AC genotypes are associated with a susceptibility to hypertension [32], while it was recently documented that C allele carriers are at an increased risk of pronounced arterial stiffening during aging, especially after the age of 55 years [33]. It is noteworthy that the incidence of RVO also increases after the age of 50 years. We recognized a significant association between *AGTR1* A1166C and CRVO but not BRVO. Demir et al [9] demonstrated an association between *AGTR1* A1166C SNP and BRVO but not CRVO. Ethnicity differences and the small number of BRVO patients, as well as the higher number of hypertensive subjects in our studied groups compared to Demir et al, [9], could account for the disparity of the results.

Adiponectin is considered to exert several effects on vascular structure and function, including decrease in endothelial thickening [34] induction of arterial vasodilation [35] and inhibition of the expression of adhesion molecules [36–38]. The serum adiponectin concentration has a strong genetic component, with heritability estimated at 88% [39]. However, associations between adiponectin gene locus variants and adiponectin concentration in population studies have not always been confirmed, probably due to co-regulation by nutrition, hormones, inflammatory status, medication or technical issues [39]. Low circulating adiponectin level has been associated with an increased risk of arterial stiffness progression [40], supporting the concept that plasma adiponectin is a vascular protective hormone. On the contrary, higher plasma adiponectin concentration in populations with increased risk of cardiovascular events was associated with increased incidence of cardiac events [41], and moreover, adiponectin has been inversely associated with retinal arteriolar caliber measured by retinography in elderly hypertensive participants, suggesting that plasma adiponectin may be a marker of vascular disease or dysfunction in high-risk populations [42]. Regarding the *adiponectin* + 276 G/T polymorphism, adiponectin concentration has been shown to progressively increase from GG homozygotes to heterozygotes and TT homozygotes, but the statistical significance of this association has been reported to be marginal [39]. In our study, we did not identify an association between *adiponectin* + 276 G/T polymorphism and the incidence of RVO in the overall age group. Stratification by age, however, allowed the detection of a statistically significant association in the higher risk group consisting of older, hypertensive participants. Since T allele is associated with increased plasma adiponectin and there is an increased association between adiponectin and retinal vascular changes with age [41, 42], its presence could contribute to a higher prevalence of RVO only in the older, high-risk subjects of our studied group. These findings could support the suggestion that age modifies the association between plasma adiponectin and arteriole caliber [42], and furthermore, that this association could predispose to RVO. We did not, however, measure the plasma adiponectin or retinal vessel caliber in our studied subjects to confirm this suggestion and this is a limitation of our report.

MMP2 has been shown to be implicated in atherogenesis [43] and platelet function [44] both important in the pathophysiology of RVO. One previous study has associated *MMP2*-1306C/T polymorphism with the risk of RVO [11]. We did not reveal an association. However, *MMP2*-1306C/T polymorphism shows significant differences between ethnic groups [45, 46], and in agreement with that, genotype distribution of *MMP2*-1306C/T polymorphism in our control group varied significantly from the one reported in Ortak et al study [11]. T allele frequency of *MMP2* polymorphism for instance was lower in Turkish population (9%) [11], compared to the Greek population (29.88%).

Our study did not reveal any association between *VKORC1* G-1639A polymorphism and the prevalence of RVO as well. This finding aligns with the results reported by Weger et al. [10], but it comes in contrast to one reported by Ortak et al [15], where an association between CC and GG genotypes of the *VKORC1* C1173T, G-1639A SNPs and RVO was found. The frequency of GG genotype, however, was 40.2% in our control group similar to 36.9% reported in Weger et al study but different from 21% reported in Ortak et al study [15], emphasizing probably the differences between ethnicities, which could be responsible for the conflicting results.

This study has some limitations. Only Greek subjects were included in our report, and it has been shown that gene variants vary between populations due to ethnic differences. We did not identify any significant associations between the SNPs of *MMP2* and *VKORC1* and RVO. However, only one polymorphism of these genes was evaluated in this study. Moreover, due to a relatively small number of patients and controls, our results should be confirmed in larger cohorts. Specifically, regarding the BRVO group, no definite conclusions can be drawn from these data.

In conclusion, it seems that *AGTR1* A1166C and *Gpla/lla* C807T/G873A polymorphisms are likely predisposing risk factors for CRVO. Moreover, their role on the prevalence of RVO was identified to be stronger in patients younger than 75 y/o, indicating that the genetic susceptibility determined by the *AGTR1 and Gpla/lla* background may be more pronounced, when other risk factors are not as prevalent as in older subjects. On the other hand, *Adiponectin* + 276 G/T SNP is likely to predispose to RVO in older subjects; however, further studies are required with larger sample sizes to verify these findings.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Informed consent Informed consent was obtained from all individual participants included in the study.

References

1. Cugati S, Wang JJ, Rochtchina E, Mitchell P (2006) Ten-year incidence of retinal vein occlusion in an older population: the Blue Mountains Eye Study. Arch Ophthalmol 124(5):726–732. https://doi.org/10.1001/archopht.124.5.726
2. Rehak J, Rehak M (2008) Branch retinal vein occlusion: pathogenesis, visual prognosis, and treatment modalities. Curr Eye Res 33(2):111–131. https://doi.org/10.1080/02713680701851902
3. Ehlers JP, Fekrat S (2011) Retinal vein occlusion: beyond the acute event. Surv Ophthalmol 56(4):281–299. https://doi.org/10.1016/j.survophthal.2010.11.006
4. Hayreh SS (1994) Retinal vein occlusion. Indian J Ophthalmol 42(3):109–132
5. Frangieh GT, Green WR, Barraquer-Somers E, Finkelstein D (1982) Histopathologic study of nine branch retinal vein occlusions. Arch Ophthalmol 100(7):1132–1140
6. Rosendaal FR, Reitsma PH (2009) Genetics of venous thrombosis. Journal of thrombosis and haemostasis : JTH 7(Suppl 1):301–304. https://doi.org/10.1111/j.1538-7836.2009.03394.x
7. Russo PD, Damante G, Pasca S, Turello M, Barillari G (2015) Thrombophilic mutations as risk factor for retinal vein occlusion: a case-control study. Clin Appl Thromb Hemost 21(4):373–377. https://doi.org/10.1177/1076029614522544
8. Janssen MC, den Heijer M, Cruysberg JR, Wollersheim H, Bredie SJ (2005) Retinal vein occlusion: a form of venous thrombosis or a complication of atherosclerosis? A meta-analysis of thrombophilic factors. Thromb Haemost 93(6):1021–1026. https://doi.org/10.1160/TH04-11-0768
9. Demir S, Ortak H, Benli I, Alim S, Butun I, Gunes A, Ates O (2015) Genetic association between arterial stiffness-related gene polymorphisms in Brvo and Crvo patients in a Turkish population. Retina 35(10):2043–2051. https://doi.org/10.1097/IAE.0000000000000580
10. Weger M, Steinbrugger I, Renner W, Poschl EM, Brockmann T, Rabensteiner DF, El-Shabrawi Y, Haas A (2013) Role of the vitamin K epoxide reductase complex subunit 1 (VKORC1)–1639G>A gene polymorphism in patients with retinal vein occlusion. Curr Eye Res 38(12):1278–1282. https://doi.org/10.3109/02713683.2013.820329

11. Ortak H, Demir S, Ates O, Sogut E, Alim S, Benli I (2013) Association of MMP2-1306C/T and TIMP2G-418C polymorphisms in retinal vein occlusion. Exp Eye Res 113:151–155. https://doi.org/10.1016/j.exer.2013.06.009
12. Weger M, Renner W, Steinbrugger I, Cichocki L, Temmel W, Stanger O, El-Shabrawi Y, Lechner H, Schmut O, Haas A (2005) Role of thrombophilic gene polymorphisms in branch retinal vein occlusion. Ophthalmology 112(11):1910–1915. https://doi.org/10.1016/j.ophtha.2005.05.019
13. Salomon O, Moisseiev J, Vilganski T, Dardik R, Kurtz S, Shpringer E, Steinberg DM, Rosenberg N (2006) Role of five platelet membrane glycoprotein polymorphisms in branch retinal vein occlusion. Blood Coag Fibrinol 17(6):485–488. https://doi.org/10.1097/01.mbc.0000240922.57683.a8
14. Dodson PM, Haynes J, Starczynski J, Farmer J, Shigdar S, Fegan G, Johnson RJ, Fegan C (2003) The platelet glycoprotein Ia/IIa gene polymorphism C807T/G873A: a novel risk factor for retinal vein occlusion. Eye 17(6):772–777. https://doi.org/10.1038/sj.eye.6700452
15. Ortak H, Sogut E, Demir H, Ardagil A, Benli I, Sahin S (2012) Predictive value of the vitamin K epoxide reductase complex subunit 1 G-1639A and C1173T single nucleotide polymorphisms in retinal vein occlusion. Clinical & experimental ophthalmology 40(7):743–748. https://doi.org/10.1111/j.1442-9071.2012.02780.x
16. Johns MW (1991) A new method for measuring daytime sleepiness: the Epworth sleepiness scale. Sleep 14(6):540–545
17. Silva GE, Vana KD, Goodwin JL, Sherrill DL, Quan SF (2011) Identification of patients with sleep disordered breathing: comparing the four-variable screening tool, STOP, STOP-Bang, and Epworth Sleepiness Scales. J Clin Sleep Med 7(5):467–472. https://doi.org/10.5664/JCSM.1308
18. Singh A, Srivastava N, Amit S, Prasad SN, Misra MP, Ateeq B (2018) Association of AGTR1 (A1166C) and ACE (I/D) polymorphisms with breast cancer risk in North Indian population. Transl Oncol 11(2):233–242. https://doi.org/10.1016/j.tranon.2017.12.007
19. Fang WL, Zhou B, Wang YY, Chen Y, Zhang L (2010) Analysis of adiponectin gene polymorphisms in Chinese population with systemic lupus erythematosus. J Biomed Biotechnol 2010:401537. https://doi.org/10.1155/2010/401537
20. Li Y, Sun DL, Duan YN, Zhang XJ, Wang N, Zhou RM, Chen ZF, Wang SJ (2010) Association of functional polymorphisms in MMPs genes with gastric cardia adenocarcinoma and esophageal squamous cell carcinoma in high incidence region of North China. Mol Biol Rep 37(1):197–205. https://doi.org/10.1007/s11033-009-9593-4
21. Dubovyk YI, Harbuzova VY, Ataman AV (2016) G-1639A but not C1173T VKORC1 gene polymorphism is related to ischemic stroke and its various risk factors in ukrainian population. Biomed Res Int 2016:1298198. https://doi.org/10.1155/2016/1298198
22. Jaulim A, Ahmed B, Khanam T, Chatziralli IP (2013) Branch retinal vein occlusion: epidemiology, pathogenesis, risk factors, clinical features, diagnosis, and complications. An update of the literature. Retina 33(5):901–910. https://doi.org/10.1097/IAE.0b013e3182870c15
23. Harsfalvi J, Stassen JM, Hoylaerts MF, Van Houtte E, Sawyer RT, Vermylen J, Deckmyn H (1995) Calin from Hirudo medicinalis, an inhibitor of von Willebrand factor binding to collagen under static and flow conditions. Blood 85(3):705–711
24. Kunicki TJ, Kritzik M, Annis DS, Nugent DJ (1997) Hereditary variation in platelet integrin alpha 2 beta 1 density is associated with two silent polymorphisms in the alpha 2 gene coding sequence. Blood 89(6):1939–1943
25. Kritzik M, Savage B, Nugent DJ, Santoso S, Ruggeri ZM, Kunicki TJ (1998) Nucleotide polymorphisms in the alpha2 gene define multiple alleles that are associated with differences in platelet alpha2 beta1 density. Blood 92(7):2382–2388
26. Brasier AR, Recinos A 3rd, Eledrisi MS (2002) Vascular inflammation and the renin-angiotensin system. Arterioscler Thromb Vasc Biol 22(8):1257–1266
27. Benigni A, Cassis P, Remuzzi G (2010) Angiotensin II revisited: new roles in inflammation, immunology and aging. EMBO Mol Med 2(7):247–257. https://doi.org/10.1002/emmm.201000080
28. Osterop AP, Kofflard MJ, Sandkuijl LA, ten Cate FJ, Krams R, Schalekamp MA, Danser AH (1998) AT1 receptor A/C1166 polymorphism contributes to cardiac hypertrophy in subjects with hypertrophic cardiomyopathy. Hypertension 32(5):825–830
29. Takami S, Katsuya T, Rakugi H, Sato N, Nakata Y, Kamitani A, Miki T, Higaki J, Ogihara T (1998) Angiotensin II type 1 receptor gene polymorphism is associated with increase of left ventricular mass but not with hypertension. Am J Hypertens 11(3 Pt 1):316–321
30. Alavi-Shahri J, Behravan J, Hassany M, Tatari F, Kasaian J, Ganjali R, Tavallaie S, Sabouri S, Sahebkar A, Oladi M, Mirhosseini N, Shakeri MT, Montaser-Kouhsari S, Omidvar Tehrani S, Ghayour-Mobarhan M, Visvikis-Siest S, Ferns G (2010) Association between angiotensin II type 1 receptor gene polymorphism and metabolic syndrome in a young female Iranian population. Arch Med Res 41(5):343–349. https://doi.org/10.1016/j.arcmed.2010.06.007
31. Plat AW, Stoffers HE, de Leeuw PW, van Schayck CP, Soomers FL, Kester AD, Aretz K, Kroon AA (2009) The association between arterial stiffness and the angiotensin II type 1 receptor (A1166C) polymorphism is influenced by the use of cardiovascular medication. J Hypertens 27(1):69–75
32. Yang Y, Tian T, Lu J, He H, Xing K, Tian G (2017) A1166C polymorphism of the angiotensin II type 1 receptor gene contributes to hypertension susceptibility: evidence from a meta-analysis. Acta Cardiol 72(2):205–215. https://doi.org/10.1080/00015385.2017.1291211
33. Benetos A, Giron A, Joly L, Temmar M, Nzietchueng R, Pannier B, Bean K, Thomas F, Labat C, Lacolley P (2013) Influence of the AGTR1 A1166C genotype on the progression of arterial stiffness: a 16-year longitudinal study. Am J Hypertens 26(12):1421–1427. https://doi.org/10.1093/ajh/hpt141
34. Matsuda M, Shimomura I, Sata M, Arita Y, Nishida M, Maeda N, Kumada M, Okamoto Y, Nagaretani H, Nishizawa H, Kishida K, Komuro R, Ouchi N, Kihara S, Nagai R,

Funahashi T, Matsuzawa Y (2002) Role of adiponectin in preventing vascular stenosis. The missing link of adipovascular axis. J Biol Chem 277(40):37487–37491. https://doi.org/10.1074/jbc.m206083200
35. Fernandez-Real JM, Castro A, Vazquez G, Casamitjana R, Lopez-Bermejo A, Penarroja G, Ricart W (2004) Adiponectin is associated with vascular function independent of insulin sensitivity. Diabetes Care 27(3):739–745
36. Yokota T, Oritani K, Takahashi I, Ishikawa J, Matsuyama A, Ouchi N, Kihara S, Funahashi T, Tenner AJ, Tomiyama Y, Matsuzawa Y (2000) Adiponectin, a new member of the family of soluble defense collagens, negatively regulates the growth of myelomonocytic progenitors and the functions of macrophages. Blood 96(5):1723–1732
37. Arita Y, Kihara S, Ouchi N, Maeda K, Kuriyama H, Okamoto Y, Kumada M, Hotta K, Nishida M, Takahashi M, Nakamura T, Shimomura I, Muraguchi M, Ohmoto Y, Funahashi T, Matsuzawa Y (2002) Adipocyte-derived plasma protein adiponectin acts as a platelet-derived growth factor-BB-binding protein and regulates growth factor-induced common postreceptor signal in vascular smooth muscle cell. Circulation 105(24):2893–2898
38. Ouchi N, Kihara S, Arita Y, Nishida M, Matsuyama A, Okamoto Y, Ishigami M, Kuriyama H, Kishida K, Nishizawa H, Hotta K, Muraguchi M, Ohmoto Y, Yamashita S, Funahashi T, Matsuzawa Y (2001) Adipocyte-derived plasma protein, adiponectin, suppresses lipid accumulation and class A scavenger receptor expression in human monocyte-derived macrophages. Circulation 103(8):1057–1063
39. Menzaghi C, Trischitta V, Doria A (2007) Genetic influences of adiponectin on insulin resistance, type 2 diabetes, and cardiovascular disease. Diabetes 56(5):1198–1209. https://doi.org/10.2337/db06-0506
40. Youn JC, Kim C, Park S, Lee SH, Kang SM, Choi D, Son NH, Shin DJ, Jang Y (2013) Adiponectin and progression of arterial stiffness in hypertensive patients. Int J Cardiol 163(3):316–319. https://doi.org/10.1016/j.ijcard.2011.06.061
41. Wilson SR, Sabatine MS, Wiviott SD, Ray KK, De Lemos JA, Zhou S, Rifai N, Cannon CP, Morrow DA, Group TS (2011) Assessment of adiponectin and the risk of recurrent cardiovascular events in patients presenting with an acute coronary syndrome: observations from the Pravastatin OratorVastatin Evaluation and Infection Trial-Thrombolysis in Myocardial Infarction 22 (PROVE IT-TIMI 22). Am Heart J 161(6):1147–1155 e1141. https://doi.org/10.1016/j.ahj.2011.02.014
42. Beltrami-Moreira M, Qi L, Maestri MK, Fuchs FD, Pakter HM, Moreira LB, Gus M, Matte US, Tavares AM, Oliveira MM, Fuchs SC (2015) Association between plasma adiponectin and arteriolar vessel caliber among elderly hypertensive subjects. J Am Soc Hypertens JASH 9(8):620–627 e621. https://doi.org/10.1016/j.jash.2015.05.008
43. Li Z, Li L, Zielke HR, Cheng L, Xiao R, Crow MT, Stetler-Stevenson WG, Froehlich J, Lakatta EG (1996) Increased expression of 72-kd type IV collagenase (MMP-2) in human aortic atherosclerotic lesions. Am J Pathol 148(1):121–128
44. Fernandez-Patron C, Martinez-Cuesta MA, Salas E, Sawicki G, Wozniak M, Radomski MW, Davidge ST (1999) Differential regulation of platelet aggregation by matrix metalloproteinases-9 and -2. Thromb Haemost 82(6):1730–1735
45. Srivastava P, Lone TA, Kapoor R, Mittal RD (2012) Association of promoter polymorphisms in MMP2 and TIMP2 with prostate cancer susceptibility in North India. Arch Med Res 43(2):117–124. https://doi.org/10.1016/j.arcmed.2012.02.006
46. Zhang LY, Ren KW (2011) Meta-analysis of MMP2-1306T allele as a protective factor in digestive cancer. Arch Med Res 42(3):239–243. https://doi.org/10.1016/j.arcmed.2011.04.013