A Retrospective Cross-sectional Study on Activated Protein C Resistance (Factor V Leiden): an Independent, Gender-dependent Risk Factor for Venous Thromboembolism

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Research

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Abstract

Background: Activated protein C resistance (APCR) due to factor V R506Q (Leiden) mutation is a major risk factor in patients with venous thromboembolism. The present study investigated the symptoms patterns and the risk for venous thromboembolism regarding multiple clinical, laboratory, and demographic properties in APCR patients.

Material and Methods: A retrospective cross-sectional analysis was conducted on a total of 288 APCR patients with age interval ranging between 1 to 80 years. In addition, 288 control samples, reported healthy after confirmatory tests, were also randomly selected. Demographic information, clinical manifestations, family and treatment history were recorded, and specific tests applied.

Results: APCR was found to be 2.3 times significantly more likely in men (OR: 2.1, $p < 0.05$) than women. The risk of DVT and PE in APCR patients was 4.5 and 3.2 times more than the normal group, respectively ($p < 0.05$). However, APCR could not be an independent risk factor for arterial thrombosis and pregnancy complications. Moreover, patients were evaluated for thrombophilia panel tests and showed significantly lower protein C and S than the control group and patients without DVT ($p<0.0001$).

Conclusion: Factor V Leiden mutation and APCR abnormality are noticeable independent risk factors for venous thromboembolism. Screening strategies for factor V Leiden mutation in patients undergoing surgery, oral contraceptive medication, and pregnancy cannot be recommended, but a phenotypic test for activated protein C resistance should be endorsed in patients with venous thromboembolism.

1. Introduction

Thrombophilia, the most common hematologic disease, is characterized by blood coagulation abnormalities that increases the risk of thrombosis events [1]. Venous Thrombosis (VT) is the third cause of death due to cardiovascular diseases [2]. VT occurs in two general forms known as pulmonary embolism (PE) and deep vein thrombosis (DVT) [3, 4]. Common predisposing factors include aging, surgery, pregnancy, cancer, recent myocardial infarction, hormone therapy in females, prolonged inertia and genetic factors. The latter genetic causes comprise non-O-blood groups, genetic mutations such as G20210A in prothrombin (PTM) gene, deficiencies in protein C (PC), protein S (PS), and antithrombin III (AT-III) as well as activated protein C resistance (APCR) associated with factor V Leiden (FVL) [2, 5–7]. An estimated 64 percent of patients with venous thromboembolism have APCR as the most common associated clotting abnormality [8].

APCR is a hemostatic disorder characterized by the lack of adequate anticoagulant response to activated protein C (APC). More than 95% of cases with APCR abnormality are due to FVL mutation. Dahlback et al in 1993 reported the first case of APC resistance created by FV Leiden and in 1994; Bertina and colleagues discovered a single point mutation in the FV gene [7] [9]. APC, along with protein S as a cofactor, degrades Factor Va and Factor VIIIa by cleaving three arginine sites (R306, R506, and R679). The substitution mutation at G1691A results into an amino acid alteration (R506Q), leading to increased

thrombin generation and a hypercoagulable state [9, 10]. The risk of thrombosis was reported to increase by 5 to 10 fold in patients with heterozygous FVL mutations while the increase was nearly 80 fold in homozygotes [7].

APCR screening tests are based on the anticoagulant activity of APC and include modified activated partial thromboplastin time (aPTT), prothrombin time (PT) and snake Russell Viper Venom (RVV) time for patients on heparin and with a lupus anticoagulant (LA). A minimal prolongation of plasma clotting times by exogenous APC in the presence of FVL characterizes the APC-resistant phenotype. In addition, the R506Q mutation genetic defect can be unraveled by DNA techniques [10, 11]. A number of demographic, ethnic, and coagulation factors can influence diagnostic tests and patients' clinical phenotype. Previous literature suggested a possible correlation between levels of factors V, VIII and IX with APCR parameters [12]. Protein S, protein C and antithrombin III levels may also affect APCR, however, they have not been evaluated before [13].

To date, the majority of studies on this disease have been focused on the genotype component. However, clinical manifestations and disease characteristics have not been assessed sufficiently, whether independently or in correlation. Given the above background, and due to concerns regarding assay limitations that could adversely affect clinical diagnosis, we aimed to evaluate inherited APCR, in comparison to normal controls (APCR-negative group). The APCR diagnostic and clinical parameters that were studied include the levels of thrombophilia panel, the frequency of adverse thrombotic outcomes, the frequency of adverse pregnancy outcomes associated with FV Leiden, and the influence of gender and age.

2. Material And Methods

2.1. Study design and patients’ enrollment:

This study was designed as a retrospective cross-sectional survey conducted at the Coagulation Center of the Iranian Blood Transfusion Organization (IBTO) in Tehran, Iran. This center is a regional referral center for specialized coagulation testing and rare bleeding disorders. During the period from 2013 to 2018, a total of 288 patients with confirmed APCR abnormality were recruited to our study. Patients with the following criteria were included: 1) thrombotic symptoms and 2) with a reduced activity of APC, confirmed by APCR tests. Patients excluded from the study were those having other hemostatic bleeding and thrombotic disorders such as an inherited deficiency of protein C, protein S, AT-III or FVIII as well as those who received anticoagulants (warfarin, heparin, etc…), or antiplatelet (aspirin, clopidogrel, etc...) therapies at least 72 prior to blood sampling. Patients then undertook a detailed history assessment including demographical and clinical characteristics, age at first symptom, patient's chief complaint, familial history of thrombotic disorders, and their blood groups. In addition, clinical characteristics and laboratory investigations were determined. The healthy control group enrolled in this study included a total of 288 patients who were reported healthy after confirmatory tests. This study was approved by the

Medical Ethical Committee and the Institute Review Board (IRB) at IBTO. A written consent form was obtained and signed by all study participants for collection of blood samples.

2.2. Thrombophilia episodes in the year preceding the inclusion in the study:

Before sample collection and in the year preceding the inclusion in the study, the patient's detailed history was obtained from all participants. This included demographic data, chief complaint, familial history of bleeding and thrombotic disorders, anticoagulants and antiplatelet drugs used, clinical phenotype and thrombotic symptoms, pregnancy complications (such as abortion, eclampsia, preeclampsia, stillbirth, infertility, abortion trimester) and a detailed clinical history of 12 bleeding episodes based on ISTH scoring.

2.3. Laboratory Work-up:

Peripheral venous blood was collected in 3.2% (0.105M) sodium citrated tubes and centrifuged twice at 2,200 x g for 10 minutes at room temperature to obtain citrated platelet-poor plasma which was then stored at − 80°C. APCR abnormality was tested on a group of patients being screened for thrombophilia. As a principal challenge, complete assessments were performed in order to classify the patients' possible disorders and to discriminate between healthy subjects from mild thrombophilic disorders. Standard routine diagnostic assays were performed such as PT, International Normalized Ratio (INR), and aPTT. Presence of LA was confirmed with LA profile and PTT-LA and an abnormal dilute Russell Viper Venom (dRVV) time that demonstrated modification after the addition of the phospholipid rich dRVVT (LA Test and LA Confirm; Gradipore, Australia). Protein C activity was measured by chromogenic protein C method (Chromogenix, USA) with the normal range being 70 ± 140%. On the other hand, Protein S activity was performed using the Elisa method (protein S normal range is 55 ± 150%). Finally, APC resistance was measured using the APCR test according to the manufacturer's instructions (Pefakit APC-R FVL, Pentapharm, Basel, CH). To finalize the diagnosis, we also assessed the levels of other factors including Factor VIII, AT-III and fibrinogen. FVIII and AT-III were assayed using deficient plasma (Diagnostica Stago, Asnieres, France) whereas Von clauss method was used for Fibrinogen assessment.

2.4. Statistical analysis:

Descriptive analyzes were carried out and reported as frequencies and proportions (N, %). Data were analyzed by Kolomogorov-Smirnov test for normality checking. On the other hand, Spearman Correlation, Mann-Whitney U, ANOVA and Kruskal-wallis tests, and then uni and multi variable logistic regression were used to assess the relationship between risk factors and signs and symptoms. All tests were two-sided with the type I error rate fixed at 0.05 and the significant level for univariable and multivariable analyses assigned 0.25 and 0.05, respectively. Computations were performed using SAS (version 9.4; SAS Institute Inc, Cary, NC, USA) and SPSS for windows (Version 19) (SPSS Inc., Chicago, IL, USA).

Table 1
Characteristics of the APCR patients.

Variable	Median or Mean ± SD	Frequency (%)
Sex		
Female	-	424 (73.6%)
Male		152 (26.4%)
Age	35.34 ± 12.88	-
Familial History	-	167 (31%)
Type of Disease		
Bleeding	-	5 (0.9%)
Thrombophilia		540 (93.8%)
Bleeding and Thrombophilia		16 (2.8%)
Laboratory Work-up		
PT	14.15 ± 3.69	-
APTT	30.46 ± 3.28	
Protein C	117.76 ± 36.61	
Protein S	77.15 ± 22.91	
Anti-Thrombin	100.42 ± 14.30	
APCRPenta	2.57 ± 1.26	
Clinical Manifestation		
AT-III	-	37 (6.4%)
DVT		149 (25.9%)
PE		39 (6.3%)
Abortion		199 (33.3%)
TUS		39 (6.8%)
Abortion Time		
First-trim	-	163 (28.3%)
Second-trim		24 (4.2%)
Third-trim		5 (0.9%)

3. Results

A total of 285 FVL patients were investigated in this study of whom189 patients (65.6%) were females and the remaining (99 patients) were males (34.4%) (Table 1). Patients age ranged from 1 to 80 years old with a mean of 37.87 ± 13.67 years while the control group age ranged from 2 to 84 years old with a mean of 33.58 ± 11.8 years. Patients with familial history of thrombosis were ~ 37.7 % whereas most of the patients (66.3%) were new cases with no familial history. Demographic data and their analysis are presented in Table 1.

3.1. Demographic characteristics of patients with FV Leiden abnormality and their changes in line with various risk factors:

With regards to the presence or absence of FVL, this study contained the two groups: APCR and Normal population. There was a significant correlation between gender and APCR abnormality. In fact, despite the significantly higher frequency of women in both groups, the ratio of women in the normal group was significantly (***, $p < 0.0001$) higher than that in the APCR patients (55.2% normal versus 44.8% APCR) (Table 2). Moreover, using the uni- and multivariate logistic regression model, APCR was found to be ~ 2.3 fold significantly higher in men than women, where female sex was a protective variable that remained significant after adjusting for other variables (OR: 2.1, $p < 0.05$) (Table 3). However, the mean APC ratio showed no statistically significant difference in sex subgroups between females (1.46 ± 0.1), compared to males (1.45 ± 0.2). The mean age of women was significantly lower than men, owing to the adjustment for other variables in a multivariate analysis. Patients age ranged from 1–80 years (median 37), however, about 58.6% of patients were in the age interval of 20 to 50 years whereas 20% were between 50 to 60 years. Regarding the autosomal recessive inheritance of this disease, the familial history of patients was evaluated in APCR patients and controls. Indeed, the APCR abnormality was 1.2 times more likely to occur in individuals with familial history (Odds ratio of 1.27, and a 75% CI of 0.63 to 0.87) (Table 3). As shown in this table, sex, young age, and familial history of thrombosis were independent predictive factors for FV Leiden.

Table 2
Demographic, laboratory, and clinical status of patients with APCR abnormality and normal confirmed final diagnosis. (* p < 0.05, ** p < 0.01, *** p < 0.001)

Variable	Study Population		P-value
	APCR	Normal	
Sex			
Female	189 (44.8%)	233 (55.2%)	0.000***
Male	99 (65.6%)	52 (34.4%)	
Age (mean ± SD and range)	37 (1–80)	33.5 (2–84)	0.001***
Familial History of Thrombosis	87 (52.1%)	80 (47.9%)	0.18
Type of Disease			
Bleeding	5 (100%)	0 (0.0%)	0.99
Thrombophilia	261 (48.3%)	279 (51.7%)	
Bleeding and Thrombophilia	7 (43.8%)	9 (51.7%)	
Laboratory Findings			
PT	15.32 ± 4.14	13.13 ± 0.74	0.008***
APTT	36.44 ± 9.34	34.63 ± 3.26	0.92
Protein C	94.84 ± 46.58	125.89 ± 28.08	0.000***
Protein S	64.43 ± 29.96	82 ± 20.10	0.000***
Anti-Thrombin	97.58 ± 18.36	98.52 ± 11.64	0.189
APCRPenta	1.47 ± 0.19	3.91 ± 0.78	0.000***
Pregnancy Complications			
Abortion	65 (33.9%)	127 (66.1%)	0.000***
Eclampsia	0	0	-
Preeclampsia	3 (60%)	2 (40%)	0.25
Stillbirth	8 (42.1%)	11 (57.9%)	0.23
Infertility	4 (26.7%)	11 (73.3%)	0.06

Variable	Study Population		P-value
	APCR	Normal	
Abortion Time			
First Trim	51 (31.3%)	112 (68.7%)	0.17
Second Trim	11 (45.8%)	13 (54.2%)	
Third Trim	3 (60%)	2 (40%)	
Thrombotic Complications			
DVT	113 (75.8%)	36 (24.2%)	0.000***
PE	27 (75%)	9 (25%)	0.001***
AT-III	16 (43.2%)	21 (56.8 %)	0.24

3.2. Laboratory assessments:

Results showed significant differences in laboratory findings between patients and controls. Overall, most APCR patients showed normal PT and aPTT with; respectively, only 10.8% and 5.6% of patients harboring abnormal interpretations. Similarly, all normal controls also revealed normal PT and aPTT results; however, their mean values were significantly lower than those in the APCR group (Table 2). Therefore, we measured the association of PT and PTT results with APCR abnormality by uni- and multivariable regression model. Increased PT in the APCR group was 1.43 more likely than the control group, which remained significant after adjustment for other variables in a multivariate analysis (Table 3). In addition, the risk of APCR diagnosis was increased threefold in prolonged PTT situations (Table 3). Then, the patients were evaluated for thrombophilia panel tests; protein C, protein S and anti-thrombin. Interestingly, results showed that FV Leiden patients had statistically significant lower PC and PS levels than the control group (***, $p < 0.0001$), however, with no significant difference in anti-thrombin levels (Table 2). Using the logistic regression model, the thrombophilia panel was also identified as a protective risk factor and was highly statistically significant (***, $p < 0.0001$) in univariate analysis (Table 3). Moreover, APCR patients experiencing DVT episodes had significantly lower levels of PC ($p = 0.003$), PS ($p = 0.000$) and, anti-thrombin III (0.007) than patients without DVT. On the other hand, all thrombophilia tests including PC, PS and anti-thrombin had a strong negative correlation with PT and aPTT results in both groups that were statistically significant in APCR patients only (data not shown). FV Leiden patients also exhibited significantly lower levels of APCRPenta, used as a confirming test. The results of this test did not correlate with any of the laboratory tests, the thrombophilia panel tests and; hence, had no significant changes over the patient's lifetime.

3.3. Thrombophilia panel and screening tests according to age and gender:

Since APCR abnormality, compared to the normal group, revealed a varied pattern in the age of manifestations and sex subgroups, therefore, changes in laboratory tests and thrombophilia panel changes were evaluated at different ages and assessed in sex subtypes. Thrombophilia panel in APCR patients showed a negative correlation with age, however, this was not statistically significant. There was no significant correlation between screening tests and sex subgroups except PrC that female showed a significant higher mean rank range than male (P = 0.000).

Table 3
Crude and adjusted differences in APCR diagnosis according to demographic, laboratory and clinical parameters.

Variable	Univariable		Multivariable	
	Adjusted OR (75% CI)	P-value	Adjusted OR (95% CI)	P-value
Sex				
Female	1	-	-	-
Male	2.34	0.000*	**2.13**	**0.05****
Age (mean ± SD and range)	1.02	0.001*	1	0.92
Familial History of Thrombosis	1.27	0.18*	1.63	0.24
Laboratory Findings				
PT	1.43	0.001*	0.98	0.94
APTT	3.03	0.08*	0.99	0.97
Protein C	0.97	0.000*	0.99	0.20
Protein S	0.95	0.000*	0.98	0.10
Anti-Thrombin	0.98	0.09*	1	0.70
APCRPenta	0.00	0.95	-	-
Thrombotic Complications				
DVT	4.52	0.000*	**3.53**	**0.006****
PE	3.2	0.003*	3.25	0.19
AT-III	0.74	0.39	1.98	0.48
Abortion	0.37	0.000*	0.82	0.67
Abortion Time				
First Trim	-	0.185*		
Second Trim	1.85	0.162*		
Third Trim	3.29	0.199*		

3.4. Clinical manifestations:

Patients were divided into 3 groups according to their type of symptoms: bleeding disorder, thrombophilia, and bleeding and thrombophilia. More than 94% of subjects were thrombophilic and about

3% were thrombophilic-hemorrhagic. In the APCR group, 5 patients had just bleeding symptoms such as menorrhagia, coetaneous bleeding, epistaxis, GI bleeding, bleedings after tooth extraction and surgery. In fact, one patient had a total score of 21, three patients with a score of 3 and one patient with a score of 2.

The frequency of each clinical symptom was assessed and compared between the two groups. The most common manifestations in patients with APCR abnormality were DVT (39.2%) followed by abortion (22.6%) and PE (9.4%) (Fig. 1). Other reported bleeding symptoms were arterial thrombosis (AT, 11.8%) and thrombosis in unusual sites (TUS) (7.4%). About 75% of DVT and PE were seen in APCR patients and the rest of them in the normal control. The frequency of DVT and PE in the APCR group was significantly higher than the control group. Moreover, in the uni- and multivariate logistic regression model, the risk of DVT and PE in APCR patients was 4.5 and 3.2 times higher than the normal group, respectively. This risk remained significant after adjusting for other variables (Table 3).

Abortion, the second most common symptom in APCR patients, was the most common symptom in normal patients who showed ~ 66% of all abortions. Abortion in both groups occurred most frequently in the first trimester of pregnancy and the two groups did not show a significant difference in abortion trimester (Tables 2 and 3). Other pregnancy complications such as eclampsia, pre-eclampsia, stillbirth, and infertility were also assessed and there was no significant difference between the two groups (Table 2).

We then investigated separately all thrombotic symptoms in different demographic groups. Chi-Square test results revealed significant correlations between DVT and PE symptoms and sex subgroups. Despite the higher frequency of women, the incidence of DVT and PE symptoms was significantly ($p = 0.01$) higher in men (Fig. 2). According to the results obtained from the logistic regression model, the risk of DVT and PE was 1.5-fold and 1.7-fold significantly ($p = 0.01$) higher in men than women, respectively (95% CI: 0.31–0.85 and 95% CI: 0.17–0.84) (Table 3).

Finally, the incidence of symptoms at age intervals in both normal and APCR groups was then investigated. The age of all symptoms in APCR patients occurred at a young age. The inter-quartile range or the mean age dispersion of symptoms is low for the FV Leiden patients and ranged from 30 to 50 years for all symptoms. Results showed that individuals with Arterial Thrombosis and DVT in APCR patients had a mean age higher than normal controls that significant for DVT ($P = 0.003$) (Fig. 3).

4. Discussion

APCR is the most common finding in individuals with familial thrombophilia as a heightened propensity to, or increased risk of, thrombosis [1]. The inheritance of this abnormality is autosomal recessive and its frequency varies between countries. The prevalence of FVL in some parts of the world, such as Japan and Africa, is 0%; however, it is 5–10% in Europe. In Tehran, a province with different nations in Iran, the prevalence was reported to be ~ 5.5% [7, 11, 14, 15]. Consanguineous marriages in Iranian culture play an important role in the development of hereditary disorders [16]. Therefore, we focused in this study on the

demographic, clinical and laboratory characteristics as well as on estimating the risk of venous thromboembolism in patients showing resistance to APC in an Iranian population.

APCR abnormality is associated not only with genetic mutations but also with a number of factors including sex, age, anticoagulant and antiplatelet agents. Previous studies have shown that APCR along with the female gender appeared to increase the risk of thrombosis and that the mean APC ratio is significantly lower in females than in males [17]. Interestingly, in the present study, despite the higher frequency of women in both APCR and control groups, more than 65% of men were resistant to APC and this APCR abnormality was found to be significantly more likely in men than women, in contradiction to other previous reports [18, 19]. Indeed, although twice as many females than males were included in the study; however, we found a gender-related difference for APCR which was more likely to develop in men. This could reflect the fact that women are generally more investigated then men for inherited thrombophilia [20]. We suggest that a comprehensive study should be performed on the gender-related differences of APCR abnormality, which is beyond the scope of this study. On the other hand, the onset of thrombosis in the APCR group occurred at a young age, with a narrower range of 20–50 years, than the control group. Our data is in accordance with previous studies which showed that APCR is a significant thrombosis cause in younger individuals [21, 22].

Here, we describe the association of resistance to activated protein C with venous thromboembolism. Participants bearing APCR abnormality had a clearly significant increased risk for venous thromboembolism. The age and sex-adjusted incidence odds ratio for the first episode of venous thromboembolism was 3.53, consistent with the data reported by Ridker and Francesco [23, 24]. The most common manifestations in patients with APCR abnormality were DVT followed by abortion and PE. Other reported symptoms were arterial thrombosis (AT) and TUS. Despite the higher frequency of women, the second thrombotic symptom was miscarriage (22%) along with pregnancy complications (5%), accounting together for 27% of all symptoms. This percentage in the APCR group was significantly lower than that in the normal controls women which harbored more than 66% of all abortions. Previous studies reported that APC-resistant women experience their first thrombotic event at fertility, associated with both oral contraceptive use and pregnancy [25–27]. The authors recommended that women should be screened for FV mutations when becoming pregnant or before oral contraceptive use. This may in part explain that one of the relevant risk factors associated with the first event are oral contraceptive medication, pregnancy, and postpartum because women also had a lower average age than men [27]. However, given that abortions and pregnancy complications were higher in normal controls, FVL cannot be considered independently the cause of these symptoms in APCR patients, but this is rather multifactorial.

In general, only 11.8% and 9% of total events in APC-resistant patients were arterial thrombosis and PE. The association of AT with resistance to APC is a controversial issue. In our study, the mean age of the patients with AT was higher than other symptoms and also higher than the control group. In accordance, a previous study revealed that AT has been associated with other risk factors at an advanced age [28]. All thrombotic symptoms were also compared in sex subgroups when gynecological symptoms were

omitted. As mentioned above, despite the significantly higher frequency of women investigated, APCR was found to be significantly more likely in men. Moreover, thrombotic complications including DVT, PE, and AT were significantly higher in men than women. This could indicate a clinically relevant gender difference and reflect an increased DVT and PE risk in men, highlighting the low risk of venous thromboembolism caused by oral contraceptive use in female carriers [29].

Further investigations on our APCR population revealed significantly lower levels of protein C in male than female; however, no significant gender-related differences were found in the normal population (data not shown). Deficiencies and functional abnormalities of anti-thrombin III, protein C, and protein S are well-recognized causes of thrombotic events and familial thrombotic disease [30, 31]. In our study, levels of free protein C and protein S were significantly reduced in APCR patients, compared to the control group. Moreover, APCR patients experiencing DVT episodes also had significantly lower levels of protein C, protein S and anti-thrombin III than patients without DVT. In contrast, levels of factor VIII were higher in APCR patients with DVT episodes than those without it. The increased risk of thrombosis in APCR patients could be attributed to a hypercoagulable state caused by decreased levels of thrombophilia panel. However, the underlying mechanism of these changes in APCR patients has not been well studied and the importance of the plasma levels thrombophilia panel on thrombotic complications in APCR patients should be further evaluated.

5. Conclusion

In conclusion, patients with phenotypic resistance to APC have an increased risk for venous thromboembolism. Therefore, we suggest a phenotypic test for APCR in patients with venous thromboembolism. In addition, patients with a reduced response to APC should be further evaluated for factor V Leiden mutation. Moreover, some relevant risk factors associated with the first event are oral contraceptive medication, pregnancy, and postpartum. This study suggests that women experience their first thrombotic event at a younger age; however, factor V Leiden cannot be independently the cause of abortion and pregnancy complications in APCR patients. Finally, thrombotic events are found to be significantly more likely in men. It could be proposed that the increased risk of thrombosis in APCR patients could be attributed to a hypercoagulable state caused by decreased levels of thrombophilia panel.

List Of Abbreviations

Venous Thrombosis (VT), pulmonary embolism (PE), deep vein thrombosis (DVT), prothrombin (PTM), protein C (PC), protein S (PS), antithrombin III (AT-III), factor V Leiden (FVL), activated protein C (APC), activated Partial Thromboplastin Time (aPTT), prothrombin time (PT), Russell Viper Venom (RVV), lupus anticoagulant (LA), Iranian Blood Transfusion Organization (IBTO), Medical Ethical Committee and the Institute Review Board (IRB), International Normalized Ratio (INR), dilute Russell Viper Venom (dRVV)

Declarations

Ethics approval and consent to participate

All procedures have been approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.

Consent for publication

Informed consent was signed prior to participation in the study.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors contribution

Abbas Khosravi designed the study. Vahideh Takhviji, Asma Maleki, Sanaz Hommayoun, Mohammadreza Tabatabaei, Seyed Esmaeil Ahmadi and Omid Kiani Ghalesardi conceived and carried out the experiments. Afshin Davari, Ebrahim Azizi and Kazem Zibara analyzed the data. Mina Farokhian and Maral Soleimani validated the data. Vahideh Takhviji and Kazem Zibara wrote the paper. All authors discussed the results and commented on the manuscript.

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Figures

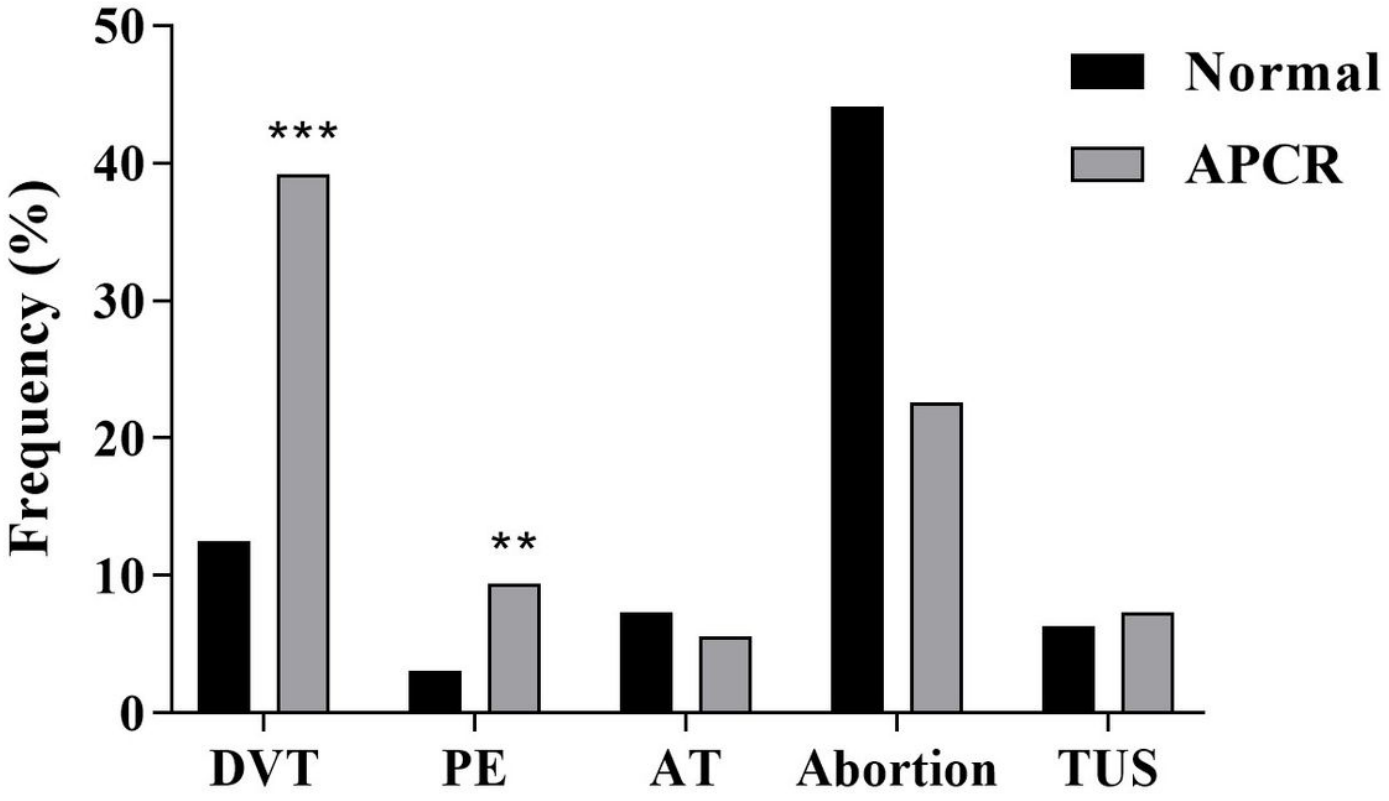

Figure 1

Clinical manifestations of confirmed APCR patients compared to control group in the year preceding inclusion in the study. (DVT: Deep Vein Thrombosis, PE: Pulmonary Embolism, AT: Arterial Thrombosis, TUS: Thrombosis in Unusual Sites) (* $p<0.05$, ** $p<0.01$, *** $p<0.001$)

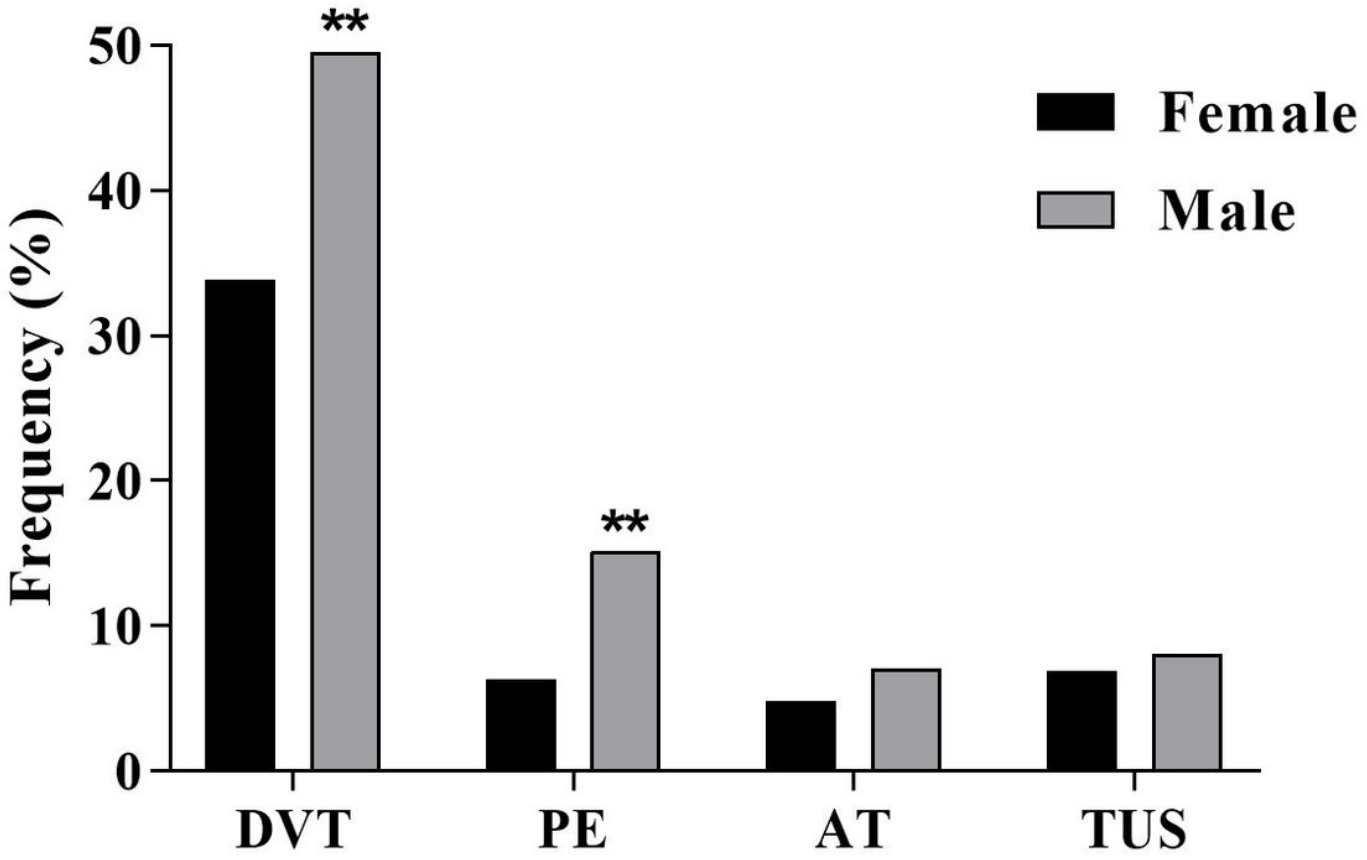

Figure 2

Comparison of thrombotic symptoms among males and females in APCR patients. (DVT: Deep Vein Thrombosis, PE: Pulmonary Embolism, AT: Arterial Thrombosis, TUS: Thrombosis in Unusual Sites) (* $p<0.05$, ** $p<0.01$, *** $p<0.001$)

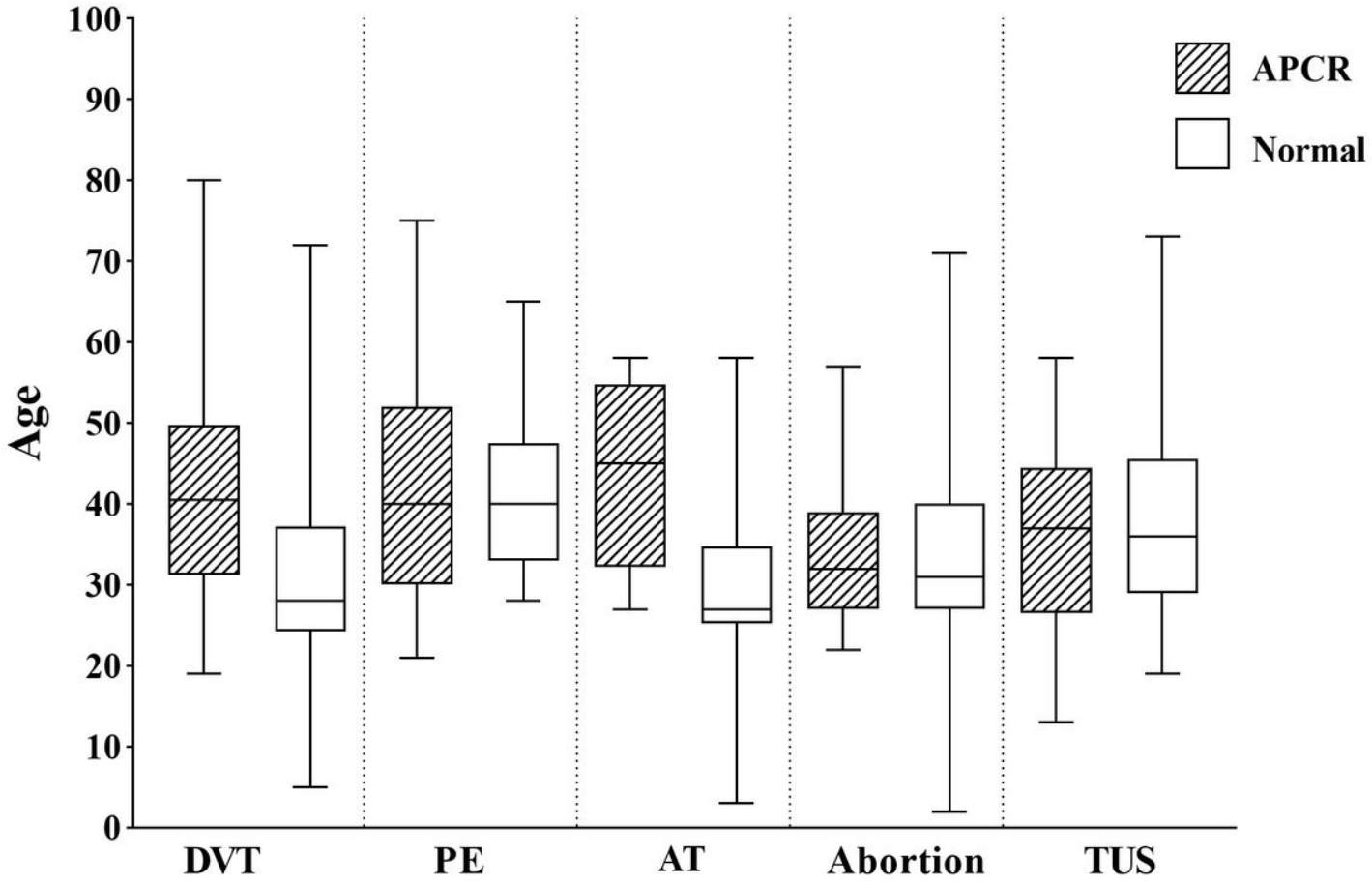

Figure 3

Pattern of thrombotic symptoms in APCR patients and control group in different ages. (DVT: Deep Vein Thrombosis, PE: Pulmonary Embolism, AT: Arterial Thrombosis, TUS: Thrombosis in Unusual Sites)