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Blood Cell Parameters Combined with Inflammatory Markers in the Early Diagnosis of Pulmonary Embolism

Dongfu Li, Mengqi Yuan*, Huijun Yang

Respiratory Department, Shijiazhuang people's Hospital, Shijiazhuang, 050000, China

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ABSTRACT

Venous thrombosis is a semi-solid formation of blood components that coalesce in the venous system, and the pathological process of its formation is called venous thrombosis. The deep veins of the lower extremities are a common site of prevalence, and the clinical diagnosis of lower extremity deep vein thrombosis can occur independently or as a complication of other diseases. There is a clear link between inflammation and coagulation/anticoagulation, with inflammatory mechanisms upregulating pro-inflammatory factors, downregulating natural anticoagulant substances, and inhibiting fibrinolytic activity; systemic inflammation is a strong pro-thrombotic stimulus; and in vivo, natural anticoagulant substances not only prevent thrombosis, but also deter inflammatory processes. The interconnection between inflammation and coagulation plays an important role in venous thrombosis. In this study, we analyzed the relationship between inflammatory markers CRP and Fg, FVIII:C and FIX:C by measuring plasma CRP concentration, Fg level, FVIII:C and FIX:C levels in patients with DVT diagnosed by ultrasound, and explored the role and mechanism of inflammatory response and coagulation factor abnormalities and the interaction between them in the development of DVT. In this paper, human blood DNA was extracted by phenol-chloroform-isoamyl alcohol extraction, and CRP 1059G/C gene polymorphism was detected by polymerase chain reaction-restriction enzyme segment length polymorphism (PCR-RFLP) nucleotide typing technique, and the genotypes of each subject were distinguished according to the bands seen by gel electrophoresis, and the frequency of each genotype was counted. Plasma CRP concentrations were measured by immunoturbidimetric assay, FVIII:C and FIX:C levels were measured by phase I assay, and plasma Fg levels were measured by coagulation assay in 59 cases (38 males and 21 females, aged 21-82 years, mean 49.67±11.12 years) and 26 controls (17 males and 9 females, aged 32-67 years, mean 50.13±8.96 years). The above indexes were compared between the two groups, and the correlation between CRP and FVIII:C, FIX:C and Fg was analyzed. Polymerase chain reaction-restriction enzyme segment length polymorphism nucleotide typing technique was used to detect the relationship between CRP 1059G/C gene polymorphism and DVT, to further search for risk factors of venous thrombosis, thus providing new ideas for the future prevention and treatment of this disease in clinical practice.

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Introduction

Venous thrombosis is a multifactorial disease. Virchow first noted in the mid-19th century that the causes of thrombosis could be divided into factors affecting blood flow (e.g., blood stasis), factors affecting blood components (e.g., hypercoagulable state), and factors affecting the vessel wall (e.g., atherosclerosis). Of these, blood stasis is the main risk factor for venous thrombosis (1). In recent years it has been found that inflammation and thromboembolic disease often go hand in hand, and it has been suggested that inflammation may also be a risk factor for VT (2). A large number of pathological and serological studies have provided strong evidence for the association between inflammation and VT, and have provided insight into the involvement of inflammation in the pathogenesis of VT (3). Clinical studies have looked at the inflammatory response to iliofemoral deep vein thrombosis by gadolinium-enhanced magnetic resonance venography (Gd-MRV) and found that the magnetic resonance scan around the thrombus in the affected limb of acute venous thrombosis was significantly more enhanced than the normal control side, confirming the presence of inflammatory exudation on the affected side (4-6). In addition, in a large number of animal experiments, there were findings of neutrophilic exudation in the vessel wall early in the induction of thrombosis by histopathological observation and leukocyte morphometry, followed by the appearance of

* Corresponding author. Email: ymq0116 @126.com

monocytes/macrophages and lymphocytes, while the elevation of cytokines occurred after thrombosis (7).

The plasma concentrations of three inflammatory mediators, interleukin-6, monocyte chemotactic protein-1, and interleukin-8, were observed to be significantly higher in patients with recurrent venous thrombosis than in normal healthy subjects (8). In addition, tumor necrosis factor, IL-6, MCP-I, and IL-8 were also significantly elevated in baboons, an animal model of DVT, and increased and peaked at different time points, respectively. The elevated levels of these inflammatory cytokines in venous thrombosis further confirm the presence of inflammatory phenomena (9). In addition, elevated levels of inflammatory response substances such as adhesion molecules and selectins have been found in some animal experiments, again confirming the correlation between inflammation and thrombosis. However, whether this inflammatory response occurs in VT is a cause or a consequence is still under investigation (10-12). The coagulation process in vivo is initiated by the tissue factor pathway and truncated by the amplification pathway. Tissue factor (TF) is a transmembrane glycoprotein that plays a key role in initiating the coagulation process as a surface receptor for coagulation factor VIIa (13). Under normal conditions, monocytes and endothelial cells have little to no TF activity on their surfaces, but increased TF expression in response to a number of stimulating factors causes a shift in hemostatic homeostasis that favors coagulation and thrombosis (14). Many inflammatory cytokines, including IL-1, IL-6 and TNF, are effective inducers of TF upregulation (15). It has been suggested that clinical and experimental findings in many different diseases have shown that cytokines play a key role in the physiopathological process of abnormal hemostasis through the mechanism causing the adverse coagulation state of the body, such as coagulopathies observed during sepsis, hepatic veno-occlusive disease after bone marrow transplantation, pro-thrombotic state of atherosclerotic vessels, deep vein thrombosis after major abdominal surgery, and thrombotic tendency in cancer patients (16). Cytokines are involved in the pathology of diseases such as thrombotic tendency of cancer patients (17-19).

The inflammatory response of neutrophil and monocyte rolling, adhesion, activation, and exudation in the presence of venous wall cytokines also leads to a waterfall response of venous thrombosis in which not only monocytes express increased TF, but also cathespin G released by neutrophils. Cathespin G causes endothelial cell damage exposing venous vessels the cathespin G causes collagen in the subendothelial tissues of the exposed venous vessels, thus causing thrombosis. In this study, we analyzed the relationship between the inflammatory markers CRP and Fg, FVIII:C and FIX:C by measuring plasma CRP concentration, Fg level, FVIII:C and FIX:C. We investigated the role and mechanism of inflammatory response and coagulation factor abnormalities and their interaction in the development of DVT. Plasma CRP concentrations were significantly elevated in patients with DVT, indicating that DVT is closely related to inflammation and that elevated plasma CRP levels may be a predictor of DVT development. It was further confirmed that elevated plasma levels of Fg, FVIII:C and FIX:C are important risk factors for DVT. plasma levels of Fg, FVIII:C and FIX:C are associated with the inflammatory state of the body, and the inflammatory response exerts procoagulant effects by elevating coagulation factors, which may be one of the mechanisms of DVT. The frequency of CRP 1059G/C allele mutation in patients with DVT was not significant compared with normal controls, and whether this gene polymorphism is a genetic risk factor for DVT formation remains to be investigated.

Methods

Clinical information

A total of 61 cases were selected from outpatients and inpatients with DVT from March 2017 to December 2019, of whom 39 were male and 22 were female, aged 16-82 years, with a mean of 49.64± 11.20 years; 60 healthy subjects were selected as the control group, of whom 38 were male and 22 were female, aged 30-71 years, with a mean of 51.54±9.63 years. The patients with DVT in the case group underwent vascular color Doppler ultrasonography to confirm the diagnosis (as shown in Table 1), which was performed by two experienced physicians.

In patients with venous thrombosis and healthy controls, 5 ml of fasting elbow venous blood was collected in the morning, anticoagulated with 109 mmol/L sodium citrate (V/V=9:1), centrifuged at 4℃

for 15 minutes at 3000 rpm, plasma was separated from cells, and part of the plasma was directly measured for Fg, while the remaining part was divided and stored in a refrigerator at -800C, and rapidly re-dissolved in a water bath at 37℃ during measurement.

Table 1: Case information

Case group	Gender	Number of people	Age
Case group	Male	39	46.64±10.20
	Female	22	53.64±9.20
Health Group	Male	38	48.54± 9.23
	Female	22	53.54± 8.32

Plasma CRP concentration was measured by immunoturbidimetric method and plasma Fg content was measured by coagulation method. The results were analyzed by fully automated biochemical analyzer.

(i) Reagent Reconstitution: Dissolve each spent FVIII plasma in 1 ml of distilled water and keep it at 15~250C for at least 15 minutes before use, then shake well until dissolved.

(ii)The standard plasma was diluted with imidazole buffer at 1:10, 1:20, 1:40, 1:80 and 1:160, and the samples were mixed with 0.1 ml of each of matrix plasma, ceruloplasmin suspension and 5% white clay saline suspension of FVIII plasma, and pre-warmed at 37℃ for 2 minutes, and 0.05 mol/L CaCl2 solution was added for 0.1 ml, and the coagulation time was recorded on a semi-automatic. The clotting time was recorded and detected by a semi-automatic hemagglutinator.

(iii) Plot the curve on double logarithmic curve paper with the dilution (1:10 as 100%) as the horizontal coordinate and the solidification time as the vertical coordinate.

(iv) The anticoagulated plasma of sodium citrate to be tested was taken in a water bath and diluted with imidazole buffer at 1:20, respectively, and the assay was performed as above.

(v) The activity of the plasma to be tested was then obtained by checking the standard curve against its clotting time and multiplying by 2.

2.2 Inflammatory marker reference methods

(i) Take 700-800μl of isolated cells in 1.5ml Eppendorf tubes

Add ddH_2O to 1.5 ml → shake → leave for 5 min → centrifuge at 8000 r/min for 10 min → discard the supernatant → continue adding ddH2O to 1.5 ml. repeat this step 2~3 times.

(ii) Aspirate the residual supernatant and keep only the leukocyte layer at the bottom of the tube in an Eppendorf tube → Shake → Add 500μl of leukocyte lysate and 10μl of PK (20ug/ml) → Place in a 50° C water bath overnight (12 hours), shaking the Eppendorf tube frequently.

(iii) Add 0.5 ml of Tris equilibrium phenol→Gently invert and mix for 5 minutes→ Centrifuge at 5000 r/min for 10 minutes→Aspirate the supernatant into another Eppendorf tube→Repeat the process with an equal amount of Tris equilibrium phenol (about 0.5 ml).

(iv) Add 500 μl of chloroform:isoamyl alcohol (24:1) to the aspirated supernatant → mix gently upside down for 5 min → centrifuge at 5000 r/min for 10 min → aspirate the supernatant → add 50 μl of 3Mol NaAc and 450 μl of isopropanol → mix gently → flocculent precipitate appears.

(v) Flocculent precipitate centrifugation 10000rpm ×8min→discard supernatant→add 70% ethanol 1ml, invert and mix several times→centrifugation 5000rpm ×10min, wash 2 times if necessary.

(vi) After evaporation of ethanol, dissolve in 100μl ddH2O and store at -20℃. The extracted DNA was analyzed by spectrophotometer and the OD260/OD280 ratio was measured to observe the nucleic acid concentration and purity. Those with a ratio between 1.6 and 1.8 were selected as PCR amplification templates.

The specific extraction method is shown in Figure 1.

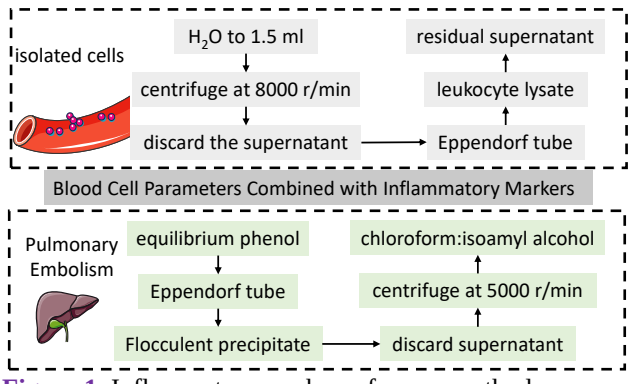

Figure 1. Inflammatory marker reference methods

The data were analyzed and processed using SPSS17.0 statistical software. The data were analyzed by the SPSS17.0 statistical software. Genotypes and allele frequencies were counted by frequency counting method, and chi-square test was used for frequency comparison, and P<0.05 was considered statistically significant.

Experimental results

We measured plasma CRP, Fg, FVIII:C, and FIX:C levels in 59 patients with DVT and 26 normal controls, and the results were as follows. The correlation coefficients were 0.432, 0.571, and 0.544, respectively, with P values less than 0.01 (as shown in Table 2).

Table 2: Measurement results of each index in the case and control groups Case group (A), Control group (B)

Group	Number of cases	CRP (mg/dl)	Fg(g/L)	FVIII:C(%)	FIX:C(%)
A	59	2.67±0.91	2.73±0.36	96.71±28.10	61.01±23.6
B	26	0.47±0.02	2.68±0.61	82.67±0.14	70.67±0.11

CRP 1059G/C means that the G in exon 1059 of CRP gene is replaced by C. The DNA fragment containing this mutated site is amplified by PCR using primers A and B. The length of the product is 744 bp. The normal unmutated amplified fragment had two restriction endonuclease MaeIII sites and three fragments of 310 bp, 233 bp and 201 bp, respectively; when the G mutation was changed to C, one MaeIII site was lost and only two fragments of 434 bp and 310 bp appeared after enzymatic cleavage. The genotypes can be classified according to alleles: wild type G/G (310bp, 233bp and 201bp bands are visible), heterozygous G/C (310bp, 233bp, 201bp and 434bp bands are visible), and pure mutant C/C (only 434bp and 310bp bands are visible). The genotypes of the subjects were distinguished by the bands seen in gel electrophoresis, and the frequencies of each genotype were counted. The distribution frequencies of GG, GC genotypes and G and C alleles in 60 normal controls were 88.3%, 6.7%, 91.7% and 8.3%, which were in accordance with Hardy-Weinberg equilibrium (x2=0.017, P=0.897), and the distribution trends of 1059G/C genotypes and alleles in the DVT and control groups were not significant (P > 0.05) (see Table 3 for results).

Table 3: Distribution of CRP 1059G/C genotypes and alleles in the case and control groups

Gene type	Case group (n=61)	Control group(n=60)	p
GG	86.9%	88.3%	
GC	8.2%	6.7%	0.897
CC	4.9%	5.0%	
Allele frequency %			
G	90.98	91.67	
C	9.02	8.33	

Currently, the risk factors for venous thrombosis are often classified as genetic or acquired, and the following genetic defects have been identified: PC deficiency, protein S (PS) deficiency, coagulation factor V (FV) Leiden mutation, hyperfibrinogenemia, abnormal fibrinogenemia, prothrombinogen G20210A abnormalities, hyperhomocysteinemia, and high levels of FVIII, FIX, FXI, and fibrinolytic inhibitors of thrombin activation play an important role in venous thrombosis. Statistically, at least 20% of patients with a first episode of venous thrombosis may have genetic alterations. Acquired risk factors include bed rest, fractures, surgery, trauma, long-term immobilization, malignancy, hormone replacement therapy, pregnancy, puerperium, and oral contraceptives. In addition, endothelial cell insufficiency, vitamin K deficiency and some organ diseases that affect coagulation factor production, such as liver diseases, are risk factors for venous thrombosis. In recent years, inflammation has been found to be closely related to venous thrombosis. The role of inflammation in the pathogenesis of atherosclerosis is well established. It is well known that thrombosis is often accompanied by an inflammatory response and that thrombosis is common at the site of inflammation.

The question of whether inflammation is also a risk factor for venous thrombosis has been extensively investigated (20). The first animal models and in vitro experiments revealed histopathological observations and leukocyte morphometry that revealed neutrophil exudation from the vessel wall in the early stages of thrombosis induction, followed by the appearance of inflammatory material such as monocytes/macrophages and lymphocytes, followed by an increase in plasma cytokine levels (21). The

presence of inflammatory exudate in the venous wall of patients with DVT was confirmed using gadolinium-enhanced magnetic resonance venography (Gd-MRV). These studies provide an objective pathological basis for the close association between venous thrombosis and inflammation. Since elevated levels are 1.9 times more likely than non-elevated individuals to develop first-time venous thrombosis, and since elevated levels are associated with recurrence of venous thrombosis, it is believed that elevated IL-8 plays a causal role in venous thrombosis and is a risk factor for venous thrombosis. In addition, studies have measured significantly higher WBC counts and CRP concentrations in patients with DVT, as well as a number of acute inflammatory responses such as adhesion molecules and selectins, thus confirming the involvement of inflammatory responses in the pathophysiology of venous thrombosis.

Example calculation results and analysis

Discussion of experimental results

In the case of thrombosis, these studies further clarify previously unrecognized risk factors and also facilitate the estimation of the transition from asymptomatic to symptomatic thrombosis. A number of inflammatory or immune proteins in the body have been investigated as potential risk factors for venous thrombosis in terms of etiology and pathogenesis. CRP is recognized as one of the most valuable acute chronotropic reactive proteins and is a normal protein fraction in human plasma. It is a sensitive and reliable indicator of the inflammatory state of the body, participating in local or systemic inflammatory responses. Inflammatory cells such as granulocytes and macrophages activated during the acute phase produce large amounts of cytokines such as IL-6, IL-1, TNF, etc. that stimulate the synthesis of CRP in liver epithelial cells, and the levels can increase dramatically up to 1000-fold.

CRP levels are significantly correlated with the appearance of inflammation and its severity, and rapidly return to normal when the disease is remitted, providing a pioneering clinical predictor. Recent studies have revealed that CRP levels directly correlate with the course of coronary heart disease, independently predict the occurrence of near- and long-term cardiac events and predict mortality in coronary heart disease, and suggest that CRP is the strongest predictor of coronary events among inflammatory markers. Recent studies suggest that CRP, unlike other inflammatory mediators, is not only a hallmark product of inflammation but also plays an important role in thrombus formation. In vitro, CRP induces the expression of tissue factor in monocytes and macrophages; upregulates intercellular adhesion molecule-1 (ICAM-1), vascular endothelial cell adhesion molecule-1 (VCAM-1) and E-selectin; activates the coagulation and complement systems, leading to imbalance of coagulation and fibrinolytic mechanisms and increasing thrombus formation. Therefore, it is assumed that CRP also has a role in the pathogenesis of venous thrombosis. Therefore, CRP has been considered as a candidate indicator of risk factors associated with the development of venous thrombosis. In the present study, plasma CRP concentrations were measured in patients with DVT, and the plasma levels were significantly higher in this group than in the control group [(2.67±0.91):(0.47±0.02)(mg/dl)], further confirming the close correlation between inflammation and venous thrombosis, and that plasma CRP may be an important predictor of DVT.

Blood cell parameters combined with inflammatory markers for the diagnosis of pulmonary embolism

The recent NHLBI FHS study found that genetic factors play a 39% role in serum CRP levels and that genes, as a relatively stable indicator, may be more convincing in explaining whether it is a causal problem. In addition, venous thrombosis being a polygenic disease, the search for genetic markers has been of great interest. The gene sequence of human CRP was identified as early as 1985 and belongs to the pentasomal protein Oligomeric calcium-binding protein. The G→C mutation in exon 2 of CRP at position 1059 was suggested to be associated with the development of coronary artery disease. The 1059G/C polymorphism in CRP gene was found to be associated with reduced serum CRP levels in 1452 cases with 8.6 years of follow-up. Based on the large number of studies that concluded that CRP levels are associated with venous thrombosis, it was suggested that the 1059G/C gene polymorphism may be a candidate gene for venous thrombosis.

Therefore, the first genetic epidemiological study was conducted to investigate the association between CRP gene polymorphisms and VTE, and it was found that CRP gene polymorphisms were not significantly associated with VTE in the Caucasian male population. Because of the genetic and population differences, the present study examined the CRP 1059G/C gene polymorphism in a population of Hebei, and found that the 1059G/C gene polymorphism also existed in this population, with the frequency of the 1059C allele being 9.02%, but this gene polymorphism did not correlate with the occurrence of DVT, which is consistent with the study in the white male population. The results were consistent with those in the white male population. The reason for this may be that the case sample was small and not representative of the population, so further large-scale clinical studies and exploration of whether the CRP 1059G/C gene polymorphism is a genetic risk factor for venous thrombosis is still needed. In this regard, it has been explained that the 1059G/C mutation may be due to the fact that it does not change the amino acid sequence, but only affects the level of CRP expression by associating other unidentified CRP functional mutations or nearby genes. Moreover, sometimes genetic polymorphisms themselves may not have a direct effect on the disease but affect the susceptibility of individuals to environmental risk factors, hence the current growing awareness of the role of gene-environment factor interactions in disease susceptibility.

There is a close and extensive link between inflammation and the coagulation system. The activated coagulation system interacts with the inflammatory response through multiple links, while monocytes, platelets, macrophages and endothelial cells are also involved, together intertwining into a complex network system that ultimately leads to thrombotic events. Several mediators involved in the coagulation response play an important regulatory role in the inflammatory response, and changes in their levels will significantly affect the inflammatory response. Natural anticoagulant substances have a unique anti-inflammatory activity, a biological activity that is distinct from their anticoagulant activity. Antithrombin has been found to have an inhibitory effect on endotoxin-induced IL-6 production in individual nucleated cells and vascular endothelial cells. Thrombin is the terminal enzyme of the coagulation waterfall and has multiple functions, not only by activating PC as an anticoagulant but also by inducing the expression of E-selectin, P-selectin and platelet-activating factor (PAF), which promotes the aggregation and adhesion of platelets and activated neutrophils and enhances the interaction between endothelial cells and neutrophils and participates in the inflammatory response。As shown in Figure 2. FVIII:C and FIX:C in the DVT group are higher than in the control group.

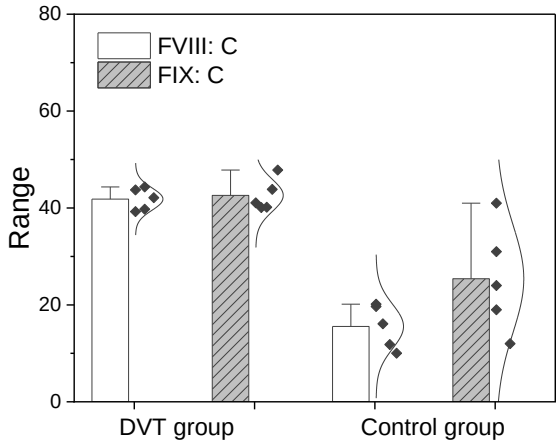

Figure 2: DVT group compared with control group FVIII:C and FIX:C

First of all, inflammation may play a key role in the process of venous thrombosis by activating monocytes and endothelial cells to release cytokines and chemokines that participate in the activation of the coagulation system and contribute to a hypercoagulable state in the body. One study measured a significant increase in the levels of FVIII, FIX, FX and FXI during human endotoxemia. In another study, IL-8 was found to be a risk factor for venous thrombosis when comparing 474 patients with DVT for the first time with the same number of normal controls. These provide a strong basis for the notion that the body has procoagulant properties in inflammatory states. The most studied mechanism of the pro-thrombotic role of cytokines such as TNF, IL-6, interleukin-1 (IL-1), and IL-8 is elevated in inflammatory states. The coagulation process in vivo is initiated by the tissue factor pathway and truncated by the amplification pathway.

Correlation analysis

Further studies found that plasma levels of FVIII:C were positively associated with the risk factor for venous thrombosis, and that each 10 IU/dL increase in FVIII:C was associated with an approximately 24% increase in the chance of recurrent deep vein thrombosis. When comparing the group with FVIII levels ≤100 U/dL to the group with ≥150 U/dL, the latter group had a 6-fold higher risk of thrombosis than the former group. Elevated FIX levels are an independent risk factor for the first episode of DVT, and elevated plasma FIX levels can increase the risk of VTE, and epidemiology shows that in 20% of patients with thrombosis, elevated FIX increases the risk by a factor of 2. In addition, high levels of FIX increase the risk of recurrence in patients with high FVIII; in fact, the relative risk of recurrence is up to 6 times higher in patients with both elevated FVIII and FIX, as shown in Figure 3. However, the mechanisms responsible for the risk of venous thrombosis due to elevated levels of these coagulation factors, FVIII, FIX, and Fg, are not known. One study concluded that FIX and Fg promote thrombosis by increasing prothrombin.

It has been analyzed that thrombin induces and increases the production of inflammatory markers such as pro-inflammatory cytokines by endothelial cells and monocytes pathologically, perhaps as a result of FVIII-related thrombin production, and that environmental factors such as hormone replacement therapy and other physiologic causes of elevated FVIII levels may further increase thrombin production and promote thrombosis. Prothrombin, the terminal enzyme of the coagulation waterfall, may not only act as an anticoagulant by activating PC, but may also be involved in the inflammatory response. Thus, inflammation and coagulation interact through multiple links, causing a disruption of the original balance between coagulation and anticoagulation, fibrinolysis and antifibrinolysis, and inflammation and anti-inflammation, creating an auto-amplified cascade effect that may be involved in the pathophysiological process of venous thrombosis.

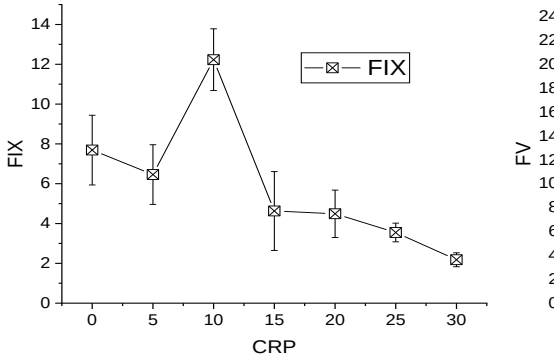

Figure 3: Scatter plot of CRP and FIX correlation analysis

In this study, plasma Fg, FVIII:C and FIX:C levels were measured, and these coagulation factors were found to be significantly higher in patients with deep vein thrombosis than in controls [Fg(2.73± 0.36):(2.68± 0.61)(g/l); FVIII:C(96.71± 28.10):(82.67±0.14)(%); FIX:C(61.01± 23.60):(70.67±0.11)(%)], further confirming that elevated levels of these coagulation factors are one of the important risk factors for venous thrombosis. The correlation between CRP, a marker of inflammation, and the levels of Fg, FVIII:C, and FIX:C in the DVT group was found to be significant, with correlation coefficients of 0.432, 0.571, and 0.544, respectively. The plasma levels of Fg, FVIII:C and FIX:C, especially FVIII:C, are not only genetically related but also influenced by the inflammatory status of the body, as shown in Figure 4. Thus, we can suggest that the pro-thrombotic mechanism of inflammation may include the pro-coagulant effect through the elevated coagulation factor pathway. This further illustrates the complex interactions between coagulation proteins and inflammatory markers arising from the interaction of genetic and environmental factors. The role of inflammation in the pathogenesis of atherosclerosis is well established. It is well known that thrombosis is often accompanied by an inflammatory response and that thrombosis is common at the site of inflammation.

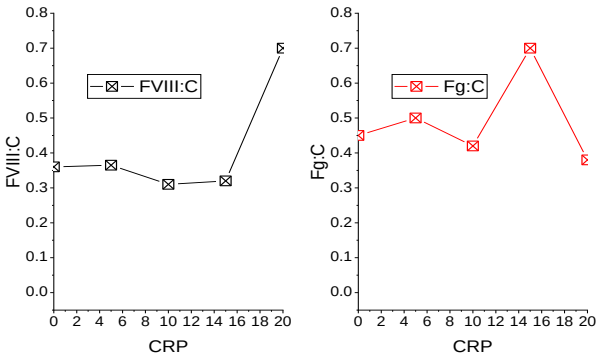

Figure 4: Correlation between CRP and level of FVIII:C

In conclusion, venous thrombosis is a multifactorial disease occurring under the combined effect of genetic and acquired risk factors driven by each other, and the interactions between genes and genes and gene-environment have an important role in inflammation and coagulation-activated venous thrombosis.

Conclusion

Bacterial infections are prevalent in outpatient clinics, emergency departments, wards and ICUs, and often pose a significant therapeutic challenge as concomitant diseases that accelerate deterioration. Due to the atypical symptoms of bacterial infections, inconspicuous primary lesions and easy diffusion through the bloodstream, and the variety and complexity of infectious bacteria, clinical laboratories and researchers at home and abroad have been devoted to exploring reasonable, effective and economical diagnostic methods and laboratory markers with high sensitivity and specificity. In this study, we analyzed the relationship between the inflammatory markers CRP and Fg, FVIII:C and FIX:C by measuring plasma CRP concentration, Fg level, FVIII:C and FIX:C. We investigated the role and mechanism of inflammatory response and coagulation factor abnormalities and their interaction in the development of DVT. In this paper, human blood DNA was extracted by phenol-chloroform-isoamyl alcohol extraction, and CRP 1059G/C gene polymorphism was detected by polymerase chain reaction-restriction enzyme segment length polymorphism (PCR-RFLP) nucleotide typing technique, and the genotypes of each subject were distinguished according to the bands seen by gel electrophoresis, and the frequency of each genotype was counted. Plasma CRP concentrations were measured by immunoturbidimetric assay, FVIII:C and FIX:C levels were measured by phase I assay, and plasma Fg levels were measured by coagulation assay in 59 cases (38 males and 21 females, aged 21-82 years, mean 49.67±11.12 years) and 26 controls (17 males and 9 females, aged 32-67 years, mean 50.13±8.96 years). The above indexes were compared between the two groups, and the correlation between CRP and FVIII:C, FIX:C and Fg was analyzed. Polymerase chain reaction-restriction enzyme segment length polymorphism nucleotide typing technique was used to detect the relationship between CRP 1059G/C gene polymorphism and DVT, to further search for risk factors of venous thrombosis, thus providing new ideas for the future prevention and treatment of this disease in clinical practice.

Conflict of Interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work.

Acknowledgment

Correlation of blood cell parameters, inflammatory markers and biochemical markers in patients with pulmonary embolism No 191200963.

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