The importance of laboratory testing to confirm hereditary thrombophilia and for prophylaxis of thrombotic episodes

Importanța testării de laborator pentru confirmarea trombofiliei ereditare și profilaxia episoadelor trombotice

Gabriela Jimborean[1], Paraschiva Postolache[2], Alpar Csipor[3], Edith Simona Ianoși[1]

1. University of Medicine and Pharmacy, Department of Pneumology, Târgu-Mureș, Romania

2. "Grigore T. Popa" University of Medicine and Pharmacy, Iași, Medical Department, Rehabilitation Clinical Hospital, Iași

3. Clinic of Pneumology, County Hospital Clinic Mureș, Romania

Corresponding author:
Paraschiva Postolache
E-mail: par.postolache@umfiasi.ro

Abstract

We present the case of a complicated pulmonary embolism in a young patient with factor V (factor V Leiden) (FVL) mutation - induced thrombophilia, aiming to emphasise the role of laboratory testing for thrombophilia in patients with deep vein thrombosis (DVT). A 22-year-old female (non-smoker, with chronic use of contraceptives) was admitted in the Pulmonary Clinic with a brutal episode of thoracic pain and fever. One month before, the patient was diagnosed with an extended femoral DVT. She was treated with anticoagulants and an arterial-venous shunt for vein patency preservation was placed. In our clinic, we found on CT scan a pulmonary infarction with pleural collection. The bacteriology of pleural fluid revealed Staphylococcus aureus. Targeted therapy was instituted (antibiotic guided by antibiogram, anticoagulants and anti-inflammatory drugs). We performed a large panel of tests to detect the etiology of thrombophilia. Factor V (FVL) mutation was found. The patient improved under treatment, and was discharged with recommendation for surgical reduction of the arterial-venous shunt, anticoagulants and contraceptives cessation. She was advised to permanently wear elastic stockings and avoid smoking and prolonged limbs immobilisation. A future pregnancy will need to be closely monitored, given the risk for pregnancy-associated thrombosis (repeated DVT, miscarriages, pre-eclampsia, foetal growth stop). ***Conclusion.*** *Sudden thrombosis occurring in young patients benefit from an extended laboratory investigation for hereditary thrombophilia. In these patients, antepartum and postpartum anticoagulation therapy should be a long-term prophylaxis method against thromboembolism and pregnancy complications.*

Keywords: ***thrombophilia, pulmonary infarction, factor V Leiden, prophylaxis***

Rezumat

Prezentăm un caz de embolie pulmonară complicată, la o pacientă tânără cu trombofilie indusă de mutația factorului V (factor V Leiden), pentru a sublinia importanța testării de laborator a trombofiliei la pacienții cu tromboză venoasă profundă (TVP). O pacientă de 22 de ani (nefumătoare, consumatoare de anticoncepționale) a fost internată în Clinica de Pneumologie pentru un episod brutal de durere toracică și febră. Pacienta a fost diagnosticată cu o lună înainte cu TVP femurală extinsă. S-au instituit tratament anticoagulant și un șunt arteriovenos pentru menținerea patenței venoase și prevenirea extensiei proximale a TVP. La internarea în Clinica de Pneumologie din Târgu-Mureș, CT-ul toracic a evidențiat un infarct pulmonar stâng cu o colecție mică pleurală. Examinarea bacteriologică din lichidul pleural a relevat un stafilococ auriu. Se instituie tratament antibiotic conform antibiogramei, anticoagulante și antiinflamatoare. Investigația largă a cauzelor trombofiliei decelează mutația factorului V (factor Leiden). După o evoluție favorabilă sub tratamentul conservator, pacienta se externează cu recomandarea de reducere chirurgicală a șuntului arteriovenos, tratament anticoagulant de lungă durată, excluderea anticoncepționalelor, purtarea permanentă a ciorapilor elastici, evitarea imobilizării prelungite a membrelor și evitarea fumatului. O sarcină viitoare va fi atent monitorizată, având în vedere riscul trombotic asociat sarcinii (tromboza venoasă profundă a mamei, avorturi spontane, preeclampsie sau opriri în creșterea fătului). Pe parcursul sarcinii, pacienta va beneficia de terapie anticoagulantă cu preparate heparinice. ***Concluzii.*** *Toate cazurile de TVP apărute la tineri trebuie investigate pentru evaluarea cauzelor genetice ale trombofiliei. Tratamentul anticoagulant este recomandat antepartum și post-partum la pacientele cu trombofilie pentru prevenirea complicațiilor tromboembolice asociate sarcinii.*

Cuvinte-cheie: ***trombofilie, infarct pulmonar, factor V Leiden, profilaxie***

Introduction

The three major causes of thrombosis, described by the german pathologist Rudolph Virchow (1821-1902), consist in Virchow' triad: blood hypercoagulability, vascular endothelial injury, and stasis or turbulence of the blood flow.

Hypercoagulability (thrombophilia) may be frequently due to genetic or acquired factors[1-3]. Among the genetic factors, there are: deficiency in anti-thrombin III, deficiency in protein C or S by gene mutations, factor V Leiden mutation(genetic mutation in F5 gene at 1691 position)[4], elevated clotting factor levels (Factors VIII, IX, XI), mutation of G20210A prothrombine gene (≥50% of hereditary thrombophilia)[5], mutation of metylen-tetra-hydro-folatreductase (MTHFR) and hyper-homocysteinemia. Alone or associated with the hereditary factors may act acquired factors[6-10] such as: smoking, myocardial infarction, obesity, infection, autoimmune diseases (collagen diseases and antiphospholipid syndrome)[5,8,10], chronic use of contraceptives, malignant

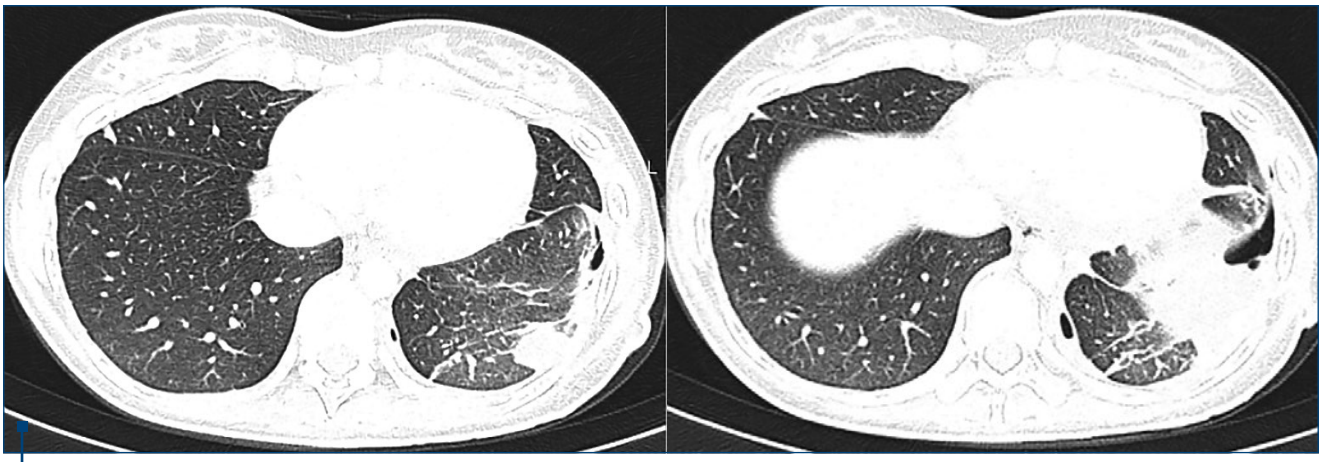

Figure 1. *Thoracic scan (pulmonary window): posterior left inferior lobe pulmonary infarction + small pneumothorax; small opacity in the right medium lobe*

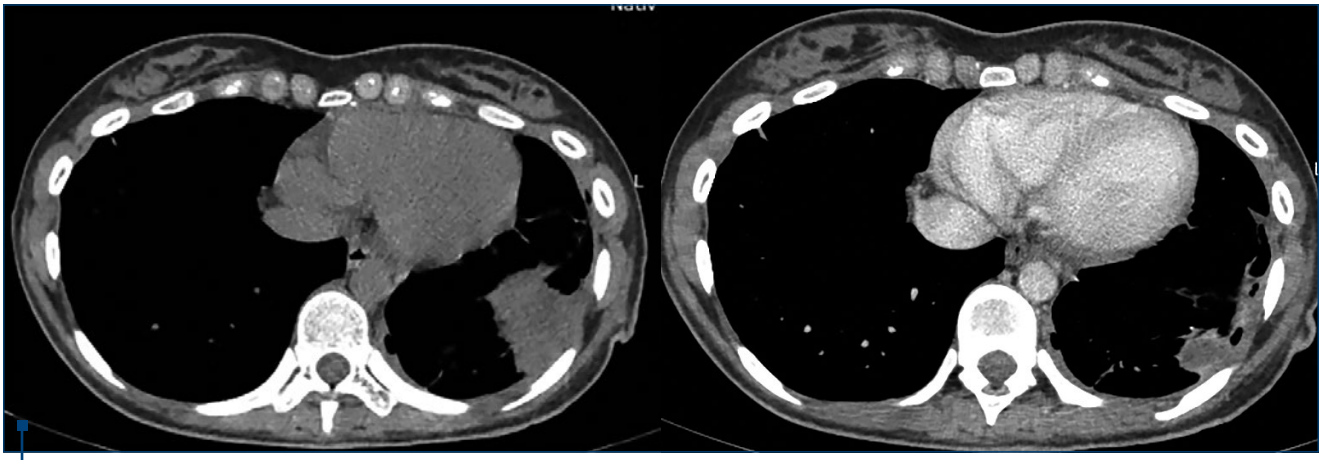

Figure 2, *a and b. Thoracic scan (mediastinal window): posterior left inferior lobe pulmonary infarction*

tumours[7], burns and polytrauma[11], pregnancy and labor, sickle-cell disease, essential thrombocytosis, *polycytemia vera* or leukaemia, abdominal or lower limb surgery.

Vascular injury can also be encountered in several conditions: long period of legs immobility (bed rest due to severe diseases or after surgery, orthopaedic diseases, fractures, stroke, devices for bones contention), inflammation, infection, burns, vessels trauma and implants[11,12].

Blood stasis may occur in common conditions: varicosities of the veins and vein compression, venous insufficiency, long-distance travel, chronic heart failure, atrial fibrillation, mitral stenosis, cardiac tumours, obesity[12,13].

The majority of hereditary thrombophilia cases are mild and affected individuals never develop abnormal clots. Hereditary thrombophilia may be associated with acquired comorbidities (heart failure, pregnancy, leg injuries, surgery, infection, advanced age, contraceptives, tumours or obesity) and the risk for thrombosis may consequently strongly increase. All the risk factors need to be identified and eliminated as soon as possible, and the patients benefit from anticoagulation prevention treatment.

The possible consequences of thrombophilia are: recurrent DVT, pregnancy complication (DVT in pregnancy, recurrent abortions, preeclampsia, delay of intrauterine growth, death of the foetus, premature birth)[14] and early coronary disease in patients under 50 years of age[12,13].

Factor V Leiden (FVL) represents an abnormal type of V factor which becomes resistant to the action of activated protein C - aPC (FVL cannot be easily degraded by aPC). FVL is a pro-coagulant that increases trombine blood levels. It is slowly inactivated (compared to the normal type) and it can contribute to the production of large quantities of trombine[15]. FVL is the specific name of the V factor gene mutation (chromosome 1q23) which controls the synthesis[4,16]. FVL is the most common genetic disorder of the blood coagulation, accounting for about 50% from all hereditary thrombophilia cases. Its prevalence is 5-7% in the general Caucasian population[17].

The thrombosis risk in FVL is 10% for heterozygote and 40% for homozygote individuals[17]. Sixty percent of pregnant women with DVT have FVL. The thrombotic risk increases with the association of other hereditary disorders or other pro-coagulants factors (from the "Virchow" triad).

Case report

A 22-year-old female patient (non-smoker) was admitted in the Pulmonology Clinic of Târgu-Mureș for high fever (38.5°C), dry cough, weakness and sudden thoracic pain.

One month before, the patient had been admitted in another hospital for an extended DVT of the main left femoral vein confirmed by Doppler ultrasound. The patient started an anticoagulant treatment with low molecular weight heparin, followed by oral anticoagulants (acenocumarol 2 mg/day). An arterio-venous shunt was performed in the Vascular Surgery Unit to ensure vein patency and to prevent clinically significant pulmonary embolism (PE) (in the absence of the inferior vena cava filters). The patient was a non-smoker, but she chronically used oral contraceptives.

In our clinic, a thoracic CT scan showed posterior left lobe pulmonary infarction with pleural collection, thickening of the pleura and a small pneumothorax (Figures 1, 2 and 3). The abdominal scan sections showed splenomegaly.

The thoracentesis revealed a sero-hematic, turbid fluid. The pleural fluid was an exudate, glucose 56 mg%, rich in neutrophils and mesothelial cells, without tumoral cells. The bacteriology exam of the pleural fluid revealed an infection with *Staphylococcus aureus*, and thus targeted antibiotic therapy was instituted according to the antibiogram. Acid fast bacilli in the pleural fluid were smear negative (Ziehl-Neelsen stain) and no growth on Löwenstein-Jensen medium for mycobacterium was present after two months.

We performed a large panel of laboratory tests to confirm the suspicion of hereditary thrombophilia and to diagnose the type of genetic disorder: test of resistance to activated protein C (aPC), DNA analysis for factor V Leiden and for prothrombin mutation, deficit of C protein or S protein or anti-thrombin III, genetic analysis for mutation of protein C, S. A heterozygous factor V mutation was evidenced (factor V Leiden).

Blood culture, basic haematology tests and immunologic tests for collagen diseases (antinuclear antibodies, antiphospholipidic and anticardiolipin antibodies, anti-beta2 glycoprotein 1, lupus inhibitor) were negative. The International Normalized Ratio at the admission in our clinic was 2.80.

The treatment was complex: antibiotics (cephalosporin and aminoglycosides corresponding to the germs drug sensibility), low molecular weight heparins (enoxaparin) followed by acenocumarol (anti-vitamin K; AVK) for long term, evacuation of the pleural collection and anti-inflammatory drugs. The patient was also under a close thoracic surgery surveillance, but given the complete resorbtion of the fluid by conservatory approach, she didn't need a drain insertion.

After three weeks of favourable evolution, the patient was discharged with the recommendation for surgical reduction of the arterial venous shunt, oral anticoagulants and cessation of contraceptives. We also recommended permanently elastic stockings, avoidance of smoking and of prolonged limbs immobilization.

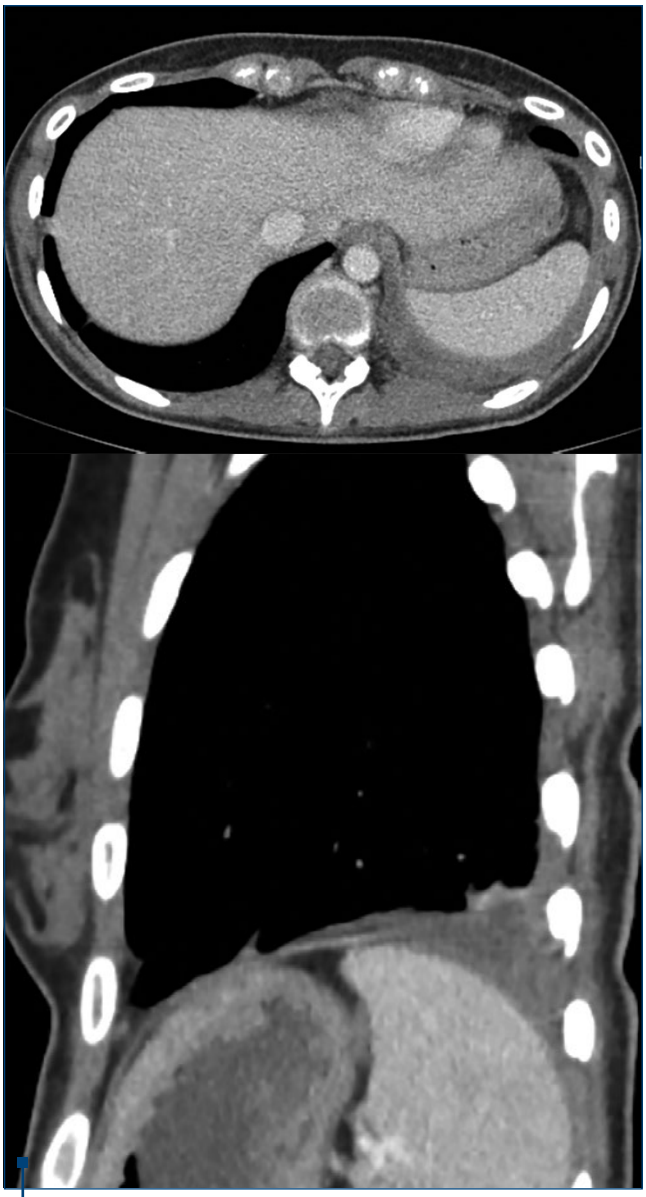

Figure 3. *Thoracic scan (mediastinal windows): a. Thoracic transversal section – posterior small basal pleural collection; b. Thoracic sagittal plan – posterior left basal pleural effusion, pleural thickening*

A future pregnancy will need to be closely monitored because of the pregnancy-associated thrombosis risks (DVT of the mother, miscarriages, preeclampsia and impaired foetal growth). During pregnancy, the patient will benefit of anticoagulant heparin therapy.

Discussion

The sudden onset of DVT in our young patient determined us to search for an hereditary thrombophilia. Screening for specific hereditary thrombophilia is recommended by several literature data in a large array of clinical conditions: DVT or arterial accidents (myocardial infarction or stroke) under age of 50 and especially in small children[4,18-21], family medical history of DVT or thrombophilia, DVT in other sites than "usual inferior members" (brain, liver and lungs, spleen)[2,16,21], recurrent

thrombosis (with no apparent cause), DVT or other pregnancy complication (recurrent abortion, preeclampsia, placental abruption, delay of intrauterine growth, death of the foetus, premature birth), heavy smokers, female with DVT and contraceptive, use of tamoxifen[21] etc.

In our case, the pulmonary embolism was considered a delayed complication of the DVT in the context of the large extension of DVT, thrombophilia and contraceptives use.

We interpreted the small pneumothorax and the pleural collection like a complication of the lung infarction. The ischemic process with necrosis increases vascular permeability and releases vasoactive cytokines[22]. The *staphylococcus* found in the liquid was considered a subsequent infection in the long-lasting pleural effusion associated to the lung infarction (ischemic necrosis area with low immunity).

Contraceptive drugs use was an additional risk factor for clinical thrombosis, developing in female patients carrying a genetic disorder.

Several laboratory tests are now available to diagnose hereditary thrombophilia. In our case, the lab investigation was crucial for the diagnosis of thrombophilia. The right etiological diagnosis permitted the accurate treatment and lifestyle recommendation for future thrombosis prevention (mostly in special condition – pregnancy, surgery, and immobilization). Complicated DVT with pulmonary embolism (DVT in a "high position" – main femoral vein in our case) has to be carefully and closely watched by clinic examination and by venous Doppler ultrasound examination.

We provided a targeted medical education (for the patient and family) with practical information for thrombosis prevention. Long-term anticoagulant treatment (3-6 months) will be recommended to reduce the risk for complications and clotting events.

After the complete restoration of vein patency, the surgical reduction of the A-V shunt under antithrombotic treatment will be needed.

The patient has to abandon for good the contraceptives use, to avoid smoking and inform other physicians (surgeon, gynaecologist, and anaesthetist) about the presence of the FVL disorder. Elastic stockings will be used especially when standing for long periods of time. She will avoid prolonged limbs immobilization, weight gain, leg trauma and "contact sports". Regular physical exercise could prevent vein stasis. Antepartum and postpartum anticoagulation (heparin) therapy will achieve prophylaxis against thrombosis and pregnancy complication.

Conclusion

Any thrombosis occurring in young patients should trigger laboratory investigations for thrombophilia. Screening for specific hereditary thrombophilia is recommended in some clinical conditions, especially in complicated, repeated DVT in young people without other evident recent risk factors, heavy smoking or using oral contraceptives. Contraceptive drugs use is a frequent risk factor for clinical thrombosis in female patients with this genetic disorder. Complicated DVT with pulmonary embolism has to be carefully long-term followed-up (by clinical examination and venous Doppler investigation).

Targeted medical education (for patient and family) regarding lifestyle is important to avoid additional risk factors for thrombosis. Antepartum and postpartum anticoagulation therapy has to be included for prophylaxis against thromboembolism and pregnancy complication. ■

References

1. Crowther MA, Kelton JG. Congenital thrombophilia states associated with venous thrombosis: a qualitative overview and proposed classification system. *Ann. Intern. Med*. 138 (2)/2003: 128–34.
2. Baglin T, Gray E, Greaves M, Hunt BJ, Keeling D, Machin S, Mackie I, Makris M, Nokes T, Perry D, Tait RC, Walker I, Watson H. Clinical guidelines for testing for heritable thrombophilia. *Brit J Haematol*. 2010; 149(2):209–220.
3. Seligsohn U, Lubetsky A. Genetic susceptibility to venous thrombosis. *New Engl J Med* 2001; 344:1222-1231.
4. Vandenbroucke JP, Koster T, Briet E, Reitsma PH, Bertina RM, Rosendaal FR. Increased risk of venous thrombosis in OC users who are carriers of factor V Leiden mutation. *Lancet* 1994; 344:1453-157.
5. Reshetniak TM, Patrushev LI, Stukacheva EA, Miroshnikov AI, Tikhonova TL, Nasonov EL, Alekberova ZS. Leiden, G20210A mutations in prothrombin gene and antiphospholipid antibodies in systemic lupus erythematosus and antiphospholipid syndrome, *TerArkh*. 2000; 72(5):34-8.
6. Malone PC, Agutter PS. The aetiology of deep venous thrombosis. *QJM* 2006; 99:581–593.
7. Sørensen HT, Lene Mellemkjær L, Steffensen FH, Olsen JH, Nielsen GL. The Risk of a Diagnosis of Cancer after Primary Deep Venous Thrombosis or Pulmonary Embolism, *N Engl J Med* 1998; 338:1169-117.
8. DeSancho MT, Berlus N, Christos PJ, Rand J. Risk factors for clinical manifestations in carriers of Factor V Leiden and prothrombin gene mutations. *Blood Coagul Fibrinolysis*, 2010; 21(1):11-5.
9. Rosendaal FR. Risk Factors for Venous Thrombosis: Prevalence, Risk, and Interaction, Seminars in Hematology, Vol 34 NoS (July) 1997 pp 171 187.
10. Chopra N, Koren S, Greer WL, Fortin PR, Rauch J, Fortin I, Senécal JL, Docherty P, Hanly JG. Factor V Leiden, prothrombin gene mutation, and thrombosis risk in patients with antiphospholipid antibodies. *J Rheumatol*. 2002 Aug; 29(8):1683-8.
11. Sevit S, Gallagher N. Venous thrombosis and pulmonary embolism. A clinico-pathological study in injured and burned patients. *British Journal of Surgery*, 48/211, 2005.
12. Kenneth M Moser, Peter F Fedullo, Judith K Litte John, Rebekah Crawford. Frequent Asymptomatic Pulmonary Embolism in Patients with Deep Venous Thrombosis, *JAMA*, January 19, 1994, Vol. 271, No. 3.
13. Lip GYH, Nieuwlaat R, Pisters R, Lane DA, Crijns HJGM. Refining Clinical Risk Stratification for Predicting Stroke and Thromboembolism in Atrial Fibrillation Using a Novel Risk Factor-Based Approach: The Euro Heart Survey on Atrial Fibrillation, *Chest* 2010; 137; 263-272.
14. Kujovich JL. Thrombophilia and pregnancy complications. Am J Obstet Gynecol. 2004 Aug; 191(2):412-24.
15. Shaw DM. Factor V Leiden: An Overview. InClin Lab Sci 2006; 19(4): 218.
16. Centers for Disease Control and Prevention. Genomics, Factor V Leiden and Venous Thrombosis Objectives, 2010.
17. Scully M, Graham S, Peshkin B**.** Factor V Leiden - Associated Thrombosis. 2013 In: Levy H, Lopresti L, Seibert D, editors. GeneFacts Lutherville, MD: NCHPEG; 2007-2013.
18. Raskob GE, Comerota AJ. Antithrombotic and Thrombolytic Therapy, 8th Ed: ACCP Guidelines**:** antithrombotic and thrombolytic therapy, 8th Ed: ACCP guidelines, *Chest Journal*, June 2008.
19. Geerts WH, Bergqvist D, Pineo GF et al. Prevention of venous thromboembolism: ACCP evidence-based clinical practice guidelines (8th Edition). *Chest* 2008; 133:381S–453S.
20. Berg A et al**.** Recommendations from the EGAPP working group: routine testing for factor V Leiden (R506Q) and prothrombin (20210G>A) mutations in adults with history of idiopathic venous thromboembolism and their adult family members. *Genet Med*. 2011; 13(1):67-76.
21. Grody WW, Griffin JF, Taylor AK, Korf BR, HeitJA(ACMG F V L Working Group)American College of Medical Genetics Consensus Statement on Factor V Leiden Mutation Testing 2007, http://www.acmg.net/resources / policies/pol-009.asp.
22. Light RW. Pleural effusion due to pulmonary emboli. *Curr Opin Pulm Med*. 2001 Jul; 7(4):198-201.