การศึกษาการใช้ ดีไดเมอร์และทรอมบิน-แอนติทรอมบินคอมเพล็ก ในการวินิจฉัยลิ่มเลือดในห้องหัวใจเอเตรียมซ้ายในผู้ป่วยลิ้นไมตรัลตีบ

นายเรวัตร พันธุ์กิ่งทองคำ

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรมหาบัณฑิต
สาขาวิชาการพัฒนาสุขภาพ
คณะแพทยศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย
ปีการศึกษา 2547

ISBN: 974-17-6073-6

THROMBIN-ANTITHROMBIN COMPLEX AND D-DIMER FOR DETECTION OF LEFT ATIRAL THROMBUS IN THE PATIENTS WITH MITRALSTENOSIS

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A Thesis Submitted in Partial Fulfillment of the Requirements
for the Degree of Master of Science in Health Development
Faculty of Medicine
Chulalongkorn University
Academic year 2004

ISBN: 974-17-6073-6

Title	Thrombin-antithrombin complex and D-dimer for detection of left atrial thrombus in the patients with mitral stenosis
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เรวัตร พันธุ์กิ่งทองคำ: การศึกษาการใช้ ดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็ก ในการวินิจฉัยลิ่มเลือดในห้องหัวใจเอเตรียมซ้ายในผู้ป่วยลิ้นไมตรัลตีบ (THROMBIN-ANTITHROMBIN COMPLEX AND D-DIMER FOR DETECTION OF LEFT ATRIAL THROMBUS IN THE PATIENTS WITH MITRAL STENOSIS) อาจารย์ที่ปรึกษา: ศ.นพ. กัมมันต์ พันธุมจินดา, พบ., วว. (อายุรศาสตร์ทั่วไป), วทม. , อาจารย์ที่ปรึกษาร่วม: ศ. นพ. วิษณุ ธรรมลิขิตกุล, พบ., วว. (อายุรศาสตร์ทั่วไป), ศ. นพ. ปิยทัศน์ ทัศนาวิวัฒน์, พบ., วว. (อายุรศาสตร์ทั่วไป), วทม.

วัตถุประสงค์: เพื่อทราบความแม่นยำของดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็กในการวินิจฉัยลิ่มเลือดในห้องหัวใจเอเตรียมซ้ายในผู้ป่วยลิ้นไมตรัลตีบ

รูปแบบการศึกษา: ผู้ป่วยจำนวน 120 รายที่ได้รับการรักษาในแผนกผู้ป่วยนอกและคลินิกโรคหัวใจซึ่งได้รับการวินิจฉัยว่าเป็นโรคลิ้นไมตรัลตีบโดยอาศัยการตรวจคลื่นสะท้อนหัวใจจะถูกคัดเลือกเข้าสู่การศึกษานี้ โดยผู้ป่วยทุกคนได้รับการตรวจคลื่นสะท้อนหัวใจทางหลอดอาหารเพื่อวินิจฉัยว่ามีลิ่มเลือดในห้องหัวใจเอเตรียมซ้าย รวมทั้งได้รับการตรวจปริมาณดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็กจากเลือด แล้วนำข้อมูลมาวิเคราะห์ความแม่นยำ (ความไวและความจำเพาะ) ของการตรวจทั้ง 2 ชนิดนี้ในการวินิจฉัยลิ่มเลือดในห้องหัวใจเอเตรียมซ้าย

สถานที่ทำการวิจัย: โรงพยาบาลศิริราช ซึ่งเป็นโรงพยาบาลระดับตติยภูมิ

วิธีการศึกษา: การวิจัยเชิงวิเคราะห์ชนิด cross-sectional

ผลการศึกษา: ความไวและความจำเพาะของดีไดเมอร์ในการวินิจฉัยลิ่มเลือดในหัวใจเอเตรียมซ้ายของผู้ป่วยลิ้นไมตรัลตีบที่จุดตัด $\leq$ 500 นาโนกรัมต่อมิลลิลิตร มีค่า 80% (95% CI, 76.4% - 83.7%) และ 66% (95% CI, 68.68% - 70.32%) ตามลำดับ ส่วนความไวและความจำเพาะของทรอมบิน-แอนติทรอมบิน-คอมเพล็กที่จุดตัด $\leq$ 3 ไมโครกรัมต่อลิตร มีค่า 75% (95% CI,71.1% - 79%) และ 60% (95% CI, 557% - 64.4%) ตามลำดับ ค่าการทำนายบวกและลบต่อการพบลิ่มเลือดในห้องหัวใจเอเตรียมซ้ายของดีไดเมอร์ที่จุดตัด$\leq$ 500 นาโนกรัมต่อมิลลิลิตรมีค่า 32.5% และ 94.1% ตามลำดับ ดีไดเมอร์มีความแม่นยำในการวินิจฉัยภาวะดังกล่าวได้ดีกว่าทรอมบิน-แอนติทรอมบิน-คอมเพล็ก โดยพื้นที่ใต้กราฟของดีไดเมอร์มีค่ามากกว่าของทรอมบิน-แอนติทรอมบิน-คอมเพล็ก

สรุป: การตรวจดีไดเมอร์และทรอมบิน-แอนติทรอมบิน-คอมเพล็กในเลือด สามารถช่วยในการวินิจฉัยลิ่มเลือดในหัวใจเอเตรียมซ้ายของผู้ป่วยลิ้มไมตรัลตีบ โดยดีไดเมอร์มีความแม่นยำกว่าทรอมบิน-แอนติทรอมบิน-คอมเพล็ก

ลายมือชื่อนิสิต.....

สาขาวิชา การพัฒนาการสุขภาพ ลายมือชื่ออาจารย์ที่ปรึกษา.....

ปีการศึกษา 2547 ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.....

457 54301 30 : MAJOR HEALTH DEVLOPMENT

KEYWORDS: THROMBIN-ANTITHROMBIN COMPLEX/D-DIMER/LEFT ATRIAL THROMBUS/MITRAL STENOSIS.
REWAT PHANKINGTHONGKUM: THROMBIN-ANTITHROMBIN COMPLEX FOR DETECTION OF LEFT ATRIAL THROMBUS IN THE PATIENTS WITH MITRAL STENOSIS. THESIS ADVISOR: PROF. KAMMANT PANTHUMJINDA, M.D., M.Sc., THESIS CO-ADVISOR: PROF. PYATAT TASANAWIWAT, M.D., M.Sc., PROF. VISANU THAMLIKITKUL, M.D., M.Sc. pp. ISBN: 974-17-6073-6 .

Objective: To determine the diagnostic performance of the peripheral blood thrombin-antithrombin complex and D-dimer levels in detection of of left atrial thrombus in the patients with mitral stenosis

Design: Cross-sectional study

Setting: Siriraj Hospital

Method: 120 consecutive adult patients with mitral stenosis diagnosed by transthoracic echocardiography from heart clinic and Out-patient Department at Siriraj Hospital were recruited for this study. All patients underwent both transesophageal echocardiography (TEE) and peripheral blood tests for thrombin-antithrombin complex and D-dimer levels. Both tests were performed by independent experienced technicians who were blinded to TEE result which was considered as the gold standard for detection of the left atrial thrombus.

Result: The sensitivity and specificity of D-dimer for left atrial thrombus detection at the cut-off point of $\leq$ 500 ng/ml were 80% (95% CI, 76.4% - 83.7%) and 66% (95%CI, 68.7% - 70.3%), whereas the sensitivity and specificity of TAT for left atrial thrombus detection at the cut-off point of $\leq$ 3 μg/l were 75% (95%CI, 71.1% - 79%) and 60% (95%CI, 55.7% - 64.4%) respectively. The positive and negative predictive values for left atrial thrombus detection of D-dimer at the cut-off point of $\leq$ 500 ng/ml were 32.5% and 94.1% respectively, while those of TAT at the cut-off point of $\leq$ 3 μg/l were 27.8% and 92.1% respectively. The overall diagnostic performance of D-dimer was significantly better than TAT.

Conclusion: D-dimer and TAT could be used as the tool for detection of left atrial thrombus in the patients with mitral stenosis. D-dimer has better diagnostic performance for detection of left atrial thrombus in these patients better than TAT.

Student's signature……..

Field of study Health Development Advisor's signature……..

Academic year 2003 Co-advisor's signature…….

Acknowledgements

The author is grateful to Professor Visanu Thamlikitkul and Professor Pyatat Tatsanavivat for their academic advice, helpful comments and encouragement and to Dr. Chulaluk Komoltri for her statistical advice and assistance. The author would also like to thank Dr. Nisarat Opartkiattikul for laboratory support of D-dimer and thrombin-antithrombin complex (TAT). Furthermore, the author wishes to acknowledge all staffs of THAICERTC for their valuable comments in the part of research proposal.

Finally I would like to thank the Faculty of Medicine Siriraj hospital for financial support of this project.

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CHAPTER 1

INTRODUCTION

Rationale and background

Mitral stenosis caused by rheumatic carditis is still prevalent and has been a major health problem in Thailand. The true incidence of this disease in Thailand remains unknown but there have been about 900 cases of patients with mitral stenosis among patients underwent echocardiography at Siriraj hospital per year. Thromboembolism was one of the major detrimental morbidity of mitral stenosis with the average incidence of 15-20% at some point during the course of the disease.[1,2] The cause of thromboembolism is related to the left atrial thrombus. The true prevalence of left atrial clot from the autopsy study and surgical study was 27% and 23% respectively.[3, 4] The prevalence of left atrial thrombus in these patients from Siriraj experience was 17-30% (unpublished data). Left atrial thrombus was commonly found in mitral stenosis with atrial fibrillation and it was associated with systemic thromboembolism.

In the past, the diagnosis of the left atrial thrombus in the patients with mitral stenosis depends on the surgical approach or autopsy. Subsequently, transthoracic echocardiography which is a non-invasive method was developed rendering the diagnosis of left atrial thrombus in these patients easier. However, the sensitivity and the specificity of this equipment were fair.[5] Therefore, transesophageal echocardiography (TEE) was developed and it was demonstrated that to date TEE is one of the best non-invasive diagnostic tools to detect left atrial thrombus in mitral stenosis. The sensitivity and specificity of TEE for detection of left atrial clot were 100% (95%CI 74% - 99%) and 99% (95%CI 97% - 99.9%) respectively [6]. However, this procedure was not available in general hospitals due to lacking the skillful performer and expensive equipment. In addition, the procedure could also cause inconvenience and complications. Therefore, the simple tool to detect the left atrial thrombus should be further investigated.

The mechanisms of left atrial thrombus formation were related with blood stasis, endocardial injury and atrial fibrillation. When there is the obstruction of left ventricular inflow, the pressure of left atrium is gradually increased and finally can cause the endocardial injury inside the left atrium. The result of the injury promotes the adhesion of platelets and finally platelet aggregation occurs. Subsequently, the process of stimulation of coagulation cascade takes place by using the platelet surface due to accumulation of the prethrombotic substances within the left atrium. Finally, the left atrial thrombus occurs. However, the thrombus can be dissolved by the process of systemic fibrinolysis. The consequence of fibrinolysis leads to the D-dimer formation.

D-dimer is a degradation products formed when cross-linked fibrin contained within a thrombus is proteolyzed by plasmin. It may be produced in many illness and conditions associated thrombosis such as venous thrombosis, severe infection, malignancy, disseminated intravascular coagulation, heart failure, renal failure, acute myocardial infarction, acute stroke, connective tissue disease, liver disease, pregnancy, postoperative state, trauma, smoking, recent bleeding and advanced age.[7] Previous studies demonstrated that the sensitivity and specificity of D-dimer detected by ELISA technique with the cutoff level of $\leq$ 500 ng/ml for the diagnosis of pulmonary embolism was 91-93% and 25-42% respectively.[7, 9] Additionally, it was demonstrated that the sensitivity and specificity of this assay could detect deep vein thrombosis with the sensitivity and specificity of 97% and 23% respectively.[10, 11]

In addition, during the process of thrombus formation, the thrombin is generated and thrombin will combine with the antithrombin to form the thrombin-antithrombin complex (TAT). The concentration of TAT can be measured quantitatively by ELISA method. The conditions or diseases provided the abnormal increased concentration of TAT in the circulation are related to the thrombotic events such as disseminated intravascular coagulation (DIC) patients, multiple trauma, septicemia, preeclampsia, liver dysfunction, malignancy and the patients receiving heparin or fibrinolytic agents.[12-14]

From the previous study, it was demonstrated that the level of D-dimer and TAT from the peripheral blood and in the left atrium was abnormally high in the patients with mitral stenosis compared wit the normal person especially those with atrial fibrillation.[15]

It was demonstrated from a previous study that D-dimer detected by ELISA technique could be used to diagnose free floating thrombi in the left atrium of the patients with mitral stenosis with the sensitivity and specificity of 61% and 93.3% The cutoff level of D-dimer in this study was $\leq$ 300 ng/ml.[16] Therefore, D-dimer and TAT could be considerably used as a simple, non-invasive tool for detection of left atrial thrombus formation in the patients with mitral stenosis.

CHAPTER 2

REVIEW OF RELATED LITERATURE

2.1 Literature search strategy:

The literature search strategy used to obtain the information for this review included the Pub-MED database, Cochrane library and the reference list of related articles, abstracts presented in academic conferences. The keywords were **mitral stenosis**, **left atrial thrombus**, **D-dimer**, and **thrombin-antithrombin complex (TAT)**. The literature search covered the time period 1980 – 2003.

2.2 Literature review

Mitral stenosis defined as a mechanical obstruction of of blood flow between the left atrium and the left ventricle, is caused by the abnormal metal valve function. In most of the adult patients, the cause of mitral stenosis is previous rheumatic carditis.[17] About 60% of patients with rheumatic mitral valve disease do not give a history of rheumatic fever or chorea and about 50% of patients with acute rheumatic carditis do not have a clinical valvular heart disease. The other causes of mitral stenosis are uncommon. These include congenital mitral stenosis, infective endocarditis, SLE, carcinoid heart disease, endomyocardial fibrosis, rheumatoid arthritis and massive mitral valve annular calcification.[18] The other conditions or diseases that can cause obstruction of left ventricular inflow include left atrial myxoma, massive left atrial ball thrombus and cor triatriatum.

Acute rheumatic carditis causes pancarditis involving the pericardium, myocardium and endocardium. In the tropical and subtropical climates including Thailand, the latent period

is often shorter than in the developing countries.[1] Repeated episodes of carditis rendering to the development of mitral stenosis that is characterized by the deposition of fibrous tissue. There is the fusion of commissures, cusps or chordae, or combination of these structures. Finally, the deformed valve is prone to fibrosis and calcification due to the abnormal blood flow around the valve. The lesions along the line of closure result in fusion of the commissures and contracture as shortening and thickening of the valve leaflets. The chordal lesions are manifested as shortening and fusion of these structures. The combination of commissural infusion, valve contracture and fusion of chordae tendineae results in a narrow, funnel-shaped orifice, which restricts the flow of blood from the left atrium to the left ventricle. The left atrium is enlarged and hypertrophied as a consequence of left atrial hypertension.

The major consequence of the obstruction is stasis of blood in left atrium which might lead to local prethrombotic state due to accumulation of circulating prothrombotic substance as demonstrated in some previous studies.[4, 5, 7-9] Recently, our study confirmed the accumulation of thrombin-antithrombin complex (TAT), F1+2, and D-dimer, the markers of prethrombotic state, in the left atrium of the patients with mitral stenosis.[15] In addition, we also demonstrated that the level of TAT in peripheral blood of the patients with mitral stenosis was significantly higher than those of the normal subject. If the abnormal physiology persisted and gradually increased to the circumstances overwhelming the ability of the fibrinolytic system to maintain the hemostasis, the thrombus formation could take place in the left atrium. In addition, abnormal fibrinolysis was observed in the patients with mitral stenosis by demonstrating the increased level of PAI-I (plasminogen activator inhibitor-I) from peripheral blood of these patients.[19]

D-dimer is a specific degradation product that is formed when cross-linked fibrin contained within a thrombus is cleaved by plasmin. D-dimer may be produced in many illness and conditions associated with thrombosis and thrombolysis. These conditions include venous thrombosis, severe infection, malignancy, disseminated intravascular coagulation defect (DIC), heart failure, renal failure, acute myocardial infarction, acute stroke, connective tissue disease, liver disease, pregnancy, postoperative state, trauma,

smoking, recent bleeding and advanced age.[7] Many factors affect the blood level of D-dimer detected. Larger clots tend to produce a larger amount of D-dimer.[24] Conversely, D-dimer may not increase in patients with acute thrombosis, but impaired fibrinolytic activity.[25, 26] The level of D-dimer decreases with initiation of heparin therapy, and may be reduced by two-thirds in patients on oral anticoagulants.[27] In addition, the time between onset of acute venous thromboembolism and sample collection may affect the concentration of D-dimer detected. D-dimer may be normal in the patients with venous thromboembolism of longer than 7 days. Therefore, even though the presence of D-dimer is specific for degradation product of cross-linked fibrin, cross-linked fibrin is not specific for venous thromboembolism. Because of their ubiquitous nature, D-dimer is generally not useful in confirming venous thromboembolism.

Several methods are available for measuring D-dimer levels. D-dimer assays typically use the monoclonal antibodies that bind to epitopes on the D-dimer fragment and the resulting complexes are detected by ELISA or agglutination techniques. In the classic enzyme-linked immunosorbent assay enzyme-linked immunoassay (ELISA) technique, anti-D-dimer antibody is immobilized to a solid surface, such as a well in a test plate. The plate is then incubated with the plasma of the patient and a second anti-D-dimer antibody linked to an enzyme such as peroxidase. When enzyme substrate is added, the resulting reaction can be qualitified by a color change that is measured at a specified wave length. Enzyme-linked immunosorbent assays provide the accurate and quantitative results. This assay have the ability to detect D-dimer in concentration of 30-80 ng/ml.[25] For detection of deep venous thrombosis, the standard ELISA has a sensitivity and specificity of 97% and 23% respectively.[10, 11] However, the sensitivity of the standard ELISA for diagnosing of distal deep venous thrombosis is only 78%.[28] For pulmonary embolism, the sensitivity is 91-93% with a specificity of 25-42%.[8, 9] Even though the more sensitive than other assays this test is that it takes several hours (4 hours) to perform and the ELISA kit is designed to be used in batches. The assays produced by different manufacturers are not always interchangeable because appropriate cutoff values for the diagnosis of venous thromboembolism vary. Depending on the specificity assay, thresholds from 40 ng/ml to more than 500 ng/ml have

been reported.[29] Thus, a D-dimer level lower than 500 ng/ml does not always rule out thrombosis. However, many studies report that 500 ng/ml is the standard cutoff for all ELISAs.[30] At present, there has been prompted the development of newer assays, still based on the ELISA methodology, which can be done in minutes. Those rapid ELISAs provide quick and quantitative results. The sensitivities have been reported similar to traditional ELISAs.[31]

The other technique to detect D-dimer fragments involves agglutination of latex beads or erythrocytes (Latex agglutination). The result can't be quantified, it is either considered positive or negative. The test can be completed in a few minutes. However, these assays are more often falsely negative compared with ELISAs. Furthermore, standardizing test results is problematic in as much as the reagents used may vary from manufacturer to manufacturer. The wide range reported sensitivities among latex agglutination assays limits their clinical utility for the diagnosis of venous thromboembolism. Another method that can also be performed in a few minutes is the immunofiltration. This method uses the same monoclonal anti-D-dimer antibody twice in the testing sequence with a gold tag placed on the second antibody. The Nycocard D-dimer assay uses this technique.

New rapid immunoturbidimetric techniques have also been developed. One of these, the Liatest assay uses a suspension of latex particles of submicron size coated with 2 complementary monoclonal anti –D-dimer antibodies. Plasma with reaction buffer is added to this suspension and a biochemical analyzer measures the change in optical absorption at a specified wavelength, which is proportional to the amount of D-dimer present. The device is calibrated once a week to specified controls. The advantage over the standard latex method is that the amount of D-dimer can be quantified. Sensitivity and specificity similar to traditional ELISAs have been reported for this test.[32]

Regarding to the cutoff level of D-dimer using standard ELISA for diagnosis of pulmonary embolism, it was demonstrated that at the cutoff level of 500 ng/l the sensitivity and specificity were 91-93% and 25-42% respectively.[8, 9] Pool data on sensitivity and specificity characteristics for various type of D-dimer assays in diagnosing venous thromboembolism was reviewed by Bounameaux H et al.[33] They reported that the sensitivity

and specificity of ELISA was 95-99% and 32-46%; 78-96% and 48-72% for Latex agglutination ;and 92-100% and 29-62% for rapid ELISA (Vidas DD).

To date, no D-dimer reference standard exists, making the inter-assay correlation poor. The sensitivity of D-dimer detection is dependent on both the assay method employed and the assay-specificity kit within that same assay method. Therefore, results from one assay can't be extrapolated to another.

Due to the poor specificity of D-dimer, this test should not be used for ruling in a diagnosis of venous thromboembolism. However, the sensitivity of D-dimer for venous thromboembolism is rather high; therefore, this test should be used to rule out this condition.

Thrombin-antithrombin complex (TAT) is an additional assay of thrombin generation and thrombin neutralization. The conversion of prothrombin into active thrombin is a key event within the coagulation cascade. Thrombin acts on various physiological substrates and is inhibited by antithrombin III; this results in an inactive proteinase/inhibitor complex, the concentrations of which can be measured quantitatively by enzyme immunoassay. The clinical significance of the determination of TAT is in the diagnosis of thrombotic events. Individuals predisposed to thrombosis, and disseminated intravascular coagulation (DIC) patients, are found to have elevated concentrations of TAT. [12] The determination of TAT is also of importance in the following groups of patients: patients with multiple trauma, promyelocytic leukemia, endotoxin infusion, liver dysfunction, septicemia[13] and preeclampsia.[14] In patients with malignant diseases, levels of TAT have been found to be raised in relation to the stage of the disease. Rises in TAT concentrations have been observed in the course of heparin and fibrinolytic therapy.[34] Because TAT is stable and has half-life of approximately 15 minutes[35], immunologic methods have been developed for its quantitation in plasma. The original radioimmunoassay used a liquid-phase double-antibody method[36], whereas the commercially available method is a solid-phase ELISA.[37] The difference of the two methods is that the range of normal values differs by a factor of 100: with the radioimmunoassay the average normal concentration is 2 nmol/l; with the immunoenzymatic method, it is around 0.02 nmol/l.

Atrial fibrillation and left atrial appendage dysfunction which was frequently detected in the patients with severe long-standing mitral stenosis were other factors leading to blood stasis in the left atrium and predisposing to thrombus formation and thromboembolism.[19-24, 38] In this condition, blood flow velocity within the left atrium is reduced because of changes in atrial contraction with the loss of sinus rhythm, dilatation of the chamber, and impairment of left atrial emptying. The latter may occur because of resistance to emptying in patients with mitral stenosis, or a similar phenomenon in patients with impaired left ventricular relaxation or compliance. The incidence of stroke related to thrombus in the patients with mitral stenosis and atrial fibrillation had increased 18-fold compared with matched control subjects and the risk is 2 to 7-fold greater in patients with mitral stenosis and atrial fibrillation than in those with normal sinus rhythm.[39]

The left atrial appendage can be imaged in some patients on routine transthoracic echocardiography (TTE) using a modified short axis parasternal view and by aiming for the pulmonary artery bifurcation and angling the transducer inferiorly and laterally. However, even when the appendage is visualized, the images are generally not of sufficient resolution to diagnose the presence of a thrombus in a reliable fashion. The use of transesophageal echocardiography (TEE) allows for high quality images of the left atrial appendage in virtually every patient. The details of the anatomy of the left atrial wall are readily obtainable. This is not usually achieved with TTE. In addition, TEE could detect the left atrial thrombus filling the appendage and extending into left atrial cavity in which thrombus could not be visualized from the chest wall by a good quality transthoracic echocardiogram.

Left atrial thrombi are nearly always mural. Very rarely, left atrial thrombi are free-floating and may be seen careening around the atrium as they are repelled by the mitral leaflets (particularly in mitral stenosis). In general intracavitary atrial thrombi are imaged in subjects with markedly reduced cardiac output or in patients with extreme atrial enlargement due to rheumatic metal stenosis, and most of these patients have atrial fibrillation. Intracavitary thrombi show some predilection for the superior atrial wall between the entrance points of pulmonary veins, although the thrombi on the interatrial septum could be found, as well as adjacent to the free wall of the atrium. Thrombi, if they are of recent

origin, have a homogeneous echo reflectance which is finely granular in appearance and similar to that of hepatic tissue. In general, atrial thrombi tend to be less echodense than those in the left ventricle. Chronicity causes organization of the thrombus with and increase in echodensity. Localization to the appendage appears to be related to several factors, the most important of which is its position at the periphery of flow.[40] Other investigators have reported a reduction in appendage ejection fraction in patients with left atrial thrombus. [41] Within the appendage, thrombi may be mural or intracavitary. Mural thrombi in the appendage tend to be elongated, flat structures aligned along the wall and wider than the pectinate muscle. Intracavitary appendage thrombi are often round, and they may fill the appendage. One previous study documented about the beneficial effect of warfarin to prevent the thromboembolism in the patients with mitral stenosis and atrial fibrillation. [45] Therefore, prophylactic anticoagulant agent appeared to reduce the incidence of stroke in patients with left atrial thrombus.

Endocardial injury of the left atrium might perhaps be an important factor in the pathogenesis of mural thrombus due to creating a thrombogenic surface which produced the tissue factor as well as losing the protective mechanisms of the cardiac surface such as platelet inhibitor, prostacyclin, the effects of anticoagulants such as protein C and antithrombin III, and the fibrinolytic enzyme, tissue plasminogen activator. Activated coagulation factors or platelets, therefore, could be generated on the damaged endocardium and thrombus formation may occur.

From the extensive literature review, there was only one report on the relationship between an abnormal increase level of D-dimer and the presence of left atrial thrombus in the patients with mitral stenosis. With the use of the level of D-dimer 300 ng/dl as the cut off point, the sensitivity and the specificity of D-dimer to detect the left atrial thrombus were 61.1% (95%CI 38.6% - 79.7%) and 93.3% (95%CI 82.1% - 97.7%) respectively.[16] However, the method for left atrial thrombus detection in this study was transthoracic echocardiography. Although, there was no comparative data between TAT and D-dimer for detection of left atrial thrombus (LAT) in these patients, these markers would be other

potential alternative diagnostic tools to detect the left atrial thrombus in the patients with mitral stenosis.

CHAPTER 3

RESEARCH METHODOLOGY

3.1 Research questions:

3.1.1 Primary research question

Could peripheral blood thrombin-antithrombin complex (TAT) and D-dimer (DD) levels be used as the diagnostic tool to detect the presence of left atrial thrombus in the patients with mitral stenosis?

3.1.2 Secondary research questions

1. Which one was better between TAT and D-dimer to detect the presence of left atrial thrombus in the patients with mitral stenosis?
2. What is the ability of the combination of TAT and D-dimer to detect the presence of left atrial thrombus in the patients with mitral stenosis?
3. What is the utility of TAT and D-dimer to detect the presence of left atrial thrombus in the patients with mitral stenosis?
4. What is the ability of D-dimer and TAT to detect the presence of left atrial thrombus in the patients with mitral stenosis and atrial fibrillation?

3.2 Objectives

Primary Objectives

To determine the diagnostic performance of the peripheral blood thrombin-antithrombin complex and D-dimer levels for detection of left atrial thrombus in the patients with mitral stenosis.

Secondary Objective

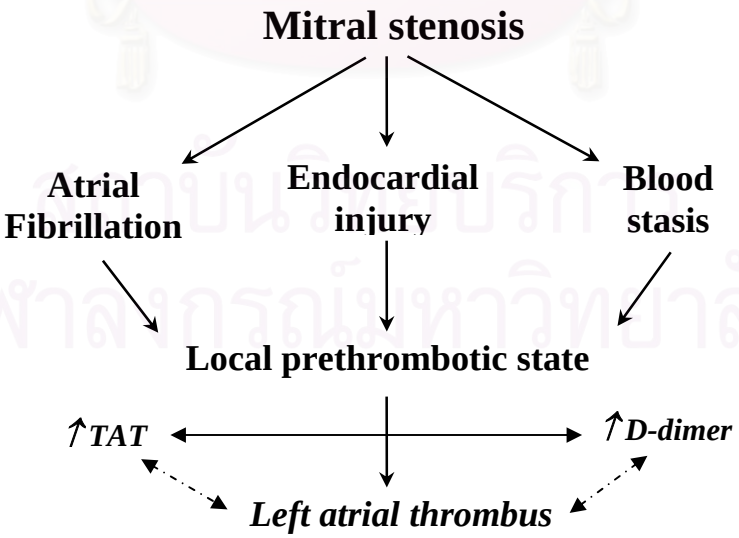
1. To determine the difference of the diagnostic performance between TAT and D-dimer.
2. To determine the ability of the combination of TAT and D-dimer for detection of left atrial thrombus in the patients with mitral stenosis
3. To determine the utility of TAT and D-dimer to detect the left atrial thrombus in the patients with mitral stenosis
4. To determine the diagnostic performance of D-dimer and TAT in detection of left atrial thrombus in the patients with mitral stenosis and atrial fibrillation?

3.3 Hypothesis:

Research hypothesis

Hypothesis: The sensitivity and the specificity of D-dimer and TAT for the diagnosis of left atrial thrombus in the patient with mitral stenosis were both 90%.

3.4 Conceptual framework:



3.5 **Keywords:** Mitral stenosis, left atrial thrombus, D-dimer, Thrombin-antithrombin complex

3.6 Operational definition:

Mitral stenosis:

Mechanical LV inflow obstruction with mitral valve area $\leq 2\ cm^2$

Thrombus:

Echo-dense mass with a clearly defined contour, smooth or irregular surface and clearly separated from endocardium

Left atrial appendage

A multi-lobulated structure in the left atrium predisposing to the left atrial thrombus formation

Left atrial spontaneous echocardiographic contrast

The dynamic swirling smoke-like echoes seen in the left atrium

3.7 Research design:

Cross-sectional study

3.8 Research method

3.8.1 Population

Target population

Adult patients with mitral stenosis

Study population

Adult patients with mitral stenosis underwent transthoracic echocardiography at Out-patient Department and Heart Clinic at Siriraj Hospital.

Inclusion criteria

1. Age > 12 years
2. Having mitral stenosis diagnosed by transthoracic echocardiography

Exclusion criteria

1. Inability to undergo transesophageal echocardiography such as esophageal obstruction (stricture or tumor), esophageal fistula or laceration or fistula, esophageal diverticulum, esophageal varices, recent esophageal or gastric surgery, upper gastrointestinal bleeding and unexplained symptoms of dysphagia or odynophagia and cervical spine instability
2. Concomitant moderate to severe mitral regurgitation or other valvular lesions that may have the impact to the prevalence of left atrial thrombus in the patients with mitral stenosis.
3. Continuing aspirin, other anti-platelet drugs and anticoagulants that may influence the value of D-dimer and TAT
4. Stopping aspirin, other anti-platelet drugs and anticoagulants that may influence the value of D-dimer and TAT less than 2 weeks
5. History of renal or liver disease, pulmonary embolism, deep vein thrombosis and malignancy diagnosed by standard technique. All these conditions may influence to the level of D-dimer and TAT

3.9 Sample size determination

Since this study was the cross sectional study (diagnostic test), the sample size estimation was calculated based on 95% confidence interval (CI) of sensitivity and specificity as follows:

$$n_1 = Z_{\alpha/2}^2 \times ss.(1-ss.) / dss^2$$

$$n_2 = Z_{\alpha/2}^2 \times sp.(1-sp.) / dsp^2$$

Where n_1 = number of the mitral stenosis patients with left atrial thrombus

n_2 = number of the mitral stenosis patients without left atrial thrombus

α = Probability of type I error = 0.05 (2 –sided)

ss = sensitivity = 0.9

sp = specificity = 0.9

dss = allowable error of sensitivity = 0.1

dsp = allowable error of specificity = 0.1

From the above formula

$n_1 = 35$ and $n_2 = 35$

From our previous data, the prevalence of left atrial thrombus, diagnosed by TEE, in patients with mitral stenosis was 30%. Therefore, 117 patients with mitral stenosis were recruited

3.10 Randomization:

None

3.11 Experimental maneuver

1. Transesophageal echocardiography

All patients underwent transesophageal echocardiography (TEE) with 3.75 MHz transducer of Hewlett Package System 5500. The procedure was done by the standard technique[6] to assess the left atrial thrombus, left atrial size, left atrial appendage function, left atrial area, left atrial spontaneous echo contrast (LASEC), mitral valve area and trans-mitral valve gradient.

A thrombus was diagnosed when an echo-dense mass with a clearly defined contour was seen within either the cardiac cavity or left atrial appendage or both. The surface of mass might be smooth or irregular.

TEE was used as the gold standard for the presence of left atrial thrombus in this study.

Echocardiographic interpretation

Two experienced cardiologists were independently reviewed and interpreted the echocardiography images, which were video tape-recorded, without the knowledge of any information of the patients and TAT and D-dimer results. When there were disagreements about the presence of left atrial thrombus between the two reviewers, the final conclusion was adjudicated by the third reviewer who experienced the transesophageal echocardiography more than 10 years.

Kappa statistic was used for evaluation of the inter-observer variation with 95 percent confidence as shown below:

			Cardiologist A	
			Thrombus	
			Present	*Absence*
Cardiologist B	Thrombus	*Present*	a	b
		Absence	c	d

Kappa index = $(P_0 - P_e) / (1 - P_e)$

Where P_0 = Observed agreement = $(a + d)/n$

P_e = Chance-expected agreement = $(R_1C_1 + R_2C_2)/n^2$

$R_1 = a+b$ $R_2 = c+d$

$C_1 = a+c$ $C_2 = b+d$

The 95% confidence interval of Kappa was determined by the following formula[43].

$$95\%CI \text{ of } \kappa = \kappa \pm 1.96 \times SE(\kappa)$$

2. Thrombin-antithrombin complex and D-dimer study

Ten milliliter of venous blood was drawn from ante-cubital vein on the same day of the trans-esophageal echocardiographic procedure for plasma thrombin-antithrombin complex and D-dimer levels determination. Enzyme immunoassay kits form DADE BEHRING (E'NOS TAT) was be used for thrombin-antithrombin complex. and D-dimer was measured by enzyme immunoassay. Both tests were done by separate experienced technicians who were blinded from another test result as well as transesophageal echocardiography results. The values of CV for D-dimer and TAT from our laboratory unit were 9.6 and 10.4 respectively.

3.12 Outcomes measurement

The following variables were measured:

3.12.1 Demographic and baseline variables

- Age (year)
- Gender
- Prior history of thromboembolism (%)
- Atrial fibrillation (%)
- Mitral valve area (cm^2)
- Left atrial size (mm)
- Left atrial area (mm)
- Right ventricular systolic pressure (mmHg)
- Left atrial spontaneous echo contrast (SEC) (%)

3.12.2 Outcome variables

- Left atrial thrombus
- Thrombin-antithrombin complex (μg/l)
- D-dimer (ng/ml)

3.13 Data Collection

The data was collected in a case record form.

3.14 Data analysis methods:

3.14.1 Demographic and baseline variables

For continuous variables, they were expressed as mean $\pm$ SD and the categorical data as percent. From the raw data, we could create the ROC curve from the SPSS program version 11 and MedCalc 7.3.0.1. Subsequently, we had to choose the optimal cut off point from the curve. Then, we obtained the best sensitivity and specificity of TAT and D-dimer to diagnose whether there was the thrombus in the left atrium of the patients with mitral stenosis. In addition, the combination of TAT and D-dimer were also further be analyzed to demonstrate whether the combination of tests would be beneficial more than separated test.

Another way to provide the diagnostic performance of TAT and D-dimer was to calculate likelihood ratio (LR) for each cut-off point as shown in the following table.

Level of TAT (ng/ml)	No of patients with thrombus (%)	No of patients without thrombus (%)	Likelihood Ratio
X	Y	Z	Y/Z
.....	.....	.....	.....
.....	.....	.....	.....
.....	.....	.....	.....
.....	.....	.....	.....

The likelihood ratio could be calculated by dividing the percentage of the patients with thrombus with the percentage of patients without thrombus for each cut-off value.

Subsequently, the sensitivity, specificity, positive predictive value and negative predictive value were further determined. We could determine the predictive value by the following formula.

$$\textbf{Positive predictive value} = \frac{\text{Sensitivity x Prevalence}}{\text{Sensitivity x Prevalence + (1-Specificity) (1-Prevalence)}}$$

$$\textbf{Negative predictive value} = \frac{\text{Specificity x (1- Prevalence)}}{\text{Specificity x (1- Prevalence) + (1-Sensitivity) Prevalence}}$$

The prevalence could be calculated by constructing the contingency 2 x 2 table as shown below:

		Thrombus		
		Present	*Absence*	
Test	*Positive*	*a*	*b*	a + b
	Negative	*c*	*d*	c + d
	Total	a + c	b + d	a+b+c+d

$$\textbf{Prevalence} = \frac{a + c}{a + b + c + d}$$

$$\textbf{Accuracy} = \frac{a + d}{a + b + c + d}$$

In summary, the results would be displayed as sensitivity, specificity and likelihood ratio for each cut-off value of TAT and D-dimer. The statistics for hypothesis testing in this study was Chi-square.

3.15 Ethical Consideration

In comparison to TEE in detection the presence of LAT, D-dimer and TAT tests are more convenient, less invasive and less expensive. The research proposal was approved by Institution Review Board of Faculty of Medicine Siriraj Hospital. Patients were thoroughly explained about the detail of the study before the informed consent was signed

3.16 Limitation of this study

The study was conducted in a university hospital, therefore the study population might be different from those in general hospitals especially the prevalence of left atrial thrombus in the patients with mitral stenosis. Therefore, the diagnostic performance of the D-Dimer and TAT in this study might not be accurately applied in other population who had a different prevalence of the left atrial thrombus. Moreover, the patients with asymptomatic deep vein thrombosis had high level of D-dimer rendering the increased false positive rate of D-dimer test.

3.17 Expected benefit and application

If D-dimer and TAT are accurate in detecting left atrial thrombus, these markers could be used in general hospitals without transesophageal echocardiography and could guide the physicians to prescribe anticoagulant properly especially in the patients with mitral stenosis who had normal sinus rhythm. In addition, we could save the budget due to the cost of this marker was less expensive than the TEE about 10 times.

3.18 Obstacles

Due to the large sample size of the study, the duration of the study lasted more than one year to complete the adequate recruitment.

CHAPTER 4

RESULTS

4.1 Demographic and baseline data

One-hundred and twenty patients diagnosed as having mitral stenosis were consecutively recruited from out patient department and heart clinic at Siriraj hospital from April 2003 to February 2004. Among these patients, 75% were females and 25% were males. The mean age was 42 years and 56.7% of the patients had atrial fibrillation. Regarding to the severity of mitral stenosis, 50.8% of cases was mild whereas moderate and severe cases were found in 43.3% and 5.8% respectively. The other baseline characteristics were shown in Table 1. The number of patients who had left atrial thrombus was 20; therefore, the prevalence of left atrial clot in our study was 16.6%. The Kappa index for the inter-observer variation of the interpretation of presence or absence of left atrial thrombus by trans-esophageal echocardiography was 1. The coefficient of variation (CV) of D-dimer and TAT from the laboratory were 6.28% and 7.11% respectively.

Table 1: Baseline characteristics of 120 patients with mitral stenosis

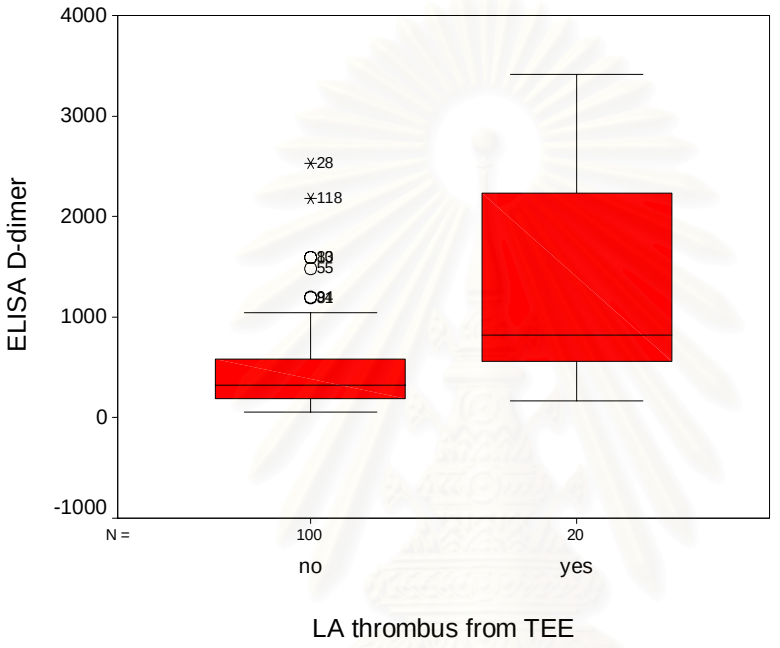
Characteristics	Value
Age (yr) (mean ± SD)	42.54 ± 11.14
Sex	
Male (%)	25
Female (%)	75
NYHA Functional Class (%)	
Class I	15

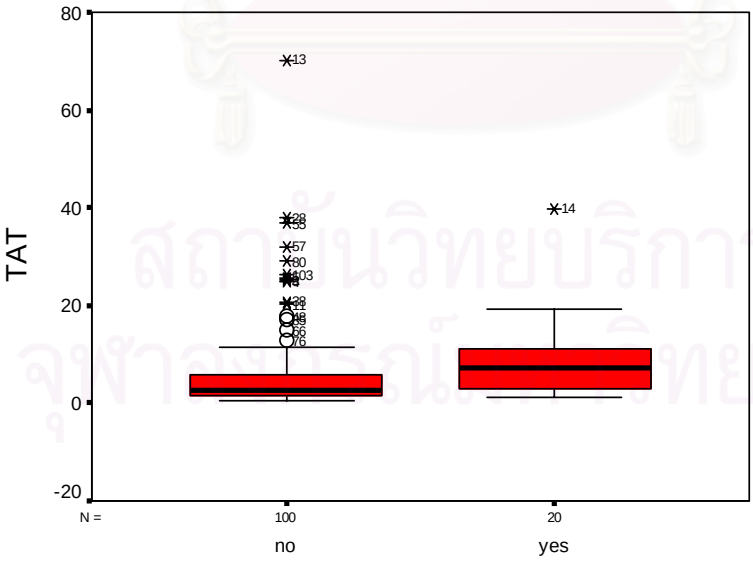
Class II	80
Class III	4.2
Class IV	0.8
Diabetes mellitus (%)	4.2
Hypertension (%)	2.5
Prior history of stroke (%)	15.8
Prior history of balloon mitral valvuloplasty (%)	35
Prior history of surgical mitral valvulotomy (%)	3.3
Atrial fibrillation (%)	56.7
Left atrial size (mm) (mean ± SD)	5.61 ± 0.87
Left atrial volume (mm^3) (mean ± SD)	131.08 ± 57.97
Left ventricular ejection fraction (%) (mean ± SD)	61.25 ± 8.54
Mitral valve score (mean ± SD)	8.58 ± 0.97
Mitral valve area (cm^2) (mean ± SD)	1.01 ± 0.30
Mean LA-LV gradient (mmHg) (mean ± SD)	10.43 5.24
Right ventricular systolic pressure (mmHg) (mean ± SD)	46.61 ± 15.77
LASEC (%)	98.3
Mitral regurgitation (%)	
Grade 0	41.7
Grade I	39.2
Grade II	19.2
Aortic regurgitation (%)	
Grade 0	56.7
Grade I	25.8
Grade II	17.5

The median of D-dimer in the patients with and without left atrial thrombus were 1331.06 and 457.06 ng/ml. respectively. The median of TAT in the patients with and without

left atrial thrombus were 7.22 and 2.59 μg/ml. respectively. The distribution of D-dimer and TAT according to the presence or absence of left atrial thrombus was shown in Figure 1.

Figure1. Distribution of D-dimer and thrombin-antithrombin complex according to the presence or absence of left atrial thrombus





4.2 Primary outcome analysis

The ability of D-dimer and TAT to identify the left atrial thrombus in the patients with mitral stenosis was assessed with ROC analysis (figure 2). The mean area under the ROC curve for D-dimer and TAT were 0.808 (95%CI, 0.726 - 0.874) and 0.683 (95%CI, 0.592 - 0.765) respectively. The difference between these areas was 0.125 (95%CI, 0.003-0.248, p = 0.045). The sensitivity, specificity, predictive value and likelihood ratio of both tests were demonstrated in Table 2 and 3.

Figure 2. ROC curves of D-dimer and TAT in all patients (n=120)

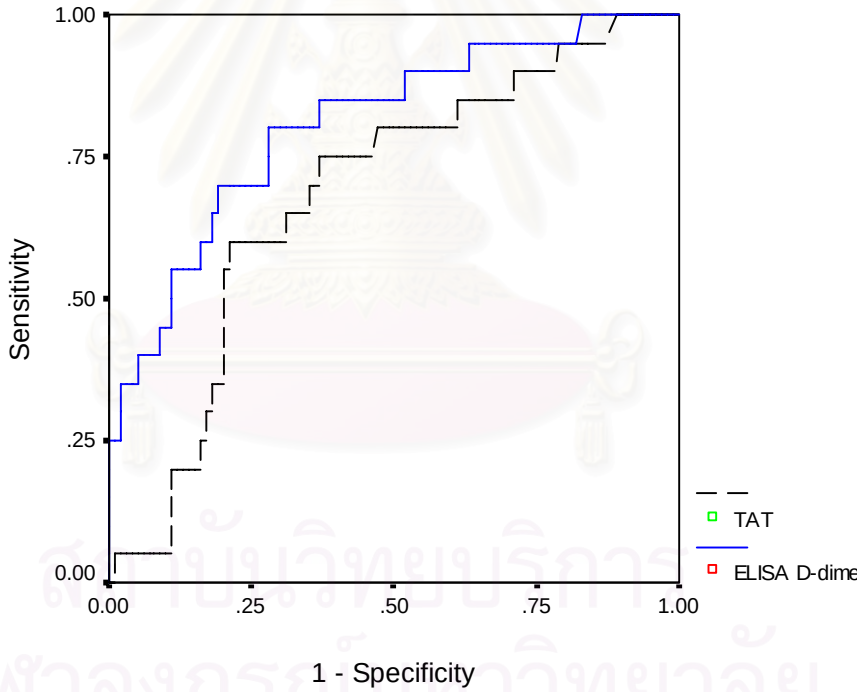


Table 2: Sensitivity, specificity, predictive values, accuracy and likelihood ratio of D-dimer for detection of left atrial thrombus in the patients with mitral stenosis

Level (ng/ml)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)	LR+	LR-
55	100	0	17	0	17	1	NA
100	100	5	17.7	100	21.15	1.06	0
150	100	12	18.8	100	26.96	1.14	0
200	95	27	21	96.3	38.56	1.30	0.19
250	90	37	22.6	94.7	46.01	1.43	0.27
300	90	46	25.4	95.7	53.48	1.67	0.22
350	85	55	27.8	94.7	60.10	1.89	0.27
400	85	60	30.3	95.1	64.25	2.13	0.25
450	80	65	31.8	94	67.55	2.29	0.31
500	80	66	32.5	94.1	68.38	2.25	0.30
550	75	72	35.4	93.3	72.51	2.68	0.35
600	70	77	38.3	92.6	75.81	3.04	0.39
650	65	82	42.5	91.9	79.11	3.61	0.43
700	60	83	41.9	91	79.09	3.53	0.48
750	55	85	42.8	90.2	79.90	3.67	0.53
800	55	89	50.5	90.6	83.22	5	0.51
1000	40	91	47.6	88.1	82.33	4.44	0.66
1200	40	95	62.1	88.5	85.65	8	0.63
1800	35	98	78.1	88	87.29	17.5	0.66
2000	25	98	71.9	86.4	85.59	12.5	0.77
2500	25	99	83.6	86.5	86.49	25	0.76
3000	10	100	100	84.4	84.70	NA	0.90

Table 3: Sensitivity, specificity, predictive value and accuracy of TAT for detection of left atrial thrombus in the patients with mitral stenosis

Level (μg/l)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)	LR+	LR-
1	100	4	17.58	1	20.32	1.04	0
1.5	95	21	19.76	95.35	33.58	1.20	0.24
2	85	39	22.20	92.70	46.82	1.39	0.38
2.5	80	49	24.32	92.29	54.27	1.57	0.41
3	75	60	27.75	92.14	62.55	1.88	0.42
3.5	70	65	29.06	91.36	65.85	2	0.46
4	65	69	30.04	90.59	68.32	2.09	0.52
6	60	78	35.84	90.49	74.94	2.73	0.52
10	30	82	25.45	85.12	73.16	1.67	0.85
15	20	87	23.96	84.15	75.61	1.54	0.92
20	5	89	8.52	82.06	74.72	0.45	1.06
30	5	96	20.38	83.15	80.53	1.25	0.99
40	0	97	0	82.86	82.17	0	1.01

When D-dimer and TAT values were chosen as range, the calculated sensitivity, specificity, predictive value and likelihood ratio are shown in table 4 and 5

Table 4: Sensitivity, specificity, predictive value and likelihood ratio of D-dimer at the cut –off point in range for detection of left atrial thrombus in the patients with mitral stenosis

Level (ng/ml)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Likelihood Ratio
55-100	0	95	0	82.57	0
100-150	0	93	0	82.27	0
150-200	5	85	6.26	81.69	0.33
200-250	5	90	9.11	82.53	0.50
250-300	0	91	0	81.95	0
300-400	5	86	6.68	81.87	0.36
400-500	5	94	14.32	83.15	0.83
500-600	10	89	15.42	83.14	0.91
600-700	10	94	25.01	83.90	1.67
700-800	5	94	14.32	83.15	0.83
800-1000	15	98	60.06	85.19	7.5
1000-1200	0	96	0	82.72	0
1200-1800	5	97	25.05	83.59	1.67
1800-2000	10	0	1.97	0	NA
2000-2500	0	99	0	83.16	99
>2500	25	99	83.37	86.81	99

Table 5: Sensitivity, specificity, predictive value and likelihood ratio of TAT for detection of left atrial thrombus in the patients with mitral stenosis

Level (μg/l)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Likelihood Ratio
≤ 1	0	96	0	82.72	0
1-1.5	5	83	5.57	81.34	0.29
1.5-2	10	82	10.02	81.96	0.56
2-2.5	5	90	9.11	82.53	0.5
2.5-3	5	89	8.35	82.37	0.45
3-3.5	5	95	16.70	83.30	1
3.5-4	5	96	20.04	83.45	1.25
4-6	5	91	10.02	82.69	0.56
6-10	30	96	60.06	87.25	7.5
10-15	10	95	28.62	84.04	2
15-20	15	98	60.06	85.19	7.5
>20	5	89	8.35	82.37	0.45

4.2 Secondary outcome analysis

If D-dimer and TAT were combined in the way of parallel test, the sensitivity and specificity could be calculated as shown in table 6.

Table 6: Diagnostic performance of D-dimer and TAT in parallel test (n=120)

Test	Sensitivity	Specificity
D-dimer ($\leq$ 500 ng/ml)	0.8	0.66
TAT ($\leq$ 3 μg/l)	0.75	0.60
D-dimer or TAT positive	0.95	0.40
D-dimer and TAT positive	0.60	0.86
D-dimer and TAT negative	0.05	0.60

In another way, if these tests were used as the way of serial test, the positive predictive value (if both tests were positive) was 47%.

In patients with atrial fibrillation, the ROC curves of D-dimer and TAT were shown in figure 3. The mean areas under the ROC curve for D-dimer and TAT were 0.903 (95%CI, 0.806 - 0.961) and 0.739 (95%CI, 0.618 - 0.838) respectively. The difference between these areas was 0.163 (95%CI, 0.029 - 0.298, p = 0.017). The sensitivity, specificity, predictive value and likelihood ratio of both tests were demonstrated in Table 7 and 8.

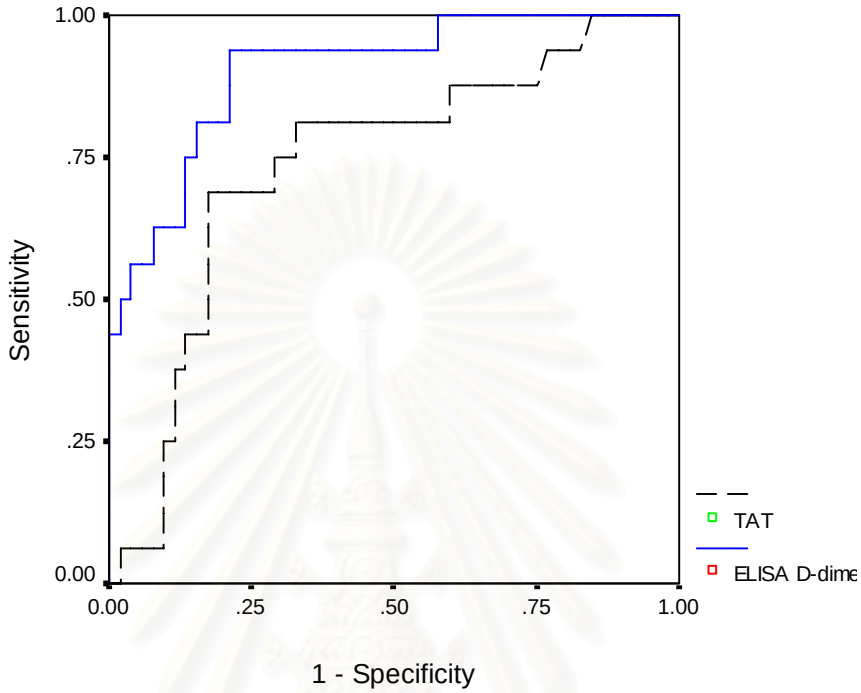
Table 7: Sensitivity, specificity, predictive value and accuracy of D-dimer for detection of left atrial thrombus in the patients with mitral stenosis and atrial fibrillation (n=68)

Level (ng/ml)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)	LR+	LR-
55	100	0	0	NA	17	1	NA
100	100	9.6	18.15	1	24.97	1.11	0
150	100	19.2	19.87	1	32.94	1.24	0
200	93.8	42.3	25.60	97	51.06	1.63	0.15
250	93.8	42.3	24.60	97	51.06	1.63	0.15
300	93.8	51.9	28.10	97.66	59.02	1.95	0.12
350	93.8	63.5	34.00	98.08	68.65	2.57	0.09
400	93.8	67.3	36.51	98.19	71.81	2.87	0.09
450	93.8	71.2	39.50	98.28	75.04	3.26	0.09
500	93.8	73.1	41.14	98.33	76.62	3.49	0.08
550	87.5	78.8	45.27	96.92	80.28	4.13	0.16
600	81.3	80.8	45.91	95.57	80.89	4.24	0.23
650	75	86.5	52.69	94.53	84.55	5.56	0.29
700	68.8	86.5	50.53	93.26	83.49	5.10	0.36
750	62.5	86.5	48.13	92.00	82.42	4.63	0.43
800	62.5	92.3	61.93	92.47	87.23	8.12	0.41
1000	50	96.2	72.50	90.56	88.35	13.16	0.52
1200	50	98.1	84.06	90.73	89.92	26.32	0.51
1800	43.8	100	100	89.87	90.45	NA	0.56
2000	31.3	100	100	87.89	88.32	NA	0.69
2500	31.3	100	100	87.89	88.32	NA	0.69
3000	12.5	100	100	85.08	85.13	NA	0.88

Table 8: Sensitivity, specificity, predictive value and accuracy of TAT for detection of left atrial thrombus in the patients with mitral stenosis and atrial fibrillation (n=68)

Level (μg/l)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)	LR+	LR-
1	100	5.8	17.55	1	21.81	1.06	0
1.5	93.8	23.1	19.65	94.89	35.11	1.22	0.27
2	87.5	39.2	22.39	93.99	47.41	1.44	0.32
2.5	81.3	48.1	23.89	92.76	53.74	1.57	0.39
3	81.3	63.5	30.87	94.42	66.52	2.23	0.29
3.5	75	67.3	31.49	93.06	68.60	2.29	0.37
4	73.3	71.2	33.78	93	71.55	2.54	0.38
6	68.8	80.8	41.80	92.81	78.76	3.58	0.39
10	37.5	86.5	35.76	87.34	78.17	2.78	0.72
15	25	88.5	30.35	85.47	77.70	2.17	0.85
20	6.3	90.4	11.62	82.79	76.10	0.66	1.04
30	6.3	96.2	14,09	83.08	77.68	0.82	1.02
40	0	98.1	24.94	83.66	80.91	1.66	0.97

Figure 3. ROC curve of D-dimer and TAT in patients with atrial fibrillation (n=68)



To investigate about the utility of D-dimer and TAT for detection of the left atrial thrombus in the patients with mitral stenosis, a clinical decision analysis model was done. The sensitivity and specificity of D-dimer and TAT to detect left atrial thrombus in this study were 80%, 66%, 75% and 60% respectively. The utility of each outcome of these tests were as following:

Test positive and clot presence	(*True positive* or clot diagnosed)
Test positive and no clot	(*False positive* or clot misdiagnosed)
Test negative and clot presence	(*False negative* or clot missed)
Test negative and no clot	(*True negative* or clot excluded)

Initially, we ranked the outcomes from best to worst as shown below:

Best

1. **Clot excluded:** The patient was free of left atrial thrombus and had not been labeled.

2. Clot misdiagnosed: The patient was free of left atrial thrombus but had been labeled and must suffer from the risk of warfarin therapy which could increase the incidence of serious bleeding and intracerebral bleeding.

3. Clot diagnosed: The patient had left atrial clot and knows it, but it could benefit from warfarin therapy which was greater than the risk from serious bleeding.

Worst

4. Clot missed: The patient had left atrial clot but it had been missed and he would not receive the benefit from warfarin therapy.

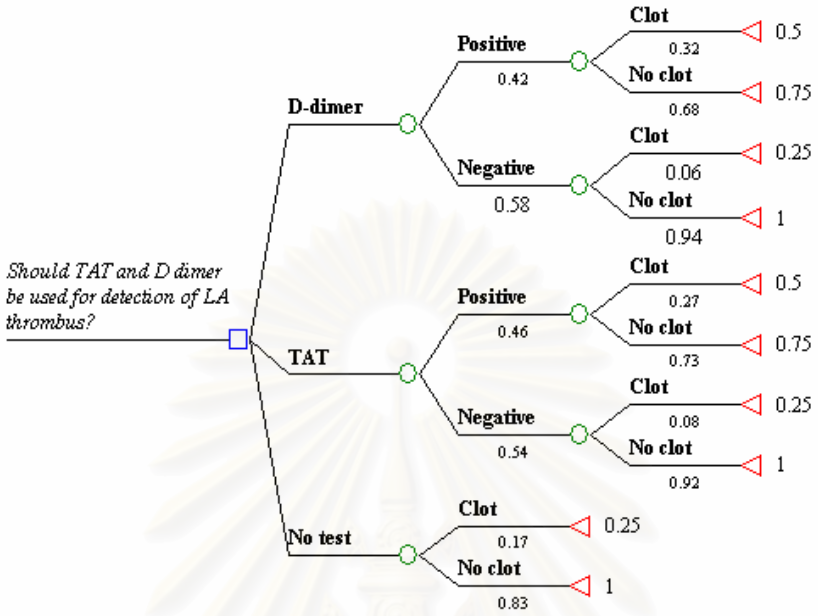
We could determine these utilities from the patient's perspective by the method of standard gamble. In this case, suppose we used this method and the utility of each outcome were shown as the followings:

Clot excluded	=	1
Clot misdiagnosed	=	0.75
Clot diagnosed	=	0.5
Clot missed	=	0.25

Analysis and interpretation for clinical decision analysis

This analysis was carried out on the basis of patient's perspective. After critical review of the literature, the decision tree was performed and the path probability was subsequently calculated as demonstrated in figure 4.

Figure 4. Clinical decision tree of D-dimer and TAT (Model 1)



From the information described above:

The sensitivity and specificity of D-dimer was 80% and 75% respectively

The sensitivity and specificity of TAT was 66% and 60% respectively

The prevalence of left atrial thrombus was 17%

For the utility outcome:

Clot excluded (True Negative) = 1

Clot misdiagnosed (False Positive) = 0.75

Clot diagnosed (True Positive) = 0.5

Clot missed (False Negative) = 0.25

From the decision tree:

The expected value of path A is 0.8353

The expected value of path B is 0.8216

The expected value of path C is 0.8725

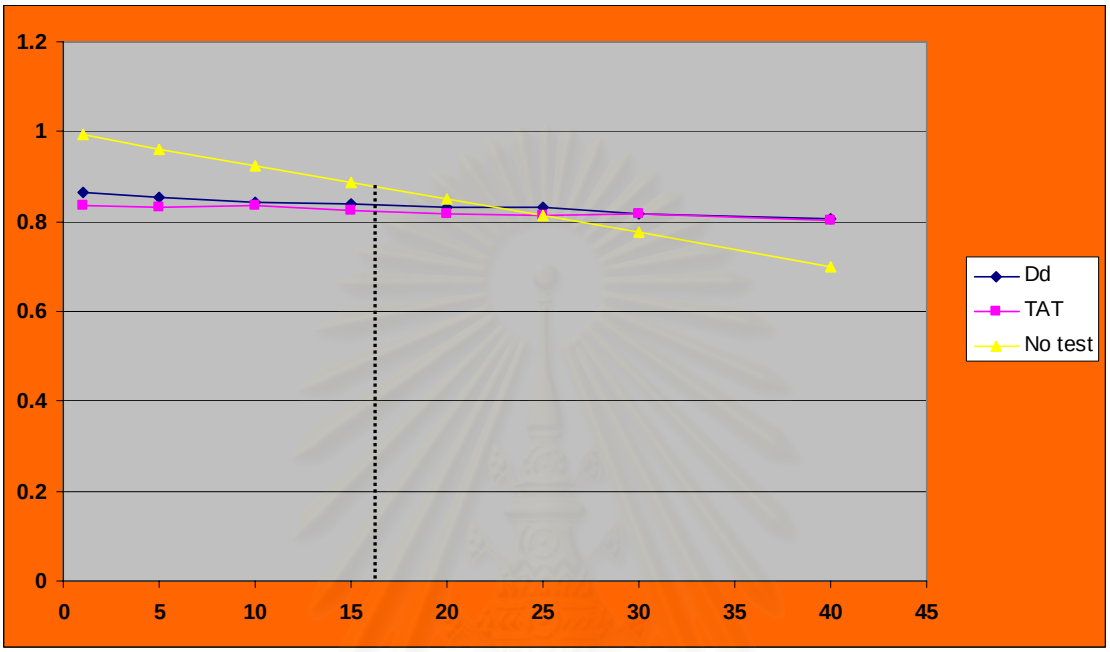
Sensitivity Analysis

We tried to challenge the vulnerability of our model decision by performing the sensitivity analysis. We varied the prevalence of left atrial thrombus from 1-40% and recalculated the expected utility and displayed in Table 9 and figure 5.

Table 9: Expected utility of D-dimer, TAT and no test for a given prevalence of left atrial thrombus (Model 1)

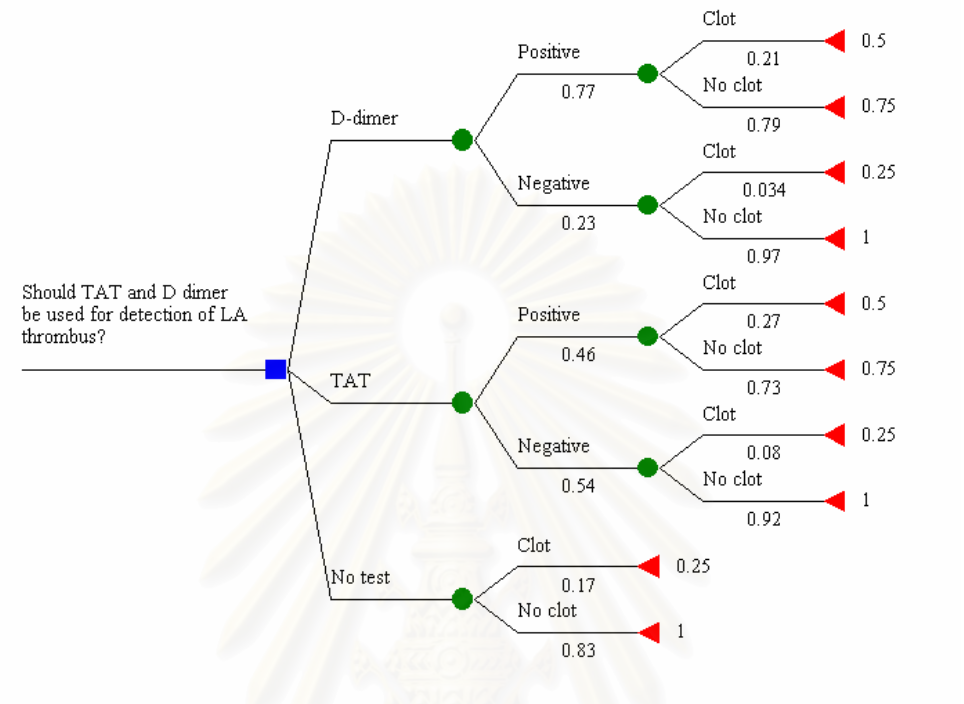
Prevalence of left atrial thrombus	D-dimer	TAT	No test
	A	B	C
1	0.8648	0.8370	0.9925
5	0.8524	0.8318	0.9625
10	0.8438	0.8361	0.9250
15	0.8381	0.8241	0.8875
20	0.8324	0.8189	0.8500
25	0.8334	0.8138	0.8125
30	0.8182	0.8180	0.7750
40	0.8068	0.8009	0.7000

Figure 5. Sensitivity analysis assessing the effect of changes in the prevalence of left atrial thrombus in the patients with mitral stenosis (Model 1)



However, if the cut-off point of $\leq$ 200 ng/ml was used, the sensitivity and specificity of D-dimer were 95% (95%CI, 91.1% - 99%) and 27% (95%CI, 23% - 31.1%) respectively. The sensitivity and specificity of TAT was similar the first model, that was 66% and 60% respectively. Then, we could make another model of decision tree as in Figure 6.

Figure 6. Clinical decision tree of D-dimer and TAT (Model 2)



From the decision tree:

The expected value of path A was 0.7620

The expected value of path B was 0.8216

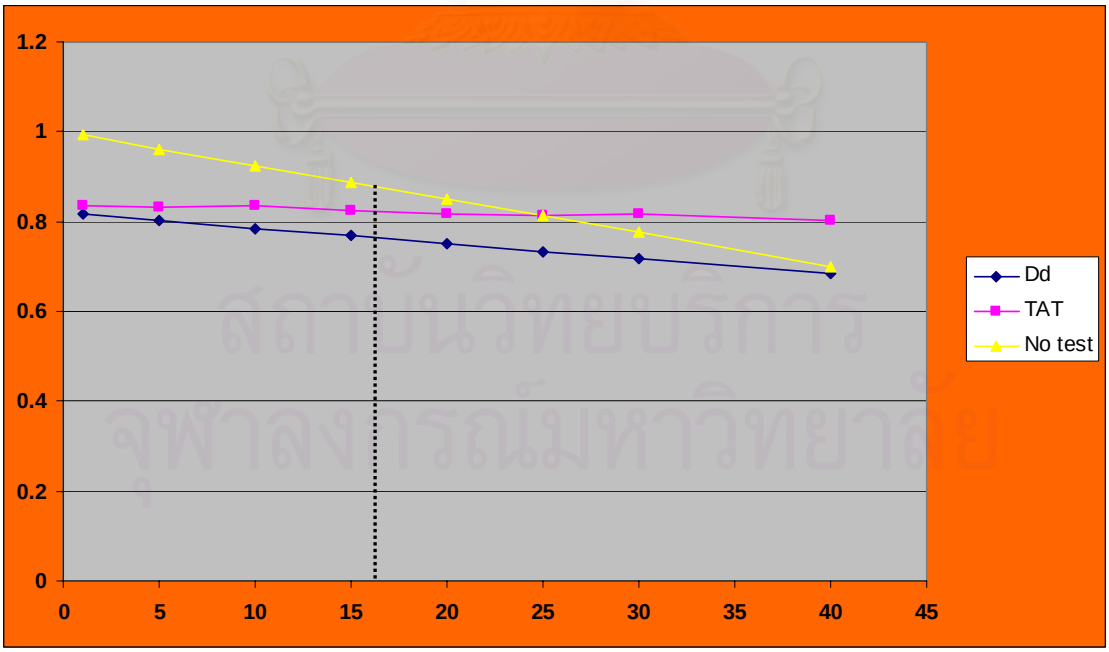
The expected value of path C was 0.8725

We tried to challenge of the vulnerability of the second model by varying the prevalence of left atrial clot as the first model and the result was shown in table 10 and figure 7

Table 10: Expected utility of D-dimer, TAT and no test for a given prevalence of LA thrombus (Model 2)

Prevalence of LA clot	D-dimer	TAT	No test
	A	B	C
1	0.8155	0.8370	0.9925
5	0.8019	0.8318	0.9625
10	0.7843	0.8361	0.9250
15	0.7685	0.8241	0.8875
20	0.7513	0.8189	0.8500
25	0.7338	0.8138	0.8125
30	0.7175	0.8180	0.7750
40	0.6878	0.8009	0.7000

Figure7. Sensitivity analysis assessing the effect of changes in the prevalence of left atrial thrombus in the patients with mitral stenosis (Model 2)



Test-threshold and test-treatment threshold

To analyze the test threshold and test-treatment threshold of model 1, the following variables were defined as below:

For D-dimer

True positive rate = 0.8

False positive rate = 0.34

False negative rate = 0.2

True negative rate = 0.66

For TAT

True positive rate = 0.75

False positive rate = 0.28

False negative rate = 0.25

True negative rate = 0.72

Test-Threshold

The utility that we measured was mortality. From the literature, it was found that the mortality in the patients with mitral stenosis who received the anticoagulant reduced 16.7% [46] when compared with those without anticoagulant treatment. The mortality from anticoagulant due to serious bleeding was 0.3% (INR = 2-3) [47]. No excess risk of death from the tests was considered.

Test-Threshold for D-dimer

$$= \frac{\text{(FP rate) (risk of inappropriate Rx) + (risk of diagnostic test)}}{\text{(FP rate) (risk of inappropriate Rx) + (TP rate) (benefit of appropriate Rx)}}$$

From this formula, the test-threshold $= \frac{(0.34)\,(0.003) + (0)}{(0.34)\,(0.003) + (0.8)\,(0.167)}$

$= 0.0076$

Test-Treatment Threshold

$$= \frac{\text{(TN rate) (risk of inappropriate Rx) - (risk of diagnostic test)}}{\text{(TN rate) (risk of inappropriate Rx) + (FN rate) (benefit of appropriate Rx)}}$$

From this formula, the test-treatment threshold for D-dimer

$$= \frac{(0.66)\,(0.003) - (0)}{(0.66)\,(0.003) + (0.2)\,(0.167)} = 0.559$$

In the similar way, we could also determine the test-threshold and test-treatment threshold of TAT which is 0.0067 and 0.1695, respectively.. For the second model, the test threshold and test treatment threshold for D-dimer were 0.0136 and 0.0884

CHAPTER 5

DISCUSSION, CONCLUSION, AND RECOMMENDATION

5.1 Discussion

From this study, it was shown that D-dimer and TAT could be used as the markers for detection of left atrial thrombus in the patients with mitral stenosis. The area under the ROC curve of D-dimer and TAT were 0.808 (95%CI, 0.726 - 0.874) and 0.683 (95%CI, 0.592 - 0.765). Regarding to the test performance, D-dimer was better than TAT by the significant difference of area under the ROC curve of both agents as demonstrated in figure 1. The optimal sensitivity and specificity of D-dimer at the cut-off point of $\leq$ 500 ng/ml were 80% (CI, 76.35-83.65) and 66% (CI, 61.68-70.32) respectively (table 2). For TAT, the optimal sensitivity and specificity at the cut-off point of $\leq$ 3 μg/l were 75% (CI, 71.05-78.95) and 60% (CI, 55.65-64.35). This finding was different from the previous study [16] that showed the sensitivity and specificity of D-dimer at the same cut-off point was 61.1% (95%CI, 38.6-79.7) and 93.3% (95%CI, 82.1-97.7). The possible explanations might be related to the small sample size of the previous study (n=63), the difference in the study population and the prevalence of left atrial thrombus. In our study, the prevalence of left atrial thrombus was 16.6%, most of the patients (50.8%) were mild to moderate mitral stenosis and 56.7% had atrial fibrillation whereas 62.2% of patients in that study had atrial fibrillation, whereas, the prevalence of left atrial thrombus and atrial fibrillation in the study of Yasaka et al was 28.6% and 62.2% respectively.16 Finally, the most important difference between these two studies was the method for detection of left atrial thrombus. Our study used TEE to detect the left atrial clot with the sensitivity and specificity of 100% and 99% [6] respectively but TTE which had the sensitivity and specificity to detect left atrial thrombus of 33-59% and 90% [18, 45, 46] was used in the study of Yasaka M et al.

Table 1 and 2 demonstrated that at the optimal cut off points of $\leq$ 500 ng/ml, the positive predictive value of D-dimer was only 32.5% but the negative predictive value was as high as 94.1%. The positive likelihood ratio and negative likelihood ratio were 2.25 and 0.30 respectively. With these results, D-dimer could be considered as a tool to rule out the left atrial thrombus in the patient with mitral stenosis.

Regarding to the utility of D-dimer and TAT, two models of clinical decision tree were created. For the first model (figure 4), the highest expected value of decision tree was in path C. Therefore, it was suggested that we should not use D-dimer and TAT for detection of left atrial thrombus in the patient with mitral stenosis if the prevalence of left atrial clot in these patients was 17%. However, when sensitivity analysis (figure 5 and table 9) was done, it could be concluded that if the prevalence of the left atrial thrombus in the patients with mitral stenosis is $\geq$ 25%, the best way is to perform the D-dimer test and if the prevalence is $\leq$ 25%, no any test is preferred.

For the second model (figure 5), the highest expected value of decision tree was still in path C. Therefore, we should not use D-dimer and TAT for detection of left atrial thrombus in the patient with mitral stenosis if the prevalence of left atrial clot in these patients was 17%. When considering about the sensitivity analysis as shown in figure 7 and table 10, we concluded that if the sensitivity and sensitivity of D-dimer at the cut-off point $\leq$ 200 ng/ml were 95% (95%CI, 91.1% - 99%) and 27% (95%CI, 23% - 31.1%) respectively, whereas the sensitivity and specificity of TAT was similar the first model, (sensitivity and specificity were 66% and 60% respectively), D-dimer should not be used as a tool for detection of left atrial thrombus regardless of the prevalence of left atrial thrombus but TAT could be considered if the prevalence of left atrial thrombus $\geq$ 25%.

When the test-threshold and test-treatment threshold for the first model were calculated, it was demonstrated that if the prevalence of left atrial thrombus was more than 0.1%, we should perform the D-dimer to determine the presence of absence of left atrial thrombus. When the value of test-treatment threshold was considered, it could be concluded that if the prevalence of left atrial thrombus was more than 56%, we should treat

the patients without performing the D-dimer test, but if the prevalence was less than 56%, we should perform the test to determine whether there was the left atrial thrombus before prescribing the treatment.

For TAT, the test-threshold and test-treatment threshold were 0.0067 and 0.1695 respectively. Therefore, it could be concluded that if the prevalence of left atrial thrombus was more than 0.1%, D-dimer should be done and if the prevalence of left atrial thrombus was more than 17%, TAT should be also performed to determine whether there was the left atrial thrombus before treating the patients with anticoagulants.

For the second model, the test-threshold and test-treatment threshold of D-dimer were 0.0136 and 0.0884 respectively. It was therefore concluded that if the prevalence of left atrial thrombus was more than 1.4%, we should perform the D-dimer test to determine the presence of absence of left atrial thrombus. If the prevalence of left atrial thrombus was more than 8.8%, we should treat the patients without performing the test, but if the prevalence was less than 8.8%, we should perform the test to determine whether there was the left atrial thrombus before prescribing the treatment.

When D-dimer and TAT were used in combination in either parallel or serial test, it was found that the diagnostic performance was not better than a single test as demonstrated by table 6. Therefore, combination of these tests was not recommended because it increased the cost without improving the diagnostic performance.

Looking at the subgroup with atrial fibrillation (n= 68), it was demonstrated that the sensitivity and specificity of D-dimer and TAT for detection of left atrial thrombus was superior to those of all patients which included the population with and without atrial fibrillation as shown in Figure 3 and table 7 and 8. For D-dimer, the sensitivity, specificity, positive predictive value and negative predictive value at the cut-off point of $\leq$ 500 ng/ml were 93.8%, 73.1%, 41% and 98.3% respectively. The sensitivity, specificity, positive predictive value and negative predictive value of TAT at the cut-off point of $\leq$ 3 μg/l were 81.3%, 63.5%, 30.87% and 94.4% respectively. The improvement of the test performance of D-dimer and TAT in this subgroup might be related to the higher prevalence of left atrial

thrombus (23.5%). However, this analysis consisted only in small sample size (n=68). The study with large sample size should be performed to confirm this result.

5.2 Conclusion

This study demonstrates that D-dimer and TAT could be used to detect the presence or absence of left atrial thrombus in the patient with mitral stenosis with the sensitivity and specificity of 80% and 66% for D-dimer and 75% and 60% for TAT. In the subgroup with atrial fibrillation, these tests provide a better diagnostic performance than those with normal sinus rhythm.

5.3 Recommendation

Based on the results of the study, it is recommended that D-dimer can be used as a simple tool for detection of left atrial thrombus in the patients with mitral stenosis. The negative predictive value of D-dimer test at the cut-off point of $\leq$ 500 ng/ml is 94.1% and at the cut-off point of $\leq$ 200 ng/ml is 96.3%. Therefore D-dimer could be also used to rule out the presence or absence of left atrial thrombus in the patients with mitral stenosis.

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APPENDICES

APPENDIX 1

Informed Consent Form and Patient Information

หนังสือยินยอมโดยได้รับการอธิบายบอกกล่าว

(Informed Consent Form)

วันที่.....เดือน.....พ.ศ.....

ข้าพเจ้า.....อายุ.....ปี

อาศัยอยู่บ้านเลขที่.....ถนน.....แขวง.....เขต.....

อำเภอ.....จังหวัด.....โทรศัพท์.....

ขอแสดงเจตนายินยอมเข้าร่วมโครงการวิจัยเรื่อง Thrombin-antithrombin complex and d-dimer for detection of left atrial thrombus in the patients with mitral stenosis

(ภาษาไทย) การศึกษาการใช้ดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็ก ในการวินิจฉัยลิ่มเลือดในห้องหัวใจเอเตรียมซ้ายของผู้ป่วยลิ้นไมตรัลตีบ

โดยข้าพเจ้าได้รับทราบรายละเอียดของดังต่อไปนี้

ชื่อผู้วิจัย: นพ. เรวัตร พันธุ์กิ่งทองคำ

คุณวุฒิ พบ., ววอ. อายุรศาสตร์, ววอ. อายุรศาสตร์หัวใจ

ตำแหน่งวิชาการผู้ช่วยศาสตราจารย์ ระดับ 8

ภาควิชา อายุรศาสตร์ โทร. 0 2419 7745-6 หรือ 01-8142943

สถานที่ทำวิจัย สาขาหทัยวิทยา ภาควิชาอายุรศาสตร์

ศูนย์โรคหัวใจสมเด็จพระบรมราชินีนาถ

ภาควิชาพยาธิวิทยาคลินิก

ผู้สนับสนุนการวิจัย คณะแพทยศาสตร์ศิริราชพยาบาล

ความเป็นมาของโครงการ

โครงการนี้เกิดจากปัญหาที่ผู้วิจัยต้องการหาวิธีใหม่ในการตรวจหาลิ่มเลือดในห้องหัวใจบนซ้าย โดยไม่ต้องใช้อุปกรณ์การตรวจคลื่นสะท้อนหัวใจชนิดที่ต้องตรวจทางหลอดอาหารของผู้ป่วย เนื่องจากการตรวจด้วยวิธีดังกล่าวทำให้ต้องเสียค่าใช้จ่ายสูงและอาจทำให้ผู้ป่วยรู้สึกไม่ค่อยสบายได้

วัตถุประสงค์ของการวิจัย

เพื่อศึกษาการใช้สารดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็กจากเลือดของผู้ป่วยในการวินิจฉัยลิ่มเลือดในห้องหัวใจเอเตรียมซ้ายในผู้ป่วยลิ้นไมตรัลตีบ

รายละเอียดที่จะปฏิบัติต่อผู้เข้าร่วมโครงการ

ผู้เข้าร่วมโครงการวิจัยทุกคนจะได้รับการเจาะเลือดประมาณ 10 มิลลิลิตร เพื่อนำไปตรวจหาสารดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็ก นอกจากนี้ในวันเดียวกันผู้ป่วยจะได้รับการตรวจคลื่นสะท้อนหัวใจทางหลอดอาหารด้วย สำหรับการตรวจวิธีหลังนี้ผู้ป่วยจะต้องงดน้ำและอาหารหลังเที่ยงคืนของวันก่อนที่จะทำการตรวจรวมทั้งต้องหยุดยาละลายลิ่มเลือดและยาต้านเกร็ดเลือดประมาณ 10 วัน (เฉพาะผู้ป่วยที่กำลังรับประทานยาดังกล่าวอยู่) เพื่อที่จะไม่กระทบกระเทือนในการแปรผลของสารดีไดเมอร์ และ ทรอมบิน-แอนติทรอมบิน คลอมเพล็ก นอกจากนี้ผู้ป่วยจะต้องไม่มีข้อห้ามต่อการตรวจด้วยวิธีการกลืนสายทางหลอดอาหารด้วย

ในวันที่ทำการตรวจคลื่นสะท้อนหัวใจทางหลอดอาหาร แพทย์จะพ่นยาชาในคอของผู้ป่วยก่อนเพื่อไม่ให้เกิดความรู้สึกไม่สบายระหว่างการตรวจ หลังจากนั้นจะใส่สายตรวจผ่านเข้าไปในหลอดอาหารเพื่อตรวจวินิจฉัยว่ามีลิ่มเลือดในห้องหัวใจหรือไม่ ลิ้นหัวใจต่างๆมีการทำงานเป็นอย่างไร โดยการตรวจกินเวลานานประมาณ 10-15 นาที โดยขณะทำการตรวจจะมีทั้งแพทย์และพยาบาลอยู่กับผู้ป่วยตลอดเวลา ในการนี้ผู้เข้าร่วมวิจัยจะได้รับการดูแลจากแพทย์ผู้เชี่ยวชาญอย่างต่อเนื่องและไม่เสียค่าใช้จ่ายในการตรวจคลื่นสะท้อนหัวใจทางหลอดอาหารรวมทั้งค่าตรวจเลือดเพื่อหาสารดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็ก หลังจากผู้ป่วยได้รับการตรวจคลื่นสะท้อนหัวใจทางหลอดอาหารเสร็จแล้ว ผู้ป่วยจะต้องนั่งพักในบริเวณที่จัดเอาไว้ก่อนอย่างน้อย 1 ชั่วโมงเพื่อสังเกตดูว่ามีอาการผิดปกติหรือไม่ ถ้าผู้ป่วยไม่อาการใดผิดปกติก็จะสามารถกลับบ้านได้ ถ้ามีภาวะแทรกซ้อนเกิดขึ้นก็จะได้รับการตรวจรักษารวมทั้งอาจจะต้องนอนรับการรักษาในโรงพยาบาลต่อไป

ประโยชน์และผลข้างเคียงที่จะเกิดแก่ผู้เข้าร่วมโครงการ

ประโยชน์ที่คาดว่าจะได้รับจากการศึกษานี้คือ ถ้าสารดีไดเมอร์และทรอมบิน-แอนติทรอมบิน คอมเพล็ก สามารถวินิจฉัยลิ่มเลือดในเอเตรียมซ้ายได้ในผู้ป่วยลิ้นหัวใจไมตรัลตีบได้ ก็สามารถนำการตรวจนี้ไปใช้ได้ในโรงพยาบาลทั่วไปที่ไม่มีการตรวจด้วยคลื่นสะท้อนเสียงทางหลอดอาหารเพื่อช่วยเป็นแนวทางในการให้ยาต้านการแข็งตัวของเลือดโดยเฉพาะผู้ป่วยที่ยังมีหัวใจเต้นปกติ นอกจากนี้จะช่วยประหยัดค่าใช้จ่ายในการวินิจฉัยลิ่มเลือดในเอเตรียมซ้ายของผู้ป่วยกลุ่มนี้ได้อีกด้วย

ความเสี่ยงที่อาจจะเกิดขึ้นจากการตรวจคลื่นสะท้อนหัวใจทางหลอดอาหารมีน้อยมาก ผู้ป่วยบางรายอาจมีความดันโลหิตสูงขึ้นหรือบางรายอาจมีความดันโลหิตลดลง บางรายอาจมีหัวใจเต้นผิด

จังหวะ ซึ่งทั้งหมดนี้จะดีขึ้นหลังจากหยุดการตรวจ โดยผู้ทำการวิจัยจะสามารถดูแลผู้ป่วยได้ตลอดระยะเวลาของการตรวจด้วยวิธีดังกล่าวและสามารถหยุดการตรวจได้ทันทีถ้ามีปัญหาเกิดขึ้น รวมทั้งมีการเตรียมพร้อมที่จะให้การรักษาอย่างทันท่วงทีถ้ามีภาวะแทรกซ้อนเกิดขึ้น

หากผู้เข้าร่วมโครงการมีสงสัยประการใดหรือเมื่อเกิดผลข้างเคียงจากการวิจัยขึ้น สามารถติดต่อกับนายแพทย์เรวัตร พันธุ์กิ่งทองคำได้ที่ สาขาหทัยวิทยา ภาควิชาอายุรศาสตร์ โรงพยาบาลศิริราช หมายเลขโทรศัพท์ 02-4197745-6 หรือ 01-8142943

หากผู้เข้าร่วมโครงการได้รับผลข้างเคียงจากการวิจัย ผู้วิจัยจะให้การรักษาตามความเหมาะสมในบางกรณีอาจต้องรับไว้รักษาในโรงพยาบาล โดยค่าใช้จ่ายต่างๆที่เกิดขึ้นจากการร่วมในโครงการวิจัย ผู้วิจัยจะเป็นผู้รับผิดชอบ

หากผู้วิจัยมีข้อมูลเพิ่มเติมทั้งด้านประโยชน์และโทษที่เกี่ยวข้องกับการวิจัยนี้ ผู้วิจัยจะแจ้งให้ผู้เข้าร่วมโครงการทราบอย่างรวดเร็วโดยไม่ปิดบัง

ผู้เข้าร่วมโครงการสามารถที่จะของดการเข้าร่วมโครงการวิจัยโดยมิต้องแจ้งให้ทราบล่วงหน้า โดยการงดการเข้าร่วมการวิจัยนี้จะไม่มีผลกระทบต่อการได้รับบริการหรือการรักษาที่ผู้เข้าร่วมโครงการจะได้รับแต่ประการใด ผู้วิจัยจะไม่เปิดเผยข้อมูลหรือผลการวิจัยของผู้ป่วยเป็นรายบุคคลต่อสาธารณชน

ข้าพเจ้าได้รับทราบและได้ซักถามผู้วิจัยจนหมดข้อสงสัยโดยตลอดแล้วและยินดีเข้าร่วมในการการวิจัย จึงได้ลงลายมือชื่อไว้เป็นหลักฐานต่อหน้าพยาน

ลงชื่อ.....ผู้ยินยอมหรือผู้แทนโดยชอบธรรม

(.....)

ลงชื่อ.....ผู้ปกครอง (กรณีที่ผู้ป่วยอายุน้อยกว่า 20 ปีริบูรณ์)

(.....)

ลงชื่อ.....หัวหน้าโครงการวิจัย

(.....)

ลงชื่อ.....พยาน

(.....)

ลงชื่อ.....พยาน

(.....)

APPENDIX 2

CASE RECORD FORM

Case No_____

CRF for TAT and D-dimer in MS Project

Sex: ☐ Male ☐ Female

Age _____ yr **Occupation:** _____

Address: _____ **Tel :** _____

Weight: _____ kg **Height:** _____ cm **BSA:** _____M^2

Echo No: _____ **Date:** ____/_____/___

I) Demographic data

1st clinical presentation

- Dyspnea ☐ Yes ☐ No
- Acute pulmonary edema (CHF) ☐ Yes ☐ No
- Chest pain ☐ Yes ☐ No
- Hoarseness ☐ Yes ☐ No
- Hemoptysis ☐ Yes ☐ No
- Palpitation ☐ Yes ☐ No
- Syncope ☐ Yes ☐ No
- Embolization ☐ Yes ☐ No

  If yes; ☐ Brain : Dx by ☐ Clinical ☐ Clinical with CT or MRI
  ☐ Extremities
  ☐ Coronary
  ☐ Others : _____

- Duration of symptoms _____ yr
- NYHA Fc on 1st presentation : ☐ I ☐ II ☐ III ☐ IV

- Previous history of embolization ☐ Yes ☐ No

  If yes; ☐ Brain ☐ Extremities ☐ Coronary ☐ Others _____

- Previous history of rheumatic fever ☐ Yes ☐ No
- Previous history of IE ☐ Yes ☐ No
- Previous history of syncope ☐ Yes ☐ No
- Previous history of stroke ☐ Yes ☐ No
- CAD risk factors

  ☐ DM ☐ HT ☐ Dyslipidemia ☐ smoking ☐ Familial History

- Concomitant disease: _____
- Prior PBMV ☐ Yes ☐ No
- Prior OMV or CMV ☐ Yes ☐ No

II) Echocardiography

- Rhythm

  ☐ NSR ☐ Coarse AF ☐ Fine AF ☐ Others:……..

  ☐ Axis…….degree

  ☐ LAE ☐ Yes ☐ No

  ☐ RAE ☐ Yes ☐ No

  ☐ RVH ☐ Yes ☐ No

  If yes: ☐ rSR ☐ qR ☐ Rs ☐ qRS

NB: Coarse AF = amplitude of wave (0.1 mv in V_1)

TTE

MVA by trace _____ cm2 MVA by Pt½ _____ cm2

(Mitral valve score _____

(Pressure gradient LA-LV: peak _____mm Hg Mean _____mmHg

(LVEF _____% RVSP _____ mmHg

(MR severity	(None	(Mild	(Moderate	(vere
(AR severity	(None	(Mild	(Moderate	(Severe
(AS severity	(None	(Mild	(Moderate	(Severe
(TR severity	(None	(Mild	(Moderate	(Severe

- TS severity ☐ None ☐ Mild ☐ Moderate ☐ Severe

TEE

☐ LA clot ☐ Yes: at ☐ LA appendage ☐ LA body ☐ IAS
☐ No

☐ LASEC ☐ Yes ☐ No

If yes; ☐ LA ☐ LV ☐ RA ☐ RV

- MR severity ☐ one ☐ Mild ☐ Moderate ☐ Severe
- AR severity ☐ None ☐ Mild ☐ Moderate ☐ Severe
- TR severity ☐ None ☐ Mild ☐ Moderate ☐ Severe

IV) LA & LAA data

1. LA size
   - 4-chamber view length_____cm width_____cm
   - Short axis view length_____cm
   - Parasternal view length_____cm
2. LAA area & LAA ejection fraction
   - Maximum LAA area:_____cm2
   - Minimum LAA area:_____cm2
   - LAA.EF_____%
3. LAA flow pattern
   ☐ high profile ☐ low profile
4. LAA velocity
   - Maximal emptying velocity _____ cm.s-1
   - Mean emptying velocity _____ cm.s-1
   - Maximal filling velocity _____ cm.s-1
   - Mean filling velocity _____ cm.s-1

V) D-dimer and TAT level

- TAT = _____ (g/l)
- D-dimer = _____ ng/ml

VITAE

Dr. Rewat Phankingthongkum was born on March 1964, 16 in Nakornsawan, Thailand. He received the Doctor of Medicine Diploma from the Faculty of Medicine, Siriraj Hospital, Mahidol University in 1989, Diplomat Thai Board of Internal Medicine from Khon kaen University in 1993 and Diplomat Thai Board of Cardiology from Siriraj Hospital, Mahidol University in 1995.

In June 2002, Dr. Phankingthongkum admitted into the Master's degree program in Health Development in Thai-CERTC, Faculty of Medicine, Chulalongkorn University, as funded by Faculty of Medicine Siriraj Hospital, Mahidol University. During this course, he did a research paper titled "Thrombin-antithrombin complex and D-dimer for detection of left atrial thrombus in the patients with mitral stenosis".

At present, he is an assistant professor in the Department of Medicine, Faculty of Medicine, Siriraj Hospital, Mahidol University.