

Appendix 13 – Literature Review on D-Dimer – completed by Dr. Adam Peets

A review of the VIDAS D-Dimer (ELISA) assay and its role in the diagnosis of pulmonary embolism and deep vein thrombosis

Abstract

Background: Attempts are being made to reduce the costs associated with the diagnosis of deep vein thrombosis (DVT) and pulmonary embolism (PE). One promising method to help rule out these diagnoses is by measuring plasma D-dimer levels, a product of clot breakdown, with a rapid, quantitative ELISA assay, VIDAS D-Dimer. However, prior to developing a clinical algorithm that uses this assay, a review of the literature needed to be undertaken to ensure that the VIDAS D-Dimer has sufficient sensitivity and negative predictive value to be used as a screening test in patients presenting with suspected DVT or PE.

Methods: A search of MEDLINE, PubMed, Cochrane and Best Evidence was undertaken to identify all relevant English-language articles. Selected articles were then critically appraised and conclusions drawn.

Results: Fifteen articles were identified. Four trials provided level one evidence, combined with one large clinical management trial, that supported the use of the VIDAS D-Dimer assay as part of a clinical algorithm to rule out DVT. For inpatients with suspected DVT, two trials (both level one evidence) determined the sensitivity and negative predictive value of the VIDAS assay to be 100%. Diagnosis of PE in outpatients was evaluated by a large clinical management trial and a level one evidence trial, both of which showed the assay was effective. There is almost a complete lack of literature evaluating the VIDAS D-Dimer assay's performance in ruling out suspected PE in inpatients. A number of situations in which the assay's performance is lower than expected were also identified.

Conclusion: It is safe to proceed with the incorporation of VIDAS D-Dimer assay into a clinical algorithm for outpatients and inpatients presenting with a suspected first episode of DVT. It is also safe to incorporate VIDAS D-Dimer in to an algorithm to rule out PE in outpatients. However, further evidence is necessary prior to using the assay as part of an algorithm to rule out PE in inpatients.

Background:

Venous thromboembolism (VTE) is a common and fatal disease. While there are as many as 260,000 patients in the U.S. in whom VTE is diagnosed and treated each year, there are at least that same number of patients in whom the correct diagnosis of VTE is never made.¹ Studies have shown that diagnosis of VTE based on clinical judgment alone is unreliable, thereby resulting in increased utilization of radiographic tests to rule out the possibility of deep vein thrombosis (DVT) and pulmonary embolus (PE). With increased strain on health care budgets and increased workloads in radiology departments, attempts are being made to develop a cost-effective and resource-conscious clinical algorithm that can be used in the diagnostic work-up of DVT and PE.²⁻¹⁹

One strategy that has gained widespread interest is to use an algorithm consisting of an initial laboratory screening test (serum D-dimer levels) combined with a clinical pretest probability score to help decrease the number of radiologic investigations required. D-dimer is a specific fibrin degradation product that is elevated in patients with VTE. There are a number of different methods available to measure D-dimer levels in the blood. The best studied and most reliable method is ELISA (Enzyme-Linked Immunosorbent Assay). Unfortunately, conventional ELISAs are not suitable for emergency situations as the turn-around time is usually greater than two hours. Recently, however, a very promising rapid ELISA, VIDAS D-Dimer (bioMerieux, Marcy l'Etoile, France), has been shown to be equivalent to conventional ELISAs.²⁰⁻²⁴ VIDAS D-Dimer is a quantitative fully automated ELISA assay with fluorescence detection performed on a dedicated immunoanalyzer, capable of producing results in approximately 50 minutes.²²

Once the results of the assay are known, a clinical algorithm can be applied to patients presenting with suspected DVT or PE. As the D-dimer assays have high sensitivity and negative predictive value (NPV), and low specificity and positive predictive value (PPV), their role in the algorithm will be to help rule out the presence of DVT or PE. van Beek et al.¹⁹ calculated that for every 2% decrease in sensitivity, one per 1000 evaluated patients with clinically suspected PE would die. Although this is probably less for DVT, it is evident that a NPV lower than 98% may be too low to safely exclude DVT or PE. Therefore, prior to the incorporation of the VIDAS D-Dimer assay into any clinical algorithm to rule out PE or DVT, there needs to be sufficient evidence to show that the sensitivity and negative predictive value of the assay are greater than or equal to 98%.

According to Sackett et al.²⁵ the proper evaluation of any diagnostic test involves three steps. The technical aspects including definition of normal values and determination of intra- and inter-assay variability should first be undertaken. The second step is testing the accuracy of the assay in a blinded fashion in consecutive patients with and without the disease. Finally, the clinical utility and validity should be ascertained. Using these three steps as a guide, this review will critically appraise the literature currently available on the VIDAS D-Dimer assay and its role in the diagnosis of pulmonary embolism and deep vein thrombosis.

Methods:

A literature search was undertaken to identify relevant English-language articles. Using the key words "deep vein thrombosis," "DVT," "pulmonary embolism," "pulmonary embolus," "PE," "venous thromboembolism," "D-dimer," "VIDAS" and "ELISA" the databases of Medline, PubMed, Cochrane and Best Evidence were searched. The bibliographies of all studies were manually searched to identify additional relevant studies. Each article was critically appraised using the method described by Sackett et al.,²⁶ and had a level of evidence assigned as per guidelines in Table 1.

Results:

The technical aspects of the VIDAS D-Dimer assay are well established. The normal value of 500 ng/ml has been validated by numerous clinical trials.^{20-22,27-29} It has been determined that this cutoff provides the highest combination of sensitivity and specificity for diagnosing DVT or PE. Many of

these same trials have also documented that the intra- and inter-assay variability is excellent, with kappa coefficients showing good to excellent concordance.^{20,21,23,24} There are also numerous trials in the literature defining the accuracy and the clinical utility and validity of the VIDAS D-Dimer assay in the diagnosis of deep vein thrombosis and pulmonary embolism. What follows is a brief description of these trials, and is divided into three different parts: trials related to DVT only, trials related to PE only, and trials that examine both DVT and PE. The most important characteristics and results of each of the trials have been summarized in Tables 2 and 3.

Trials Evaluating VIDAS D-Dimer in the Exclusion of DVT

van der Graaf et al. “Exclusion of deep venous thrombosis with D-dimer testing.”²²

Methods:

- prospective, single-center trial involving 99 consecutive outpatients.
 - inclusion criteria: clinically suspected DVT
 - exclusion criteria: treatment with anticoagulants, recent surgery (<8 days), contraindication to contrast media, refusal or inability to sign consent, inability to perform or inadequate results of ascending venography
 - protocol: thirteen different D-dimer assays (including VIDAS) compared to gold-standard venography.
- Radiologist blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS found to be 100% sensitive (95% CI 93-100) and 41% specific (95% CI 27-56). The negative predictive value was 100% (95% CI 83-100).
- likelihood ratio for a positive test result: 1.7 ; likelihood ratio for a negative test result: 0.0

Limitations:

- single-center trial, only one radiologist interpreting results of venography (although remained blinded)

Conclusions:

Level 1b evidence

Overall, an excellent study using the gold standard of venography for comparison. With 100% sensitivity and NPV, the results of this study strongly suggest that VIDAS can be used as part of a clinical algorithm to safely exclude patients with normal D-dimer levels suspected of having a DVT or PE.

Legnani et al. “Contribution of a new, rapid, quantitative and automated method for D-dimer measurement to exclude deep vein thrombosis in symptomatic outpatients.”³¹

Methods:

- prospective, single-center trial involving 99 consecutive outpatients referred to the emergency department.
 - inclusion criteria: clinically suspected DVT
 - exclusion criteria: previous episode of DVT, stable symptoms lasting more than one month, anticoagulant or fibrinolytic therapy already underway at presentation, contraindication to ascending venography
 - protocol: three different D-dimer assays (including VIDAS) compared to gold-standard venography.
- Radiologists blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 700 ng/ml to indicate a negative test, VIDAS found to be 97.4% sensitive (95% CI 92-100) and 81.7% specific (95% CI 72-91). The corresponding negative predictive value was 98% (95% CI 94-100).
- likelihood ratio for a positive test result: 5.3 ; likelihood ratio for a negative test result: 0.03

Limitations:

- only outpatients
- higher cutoff for normal value likely led to lower than expected sensitivity

Conclusions:

Level 1b evidence

Although this was a well designed trial using the gold standard of venography, the higher cutoff value for normal (700 vs 500 ng/ml) likely led to the lower sensitivity and NPV than expected. Because of this fact, this trial should not be included in the final analysis.

Table 1. Guideline for Grading Level of Evidence for Diagnostic Studies³⁹

Grade of Recommendation	Level of Evidence	Description
A	1a	Systematic review with homogeneity of level 1 diagnostic studies or a CPG [®] validation set.
	1b	Independent blind comparison of patients from an appropriate spectrum of patients all whom have undergone both the diagnostic test and the reference standard.
	1c	Diagnostic finding where sensitivity is so high that a negative result rules out the disease.
B	2a	Systematic review with homogeneity of level ≥ 2 diagnostic studies.
	2b	-independent blind comparison -non-consecutive patients or confined to a narrow spectrum of study individuals all of whom have undergone both the diagnostic test and the reference standard. -diagnostic CPG not validated on a test set.
	3b	Independent blind or objective comparison of appropriate spectrum of patients, but reference standard not applied to all study patients.
C	4	-reference standard was unobjective, unblinded or not independent. -positive and negative tests verified using separate reference standards.
D	5	-inappropriate spectrum of patients Expert opinion without explicit critical appraisal or based on physiology, bench research or "first principles."

® CPG: Clinical Practice Guideline

Legnani et al. "Comparison of new rapid methods for D-dimer measurements to exclude deep vein thrombosis in symptomatic outpatients."²¹

Methods:

- prospective, single-center trial involving 81 consecutive outpatients.
- inclusion criteria: clinically suspected first episode of DVT
- exclusion criteria: symptoms lasting greater than one month, on anticoagulation or fibrinolytic therapy
- protocol: seven different D-dimer assays (including VIDAS) compared to gold-standard venography in each patient. Radiologist blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS found to be 95.2% sensitive (95% CI 89-100) and 55.3% specific (95% CI 39-71). (Negative predictive value of 91.3% (95% CI 80-100).
- likelihood ratio for a positive test result: 2.12 ; likelihood ratio for a negative test result: 0.087

Limitations:

- 6 patients from initial 87 were excluded due to, "inability to perform ascending venography for technical reasons or inconclusive results."
- it should be noted that the authors admit that, "All the investigated methods failed to identify one patient in whom venography results, though questionable, showed the presence of proximal DVT, and two patients with distal DVT in whom a hypofibrinolytic condition was later diagnosed."

Conclusions:

Level 1b evidence

This is a well-designed trial comparing VIDAS D-Dimer to the gold standard of venography. Despite the limitations resulting from the exclusion of six patients, VIDAS did have a lower sensitivity and negative predictive value as compared to the other trials using venography. This trial puts the ability of VIDAS D-dimer to confidently rule out DVT into question.

Elias et al. "D-dimer test and diagnosis of deep vein thrombosis: a comparative study of seven assays."²³

Methods:

- prospective, single-center trial involving 171 consecutive inpatients (n=142) and outpatients (n=29) referred to a French angiology unit.
- inclusion criteria: clinically suspected first episode of DVT
- protocol: seven different D-dimer assays (including VIDAS) compared to compression U/S. Radiologist not blinded to clinical assessment and D-Dimer results, but lab technician blinded to U/S results.

Results:

- for all patients, using a threshold of ≤ 400 ng/ml to indicate a negative test, VIDAS was found to be 97% sensitive (95% CI 90-100) and 26% specific (95% CI 18-36). The corresponding negative predictive value was 93% (95% CI 74-100).
- for outpatients only, VIDAS was found to be 100% sensitive and 50% specific. The corresponding negative predictive value was 100%.
- for inpatients only, VIDAS was found to be 98% sensitive and 18% specific. The corresponding negative predictive value was 94%.
- for inpatients, likelihood ratio for a positive test result: 1.3; likelihood ratio for a negative test result: 0.12
- for outpatients, likelihood ratio for a positive test result: 1.2; likelihood ratio for a negative test result: 0.11

Limitations:

- cutoff 400 ng/mL. Therefore, falsely elevated sensitivity compared with normal 500 ng/mL cutoff
- compression U/S instead of venography used to confirm or rule out DVT
- since no venography, distal DVT may have been overlooked
- small number of outpatients.

Conclusions:

Level 1b evidence

This study demonstrates that VIDAS is effective for detecting isolated proximal DVT in outpatients. However, the assay did not demonstrate high enough sensitivity in this study to be used as a first-line screening test to rule out DVT in inpatients, or those with distal DVT.

Funfsinn et al. "Rapid D-dimer testing and pre-test clinical probability in the exclusion of deep venous thrombosis in symptomatic outpatients."⁷**Methods:**

- prospective, single-center trial involving 106 consecutive outpatients.
- inclusion criteria: clinically suspected DVT
- exclusion criteria: anticoagulants for more than 24 hrs prior to study enrollment, pregnancy, and hospitalization within last 3 days
- protocol: four different D-dimer assays compared to compression ultrasound or venography to diagnose DVT. Three month follow-up. Retrospective application of pre-test clinical probability (Wells' criteria), to determine the changes in sensitivity and specificity when used in combination with D-dimer compared to D-dimer alone. Not stated if radiologist blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 100% sensitive (95% CI 94-100) and 52.5% specific (95% CI 38.3-66.8). The corresponding negative predictive value was 100% (95% CI 90-100). When combined with the pre-test clinical probability, the specificity rose to 76.9%.
- likelihood ratio for a positive test result: 2.1; likelihood ratio for a negative test result: 0.0

Limitations:

- only outpatients
- unclear how clinicians decided whom received U/S and who received venography
- no mention how many patients received each diagnostic test
- since venography not performed in all patients, some distal DVT may have been overlooked
- retrospective, hypothetical application of pre-test probability, rather than prospectively testing the combination of D-dimer and pre-test probability

Conclusions:

Level of evidence: 1b if assume blinded, otherwise level 4

Despite this study's limitations with not applying venography to all patients, overall, it demonstrates excellent sensitivity and NPV for VIDAS in ruling out DVT. If the results of the hypothetical implementation of pre-test probability are to be believed, the increase in specificity obtained with the combined use of D-dimer and pre-test probability would lead to improved cost-effectiveness in the diagnosis of DVT.

Janssen et al. "Reliability of five rapid D-dimer assays compared to ELISA in the exclusion of deep venous thrombosis."²⁴**Methods:**

- prospective, multicenter trial involving 132 outpatients referred to emergency or outpatient departments.
- inclusion criteria: clinically suspected DVT
- exclusion criteria: non-ambulatory
- protocol: five different D-dimer assays (including VIDAS) compared to compression U/S. If either veins could not be adequately visualized or distal DVT suspected, venography performed. (total of 26 venograms performed). Not stated if radiologist blinded to clinical assessment and D-Dimer results.

Results:

-using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 100% sensitive (95% CI 97-100) and 19% specific (95% CI 7-30). The corresponding negative predictive value was 100% (95% CI 69-100).

-likelihood ratio for a positive test result: 1.2 ; likelihood ratio for a negative test result: 0.0

Limitations:

-only outpatients

-unclear if consecutive patients

-unclear how clinicians suspected distal DVT and subsequently continued on to venography.

-since venography not performed in all patients, some distal DVT may have been overlooked

Conclusions:

Level 2b evidence if assume blinded, otherwise level 4 evidence.

Despite this study's limitations, it does demonstrate a trend toward excellent sensitivity and NPV for VIDAS in ruling out DVT.

D'Angelo et al. "Evaluation of a new rapid quantitative D-dimer assay in patients with clinically suspected deep venous thrombosis."²⁷

Methods:

-prospective, single-center trial involving 103 consecutive inpatients and outpatients referred to an Italian Hospital Coagulation Service.

-inclusion criteria: clinically suspected DVT

-protocol: D-dimer measurement (VIDAS) and compression U/S performed in all patients. If initial U/S negative, serial U/S day 1 and 7. Radiologist not blinded to clinical assessment and D-Dimer results, but lab technician blinded to U/S results.

Results:

-for patients with symptoms less than 11 days (n=99), using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 100% sensitive (95% CI 79-100) and 45% specific (95% CI 34-56). The corresponding negative predictive value was 100% (95% CI 88-100).

-for all patients enrolled in the study (n=103), VIDAS was found to be 96% sensitive (95% CI 75-100) and 44% specific (95% CI 34-56). The corresponding negative predictive value was 97% (95% CI 84-100).

-for symptoms less than 11 days, likelihood ratio for a positive test result: 1.8 ; likelihood ratio for a negative test result: 0.0

Limitations:

-compression U/S instead of venography used to confirm or rule out DVT

-lack of full compressibility on U/S the sole criteria for an abnormal result

-since no venography, distal DVT may have been overlooked

Conclusions:

Level 1b evidence

An important aspect of this study was to point out that the sensitivity of VIDAS decreases with time from onset of DVT. (Due to decreased fibrin release from older clots) Overall, this study was well designed. However, by not using venography as the reference standard, some clots may have been missed, resulting in a falsely high sensitivity.

Shitrit et al. "Diagnostic value of the D-dimer test in deep vein thrombosis: improved results by a new assay method and by using discriminate levels."³⁰

Methods:

-prospective, single-center trial involving a combination of 108 outpatients and inpatients.

-inclusion criteria: clinically suspected DVT

-exclusion criteria: under 18 years old, symptoms of DVT for more than 7 days.

-protocol: four different D-dimer assays, including VIDAS D-Dimer compared to compression ultrasound.

Radiologist blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 100% sensitive and 25% specific. The corresponding negative predictive value was 100%.
- likelihood ratio for a positive test result: 1.3 ; likelihood ratio for a negative test result: 0.0

Limitations:

- unclear if consecutive patients
- since venography not performed in all patients, some distal DVT may have been overlooked

Conclusions:

Level 2b evidence

Overall, a well designed study, but lack of stating that consecutive patients were used decreases the level of evidence from 1b to 2b. Also, with the lack of comparison to the gold-standard of venography some distal DVTs may have been missed, thereby leading to a falsely elevated sensitivity and NPV.

Borg et al. "Rapid quantitative D-dimer assay and clinical evaluation for the diagnosis of clinically suspected deep vein thrombosis."³²

Methods:

- prospective, single-center trial involving 76 consecutive patients
- inclusion criteria: clinically suspected DVT
- protocol: two different D-dimer assays (including VIDAS) compared to compression U/S. Lab technician blinded to results of U/S, but not stated if radiologist blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 93.7% sensitive and 52.2% specific. The corresponding negative predictive value was 92%.
- likelihood ratio for a positive test result: 1.96 ; likelihood ratio for a negative test result: 0.12

Limitations:

- very poorly described methods and results sections. No mention of many key factors necessary to critically appraise the trial.
- one of the patients with a false negative result had had symptoms for greater than 10 days
- small number of participants
- since no venography, distal DVT may have been overlooked

Conclusions:

Level 2b evidence

The poor quality of the manuscript alone is enough to question the results obtained by the authors. Overall, this is a poor study, and should not be included in the final analysis of the effectiveness of VIDAS.

Trials Evaluating VIDAS D-Dimer in the exclusion of PE

de Moerloose et al. "Contribution of a new, rapid, individual and quantitative automated D-dimer ELISA to exclude pulmonary embolism."²⁸

Methods:

- prospective, single-center trial involving 195 consecutive outpatients referred to an emergency department in Switzerland.
- inclusion criteria: clinically suspected PE
- protocol: according to clinical algorithm, patients underwent V/Q scan, lower limb doppler U/S or pulmonary angiogram. All patients had D-dimer levels drawn. Six month clinical follow-up to evaluate for symptoms of PE. Not stated if radiologist blinded to clinical assessment and D-Dimer results.

Results:

- using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 100% sensitive (95% CI 92-100) and 37.6% specific (95% CI 30.1-45.8). The corresponding negative predictive value was 100% (95% CI 93.3-100).

-after six months of follow-up, of the 56 patients with D-dimer levels <500 ng/ml, none of the 51 evaluable patients (5 abroad) had a new suspicion of PE.

-likelihood ratio for a positive test result: 1.6 ; likelihood ratio for a negative test result: 0.0

Limitations:

-only outpatients

-limited information on patient demographics

-steps involved in diagnostic algorithm unclear

-six month follow-up by phone only

-only 52 patients (26.6%) underwent gold standard of pulmonary angiogram. Otherwise, diagnosis of PE confirmed by high probability V/Q scan or positive U/S of legs.

Conclusions:

Level 1b evidence if assume blinded, otherwise level 4 evidence

Despite the fact that not every patient underwent pulmonary angiography, all patients did have a well-validated test to rule in or out PE performed. Importantly, from a clinical standpoint, after 6 months of follow-up, none of the patients with a normal D-dimer level had a PE. Therefore, the results of this trial show that for outpatients, VIDAS can be used as part of a diagnostic algorithm to rule out PE.

Lorut et al. "A noninvasive diagnostic strategy including spiral computed tomography in patients with suspected pulmonary embolism."¹⁰

Methods:

-prospective, single-center trial involving 247 consecutive outpatients referred to emergency or outpatient departments.

-inclusion criteria: clinically suspected PE

-protocol: D-dimer was used as part of a non-invasive strategy to rule PE. Depending on the algorithm, patients had D-dimer (n=227), spiral CT (n=201), V/Q (n=178), U/S (n=56) and/or pulmonary angiogram (n=2) performed. Three month follow-up to ensure no clinical evidence of PE. Radiologist not blinded to clinical history, but lab technicians were blinded to U/S results.

Results:

-using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 98.6% sensitive. No further statistical results were mentioned in the manuscript.

-unable to calculate likelihood ratios due to lack of statistical information.

Limitations:

-19 patients excluded from final analysis (16 because study protocol was negative, 1 because they died, 1 because diagnosis of PE had already been made by angiogram and 1 because symptoms had started several months ago)

-19 other patients lost to follow-up

-difficult to fully assess D-dimer efficacy as the trial was designed to determine if the diagnostic strategy was effective in ruling out PE.

-D-dimer was compared to CT as the confirmatory test in most patients rather than pulmonary angiogram.

Conclusions:

Level 3b evidence

As this trial was specifically examining the effectiveness of a diagnostic strategy to rule out PE and was not designed to determine the sensitivity, specificity and NPV of VIDAS, and due to the fact that spiral CT was used as the gold standard, the results of this trial need to be interpreted with skepticism.

Sijens et al. "Rapid ELISA assay for plasma D-dimer in the diagnosis of segmental and subsegmental pulmonary embolism."³³

Methods:

-retrospective, single-center trial involving 342 consecutive patients

-inclusion criteria: clinically suspicion of PE, normal leg U/S and positive lung perfusion scans.

-exclusion criteria: inability to give consent, treatment with IV heparin for more than 72 hours at time of blood sampling, or surgery within past 6 weeks.

-protocol: all patients underwent pulmonary angiogram generally within 24-48 hours and at most 72 hours after suspicion for PE arose. Importantly, patients received 1-2 days of IV heparin prior to pulmonary angiogram. Not stated if radiologist blinded to clinical assessment and D-Dimer results.

Results:

-the authors split the results into those of segmental (main lung vessels) and subsegmental thrombi.

-for all pulmonary emboli:

-using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 90% sensitive, 45% specific, with a negative predictive value of 94%.

-likelihood ratio for a positive test result: 1.6 ; likelihood ratio for a negative test result: 0.2

-for segmental pulmonary emboli:

-using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 98% sensitive, 45% specific, with a negative predictive value of 99.2%.

-likelihood ratio for a positive test result: 1.8 ; likelihood ratio for a negative test result: 0.04

-for subsegmental pulmonary emboli:

-using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS was found to be 76% sensitive, 45% specific, with a negative predictive value of 94%.

-likelihood ratio for a positive test result: 1.4 ; likelihood ratio for a negative test result: 0.53

Limitations:

-retrospective design

-pre-selected population (normal leg U/S and positive V/Q scans)

-not mentioned if inpatients or outpatients

-patients were on IV heparin for 1-2 days prior to angiogram

Conclusions:

Level 2b evidence

This paper has multiple limitations that put into question the validity of the results. Despite the fact that it is a retrospective analysis of a pre-selected population with a very high pre-test probability (positive V/Q scans), it does raise an interesting question. Can VIDAS D-Dimer detect small pulmonary emboli? The results for detection of segmental PE are very similar to those results of studies using V/Q scans as the gold standard. However, in this trial VIDAS showed an unacceptably low sensitivity for diagnosing subsegmental thrombi. The authors hypothesized that the lower clot burden associated with subsegmental emboli (i.e. less D-dimer produced), in combination with the inability of V/Q scans to diagnose the smaller clots in subsegmental areas, led to the falsely elevated sensitivity and negative predictive value of VIDAS. One possible confounding factor is that the patients were on heparin for 1-2 days prior to the angiogram. It has been shown that in patients with DVT being treated with heparin, the plasma D-dimer level dropped by approximately 40% (24h) and 50% (48h). However, even with recalculating the cutoff values by taking the possible reduction in D-dimer levels due to heparin into consideration, the overall sensitivity for VIDAS only rose to 94% with a corresponding NPV of 96%. If these results are believed, this remains below the value necessary for an effective screening device. Another potential bias is that pulmonary angiography is difficult to interpret at the subsegmental level and interobserver agreement may be as low as 60% in such patients.³⁴ Overall, the poor design of this study prevents it from being included in the final analysis of this paper. However, it does raise the issue of whether VIDAS can rule out smaller subsegmental PE. This question will need to be assessed in prospective, outcome-based trials.

Wallis et al. "A comparative study of two rapid D-dimer tests for the exclusion of pulmonary embolism in symptomatic patients."³⁵

Methods:

-prospective, single-center trial involving 129 patients

-inclusion criteria: clinically suspected PE

-protocol: two different D-dimer assays (including VIDAS) compared to pulmonary angiogram.
No mention if radiologist blinded to clinical assessment and D-Dimer results.

Results:

-using a threshold of ≤ 500 ng/ml to indicate a negative test, VIDAS found to be 97% sensitive (95% CI 83-100) and 26% specific (95% CI 18-36). The corresponding negative predictive value was 96% (95% CI 81-100).

-likelihood ratio for a positive test result: 1.3 ; likelihood ratio for a negative test result: 0.12

Limitations:

-very poorly described methods and results sections. No mention of many key factors necessary to critically appraise the trial.

Conclusions:

Level of evidence: insufficient information provided

It is unfortunate that the authors did not provide further information, as this is one of the only gold-standard prospective analyses of VIDAS D-Dimer available in the literature.

Trials Evaluating VIDAS D-Dimer in the Exclusion of DVT and PE

Perrier et al “Non-invasive diagnosis of venous thromboembolism in outpatients.”⁴
Freyburger et al. “D-dimer strategy in thrombosis exclusion.”²⁰

Methods:

- prospective, single-center trial involving 100 consecutive inpatients.
- inclusion criteria: clinically suspected DVT (n=83) or PE (n=17)
- protocol: eight different D-dimer assays (including VIDAS) compared to gold-standard venography or angiography in each patient. Not mentioned if radiologist blinded to clinical assessment and D Dimer results.

Results:

- using a threshold of ≤ 550 ng/ml to indicate a negative test, VIDAS found to be 100% sensitive and 38% specific. Negative predictive value of 100%.
- likelihood ratio for a positive test result: 1.6 ; likelihood ratio for a negative test result: 0.0

Limitations:

- only inpatients
- small number of PE patients
- threshold for a negative D-dimer result higher than the cutoff used by most other trials (500 ng/ml), thereby increasing the specificity, but potentially lowering the sensitivity.

Conclusions:

Level 1b evidence if assumed blinded, level 4 evidence if unblinded

As VIDAS D-Dimer was compared to the gold standard of venography, the results of this trial strongly suggest that VIDAS can be used as part of a clinical algorithm to safely exclude the diagnosis of DVT in outpatients. Due to the small number of patients with suspected PE (n = 17), it is difficult to use the results of this trial alone to support the use of VIDAS D-Dimer to exclude PE.

Methods:

- prospective, multicenter trial involving 918 consecutive outpatients presenting to ER department
- inclusion criteria: clinically suspected PE (n=444) or DVT(n=474)
- exclusion criteria: inability or refusal to consent, anticoagulation at onset symptoms, hospital admission more than 24 hrs prior onset symptoms or during month preceding inclusion, impossible follow-up, contraindication to phlebography or angiography, expected survival less than 3 months.
- protocol: diagnostic algorithm involving pretest probability, D-dimer, venous doppler U/S, V/Q scan, and only if necessary angiography or phlebography. Patients followed for three months to assess for recurrence on clinical grounds only. Radiology and laboratory technician blinded to clinical assessment and D-Dimer results.

Results:

- two patients had false negative D-dimer tests. Negative predictive value of 99.3% (95% CI 97.5-99.9)
- using their algorithm, 10 pts who had been negative for VTE using the algorithm had an event in 3 month follow-up (none had had normal D-dimer levels) (3 month thromboembolic risk of 1.8%)

Limitations:

- designed as a trial to evaluate a diagnostic algorithm that has VIDAS as only one of the many components
- applies only to outpatients
- no radiographic confirmation of absence of DVT or PE in patients with normal D-dimer levels
- only a few underwent gold standard tests (5.7%)
- 64 patients excluded from results due diagnostic protocol not followed.

Conclusions:

Level 3b evidence

Although it is level 3b evidence because the reference standard was not applied to all patients, this was a well designed trial that demonstrates the effectiveness of VIDAS D-Dimer as part of a clinical algorithm to rule out DVT and PE in an outpatient population.

Freyburger et al. "Rapid ELISA D-Dimer testing in the exclusion of venous thromboembolism in hospitalized patients."³

Methods:

- retrospective, single-center trial involving 989 inpatients.
- inclusion criteria: hospitalized patients with suspected VTE
- exclusion criteria: surgery less than 8 days before, heavily-traumatized patients, sepsis, malignant metastases.
- protocol: according to diagnostic algorithm, patients underwent D-Dimer measurement, and then a radiologic test (U/S, V/Q scan, spiral CT, venography or angiography) depending on clinical circumstance.

Results:

-specific results on the performance of VIDAS D-dimer were not mentioned in the results section other than to say that for 32 patients that had a negative D-Dimer, but continued on to have a radiographic procedure, all were not found to have DVT or PE. Therefore, for this small group within the trial, VIDAS D-Dimer showed 100% sensitivity and negative predictive value.

Limitations:

- retrospective design
- trial was designed to evaluate a clinical algorithm involving VIDAS D-Dimer as only one of a number of steps, and was not designed to specifically evaluate the performance of VIDAS.

Conclusions:

Level 3b evidence

The results of this trial demonstrate that for inpatients, VIDAS D-Dimer can be used as part of a cost-effective clinical algorithm to help rule out DVT and PE. However, it provided very little information on the performance of the VIDAS D-Dimer assay itself.

Discussion:

D-Dimer assays provide an opportunity to decrease both the time to diagnosis of DVT or PE and to decrease the amount of radiographic investigations, especially invasive investigations, necessary in the work-up of VTE. The VIDAS D-Dimer assay has a number of characteristics that make it an ideal tool to help in the acute diagnosis and management of DVT or PE: it is fast (turnaround time of less than one hour), reasonably priced (estimates of approximately \$25 Can per test) and the results are consistent. However, the key feature of any potential diagnostic test is whether or not it will perform up to the standards required in clinical practice. In the case of D-Dimer assays, does the VIDAS D-Dimer assay have a high enough sensitivity and negative predictive value to function as an initial screening test in patients presenting with their first episode of DVT or PE? Well, that depends on the clinical situation.

There are four major categories of patients that present with a first episode of VTE:

- 1) suspected DVT in an outpatient
- 2) suspected DVT in an inpatient
- 3) suspected PE in an outpatient
- 4) suspected PE in an inpatient

Although this is certainly oversimplifying matters, it can serve as a guideline to help in analyzing the literature on the VIDAS D-Dimer assay.

Suspected DVT in an outpatient:

Of the four categories, this one has the most literature available to date. There have been nine trials^{4,7,21-24,27,30,31} examining DVT in this population, with six of these being level 1b evidence. Four of the level one trials determined that VIDAS D-Dimer has both 100% sensitivity and 100% negative predictive value (NPV) for VIDAS D-Dimer in excluding DVT in outpatients.^{7,22,23,27} The two trials by Legnani et al.^{21,31} both produced sensitivities and NPV lower than the goal of 98% as determined by van Beek et al.¹⁹ However, these trials did have limitations that may have affected

the outcome. The first of the two trials,²¹ despite being well-designed and using the gold standard of venography as a comparison, found that all of the methods used to measure D-Dimer, including conventional ELISA, fared no better than VIDAS D-Dimer. In fact the authors admit that, "All the investigated methods failed to identify one patient in whom venography results, though questionable, showed the presence of proximal DVT, and two patients with distal DVT in whom a hypofibrinolytic condition was later diagnosed."²¹ It is difficult to assess the clinical importance of these three cases given the limited information available from the article, but this trial did demonstrate that the sensitivity and NPV were lower than what is required for an effective screening test. The second Legnani trial,³¹ was again well designed with venography as the reference standard, but a D-dimer cutoff value of 700 ng/mL was used, instead of the standard 500 ng/mL. This likely explains why the sensitivity is lower than that found in other trials.

Perrier et al.⁴ conducted a clinical management trial to evaluate the utility of VIDAS D-Dimer as part of a clinical algorithm to rule out suspected first-episode of DVT. In 474 outpatients, the combination of the assay and algorithm resulted in a negative predictive value of 99.3%. This provides excellent evidence that the assay can be safely used not only by itself, but also as part of an algorithm to decrease the number of radiographic investigations required.

Therefore, despite the results of the first Legnani et al. trial, when all of the level-one evidence is combined with the clinical management trial of Perrier et al., the majority of the evidence supports the use of VIDAS D-Dimer to help rule out DVT in outpatients.

There is level one evidence in the literature to support the use of VIDAS D-Dimer as part of a clinical algorithm to rule out a suspected first episode of DVT in outpatients.

Suspected DVT in an inpatient:

There are five trials in the literature that have evaluated the VIDAS D-Dimer assay in inpatients suspected of having a DVT,^{3,20,23,27,30} with three of them being considered level one evidence.^{20,23,27} Of these three, the only one that used the gold standard, venography, was Freyburger et al.,²⁰ with results demonstrating a sensitivity and negative predictive value of 100%. The other two trials used compression ultrasound as the reference standard. The results of D'angelo et al.²⁷ were the same as the previously mentioned study, with the assay achieving 100% sensitivity and 100% negative predictive value. However, Elias et al.²³ showed that the VIDAS D-Dimer assay had a sensitivity of 98% and negative predictive value of 94% when restricted to inpatient use. Since this trial used compression U/S as the comparison standard, some of the results are less reliable than the previously mentioned gold standard comparison trial. Unfortunately, the one trial evaluating VIDAS D-Dimer as part of a clinical algorithm in inpatients, although large (n=989), was a retrospective analysis that failed to provide convincing data of the effectiveness of the algorithm.²⁰ Therefore, based mainly on the results of the Freyberger et al. trial, it is reasonable to proceed with incorporation of the VIDAS D-Dimer assay into a clinical algorithm for the diagnosis of DVT in inpatients. However, this algorithm will need to be part of a prospective clinical trial to ensure clinical applicability, reliability and cost-effectiveness prior to becoming hospital policy for ruling out DVT.

There is level one evidence in the literature to support the use of VIDAS D-Dimer as part of a clinical algorithm to rule out a suspected first episode of DVT in inpatients. However, once the algorithm is developed, it will need to be evaluated in a prospective trial, as there is no evidence in the literature examining the cost-effectiveness and clinical applicability of this topic specifically.

Suspected PE:

The literature evaluating VIDAS D-Dimer in patients with suspected first episode of PE is far more limited than for DVT. The two major trials that used angiography as the reference standard, Sijens et al.³³ and Wallis et al.,³⁴ have major flaws, including retrospective design and not stating which population of patients they were examining, and essentially need to be excluded from the analysis.

This leaves only five trials to critically appraise. However, prior to proceeding, one important finding from Sijens et al. needs to be addressed.

Despite the fact that it is level 3b evidence, and that it is a retrospective analysis of a pre-selected population with a very high pre-test probability (positive V/Q scans), the Sijens trial does raise an interesting question. Can VIDAS D-Dimer detect small pulmonary emboli? The results of this trial show that the assay's ability to detect *segmental* PE are very similar to those results of studies using V/Q scans as the gold standard (98% sensitive, 99.2% NPV). However, VIDAS showed an unacceptably low sensitivity for diagnosing *subsegmental* thrombi. This result corroborates the PIOPED investigators finding that as many as 9% of the "near normal/normal" V/Q scans appeared to have PE using pulmonary angiogram.³⁶ The authors hypothesized that the lower clot burden associated with subsegmental emboli (i.e. less D-dimer produced), in combination with the inability of V/Q scans to diagnose the smaller clots in subsegmental areas, led to the elevated sensitivity and negative predictive value seen in other trials evaluating VIDAS. As already mentioned, however, this trial has a number of limitations. One possible confounding factor is that the patients were on heparin for 1-2 days prior to the angiogram. It has been shown that in patients with DVT being treated with heparin, the plasma D-dimer level dropped by approximately 40% (24h) and 50% (48h). However, even with recalculating the cutoff values by taking the possible reduction in D-dimer levels due to heparin into consideration, the overall sensitivity for VIDAS only rose to 94% with a corresponding NPV of 96%; these results remain below the value necessary for an effective screening device. Another potential bias of the trial is that pulmonary angiography is difficult to interpret at the subsegmental level and interobserver agreement may be as low as 60% in such patients.³⁴ Overall, the poor design of this study prevents it from being included in the final analysis of this paper. However, it does raise the issues of whether VIDAS can rule out smaller subsegmental PE and if these smaller emboli are clinically important. These questions will need to be assessed in prospective, outcome-based trials prior to the full incorporation of the assay into our diagnostic algorithms.

Suspected PE in outpatients:

There has only been one trial producing level one evidence for the use of VIDAS D-Dimer in outpatients. de Moerloose et al.¹⁴ initially evaluated 195 patients for the presence of PE, and then followed patients for clinical recurrence for six months. The results of this trial showed an impressive 100 % sensitivity and NPV, with a corresponding specificity of 37.6% even after the six month follow-up. Despite being labeled as level 3b evidence due to the fact that not every patient underwent the reference standard, the clinical management trial of Perrier et al.,⁴ is a very important study that demonstrated the ability of VIDAS D-Dimer to be used as part of a clinical algorithm to rule out PE in outpatients. Therefore, although the data is limited, due to the strength of the de Moerloose et al. trial and the proven clinical utility of the assay by Perrier et al., it would be reasonable to proceed with incorporation of the assay into a clinical algorithm to rule out PE in outpatients.

There is limited level one evidence to support the use of the VIDAS D-Dimer assay as part of a clinical algorithm to help rule out suspected first episode of pulmonary embolism in outpatients.

Suspected PE in inpatients:

There is insufficient literature evaluating the ability of the VIDAS D-Dimer assay to rule out PE in inpatients at this time. The only level one evidence trial, Freyburger et al.,²⁰ has just 17 patients suspected of PE enrolled. VIDAS did perform well, with sensitivity and NPV of 100%, but these results can not be generalized due to the small number of patients involved. The other trials that examined PE in inpatients are of poor quality and should not be considered in the final analysis. Therefore, due to the almost complete lack of literature on the subject, the decision on whether or not VIDAS D-Dimer can be incorporated into a clinical algorithm for ruling out PE in inpatients should wait until more convincing evidence is available.

There is not enough evidence at this time to support the use of the VIDAS D-Dimer assay as part

Overview of limitations:

of a clinical algorithm to rule out PE in inpatients. Prospective trials need to be undertaken to address this issue.

When the inclusion criteria for the clinical algorithms are developed for each of the above categories, a number of factors will need to be taken into consideration. First, there is no literature evaluating the VIDAS D-Dimer assay in patients with suspected recurrence of a DVT or PE. Therefore, the algorithm can only be applied to patients with suspected first episodes until more data is obtained. Secondly, there are certain circumstances when D-Dimer levels are high in the absence of a DVT or PE. These include recent surgery, trauma, sepsis, malignancies, and increasing age.³⁸ This results in decreasing specificity of the assay, which in turn leads to a decrease in the overall cost-effectiveness of the algorithm. Once the algorithm is developed, especially for inpatient use as these co-morbidities are more commonly seen in hospitalized patients, there will need to be a cost analysis to ensure that the D-dimer assay is still useful as a screening test. It may become evident that proceeding directly to ultrasound, V/Q scan or CT may be more cost-effective in these patients. Thirdly, D-dimer levels are also dependent on the amount of intravascular clot burden; the larger the clot, the higher the D-dimer levels. This becomes clinically important when there is an extended period of time between the onset of the symptoms of DVT or PE and the measurement of serum D-Dimer levels (greater than 11 days²⁷) and when the initial clot burden is very small (distal DVT and subsegmental PE³³). In these cases, the algorithm will need to be adapted to take these factors into consideration, as the sensitivity of the assay may not be high enough to consistently identify the presence of a clot.

Conclusion:

The VIDAS D-Dimer assay has shown that in certain clinical situations it can be used as an effective screening tool to help rule out DVT and PE. The literature supports the incorporation of VIDAS D-Dimer assay into a clinical algorithm for outpatients and inpatients presenting with a suspected first episode of DVT. Even though there is a small amount of literature evaluating VIDAS D-Dimer and outpatient PE, it is of high quality and can be used to justify the implementation of an algorithm to rule out PE in outpatients. However, further evidence is necessary prior to using the assay as part of an algorithm to rule out PE in inpatients. It will also be important to take into consideration the limitations of the VIDAS D-Dimer assay when developing the algorithms to ensure that the cost-effectiveness and clinical reliability are not lost.

References:

1. Anderson FA Jr, Wheeler HB, Goldberg RJ, Hosmer DW, Patwardhan NA, Jovanovic B, Forcier A, Dalen JE. A population-based perspective of the hospital incidence and case-fatality rates of deep vein thrombosis and pulmonary embolism. *Arch Intern Med* 1991;151:933-38.
2. Perrier A, Bounameaux H. Cost-effective diagnosis of deep vein thrombosis and pulmonary embolism. *Thromb Haemost* 2001;86:475-87.
3. Freyburger G, Trillaud H, Labrousse S, Gauthier P, Javorschi S, Grenier N. Rapid ELISA D-dimer testing in the exclusion of venous thromboembolism in hospitalized patients. *Clin Appl Thromb/Hemost* 2000;6(2):77-81.
4. Perrier A, Desmarais S, Miron MJ, de Moerloose P, Lepage R, Slosman D, Didier D, Unger PF, Patenaude JV, Bounameaux H. *Lancet* 1999;353:190-95.
5. de Moerloose P. D-dimer assays for the exclusion of venous thromboembolism: which test for which diagnostic strategy? *Thromb Haemost* 2000;83:180-81.
6. Michiels JJ, Oortwijn, WJ, Naaborg R. Exclusion and diagnosis of deep vein thrombosis by a rapid ELISA D-dimer test, compression ultrasonography, and a simple clinical model. *Clin Appl Thromb/Hemost* 1999;5(3):171-80.
7. Funfsinn N, Caliezi C, Biasutti FD, Korte W, Z'Brun A, Baumgartner I, Ulrich M, Cottier C, Lammler B, Wuillemin WA. Rapid D-dimer testing and pre-test clinical probability in the exclusion of deep venous thrombosis in symptomatic outpatients. *Blood Coagulation and Fibrinolysis* 2001;12:165-70.
8. Michiels JJ. Rational Diagnosis of Deep Vein Thrombosis (RADIA DVT) in symptomatic outpatients with suspected DVT: simplification and improvement of decision rule analysis for the exclusion and diagnosis of DVT by the combined use of a simple clinical model, a rapid sensitive D-dimer test and compression ultrasonography (CUS). *Sem thromb Hemost* 1998;24(4):401-7.
9. Michiels JJ. Rational Diagnosis of Pulmonary Embolism (RADIA PE) in symptomatic outpatients with suspected PE: an improved strategy to exclude or diagnose venous thromboembolism by the sequential use of a clinical model, rapid ELISA D-dimer test, perfusion lung scan, ultrasonography, spiral CT and pulmonary angiogram. *Sem thromb Hemost* 1998;24(4):413-18.
10. Lorut C, Ghossains M, Horellou MH, Achkar A, Fretault J, Laaban JP. A noninvasive diagnostic strategy including spiral computed tomography in patients with suspected pulmonary embolism. *Am J Resp Crit Care Med* 2000;162:1413-18.
11. Wicki J, Perneger TV, Junod A, Bounameaux H, Perrier A. Assessing clinical probability of pulmonary embolism in the emergency ward. *Arch Intern Med* 2001;161:92-97.
12. Indik JH, Alpert JS. Detection of pulmonary embolism by D-dimer assay, spiral computed tomography, and magnetic resonance imaging. *Prog Cardvasc Dis* 2000;42(4):261-72.
13. Michiels JJ, Pattynama PMT. Exclusion and diagnosis of pulmonary embolism by a rapid ELISA D-dimer test and noninvasive imaging techniques within the context of a clinical model. *Clin Appl Throm/Hemost* 2000;6(1):46-52.
14. Perrier A, Bounameaux H, Morabia A, de Moerloose P, Slosman D, Didier D, Unger PF, Junod A. Diagnosis of pulmonary embolism by a decision analysis-based strategy including clinical probability, D-Dimer levels, and ultrasonography: a management study. *Arch Intern Med* 1996;156:531-36.
15. Houdijk, WPM. Clinical applications of D-dimer. *Dev Thromb Hemost* 2000;3:1-26.
16. Bounameaux H, de Moerloose P, Perrier A, Miron M-J. D-Dimer testing in suspected venous thromboembolism: an update. *Q J Med* 1997;90:437-42.
17. Tapson VF for the American Thoracic Society. The diagnostic approach to acute venous thromboembolism: clinical practice guidelines. *Am J Resp Crit Care Med* 1999;160:1043-66.
18. Perrier A, Bounameaux H. Diagnosis of pulmonary embolism in outpatients by sequential noninvasive tools. *Sem Thromb Hemost* 2001;27(1):25-32.
19. van Beek EJR, Schenk BE, Michel BC, van den Ende B, Brandjes DPM, van der Heide YT, Bossuyt, PMM, Buller HR. The role of plasma D-dimer concentration in the exclusion of pulmonary embolism. *Br J Haemot* 1996;92:725-32.

20. Freyburger G, Trillaud H, Labrousche S, Gauthier P, S Javorschi, Bernard P, Grenier N. D-dimer strategy in thrombosis exclusion: A gold standard study in 100 patients suspected of deep vein thrombosis or pulmonary embolism: 8 DD methods compared. *Thromb Haemost* 1998;79:32-7.
21. Legnani C, Pancini C, Palareti G, Guazzaloca G, Fortunato G, Grauso F, Golfieri, Gianpalma E, Coccheri S. Comparison of new rapid methods for D-dimer measurement to exclude deep vein thrombosis in symptomatic outpatients. *Blood Coagulation Fibrinolysis* 1997;8:296-302.
22. van der Graaf F, van den Borne H, van der Kolk M, de Wild PJ, Janssen GWT, van Uum SHH. Exclusion of Deep venous thrombosis with D-dimer testing: comparison of 13 D-dimer methods in 99 outpatients suspected of deep venous thrombosis using venography as reference standard. *Thromb Haemost* 2000;83:191-8.
23. Elias A, Aptel I, Huc B, Chale JJ, Nguyen F, Cambus JP, Bocalon H, Boneu B. D-dimer test and diagnosis of deep vein thrombosis: a comparative study of 7 assays. *Thromb Haemost* 1996;76(4):528-32.
24. Janssen MCH, Heebels AE, de Metz M, Verbruggen H, Wollersheim H, Janssen S, Schuurmans MJ, Novakova IRO. Reliability of five rapid D-dimer assays compared to ELISA in the exclusion of deep venous thrombosis. *Thromb Haemost* 1997;77(2):262-6.
25. Sackett DL, Haynes RB, Tugwell P (eds). (1985) *Clinical epidemiology: a basic science for clinical medicine*, pp 47-57. Little, Brown and Company, Boston.
26. Sackett DL, Straus SE, Richardson WS, Rosenberg W, Haynes RB. (2000) *Evidence-based medicine. How to practice and teach EBM*. Churchill Livingstone, Edinburgh.
27. D'Angelo A, D'Alessandro G, Tomassini L, Pittel JL, Dupuy G, Crippa L. Evaluation of a new rapid quantitative D-dimer assay in patients with clinically suspected deep vein thrombosis. *Thromb Haemost* 1996;75(3):412-16.
28. de Moerloose P, Desmarais S, Bounameaux H, Reber G, Perrier A, Dupuy G, Pittet JL. Contribution of a new, rapid, individual and quantitative automated D-dimer ELISA to exclude pulmonary embolism. *Thromb Haem* 1996;75(1):11-13.
29. Pittet JL, de Moerloose P, Reber G, Durand C, Villard C, Piga N, Rolland D, Comby S, Dupuy G. VIDAS D-Dimer: fast quantitative ELISA for measuring D-Dimer in plasma. *Clin Chem* 1996;42(3):410-15.
30. Shitrit D, Heyd J, Raveh D, Rudensky B. Diagnostic value of the D-dimer test in deep vein thrombosis: improved results by a new assay method and by using discriminate levels. *Thromb Res* 2001;102:125-31.
31. Legnani C, Pancini C, Palareti G, Guazzaloca G, Coccheri S. Contribution of a new, rapid, quantitative and automated method for D-dimer measurement to exclude deep vein thrombosis in symptomatic outpatients. *Blood Coagulation Fibrinolysis* 1999;10(2):69-74.
32. Borg JY, Levesque H, Cailleux N, Franc C, Hellot MF, Courtois H. Rapid quantitative D-dimer assay and clinical evaluation for the diagnosis of clinically suspected deep vein thrombosis. *Thromb Haemost* 1997;77(3):602-3.
33. Sijens PE, van Ingen HE, van Beek EJR, Berghout A, Oudkerk M. Rapid ELISA assay for plasma D-dimer in the diagnosis of segmental and subsegmental pulmonary embolism; a comparison with pulmonary angiogram. *Thromb Haemost* 2000;84:156-9.
34. Diffin DC, Leyendecker JR, Johnson SP, Zucker RJ, Grebe PJ. Effect of anatomic distribution of pulmonary emboli on interobserver agreement in the interpretation of pulmonary angiography. *AJR* 1998;171:1085-9.
35. Wallis JW, Kruij M, de Jongh-leuvenink J, Buller HR. A comparative study of two rapid D-dimer tests for the exclusion of pulmonary embolism in symptomatic patients. *Thromb Haemost* 2000;84:925.
36. PIOPED Investigators. Value of the ventilation-perfusion scan in acute pulmonary embolism: Result of the prospective investigation of pulmonary embolism diagnosis. *JAMA* 1990;263:2753-59.
37. Speiser W, Mallek R, Koppensteiner R, Stumpflen A, Kapiotis S, Minar E, Ehringer H, Lechner K. D-dimer and TAT measurement in patients with deep venous thrombosis: utility in diagnosis and judgement of anti-coagulant treatment effectiveness. *Thromb Haemost* 1990;64:196-201.

38. Raimondi P, Bongrd O, de Moerlose P, Reber G, Waldvogel F, Bounameaux H. D-dimer plasma concentration on various clinical conditions: implication for the use of this test in the diagnostic approach of venous thromboembolism. *Thromb Res* 1993;69:125-30.
39. Ball CM, Phillips RS. (2001) Evidence-based on call; acute medicine. pp 441-4. Churchill Livingstone, Edinburgh.